



**HAL**  
open science

## Virulence of Classical Swine Fever Virus Isolates from Europe and other areas during 1996 until 2007

G. Floegel-Niesmann, S. Blome, H. Gerss-Dülmer, C. Bunzenthal, V. Moennig

► **To cite this version:**

G. Floegel-Niesmann, S. Blome, H. Gerss-Dülmer, C. Bunzenthal, V. Moennig. Virulence of Classical Swine Fever Virus Isolates from Europe and other areas during 1996 until 2007. *Veterinary Microbiology*, 2009, 139 (1-2), pp.165. 10.1016/j.vetmic.2009.05.008 . hal-00520662

**HAL Id: hal-00520662**

**<https://hal.science/hal-00520662>**

Submitted on 24 Sep 2010

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

## Accepted Manuscript

Title: Virulence of Classical Swine Fever Virus Isolates from Europe and other areas during 1996 until 2007

Authors: G. Floegel-Niesmann, S. Blome, H. Gerß-Dülmer, C. Bunzenthal, V. Moennig



PII: S0378-1135(09)00259-4  
DOI: doi:10.1016/j.vetmic.2009.05.008  
Reference: VETMIC 4443

To appear in: *VETMIC*

Received date: 6-11-2008  
Revised date: 16-5-2009  
Accepted date: 28-5-2009

Please cite this article as: Floegel-Niesmann, G., Blome, S., Gerß-Dülmer, H., Bunzenthal, C., Moennig, V., Virulence of Classical Swine Fever Virus Isolates from Europe and other areas during 1996 until 2007, *Veterinary Microbiology* (2008), doi:10.1016/j.vetmic.2009.05.008

This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

1 **Virulence of Classical Swine Fever Virus Isolates from Europe and other**  
2 **areas during 1996 until 2007**

3

4 Floegel-Niesmann, G\*., Blome, S., Gerß-Dülmer, H., Bunzenthal, C., Moennig, V.

5

6 EU Reference Laboratory for CSF

7 Institute for Virology

8 University of Veterinary Medicine Hannover

9 Buenteweg 17

10 30559 Hannover

11 Tel. +49-511-953-8850

12 Fax +49-511-953-8899

13 e-mail: gundula\_niesmann@yahoo.de

14 \*corresponding author

15

16

17

18

**18 Abstract**

19 Classical Swine Fever (CSF) has caused several outbreaks in EU Member States with grave  
20 economic consequences. Several times the diagnosis of CSF was made too late partially due  
21 to non-specific clinical signs which did not raise suspicion for CSF. Virulence of CSF virus  
22 isolates (CSFV) still remains a subject of discussion and speculation as sufficient knowledge  
23 is still not available. Six uncharacterised CSFV isolates from 1996 until 2007 were assessed  
24 in animal experiments for their clinical virulence in order to broaden the knowledge about  
25 circulating CSFV and thereby assist disease eradication. A clinical (CS) and pathological  
26 score were applied and further extended by additional parameters to a modified CS (mCS)  
27 including case fatality, antibody production and leukocyte count.  
28 The unknown CSFV isolates could be classified as moderately or highly virulent. The  
29 inclusion of additional parameters, especially case fatality, into the mCS gave a more reliable  
30 classification of virulence, proving that there are clinical signs and laboratory parameters of  
31 blood which can be recognised. Therefore a subclinical course of infection is unlikely,  
32 especially in weaner pigs.

33

**34 Keywords**

35 Classical Swine Fever – virulence – clinical score – pathological lesions – differential  
36 diagnosis – case fatality.

37

**38 Introduction**

39 Classical Swine Fever (CSF), which is among the diseases notifiable to the World  
40 Organisation for Animal Health (OIE) (Anonymous, 2004), occasionally causes sporadic  
41 epidemics in EU Member States and is endemic in a number of Third Countries worldwide.  
42 Since CSF has been generally eradicated from the EU, the control measures for CSF are based  
43 on a non-vaccination policy (Anonymous, 2001). However, during the last decades several

44 reintroductions of CSF virus (CSFV) have caused epidemics of severe economic  
45 consequences (Elbers et al., 1999; Fritzemeier et al., 2000; Sandvik et al., 2000). Whether  
46 there has been a change in virulence of the virus over time is a constant subject of discussion.  
47 CSFV isolates isolated from EU Member States from 1997 and 2001 have been classified as  
48 moderately virulent (Floegel-Niesmann et al., 2003). With these strains, clinical signs may be  
49 rather non-specific and age-dependent. This made diagnosis difficult and a new CSF  
50 outbreak was often discovered too late. Textbook cases with haemorrhagic lesions like  
51 bleedings of skin, petechiae on kidney and tonsils as well as spleen infarctions (Mengling and  
52 Packer, 1969) were not frequently observed. Only lymphadenosis, and high body temperature  
53 were common features of CSF infected pigs, but for clinical and pathological diagnosis this is  
54 rather non-specific (Floegel-Niesmann et al., 2003).

55 Characterising virulence has been attempted in various ways: case fatality, clinical and  
56 pathological signs (Carbrey et al., 1980, Wood et al. 1988), observations in CSF infected  
57 animal farms (Elbers et al., 2002), characterization of CSFV in cell cultures (Kubin 1967;  
58 Mittelholzer et al., 2000) or differences in the genome (Moormann et al., 1996; Mayer et al.,  
59 2003). Most of these characterisations are restricted to individual strains and they do not  
60 apply for other CSFV strains. Whether characterisation of virulence performed about three  
61 decades ago, still applies to CSFV isolated since the vaccination policy in the EU stopped at  
62 the beginning of the nineties is therefore questionable.

63 Mittelholzer et al. (2000) started to define objective criteria for the evaluation of clinical signs  
64 using a clinical score. Floegel-Niesmann et al. (2003) extended this score by pathological  
65 signs to allow for a better discrimination and thus comparison.

66 The purpose of this study was to characterise six so far unknown CSFV, isolated in Russia,  
67 Guatemala, South Africa, the Balkan area and the most recent EU Member State Bulgaria,  
68 using an extended clinical score and an established pathological score in order to increase the

69 knowledge on virulence of CSFV which are still circulating in the pig population in different  
70 parts of the world.

71

## 72 **Material and methods**

73 Five CSFV isolated in Third Countries and one CSFV from Bulgaria were compared with two  
74 formerly characterised CSFV from EU Member States. CSF0695 was isolated in Russia from  
75 a domestic pig in 1996 and thus represents a CSFV circulating in Russia different from those  
76 circulating in the EU (Vlassova et al., 2003). CSF0650 was isolated from a domestic pig from  
77 Guatemala in 1999 and represents CSFV from Central America and The Caribbean (Pereda et  
78 al., 2005). CSF 0695 belongs to genetic subtype 1.1 whereas CSF0650 belongs to genetic  
79 subtype 1.3 (Greiser-Wilke et al., 2006). CSF0849 was isolated from domestic pigs in South  
80 Africa in 2005 (Sandvik et al., 2005). CSF0854 was isolated from domestic pigs in the  
81 Republic of Kosovo in 2006 and CSFV0870 was isolated from domestic pigs in 2007 in  
82 Croatia. CSF0864 was isolated in Bulgaria from domestic pigs in 2007. The genetic typing  
83 revealed that CSF0854, CSF0864, and CSF0870 belonged to genotype 2.3, and CSF0849  
84 belonged to genotype 2.1. Both genotypes were also isolated in EU Member States during the  
85 last decade (Greiser-Wilke et al., 2006).

86 Two CSFV from EU Member States (CSF0277 and CSF0634) have been characterised  
87 previously as moderately virulent (Floegel-Niesmann et al., 2003) and were used for  
88 comparison. CSF0277 (genetic subtype 2.1) caused the CSF epidemic in domestic pigs in  
89 1997 affecting several EU Member States. CSF0634 (genetic subtype 2.3) was isolated from a  
90 CSF outbreak in domestic pigs in 2001 in Germany and was also present in the local wild  
91 boar population for several years (Fritzemeier et al., 2000) .

92 The CSFV were cultivated on PK 15(A) cells and their virus titre determined prior to  
93 inoculation of the pigs (Anonymous, 2002). The CSF antibody titres against the homologue

94 CSFV were obtained by neutralisation test (Anonymous, 2002). Leukocyte counts on EDTA  
95 blood samples were performed according to standard haematological procedure.

96

### 97 **Animal experiments**

98 Among the duties of the EU Reference Laboratory for CSF are the characterisation of CSFV  
99 isolates from new CSF outbreaks and the production of reference material for laboratory  
100 diagnosis (Anonymous 2001). In this framework, experiments were conducted according to  
101 the German Animal Welfare Act. Serum and organ materials of the pigs were used later for  
102 inter-laboratory comparison tests and distribution of reference material, one of the main tasks  
103 of the EU Reference Laboratory. In order to obtain maximum information out of an animal  
104 experiment, several separate experiments, performed at different times, are evaluated together  
105 here. The set up of the experiments conducted by the EU Reference Laboratory is similar  
106 though not identical. Therefore some parameters do vary (e.g. number of pigs, breed and  
107 leukocyte count).

108 All pigs were kept under high containment conditions. Four groups of five eight week old  
109 German Landrace pigs were inoculated oronasally with  $10^4$  tissue culture infectious doses  
110 50% (TCID<sub>50</sub>) of the respective CSFV isolates CSF0695, CSF0650, CSF0634, and CSF0277.  
111 Four groups of four eight week old cross breed weaners (German Landrace x Pietrain) were  
112 inoculated oronasally with  $10^4$  TCID<sub>50</sub> of the respective CSFV isolates CSF0849, CSF0854,  
113 CSF0864, and CSF0870. Clinical examination and body temperature measurement were  
114 performed daily. Blood samples for haematological, serological and virological examinations  
115 were taken twice a week. Virus isolation on leukocytes and virus neutralisation tests to detect  
116 CSF antibodies were performed according to the EU Diagnostic Manual (Anonymus, 2002)  
117 and the Technical Annex accompanying it. The clinical signs were evaluated according to the  
118 clinical score developed by Mittelholzer et al. (2000) with slight modifications. Moribund  
119 animals were euthanized and a post mortem examination performed. The pathologically

120 important organs for the diagnosis of CSF were evaluated according to a pathological score  
121 developed by Floegel-Niesmann et al. (2003). The clinical and pathological scores have a  
122 scale from 0 - 3 points according to the severity of the lesion: score 0 = normal, score 3 =  
123 severe CSF symptom. For the clinical score, these parameters were assessed daily, whereas  
124 the pathological score could only be assessed on the day of euthanasia. The mean clinical  
125 score was calculated from the highest score of each animal in each group. Selecting a defined  
126 day for this calculation would be misleading, because animals which recover score lower  
127 points with progressing time whereas others are already dead. The maximum score was 27  
128 points for the clinical signs and 30 points for the pathological signs. Parameters evaluated for  
129 the clinical signs were appetite, liveliness, body tension, shape, breathing, gait, eyes, skin, and  
130 defaecation. In addition, three further parameters were included to evaluate the virulence of  
131 the four CSFV: Case fatality at three weeks post infection, leukocyte counts between 0 and 14  
132 days post infection (dpi) (Stegemann et al., 2000) and the homologue CSF antibody titre at 14  
133 dpi. They were scored as follows: Case fatality 0% = 0 points, 1-40% = 1 point, 41-80% = 2  
134 points and >80% = 3 points; leukocyte count: >10 G/l = 0 points, 8.6 – 9.9 G/l = 1 point, 6.5 –  
135 8.5 G/l = 2 points and <6.5 G/l = 3 points; homologue CSF antibody titre: >5 ND50 = 0  
136 points and <5 ND50 = 3 points. Points for these additional parameters were calculated into the  
137 clinical score (CS), presenting now a modified CS (mCS). Classification is now made as  
138 follows: > 18 points = highly virulent, > 6 points = moderately virulent, < 6 points low  
139 virulent (see Table 1) (Bunzenthall, 2003).

140

## 141 **Results**

142 Regarding the incubation period, all six so far unknown CSFV had a shorter incubation period  
143 (3-5 days) compared to CSF0634 and CSF0277 (6-8) days. The mean CS ranged between 10  
144 and 17.6 points. The lowest CS was 10 for CSF0650 and the highest score 17.6 for CSF0634  
145 (see Table 1). Regarding case fatality, in each group at least one pig died (see Table 1). Pigs

146 infected with CSF0634 and CSF0870, respectively, showed the highest case fatality of 100 %,  
147 whereas pigs infected with CSF0650 showed the lowest case fatality rate of 20 %. None of  
148 the pigs infected with CSF0634 and CSF0849 had a detectable homologue CSF antibody titre  
149 at 14 dpi. From pigs infected with CSF0277, CSF0854, CSF0864, and CSF0870, respectively,  
150 only one pig in each group started to produce CSF antibodies at 14 dpi against the homologue  
151 virus (neutralizing dose 50 % (ND<sub>50</sub>) 15-30). CSF0650 and CSF0695 both induced CSF  
152 antibody titres between 10 and 80 ND<sub>50</sub>. The leukocyte count decreased in all investigated  
153 CSFV infected pigs below 10 G/l (see Table 1). The leukocyte count for pigs infected with  
154 CSF0849, CSF0854, CSF0864, and CSF870 were not performed for technical reasons.  
155 CSF0634 and CSF0854 scored the highest pathological score (15.4 and 15.25 respectively),  
156 followed by CSF0849 (14.25). CSF0650 and CSF0277 scored rather similar (5.6 and 5  
157 respectively) the lowest pathological scores. Pathological findings were most obvious in  
158 lymphnodes upon infection with all eight CSFV isolates (see Table 2). Scores for skin lesions,  
159 representing the classical haemorrhagic picture, ranged from 0.2 in CSF0650 up to 2.5 in  
160 CSF0849.

161 The mCS (see Table 1) clearly defines CSF0634, CSF0849 and CSF0854 as highly virulent,  
162 whereas CSF0864 and CSF870 are borderline between highly and moderately virulent.  
163 CSF0650, CSF0695 and CSF0277 were classified as moderately virulent. No CSFV was  
164 classified as low virulent.

165

## 166 **Discussion**

167 The purpose of this study was to assess six so far uncharacterized CSFV isolates from 1996  
168 until 2007 for their virulence by using the recently developed pathological score and  
169 extending the existing clinical score by additional parameters. This included more parameters  
170 of clinical and laboratory diagnosis to characterise virulence more objectively and gain  
171 valuable information for clinical diagnosis of CSF and disease eradication.

172 Whether animal experiments performed by the EU Reference Laboratory giving a reference to  
173 all EU Member States should be done with inbred pigs, available from SPF-holdings or  
174 cross-breed pigs which are actually found in the farms and will meet the field virus can be  
175 discussed in several ways. When deciding for cross-breed pigs, cross-breed fattening pigs  
176 from Southern Europe will not be identical to cross-breed fattening pigs from Northern  
177 Europe either. However, looking at CSF epidemics affecting several countries, the CSFV  
178 isolate never had difficulties infecting local pig populations (Elbers et al., 1999). Also wild  
179 boars get CSF infections in different parts of Europe and the virus enters the local domestic  
180 pig population (Fritzemeier et al., 2000). Depner et al., 1997 observed breed related  
181 differences during an experimental CSF infection with a different CSFV (CSF0123). It was a  
182 single experiment and reproduction turned out to be rather difficult (data unpublished) nor has  
183 any other scientific publication been made on the subject. Therefore the use of the pig breed  
184 should not be overestimated here.

185 The standard deviation between the pigs of each infected group for the clinical and  
186 pathological is rather high. This is not surprising, when some animals in each group do  
187 recover and others die showing clinical and pathological signs. In recovering pigs, clinical  
188 signs naturally are less pronounced and pathological signs are almost absent. The standard  
189 deviations for CSF0634 are the lowest because all animals had obvious clinical signs.  
190 Regarding the introduction of the additional parameter case fatality, it did not always correlate  
191 with a high CS. Animals with the highest CS also had a high case fatality rate (CSF0634),  
192 however, CSF0870 with a case fatality rate of 100% had a clinical score of only 12.5.  
193 Furthermore CSF0277 had a lower CS than CSF0695 but a higher case fatality .  
194 Consequently, high case fatality is not necessarily associated with a high CS. Therefore case  
195 fatality is an important parameter which needs to be included, when evaluating virulence.  
196 CSF0277 had also been characterised in the marker vaccine trial performed by EU member  
197 states (Uttenthal et al., 2001). There, the case fatality differed in pigs from different countries.

198 However, a high case fatality is quite easy to observe in a pig farm and has to be regarded as  
199 an individual sign for CSF independent from clinical signs. Consequently, it is essential to  
200 include case fatality when evaluating virulence, which is achieved now in the new mCS.

201 Regarding antibody production against CSF, CSF695 and CSF650 both induced significant  
202 CSF antibody titres at 14 dpi (see Table 1). This lowers the mCS and may lead to recovery of  
203 infected animals with less obvious clinical signs. On the other hand, these CSF antibody titres  
204 would be easy to detect in laboratory diagnosis using commercial ELISAs or neutralisation  
205 tests Floegel-Niesmann et al. (2004) and helps identifying CSF infected animals which are  
206 recovering.

207 Applying the new mCS, pigs infected with CSF0634 had the highest score in all parameters  
208 and can now be characterised as highly virulent in the age class of weaner pigs. Previously  
209 CSF0634 had been characterised as moderately virulent (Floegel-Niesmann et al., 2003).

210 The CSFV classified as highly virulent in the mCS had the highest total pathological score  
211 (CSF0634, CSF0849, and CSF0854). This indicates that there is a correlation between the  
212 pathological signs and the classification of virulence in the mCS: high virulence is associated  
213 with increased pathological lesions. However, many pathological signs were non-specific  
214 with lymphadenosis dominating the picture.

215 The mCS clearly shows that there are several clinical signs and laboratory diagnostic of blood  
216 parameters which can be recognised during CSF infection and it should be possible to detect  
217 some of them during clinical investigation in an infected farm of a new CSF outbreak. With  
218 the introduction of the new mCS, speculations of mysteriously low virulent CSFV circulating  
219 in pig populations could not be sustained. A subclinical course of CSF infection in weaner  
220 pigs is therefore quite unlikely. It would be of scientific and epidemiological interest to  
221 conduct the same experiments with pigs from different age groups eg. 50 kg fattening pigs,  
222 100 kg fattening pigs and breeding sows. But animal welfare is given priority here and  
223 therefore experiments with the most susceptible age group must be sufficient. However, the

224 individual signs in other age groups may be less typical. Especially in fattening pigs, where  
225 respiratory infections are rather common, it is likely that these dominate the clinical signs as  
226 misleading secondary infections.

227

## 228 **Acknowledgement**

229 We thank Dr. Beverly Schmidt of the National Veterinary Services Laboratory in Ames, Iowa  
230 USA for CSF0650; Dr. Alexej Zabarezhny and Anastasia Vlassova from NARVAC Moscow  
231 for CSF0695; Dr. Bafti Murati from the Kosovo Veterinary and Food Agency for CSF0854;  
232 Prof. Baichev and Dr. Emilia Ivanova from the National Veterinary Services Bulgaria for  
233 CSF0864, Dr. J.P. Kitching from Veterinary Laboratory Services South Africa for CSF0849  
234 and Dr. Lorena Jemersic from the Croatian Veterinary Institute for CSF0870.

235

## 236 **Reference List**

- 237 Anonymous, 2001. Council Directive 2001/89/EC of 23 October 2001 on Community  
238 measures for the control of classical swine fever. EU Commission, Brussels.
- 239 Anonymous, 2002. Commission Decision of 1 February 2002 approving a Diagnostic manual  
240 establishing diagnostic procedures, sampling methods and criteria for evaluation of the  
241 laboratory tests for the confirmation of classical swine fever. EU Commission, Brussels.
- 242 Anonymous, 2004. Chapter 2.1.13. In: Manual for Standards of Diagnostic Tests and  
243 Vaccines. 5, Office International des Epizootie, Paris.
- 244 Bunzenthall, C., 2003. Bestimmung der Virulenz von Virusisolaten der Klassischen  
245 Schweinepest. Dissertation, School of Veterinary Medicine Hannover.
- 246 Carbrey, E. A., Stewart, W. C., Kresse, J. I., Snyder, M. L., 1980. Persistent hog cholera  
247 infection detected during virulence typing of 135 field isolates. Am.J.Vet.Res. 41, 946-  
248 949.
- 249 Depner, K. R., Hinrichs, U., Birckhardt, K., Greiser-Wilke, I., Pohlenz, J., Moennig, V.,  
250 Liess, B., 1997. Influence of breed related factors on the course of classical swine fever  
251 virus infection. Veterinary Record. 140, 506-507.

- 252 Elbers, A. R., Stegeman, A., Moser, H., Ekker, H. M., Smak, J. A., Pluimers, F. H., 1999. The  
253 classical swine fever epidemic 1997-1998 in The Netherlands: descriptive epidemiology.  
254 *Prev.Vet.Med.* 42, 157-184.
- 255 Elbers, A. R., Bouma, A., Stegeman, J. A., 2002. Quantitative assessment of clinical signs for  
256 the detection of classical swine fever outbreaks during an epidemic. *Vet.Microbiol.* 85,  
257 323-332.
- 258 Floegel-Niesmann, G., Bunzenthall, C., Fischer, S., Moennig, V., 2003. Virulence of recent  
259 and former classical swine fever virus isolates evaluated by their clinical and pathological  
260 signs. *J.Vet.Med.B Infect.Dis.Vet.Public Health.* 50, 214-220.
- 261 Floegel-Niesmann, G., Moennig, V., 2004. Quality management in reference tests for the  
262 diagnosis of classical swine fever. *Rev.Sci.Tech.* 23, 895-903.
- 263 Fritzemeier, J., Teuffert, J., Greiser-Wilke, I., Staubach, C., Schluter, H., Moennig, V., 2000.  
264 Epidemiology of classical swine fever in Germany in the 1990s. *Vet.Microbiol.* 77, 29-41.
- 265 Greiser-Wilke, I., Dreier, S., Haas, L., Zimmermann, B., 2006. [Genetic typing of classical  
266 swine fever viruses--a review]. *Dtsch.Tierarztl.Wochenschr.* 113, 134-138.
- 267 Kubin, G., 1967. [Characters of swine fever virus in vitro] In vitro Merkmale des  
268 Schweinepestvirus. *Zentralbl.Veterinarmed.B.* 14, 543-552.
- 269 Mayer, D., Thayer, T. M., Hofmann, M. A., Tratschin, J. D., 2003. Establishment and  
270 characterisation of two cDNA-derived strains of classical swine fever virus, one highly  
271 virulent and one avirulent. *Virus Res.* 98, 105-116.
- 272 Mengeling, W. L., Packer, R. A., 1969. Pathogenesis of chronic hog cholera: host response.  
273 *Am.J.Vet.Res.* 30, 409-417.
- 274 Mittelholzer, C., Moser, C., Tratschin, J., Hofmann, M. A., 2000. Analysis of classical swine  
275 fever virus replication kinetics allows differentiation of highly virulent from avirulent  
276 strains *Vet.Microbiol.* 74, 293-308.
- 277 Moormann, R. J., Van Gennip, H. G., Miedema, G. K., Hulst, M. M., Van Rijn, P. A., 1996.  
278 Infectious RNA transcribed from an engineered full-length cDNA template of the genome  
279 of a pestivirus. *J.Virol.* 70, 763-770.
- 280 Pereda, A. J., Greiser-Wilke, I., Schmitt, B., Rincon, M. A., Mogollon, J. D., Sabogal, Z. Y.,  
281 Lora, A. M., Sanguinetti, H., Piccone, M. E., 2005. Phylogenetic analysis of classical  
282 swine fever virus (CSFV) field isolates from outbreaks in South and Central America.  
283 *Virus Res.* 110, 111-118.
- 284 Sandvik, T., Drew, T., Paton, D., 2000. CSF virus in East Anglia: where from? *Vet.Rec.* 147,  
285 251.

- 286 Sandvik,T., Crooke, H., Drew, T.W., Blome, S., Greiser-Wilke, I., Moennig, V., Gous,T.A.,  
287 Ger, S., Kitching, J.A., Buehrmann, G., Brueckner, G.K., 2005. Classical Swine Fever in  
288 South Africa after 87 years' of absence. *Vet. Rec.* 157(9):267.
- 289 Stegeman, J. A., Bouma, A., Elbers, A. R., Verheijden, J. H., 2000. [The leukocyte count is a  
290 valuable parameter for detecting classical swine fever.]. *Tijdschr.Diergeneeskd.* 125, 511-  
291 518.
- 292 Uttenthal, A., Le Potier, M., Romero, L., De Mia, G. M., Floegel-Niesmann, G., 2001.  
293 Classical swine fever (CSF) marker vaccine. Trial I. Challenge studies in weaner pigs.  
294 *Vet.Microbiol.* 83, 85-106.
- 295 Vlasova, A., Grebennikova, T., Zaberezhny, A., Greiser-Wilke, I., Floegel-Niesmann, G.,  
296 Kurinnov, V., Aliper, T., Nepoklonov, E., 2003. Molecular epidemiology of classical  
297 swine fever in the Russian Federation. *J.Vet.Med.B Infect.Dis.Vet.Public Health.* 50, 363-  
298 367.
- 299 Wood, L., Brockman, S., Harkness, J. W., Edwards, S., 1988. Classical swine fever: virulence  
300 and tissue distribution of a 1986 English isolate in pigs. *Vet.Rec.* 122, 391-394.  
301  
302

**Table 1:** Incubation period, Clinical Score (CS), additional parameters leukocyte count, case fatality, homologue CSF antibody titre at 14dpi , the new modified Clinical Score (mCS) calculated out of them and the classification of virulence for all investigated CSFV.

CSFV	Mean incubation period (days)	CS (Points) +/- (Standarddeviation)	Leukocyte count (G/l)	Case fatality at 22dpi (%)	highest antibody titre (ND <sub>50</sub> ) 14dpi	mCS (Points)	Virulence
CSF0650	4-5	10 (3,5)	8,5	20	80	13,6	moderate
CSF0695	3-4	13,4 (3,5)	7,5	40	40	17,7	moderate
CSF0634	6-8	17,6 (2,7)	5,8	100	<5	26,6	high
CSF0277	6	10,2 (4,6)	9,4	60	15	15,4	moderate
CSF0849	4-5	14 (3,7)	n.d.	50	<5	19	high
CSF0854	5	15,25 (4,6)	n.d.	25	20	19,5	high
CSF0864	4	13,75 (3,9)	n.d.	75	15	18	moderate/high
CSF0870	5	12,5 (6,1)	n.d.	100	30	17,75	moderate/high

n.d.: not done

**Table 2:** Mean pathological score (PS) of organs of five or four pigs for each CSFV at day of euthanasia of pigs. Standarddeviation between pigs infected with the respective CSFV is given.

Organ/CSFV	0650	0695	0634	0277	0849	0854	0864	0870
Skin	0,2	1,2	2,2	0,4	2,5	2,25	0,5	0,75
Serosae	0	1	2	0,4	0	0,75	0,5	0
Ln. inguinales	1,4	1,6	2,4	1,2	2,5	2	1,5	2
Ln. mandibulares	2	1,4	2,4	1	2,25	2	1,5	2
Ln. ileocaecalis	1,4	1,4	1,8	0,8	1,25	1,5	1	1,75
Tonsil	0	1	0	0,2	1,25	1,75	1,25	2
Spleen	0,2	0	0,4	0	0,25	1	0,75	0,25
Kidney	0,2	1	0,8	0	1,75	1,5	0,5	0,5
Ileum	0	0,8	2,2	0,4	0,75	1,25	0,5	0,5
Respiratory system	0,2	0	1,2	0,6	1,75	1,25	0,75	0,75
Total PS	5,6	9,4	15,4	5	14,25	15,25	8,25	10,5
Standarddeviation between pigs	3,7	10,26	1,14	2,9	3,69	6,87	3,86	2,65