Differences in clinical scoring may influence the outcome of experimental African swine fever studies; harmonization needed?

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Background and objective

Survival rates and clinical scores are critical metrics in experimental African swine fever (ASF) studies. Alongside these, the timely application of humane endpoints (HEPs) is essential to safeguard **animal welfare**. However, research institutes often employ varying **clinical scoring systems and HEP criteria**, leading to differences in study outcomes. These differences pose challenges when comparing results, particularly in studies such as vaccine efficacy trials.

Materials and methods

As part of an ongoing EU project, a multicenter study was conducted at the Friedrich Loeffler Institute (FLI), Germany, and Wageningen Bioveterinary Research (WBVR), the Netherlands. The study evaluated the efficacy of three live-attenuated vaccines (A, B and C) administered orally. While both institutes used clinical scoring systems with 9 (FLI) and 10 (WBVR) parameters (Table 1) that largely overlapped, a key difference lays in the HEP application regarding body temperature. Wageningen Bioveterinary Research did not include a temperature-based HEP, while FLI applied a HEP if the body temperature exceeded 40.5°C for three consecutive days (Table 2).

Table 1. ASF-clinical score and humane endpoint (HEP)-WBVR

	1. Posture	
	0	Normal
	1	Stiffness and arched back when standing up, then 'normal'
	2	Stiffness and arched back remain when walking around
HEP	3	Muscle cramps / cramping of the muscles
	2. Body shape	
	0	Normal
	1	Sunken flanks
	2	Slimming
	3	Wasting (ribs, spinal vertebrae visible, long hair)
	3. Appetite	
	0	Normal
	1	Slow eater, does eat feed
	2	Approaches feed, tastes feed, but eats little/nothing
	3	Does not eat, no interest in feed
	4. Activity	
	0	Normal
	1	Slow, still gets up on its own, without help
	2	Slow, gets up with some help, lies down quickly
HEP	3	Stays down, doesn't get up even after some pressure
	5. Body temperature	
	0	38.0-40.0°C (for sows 37.0-40.0°C)
	1	40.1-41.0°C
	2	>41.0°C during the first 48h or <38.0°C
	3	>41.0°C longer than 48 h
	6. Vomiting	
	0	Normal
	1	Occasional vomiting (1x during observation period)
	2	Repeated vomiting (several times during observation period)
HEP	3	Bloody vomiting
	7. Breathing	
	0	Normal
	1	Increased breathing rate, occasional coughing or sneezing
	2	Pumping breathing rate, abdominal beat, frequent coughing or sneezing
HEP	3	Difficulty breathing, panting, breathing with open mouth
	8. Neurological signs	
	0	Normal
	1	Somewhat doubtful gait, deviant leg position which is slowly corrected
	2	Ataxia/weakness of the hindquarters, can still walk
HEP	3	Paralysis, can't stand anymore, not even with some help
	9. Skin (particularly on ears, muzzle, tail, legs, abdomen)	
	0	Normal
	1	Red skin
	2	White, blue/purple or other discoloration of the skin, possibly bleedings in skin
	3	Large blue/purple spots, large skin hemorrhages, skin necrosis/ulcer
	10. Exudates (eye, nose, anus)	
	0	Normal/none
	1	Thin, clear discharges from nose and/or eyes (without admixtures) or diarrea
	2	Thick discharge from nose and/or eyes (puss-like or colored, no blood)
HEP	3	Bloody discharge (fresh blood from nose, eyes or anus) or black manure (old blood)

A HEP is reached when a pig shows a clinical sign which is indicated with HEP or when a cumulative score of ≥ 10 is reached.

Results

Table 2. Comparison of ASF clinical score FLI and WBVR

FLI		WBVR	
score	0 till 4	score	0 till 3
1. bearing (posture)	0 till 4	1. posture	0 till 3 (3=HEP)
2. nutritional status	0 till 3	2. body shape	0 till 3
3. appetite	0 till 4	3. appetite	0 till 3
4. liveliness (activiy)	0 till 4	4. activity	0 till 3 (3=HEP)
body temperature	not included	5. body temperature	0 till 3
5. Defacation/vomiting	0 till 4	6. vomiting	0 till 3 (3=HEP)
6. breathing	0 till 4	7. breathing	0 till 3 (3=HEP)
7. gait (neurologic)	0 till 4	8. neurologic signs	0 till 3 (3=HEP)
8. skin	0 till 4	9. skin	0 till 3
9. eyes/ conjunctiva	0 till 4	10. exudates (eye, nose, anus)	0 till 3 (3=HEP)
Human end point (HEP)	≥10	≥10	
	score 4 for one parameter	score 3 for individual parameters (HEP)	
additional criteria HEP	body temp> 40.5°C for 72h	not applicable	
Observations/day	two observations per day	≥4: second observation afternoon	

- **WBVR**, 5 out of 30 vaccinated (n=10 per group) pigs survived the challenge and completed the study (Fig. 1). If FLI's temperature-based HEP had been applied, 3 animals would have survived
- **WBVR,** all pigs were euthanized because of **clinical score ≥ 10**, except for one pig, which was found dead in the stable
- **FLI**, 3 out of 45 pigs (n=15 per group) survived under their scoring system, but survival rates may have improved if WBVR's criteria had been used
- **FLI**, all pigs were euthanized because of **body temperature > 40.5° C for more than 72h** (Fig. 1), no pigs were found dead in the stable. **Clinical score** of these pigs were **between 2-4**

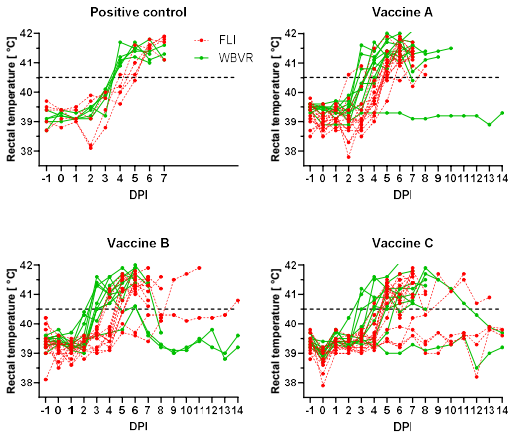

Figure 1. Comparison body temperature FLI and WBVR after ASF challenge. Non-vaccinated control animals (n=5) and *oral ASF vaccines (A, B and C) with n=10 (WBVR) and n=15 (FLI) animals per group. Animals still present at 14 DPI remained till the end of the study (21 DPI) and survived the challenge. Dotted line indicates 40,5° C, > 72 h= HEP*

Conclusion

These findings underscore the possible impact of varying ASF clinical scoring systems and HEP criteria on study outcomes. They highlight the **need to harmonize clinical scoring in ASF studies** to ensure consistent results and uphold **animal welfare**.

Considerations:

➢ different HEP for vaccinated and non-vaccinated control animals

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