

**Title:** Interventions to control PEDv (porcine epidemic diarrhea virus) in feed and feed ingredients – NPB #14-157

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**Industry Summary:** We studied the effect of low heat (60<sup>0</sup>C, 70<sup>0</sup>C, 80<sup>0</sup>C, and 90<sup>0</sup>C); high heat (120<sup>0</sup>C, 130<sup>0</sup>C, 140<sup>0</sup>C, and 145<sup>0</sup>C); irradiation; and the addition of drying agents/acidifiers on the killing of PEDV in swine feed. At low heat, we found that PEDV survival depended not only on temperature and relative humidity (RH) but also on the type of feed ingredient used. Heating feed to 60<sup>0</sup>C at any of the 3 RH levels (30%, 50%, and 70%) was not sufficient to inactivate 99.99% of PEDV. However, a combination of 90<sup>0</sup>C and 70% RH killed 99.99% PEDV within 10 to 30 minutes. At high heat, increasing amounts of virus were inactivated with increased temperature; 99.99% virus was inactivated within 10 minutes at 145<sup>0</sup>C. Virus inactivation by irradiation was dose-dependent; greater than 99.97% virus was inactivated at 50 kGy while only 90% was killed at 10 kGy. We evaluated six different additives on the inactivation of PEDV in feed and found that the addition of sugar and salt was as good in killing virus as Kemin; they all killed more than 2 log<sub>10</sub> of virus (≥99% inactivation) but it took them 21 days to do so. Ultrap, ADA, and AB were not able to achieve this level of virus kill even after 21 days. Another objective of this study was to evaluate the currently used methods of virus elution from feed and from stainless steel surfaces. Both of the currently used methods were found to be adequate in recovering PEDV. The last objective was to determine the MID<sub>50</sub> of a standardized PEDV inoculum. We found that as low as 7 infectious units (TCID<sub>50</sub>) of cell culture grown virus was able to induce disease in 11-day-old piglets.

**Keywords:** Porcine epidemic diarrhea virus, coronavirus, virus survival, swine feed, feed ingredients, irradiation, heat, MID<sub>50</sub>.

**Scientific Abstract:** This study was undertaken to determine the effect of certain interventional strategies on the inactivation of PEDV in feed and feed ingredients. The strategies tested were: low heat, high heat, irradiation, and the addition of drying agents/acidifiers. At low temperatures, the survival of PEDV depended on all three factors studied (type of feed ingredient, temperature, and RH). For example, a temperature of 60<sup>0</sup>C at any of the 3 RH levels (30%, 50%, and 70%) was not sufficient to inactivate 4 log<sub>10</sub> of PEDV. However, 4 log<sub>10</sub> of PEDV was killed at 90<sup>0</sup>C within 15 to 30 min in some of the feed ingredients. A combination of 90<sup>0</sup>C and 70% RH killed 99.99% of PEDV within 10 to 30 min except in spray-dried plasma. Virus inactivation by irradiation was dose-dependent; greater than 99.97% virus was inactivated at 50 kGy while only 90% was killed

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at 10 kGy. We studied the effect of 4 different temperatures (120<sup>0</sup>C, 130<sup>0</sup>C, 140<sup>0</sup>C, and 145<sup>0</sup>C) on PEDV-spiked feed at 5 minute intervals and found that increasing amounts of virus were inactivated with increased temperature; 99.99% virus was inactivated within 10 to 25 min depending on the temperature used; 99.99% of the virus was killed within 10 min at 145<sup>0</sup>C. The effect of six different additives on PEDV was evaluated. Surprisingly, we found that the addition of sugar and salt was as good in killing virus as Kemin; they all killed more than 2 log<sub>10</sub> of virus (≥99% inactivation) but it took them 21 days to do so. Ultrap, ADA, and AB were not able to achieve this level of virus kill even after 21 days. Another objective was to evaluate the currently used methods of virus elution from feed and from stainless steel surfaces. Both methods were found to be adequate in recovering PEDV. The last objective was to determine the MID<sub>50</sub> of a standardized PEDV inoculum. We found that as low as 7 TCID<sub>50</sub> (50% tissue culture infective dose) of cell culture grown virus was able to induce disease in 11-day-old piglets.

**Introduction:** It is now well known that PEDV is a highly important virus for the swine industry. Indications are that feed and/or feed ingredients of swine origin may play a role in the transmission of PEDV. No data are available to quantify the temperature, time, and moisture conditions necessary to inactivate PEDV. Therefore, it is necessary to find intervention strategies that can eliminate (or at least drastically reduce) PEDV, if present in feed and feed ingredients. This study was done to determine PEDV inactivation curves under various temperature, time, and moisture conditions for common swine feed ingredients. In addition, the relative effectiveness of irradiation, heat, and addition of certain chemicals on the inactivation of PEDV in feed/feed ingredients was examined. We concentrated our efforts on milled complete feed, spray dried porcine plasma, Grow-Finish premix, meat meal, meat and bone meal, blood meal, corn, soybean meal, and DDGS. The data obtained in this study should allow better decision making on choosing risk mitigation strategies including irradiation, heat treatment, chemical treatment, and/or prolonged storage.

**Objectives:** Evaluate strategies and processes to control the occurrence of PEDV in feed and feed ingredients.

### **Specific Aims:**

Aim 1. To determine time x temperature x moisture conditions necessary to inactivate PEDV in common feed ingredients used in swine diets.

Aim 2. To determine what processes (irradiation, heating, and addition of drying agents, etc.) can eliminate PEDV from feed and feed ingredients.

Aim 3: To develop a sensitive method for the detection of small amounts of PEDV in large amounts of feed and feed ingredients.

Aim 4. To determine the minimum infectious dose (MID<sub>50</sub>) of a standard inoculum of PEDV (cell culture-grown).

### **Materials & Methods:**

**General Procedures:** In this study, we used NVSL strain of PEDV. The virus was propagated and titrated in Vero cells. In all experiments, the surviving virus was eluted (recovered) in an eluent consisting of a 3% solution of beef extract in 0.05M glycine. Following elution, the eluate was lightly centrifuged to remove organic matter/debris. The supernatants were used to determine the amount of surviving virus, if any. For virus titration, serial 10-fold dilutions of the eluted virus were inoculated in Vero-81 cells contained in 96-well microtiter plates using 3 wells per dilution. The inoculated cells were examined daily for 5-7 days for the appearance of CPE (cytopathic effects). The highest dilution showing CPE was considered the end point. Virus

titers were calculated by the method of Karber (1931) and were expressed as TCID<sub>50</sub>. The amount of surviving virus was compared with the starting virus titer to calculate the amount of virus inactivated.

## **Procedures to achieve the specific aims**

*Aim 1. To determine time x temperature x moisture conditions necessary to inactivate PEDV in common feed ingredients used in swine diets.*

The impact of time x temperature x moisture on survival of PEDV was studied in nine feed ingredients as follows:

- Complete feed
- Spray dried porcine plasma
- Meat meal
- Meat and bone meal
- Blood meal
- Grow-finish premix
- Corn
- Soybean meal
- DDGS (Dried distillers grains)

To 5g aliquots of the above ingredients (in scintillation vials) was added 1mL each of PEDV. The vials were kept at four different temperatures 60°C, 70°C, 80°C and 90°C and at three different levels of relative humidity (30%, 50%, and 70%). Aliquots were removed at 0, 5, 10, 15, and 30 min followed by the addition of an eluent solution (10 mL of sterile 3% beef extract-0.05 M Glycine solution at pH 7.2) to each vial. After vortexing for a few seconds, the vials were centrifuged at 1,200 xg for 10 min. The supernatants were collected and titrated in Vero-81 cells (as given in General Procedures). The viral titers at various time points were calculated as TCID<sub>50</sub>/mL by the Karber method. The results are shown in Tables 1-27. Table 28 shows composite results from Tables 1-27.

*Aim 2. To determine what processes (irradiation, heating, and addition of drying agents, etc.) can eliminate PEDV from feed and feed ingredients.*

**Effect of irradiation on PEDV:** We spiked aliquots of complete feed with PEDV and sent them to Texas A&M where they were subjected to various doses of irradiation (0, 10, 20, 30, and 50 kGy). The samples were returned to Minnesota the same day where we eluted the virus from these samples and tested for TCID<sub>50</sub>. The results are shown in Table 29.

**Effect of high heat on PEDV:** Several 5 gm aliquots of complete feed were prepared in scintillation vials. The vials were kept in ovens at various temperatures: 120°C, 130°C, 140°C and 145°C. After the feed attained the proper temperature, the vials were removed from the oven followed by spiking them with 1mL of PEDV. The vials were re-incubated in the ovens followed by their removal at 0, 5, 10, 15, 20, 25, and 30 min. The surviving virus was eluted by adding 10 mL/aliquot of an eluent solution (3% beef extract-0.05M glycine, pH 7.2). After light centrifugation, the supernatants were decanted. Serial 10-fold dilutions of the supernatants (elutes) were prepared in MEM and then inoculated in Vero-81 cells for virus titration. The inoculated cells were incubated at 37°C and examined daily for 5-7 days for the appearance of CPE. Virus titers were calculated by the Karber method (1931) and were expressed as TCID<sub>50</sub>/mL. The results are shown in Table 30.

**Effect of additives:** Another intervention studied was the addition of drying agents and acidifiers in complete feed. For this purpose, we prepared several 5 gm aliquots of feed in scintillation vials. The following six additives were added in the amounts indicated followed by the addition of 1mL of PEDV. After thorough mixing, aliquots were removed at 0, 1, 3, 5, 7, 14, and 21 days. The surviving virus was eluted and the eluate was titrated as given above. The results are given in Table 31.

<b>Nutriadi (Ultrap):</b>	2-3 kg/ton	150 mg/5 gm of feed.
<b>Novus Intl (ADA):</b>	10 lb/ton	20 mg/5 gm of feed.
<b>AgriNutrition:</b>	1-4.5 lb/ton	10 mg/5 gm of feed.
<b>Kemin Agrifoods NA:</b>	2 kg/ton	10 mg/5 gm of feed.
<b>Sugar (sugar):</b>	-	20 mg/5 gm of feed.
<b>Salt (salt):</b>	-	20 mg/5 gm of feed.

*Aim 3: To develop a sensitive method for the detection of small amounts of PEDV in large amounts of feed and feed ingredients.*

**Method for virus detection in feed:** Currently, the feed is tested as follows: A sample of feed (50 g) is added to 200 mL of sterile PBS followed by incubation overnight at 4<sup>0</sup>C. In the morning, the mixture is centrifuged lightly and 300 µL of the supernatant is tested by RT-PCR. Simply stated, approximately 0.3 mL of the 250 mL of the feed-PBS mixture is being tested. Although this procedure seems inefficient, we found that it was superior to other methods that we proposed. We suggest the continued use of this method.

**Comparison of Swiffer, gauze, and 1-MDS filters for virus elution from stainless steel surface:** Currently, swiffers are used for swabbing surfaces for virus detection. We compared the efficacy of swiffers with that of 1-MDS filters and cotton gauze for virus elution from surfaces. For this purpose, we cut 2x2" pieces of Swiffer, 1-MDS filters and cotton gauze with sterile scissors. We applied 100 µL of PEDV on 4 corners and middle of a stainless steel sheet (a total of 5 areas per sheet). The virus was allowed to dry for 30 min. The pieces of Swiffer, 1-MDS filters or cotton gauze were wetted in elution buffer (3% beef extract with 0.05 M glycine, pH 7.2). The excess fluid was removed; the pieces were damp but not dripping wet. Using one piece of Swiffer, we wiped the surface of one whole sheet (all 5 areas). Each spot was wiped with the same piece twice each in clockwise manner, anticlockwise manner, up and down, and to-and-fro for each of the 5 spots on the sheet. 1-MDS filter and sterile gauze pieces were used for sheet numbers 2 and 3, respectively. All pieces were placed separately in 50mL tubes that contained 5 mL of eluent (3% beef extract with 0.05 M glycine, pH 7.2) per tube. After vortexing for 15 sec, the eluates were lightly centrifuged followed by virus titration as mentioned above. The results are shown in Table 32.

*Aim 4. To determine the minimum infectious dose (MID<sub>50</sub>) of a standard inoculum of PEDV (cell culture-grown).*

In a previous study, we determined the infectious dose of PEDV by inoculation of piglets with serial 10-fold dilutions of clarified homogenate of intestinal mucosa from infected piglets. A dilution of 10<sup>-8</sup> was able to produce disease in piglets. However, intestinal homogenate is not a standard inoculum and may vary in virulence from lab to lab. To solve this problem, we prepared serial 10-fold dilutions of a 'standard' virus inoculum (virus grown in Vero-81 cells) with known titer and cT value. We inoculated these dilutions orally in piglets. The results are shown in Table 33.

## Results:

### *Aim 1. To determine time x temperature x moisture conditions necessary to inactivate PEDV in common feed ingredients used in swine diets.*

Studies on the survival of PEDV in various feed ingredients under different time x temperature x moisture conditions are not available. To fill that gap, we studied the survival of PEDV in 9 different feed ingredients at 4 temperatures and 3 levels of relative humidity (RH). The results (Tables 1-28) show that the survival of PEDV depends on all three factors studied (type of feed ingredient, temperature, and RH). The consolidated results from Tables 1-27 are shown in Table 28, which indicates that a temperature of 60°C at any of the 3 RH levels was not sufficient to inactivate 4 log<sub>10</sub> (99.99% inactivation) of PEDV. On the other hand, 4 log<sub>10</sub> of PEDV was killed at 90°C within 15 to 30 minutes in some of the feed ingredients. However, a combination of 90°C and 70% RH was found to be the best; this combination killed 99.99% of PEDV within 10 to 30 min depending upon the type of feed ingredient used. However, even this combination was not enough to kill 99.99% virus in spray-dried plasma within 30 min.

### *Aim 2. To determine what processes (irradiation, heating, and addition of drying agents, etc.) can eliminate PEDV from feed and feed ingredients.*

**Irradiation:** Irradiation of feed and/or feed ingredients has been proposed as a useful method for inactivation of PEDV. To determine if this is true, we conducted an experiment in which feed was spiked with a known amount of PEDV and subjected to 4 different levels of irradiation (10, 20, 30, and 50 kGy) along with a negative control (in which feed was spiked with virus but was not subjected to irradiation). We found that virus inactivation was dose-dependent (Table 29). Greater than 99.97% virus was inactivated at 50 kGy while only 90% was killed at 10 kGy.

**High heat:** The temperature and time conditions in pelleting, spray drying, rendering, and storage are different. For rendering, temperatures are greater (120-145 °C) than pelleting (60-90°C), and storage (18-21 °C). However, time is greater in storage (1-7 days) than rendering (45-90 min) and pelleting (4-5 seconds). We studied the effect of 4 different temperatures (120°C, 130°C, 140°C, and 145°C) on PEDV-spiked feed at 5 minute intervals. The results are shown in Table 30. It is not surprising that increasing amounts of virus are inactivated with increase in temperature. At these 4 temperatures, 99.99% virus was inactivated within 10 to 25 minutes depending on the temperature used.

**Additives:** Another intervention studied was the addition of drying agents or acidifiers to virus-spiked feed samples. Six different additives were evaluated: Ultrap, ADA, AB, Kemin, sugar, and salt. The results are shown in Table 31. Surprisingly, the addition of sugar and salt was as good in killing virus as Kemin; they all killed more than 2 log<sub>10</sub> of virus (≥99% inactivation) but it took them 21 days to do so. Ultrap, ADA, and AB were not able to achieve this level of virus kill even after 21 days.

### *Aim 3: To develop a sensitive method for the detection of small amounts of PEDV in large amounts of feed and feed ingredients.*

**Testing method for PEDV in feed:** Currently, the feed is tested as follows: A sample of feed (50 g) is added to 200 mL of sterile PBS followed by incubation overnight at 4°C. In the morning, the mixture is centrifuged lightly and 300 µL of the supernatant is tested by RT-PCR. We thought that this procedure is inefficient. So we studied the use of pH manipulation to adsorb and desorb (elute) viruses from feed. However, we found that the current method gave superior results than the newly developed method and hence we suggest that the continued use of the current method.

**Comparison of wipes for recovering PEDV from stainless surfaces:** Currently, swiffers are used for swabbing surfaces for PEDV detection. We compare the efficacy of Swiffers, 1-MDS filters, and sterile gauze for virus elution from stainless steel surface. The results in Table 32 indicate that both Swiffer and gauze are adequate in eluting PEDV from stainless steel surface and that the use of 1-MDS filter does not add any value to this procedure.

*Aim 4. To determine the, minimum infectious dose (MID<sub>50</sub>) of a standard inoculum of PEDV (cell culture-grown).*

In a previous NPB-funded study, we had found that 10<sup>-8</sup> dilution of an intestinal homogenate from a PEDV-infected piglet was able to produce diarrhea in inoculated piglets. In addition, the jejunum of inoculated piglets was positive for PEDV as detected by RT-PCR. Unfortunately, this type of inoculum (intestinal homogenate from an infected piglet) cannot be standardized. Hence, we conducted an experiment to determine the highest dilution of a standard inoculum (PEDV grown in cell cultures) that could produce disease in piglets. The virus was grown in Vero-81 cells and serial 10-fold dilutions of the virus were inoculated in piglets. The results are shown in Table 33. The minimum infective dose of PEDV was found to be <6.8 TCID<sub>50</sub>.

**Discussion:** This study was undertaken to determine the effect of certain interventional strategies on the inactivation of PEDV in feed and feed ingredients. The strategies tested were: low heat (60<sup>0</sup>C, 70<sup>0</sup>C, 80<sup>0</sup>C, and 90<sup>0</sup>C at 30%, 50%, and 70% RH); high heat (120<sup>0</sup>C, 130<sup>0</sup>C, 140<sup>0</sup>C, and 145<sup>0</sup>C); irradiation; and the addition of drying agents/acidifiers. The survival of PEDV at 60<sup>0</sup>C to 90<sup>0</sup>C depended on type of feed ingredient, temperature, and RH. This is not surprising because temperature and RH have been found in many studies to influence the inactivation of several human and animal viruses. For example, a temperature of 60<sup>0</sup>C at any of the 3 RH levels (30%, 50%, and 70%) was not sufficient to inactivate 4 log<sub>10</sub> of PEDV but a combination of 90<sup>0</sup>C and 70% RH was able to do so. However, this combination was not able to reduce the virus by 99.99% in spray-dried plasma within 30 min. The reason for this observation is not known but may have to do with the protein content of plasma; protein and organic matter are known to protect viruses from inactivation. We studied the effect of 4 different high temperatures (120<sup>0</sup>C, 130<sup>0</sup>C, 140<sup>0</sup>C, and 145<sup>0</sup>C) on PEDV-spiked feed at 5 minute intervals and found that increasing amounts of virus were inactivated with increased temperature; 99.99% virus was inactivated within 10 to 25 min depending on the temperature used. This is disconcerting because even at 145<sup>0</sup>C, it took 10 minutes to kill 99.99% virus. The results on irradiation are in agreement with previous studies on other viruses; PEDV inactivation by irradiation was dose-dependent. Greater than 99.97% virus was inactivated at 50 kGy while only 90% was killed at 10 kGy. The effect of six different additives (Ultrap, ADA, AB, Kemin, sugar, and salt) on PEDV inactivation was evaluated. Surprisingly, the addition of sugar and salt was as good in killing virus as Kemin; they all killed more than 2 log<sub>10</sub> of virus (≥99% inactivation) but it took them 21 days to do so. Ultrap, ADA, and AB were not able to achieve this level of virus kill even after 21 days. Another objective of this study was to evaluate the currently used methods of virus elution from feed and from stainless steel surfaces. Both of the currently used methods were found to be adequate in recovering PEDV. The last objective was to determine the MID<sub>50</sub> of a standardized PEDV inoculum. We found that as low as 7 TCID<sub>50</sub> of cell culture grown virus was able to induce disease in 11-day-old piglets. This is in agreement with our previous belief that the infectious dose of PEDV is very low.

**Table 1.** Survival of PEDV in complete feed at four different temperatures (60<sup>0</sup>C, 70<sup>0</sup>C, 80<sup>0</sup>C, 90<sup>0</sup>C) and at 30% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60 <sup>0</sup> C	0	3.2x10 <sup>4</sup>	*NA
	5	1.1x10 <sup>4</sup>	65.63
	10	5.0x10 <sup>2</sup>	98.44
	15	3.2x10 <sup>2</sup>	99.00
	30	1.1x10 <sup>2</sup>	99.66
70 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	1.7x10 <sup>3</sup>	96.60
	10	9.1x10 <sup>2</sup>	98.18
	15	1.1x10 <sup>2</sup>	99.78
	30	6.8x10 <sup>1</sup>	99.86
80 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	1.8x10 <sup>3</sup>	96.40
	10	2.4x10 <sup>2</sup>	99.52
	15	2.4x10 <sup>2</sup>	99.52
	30	6.8 x10 <sup>1</sup>	99.86
90 <sup>0</sup> C	0	2.4 x10 <sup>4</sup>	NA
	5	1.7 x10 <sup>3</sup>	92.92
	10	1.9 x10 <sup>2</sup>	99.21
	15	7.6x10 <sup>1</sup>	99.68
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 30% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in complete feed within 10 to 15 min. More than 4 log<sub>10</sub> was inactivated at 90<sup>0</sup>C within 30 min.

**Table 2.** Survival of PEDV in spray dried plasma at four different temperatures (60<sup>0</sup>C, 70<sup>0</sup>C, 80<sup>0</sup>C, 90<sup>0</sup>C) and at 30% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	*NA
	5	3.6x10 <sup>3</sup>	92.80
	10	4.2x10 <sup>2</sup>	99.16
	15	2.4x10 <sup>2</sup>	99.52
	30	1.1x10 <sup>2</sup>	99.78
70 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	3.2x10 <sup>3</sup>	93.60
	10	1.9x10 <sup>3</sup>	96.20
	15	3.2x10 <sup>2</sup>	99.36
	30	4.2x10 <sup>1</sup>	99.92
80 <sup>0</sup> C	0	3.2x10 <sup>4</sup>	NA
	5	1.8x10 <sup>3</sup>	94.38
	10	2.4x10 <sup>2</sup>	92.50
	15	2.4x10 <sup>2</sup>	99.25
	30	6.8x10 <sup>1</sup>	99.79
90 <sup>0</sup> C	0	3.2x10 <sup>4</sup>	NA
	5	1.7x10 <sup>3</sup>	94.69
	10	2.4x10 <sup>2</sup>	99.25
	15	2.4x10 <sup>2</sup>	99.25
	30	4.2x10 <sup>1</sup>	99.87

\*NA: Not Applicable.

**Conclusion:** At 30% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in spray dried plasma within 10 to 15 min. None of the temperatures studied was able to kill 4 log<sub>10</sub> of the virus within 30 min.



**Table 3.** Survival of PEDV in meat meal at four different temperatures (60<sup>0</sup>C, 70<sup>0</sup>C, 80<sup>0</sup>C, 90<sup>0</sup>C) and at 30% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA*
	5	5.0x10 <sup>3</sup>	90.00
	10	9.1x10 <sup>2</sup>	98.18
	15	2.4x10 <sup>2</sup>	99.52
	30	1.1x10 <sup>2</sup>	99.78
70 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	2.4x10 <sup>3</sup>	95.20
	10	1.8x10 <sup>3</sup>	96.40
	15	2.4x10 <sup>2</sup>	99.52
	30	1.9x10 <sup>2</sup>	99.62
80 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	3.6x10 <sup>3</sup>	92.80
	10	2.4x10 <sup>2</sup>	99.52
	15	1.1x10 <sup>2</sup>	99.78
	30	6.8x10 <sup>1</sup>	99.86
90 <sup>0</sup> C	0	3.2x10 <sup>4</sup>	NA
	5	3.6x10 <sup>3</sup>	88.75
	10	6.8x10 <sup>1</sup>	99.79
	15	3.5x10 <sup>1</sup>	99.89
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 30% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in meat meal within 10 to 15 min. More than 4 log<sub>10</sub> of the virus was killed at 90<sup>0</sup>C within 30 min.

**Table 4.** Survival of PEDV in meat and bone meal at four different temperatures (60°C, 70°C, 80°C, 90°C) and at 30% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	3.2x10 <sup>4</sup>	*NA
	5	1.1x10 <sup>4</sup>	65.63
	10	1.7x10 <sup>3</sup>	94.69
	15	7.5x10 <sup>2</sup>	97.66
	30	3.5 x10 <sup>2</sup>	98.91
70°C	0	5.0x10 <sup>4</sup>	NA
	5	4.7x10 <sup>3</sup>	90.60
	10	4.2x10 <sup>2</sup>	91.60
	15	1.9x10 <sup>2</sup>	99.62
	30	≤1	≥99.99
80°C	0	3.2x10 <sup>4</sup>	NA
	5	3.2x10 <sup>2</sup>	99.00
	10	2.4x10 <sup>2</sup>	99.25
	15	1.1x10 <sup>2</sup>	99.66
	30	≤1	≥99.99
90°C	0	2.4x10 <sup>4</sup>	NA
	5	3.2x10 <sup>2</sup>	98.67
	10	1.1x10 <sup>2</sup>	99.54
	15	≤1	≥99.99
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 30% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in meat and bone meal within 5 to 15 min. More than 4 log<sub>10</sub> of the virus was killed at 70°C, 80°C, and 90°C within 15 to 30 min.

**Table 5.** Survival of PEDV in blood meal at four different temperatures (60<sup>0</sup>C, 70<sup>0</sup>C, 80<sup>0</sup>C, 90<sup>0</sup>C) and at 30% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA*
	5	3.7x10 <sup>3</sup>	92.60
	10	3.2x10 <sup>2</sup>	99.36
	15	3.2x10 <sup>2</sup>	99.36
	30	1.9x10 <sup>2</sup>	99.62
70 <sup>0</sup> C	0	3.2x10 <sup>4</sup>	NA
	5	1.1x10 <sup>4</sup>	65.63
	10	3.2x10 <sup>2</sup>	99.00
	15	2.4x10 <sup>2</sup>	99.25
	30	1.5x10 <sup>2</sup>	99.53
80 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	9.1x10 <sup>2</sup>	98.18
	10	2.4x10 <sup>2</sup>	99.52
	15	1.9x10 <sup>2</sup>	99.62
	30	1.1x10 <sup>2</sup>	99.78
90 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	1.7x10 <sup>3</sup>	96.60
	10	1.9x10 <sup>2</sup>	99.62
	15	1.9x10 <sup>2</sup>	99.62
	30	1.5x10 <sup>2</sup>	99.70

\*NA: Not Applicable.

**Conclusion:** At 30% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in blood meal within 10 min at any of the 4 temperatures studied. None of the temperatures was able to kill 4 log<sub>10</sub> of the virus within 30 min.

**Table 6.** Survival of PEDV in Grow-Finish premix at four different temperatures (60<sup>0</sup>C, 70<sup>0</sup>C, 80<sup>0</sup>C, 90<sup>0</sup>C) and at 30% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60 <sup>0</sup> C	0	3.2x10 <sup>4</sup>	*NA
	5	3.6x10 <sup>3</sup>	88.75
	10	3.2x10 <sup>2</sup>	99.00
	15	3.2x10 <sup>2</sup>	99.00
	30	1.5x10 <sup>2</sup>	99.53
70 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	1.8x10 <sup>3</sup>	96.40
	10	3.2x10 <sup>2</sup>	99.36
	15	2.4x10 <sup>2</sup>	99.52
	30	1.1x10 <sup>2</sup>	99.78
80 <sup>0</sup> C	0	3.2x10 <sup>4</sup>	NA
	5	1.7x10 <sup>3</sup>	94.69
	10	1.9x10 <sup>2</sup>	99.41
	15	1.1x10 <sup>2</sup>	99.66
	30	6.8x10 <sup>1</sup>	99.79
90 <sup>0</sup> C	0	3.2x10 <sup>4</sup>	NA
	5	3.2x10 <sup>2</sup>	99.00
	10	1.9x10 <sup>2</sup>	99.41
	15	1.5x10 <sup>2</sup>	99.53
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 30% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in Grow-Finish premix within 5 to 10 min. More than 4 log<sub>10</sub> of the virus was killed at 90<sup>0</sup>C within 30 min.

**Table 7.** Survival of PEDV in corn at four different temperatures (60°C, 70°C, 80°C, 90°C) and at 30% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	3.2x10 <sup>4</sup>	*NA
	5	3.6x10 <sup>3</sup>	88.75
	10	3.2x10 <sup>2</sup>	99.00
	15	2.4x10 <sup>2</sup>	99.25
	30	1.5x10 <sup>2</sup>	99.53
70°C	0	2.4x10 <sup>4</sup>	NA
	5	3.2x10 <sup>3</sup>	86.67
	10	3.2x10 <sup>2</sup>	98.67
	15	3.2x10 <sup>2</sup>	98.67
	30	8.3x10 <sup>1</sup>	99.65
80°C	0	3.2x10 <sup>4</sup>	NA
	5	4.2x10 <sup>3</sup>	86.88
	10	3.2x10 <sup>2</sup>	99.00
	15	2.4x10 <sup>2</sup>	99.25
	30	6.8x10 <sup>1</sup>	99.79
90°C	0	3.2x10 <sup>4</sup>	NA
	5	1.7x10 <sup>3</sup>	94.69
	10	2.4x10 <sup>2</sup>	99.25
	15	6.8x10 <sup>1</sup>	99.79
	30	6.8x10 <sup>1</sup>	99.79

\*NA: Not Applicable.

**Conclusion:** At 30% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in corn within 10 to 15 min at any of the 4 temperatures studied. None of the temperatures was able to kill 4 log<sub>10</sub> of the virus within 30 min.

**Table 8.** Survival of PEDV in Soybean meal at four different temperatures (60<sup>0</sup>C, 70<sup>0</sup>C, 80<sup>0</sup>C, 90<sup>0</sup>C) and at 30% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60 <sup>0</sup> C	0	3.2x10 <sup>4</sup>	*NA
	5	1.7x10 <sup>3</sup>	94.69
	10	2.4x10 <sup>2</sup>	99.25
	15	5.0x10 <sup>2</sup>	98.44
	30	1.1x10 <sup>2</sup>	99.66
70 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	1.8x10 <sup>3</sup>	96.40
	10	2.4x10 <sup>2</sup>	99.52
	15	1.9x10 <sup>2</sup>	99.62
	30	1.1x10 <sup>2</sup>	99.78
80 <sup>0</sup> C	0	3.2x10 <sup>4</sup>	NA
	5	1.8x10 <sup>3</sup>	94.38
	10	3.2x10 <sup>2</sup>	99.00
	15	1.9x10 <sup>2</sup>	99.41
	30	6.8x10 <sup>1</sup>	99.79
90 <sup>0</sup> C	0	2.4x10 <sup>4</sup>	NA
	5	9.1x10 <sup>2</sup>	96.21
	10	2.4x10 <sup>2</sup>	99.00
	15	1.5x10 <sup>2</sup>	99.38
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 30% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in Soybean meal within 10 min. More than 4 log<sub>10</sub> of the virus was killed at 90<sup>0</sup>C within 30 min.

**Table 9.** Survival of PEDV in DDGS at four different temperatures (60°C, 70°C, 80°C, 90°C) and at 30% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	3.2x10 <sup>4</sup>	*NA
	5	3.7x10 <sup>3</sup>	88.44
	10	3.2x10 <sup>2</sup>	99.00
	15	2.4x10 <sup>2</sup>	99.25
	30	1.5x10 <sup>2</sup>	99.53
70°C	0	5.0x10 <sup>4</sup>	NA
	5	1.8x10 <sup>3</sup>	96.40
	10	2.4x10 <sup>2</sup>	99.52
	15	2.4x10 <sup>2</sup>	99.52
	30	1.1x10 <sup>2</sup>	99.78
80°C	0	3.2x10 <sup>4</sup>	NA
	5	3.6x10 <sup>3</sup>	88.75
	10	2.4x10 <sup>2</sup>	99.25
	15	1.9x10 <sup>2</sup>	99.41
	30	6.8x10 <sup>1</sup>	99.79
90°C	0	3.2x10 <sup>4</sup>	NA
	5	5.0x10 <sup>2</sup>	98.44
	10	1.9x10 <sup>2</sup>	99.41
	15	1.1x10 <sup>2</sup>	99.66
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 30% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in DDGS within 10 min. More than 4 log<sub>10</sub> of the virus was killed at 90°C within 30 min.

**Table 10.** Survival of PEDV in complete feed at four different temperatures (60<sup>0</sup>C, 70<sup>0</sup>C, 80<sup>0</sup>C, 90<sup>0</sup>C) and at 50% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	*NA
	5	1.7x10 <sup>4</sup>	66.00
	10	5.0x10 <sup>2</sup>	99.00
	15	3.2x10 <sup>2</sup>	99.36
	30	3.2x10 <sup>2</sup>	99.36
70 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	3.6x10 <sup>3</sup>	92.80
	10	4.2x10 <sup>2</sup>	99.16
	15	2.4x10 <sup>2</sup>	99.52
	30	1.5x10 <sup>2</sup>	99.70
80 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	1.6x10 <sup>4</sup>	68.00
	10	2.4x10 <sup>2</sup>	99.52
	15	3.2x10 <sup>2</sup>	99.36
	30	6.8x10 <sup>1</sup>	99.86
90 <sup>0</sup> C	0	3.2x10 <sup>4</sup>	NA
	5	3.5x10 <sup>3</sup>	89.06
	10	1.9x10 <sup>2</sup>	99.41
	15	3.2x10 <sup>2</sup>	99.00
	30	6.8x10 <sup>1</sup>	99.79

\*NA: Not Applicable.

**Conclusion:** At 50% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in complete feed within 10 min at all four temperatures. None of the temperatures studied was able to kill 4 log<sub>10</sub> of the virus within 30 min.



**Table 11.** Survival of PEDV in spray dried plasma at four different temperatures (60°C, 70°C, 80°C, 90°C) and at **50%** relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	5.0x10 <sup>4</sup>	*NA
	5	1.7x10 <sup>4</sup>	66.00
	10	3.2x10 <sup>3</sup>	93.60
	15	1.9x10 <sup>3</sup>	96.20
	30	5.0x10 <sup>2</sup>	99.00
70°C	0	5.0x10 <sup>4</sup>	NA
	5	1.6x10 <sup>4</sup>	68.00
	10	9.1x10 <sup>2</sup>	98.18
	15	1.9x10 <sup>2</sup>	99.62
	30	1.9x10 <sup>2</sup>	99.62
80°C	0	5.0x10 <sup>4</sup>	NA
	5	1.6x10 <sup>4</sup>	68.00
	10	8.3x10 <sup>2</sup>	98.34
	15	1.9x10 <sup>2</sup>	99.62
	30	6.8x10 <sup>1</sup>	99.86
90°C	0	3.2x10 <sup>4</sup>	NA
	5	1.9x10 <sup>3</sup>	94.06
	10	2.4x10 <sup>2</sup>	99.25
	15	1.5x10 <sup>2</sup>	99.53
	30	6.8x10 <sup>1</sup>	99.79

\*NA: Not Applicable.

**Conclusion:** At 50% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in spray dried plasma within 10 to 15 min. None of the temperatures studied was able to kill 4 log<sub>10</sub> of the virus within 30 min.

**Table 12.** Survival of PEDV in meat meal at four different temperatures (60°C, 70°C, 80°C, 90°C) and at **50%** relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	5.0x10 <sup>4</sup>	*NA
	5	5.0x10 <sup>3</sup>	90.00
	10	6.8x10 <sup>2</sup>	98.64
	15	3.2x10 <sup>2</sup>	99.36
	30	3.2x10 <sup>2</sup>	99.36
70°C	0	3.2x10 <sup>4</sup>	NA
	5	1.8x10 <sup>3</sup>	94.38
	10	3.2x10 <sup>2</sup>	99.00
	15	1.1x10 <sup>2</sup>	99.66
	30	≤1	≥99.99
80°C	0	2.4x10 <sup>4</sup>	NA
	5	3.2x10 <sup>3</sup>	86.67
	10	3.7x10 <sup>2</sup>	98.46
	15	6.8x10 <sup>1</sup>	99.72
	30	≤1	≥99.99
90°C	0	5.0x10 <sup>4</sup>	NA
	5	9.1x10 <sup>2</sup>	98.18
	10	6.8x10 <sup>1</sup>	99.86
	15	6.8x10 <sup>1</sup>	99.86
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 50% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in meat meal within 10 to 15 min. More than 4 log<sub>10</sub> was inactivated at 3 temperatures within 30 min.

**Table 13.** Survival of PEDV in meat and bone meal at four different temperatures (60°C, 70°C,

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	5.0x10 <sup>4</sup>	*NA
	5	1.8x10 <sup>4</sup>	64.00
	10	1.9x10 <sup>3</sup>	96.20
	15	1.8x10 <sup>3</sup>	96.40
	30	3.2x10 <sup>2</sup>	99.36
70°C	0	5.0x10 <sup>4</sup>	NA
	5	1.8x10 <sup>4</sup>	64.00
	10	1.8x10 <sup>3</sup>	96.20
	15	8.3x10 <sup>2</sup>	98.34
	30	3.7x10 <sup>2</sup>	99.26
80°C	0	5.0x10 <sup>4</sup>	NA
	5	8.3x10 <sup>3</sup>	83.40
	10	9.1x10 <sup>2</sup>	98.18
	15	2.4x10 <sup>2</sup>	99.52
	30	1.1x10 <sup>2</sup>	99.78
90°C	0	5.0x10 <sup>4</sup>	NA
	5	1.6x10 <sup>4</sup>	68.00
	10	1.9x10 <sup>2</sup>	99.62
	15	9.1x10 <sup>1</sup>	99.82
	30	9.1x10 <sup>1</sup>	99.82

\*NA: Not Applicable.

**Conclusion:** At 50% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in meat and bone meal 15 to 30 min. None of the temperatures studied was able to kill 4 log<sub>10</sub> of the virus within 30 min.

**Table 14.** Survival of PEDV in blood meal at four different temperatures (60<sup>o</sup>C, 70<sup>o</sup>C, 80<sup>o</sup>C, 90<sup>o</sup>C) and at **50%** relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60 <sup>o</sup> C	0	5.0x10 <sup>4</sup>	*NA
	5	1.6x10 <sup>4</sup>	68.00
	10	2.4x10 <sup>3</sup>	95.20
	15	1.1x10 <sup>3</sup>	99.78
	30	6.8x10 <sup>2</sup>	99.86
70 <sup>o</sup> C	0	3.2x10 <sup>4</sup>	NA
	5	7.8x10 <sup>3</sup>	75.63
	10	1.8x10 <sup>3</sup>	94.38
	15	1.8x10 <sup>3</sup>	94.38
	30	4.2x10 <sup>2</sup>	98.69
80 <sup>o</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	1.8x10 <sup>4</sup>	64.00
	10	1.8x10 <sup>3</sup>	96.40
	15	9.1x10 <sup>2</sup>	98.18
	30	5.0x10 <sup>2</sup>	99.00
90 <sup>o</sup> C	0	3.2x10 <sup>4</sup>	NA
	5	5.0x10 <sup>2</sup>	98.44
	10	1.9x10 <sup>2</sup>	99.41
	15	≤1	≥99.99
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 50% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in blood meal 10 to 30 min. More than 4 log<sub>10</sub> of the virus was killed within 30 min at 90<sup>o</sup>C.

**Table 15.** Survival of PEDV in Grow finish premi at four different temperatures (60<sup>0</sup>C, 70<sup>0</sup>C, 80<sup>0</sup>C, 90<sup>0</sup>C) and at **50%** relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA*
	5	1.8x10 <sup>4</sup>	64.00
	10	3.7x10 <sup>3</sup>	92.60
	15	1.8x10 <sup>3</sup>	96.40
	30	3.2x10 <sup>2</sup>	99.36
70 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	1.6x10 <sup>4</sup>	68.00
	10	8.3x10 <sup>2</sup>	98.34
	15	2.4x10 <sup>2</sup>	99.52
	30	2.4x10 <sup>2</sup>	99.52
80 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	1.6x10 <sup>4</sup>	68.00
	10	1.7x10 <sup>3</sup>	96.60
	15	8.3x10 <sup>2</sup>	98.34
	30	2.4x10 <sup>2</sup>	99.52
90 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	1.6x10 <sup>4</sup>	68.00
	10	1.7x10 <sup>3</sup>	96.60
	15	2.4x10 <sup>2</sup>	99.52
	30	1.1x10 <sup>2</sup>	99.78

\*NA: Not Applicable.

**Conclusion:** At 50% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in Grow-Finish premix within 15 to 30 min. None of the temperatures studied was able to kill 4 log<sub>10</sub> of the virus within 30 min.

**Table 16.** Survival of PEDV in corn at four different temperatures (60°C, 70°C, 80°C, 90°C) and at **50%** relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	5.0x10 <sup>4</sup>	*NA
	5	1.9x10 <sup>4</sup>	62.00
	10	2.4x10 <sup>3</sup>	95.20
	15	1.9x10 <sup>3</sup>	96.20
	30	5.0x10 <sup>2</sup>	99.00
70°C	0	3.2x10 <sup>4</sup>	NA
	5	9.1x10 <sup>3</sup>	71.56
	10	1.8x10 <sup>3</sup>	94.38
	15	5.0x10 <sup>2</sup>	98.44
	30	3.2x10 <sup>2</sup>	99.00
80°C	0	5.0x10 <sup>4</sup>	NA
	5	3.6x10 <sup>3</sup>	92.80
	10	2.4x10 <sup>2</sup>	99.52
	15	1.1x10 <sup>2</sup>	99.78
	30	≤1	≥99.99
90°C	0	3.2x10 <sup>4</sup>	NA
	5	1.7x10 <sup>3</sup>	96.60
	10	1.9x10 <sup>2</sup>	99.62
	15	6.8x10 <sup>1</sup>	99.86
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 50% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in corn within 10 to 30 min. More than 4 log<sub>10</sub> of the virus was killed within 30 min at 80°C and 90°C.

**Table 17.** Survival of PEDV in Soybean meal at four different temperatures (60°C, 70°C, 80°C, 90°C) and at 50% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	5.0x10 <sup>4</sup>	*NA
	5	1.8x10 <sup>4</sup>	64.00
	10	1.8x10 <sup>3</sup>	96.40
	15	5.0x10 <sup>2</sup>	99.00
	30	1.5x10 <sup>2</sup>	99.70
70°C	0	5.0x10 <sup>4</sup>	NA
	5	1.8x10 <sup>4</sup>	64.00
	10	2.4x10 <sup>3</sup>	95.20
	15	3.2x10 <sup>2</sup>	99.36
	30	1.5x10 <sup>2</sup>	99.70
80°C	0	3.2x10 <sup>4</sup>	NA
	5	7.6x10 <sup>3</sup>	76.25
	10	4.2x10 <sup>2</sup>	98.69
	15	2.4x10 <sup>2</sup>	99.25
	30	6.8x10 <sup>1</sup>	99.79
90°C	0	3.2x10 <sup>4</sup>	NA
	5	1.7x10 <sup>4</sup>	NA
	10	3.2x10 <sup>2</sup>	99.00
	15	2.4x10 <sup>2</sup>	99.25
	30	6.8x10 <sup>1</sup>	99.79

\*NA: Not Applicable.

**Conclusion:** At 50% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in Soybean meal 15 to 30 min. None of the temperatures studied was able to kill 4 log<sub>10</sub> of the virus within 30 min.

**Table 18.** Survival of PEDV in DDGS at four different temperatures (60°C, 70°C, 80°C, 90°C)

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	5.0x10 <sup>4</sup>	*NA
	5	1.8x10 <sup>4</sup>	64.00
	10	9.1x10 <sup>2</sup>	98.18
	15	3.2x10 <sup>2</sup>	99.36
	30	2.4x10 <sup>2</sup>	99.52
70°C	0	3.2x10 <sup>4</sup>	NA
	5	5.0x10 <sup>3</sup>	84.38
	10	1.8x10 <sup>3</sup>	94.38
	15	4.2x10 <sup>2</sup>	98.69
	30	3.7x10 <sup>2</sup>	98.84
80°C	0	5.0x10 <sup>4</sup>	NA
	5	3.6x10 <sup>3</sup>	92.80
	10	3.7x10 <sup>2</sup>	99.26
	15	2.4x10 <sup>2</sup>	99.52
	30	1.1x10 <sup>2</sup>	99.78
90°C	0	5.0x10 <sup>4</sup>	NA
	5	1.6x10 <sup>4</sup>	68.00
	10	8.3x10 <sup>2</sup>	98.34
	15	2.4x10 <sup>2</sup>	99.52
	30	1.1x10 <sup>2</sup>	99.78

\*NA: Not Applicable.

**Conclusion:** At 50% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in DDGS 15 to 30 min. None of the temperatures studied was able to kill 4 log<sub>10</sub> of the virus within 30 min.



**Table 19.** Survival of PEDV in complete feed at four different temperatures (60°C, 70°C, 80°C, 90°C) and at 70% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	3.2x10 <sup>4</sup>	*NA
	5	1.8x10 <sup>4</sup>	43.75
	10	5.0x10 <sup>2</sup>	98.44
	15	5.0x10 <sup>2</sup>	98.44
	30	3.2x10 <sup>2</sup>	99.00
70°C	0	5.0x10 <sup>4</sup>	NA
	5	1.6x10 <sup>4</sup>	68.00
	10	3.2x10 <sup>2</sup>	99.36
	15	1.9x10 <sup>2</sup>	99.62
	30	1.1x10 <sup>2</sup>	99.78
80°C	0	5.0x10 <sup>4</sup>	NA
	5	3.6x10 <sup>3</sup>	92.80
	10	1.9x10 <sup>2</sup>	96.20
	15	3.2x10 <sup>1</sup>	99.94
	30	≤1	≥99.99
90°C	0	5.0x10 <sup>4</sup>	NA
	5	1.7x10 <sup>3</sup>	96.60
	10	≤1	≥99.99
	15	≤1	≥99.99
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 70% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in complete feed within 10 to 30 min. More than 4 log<sub>10</sub> of the virus was killed at 80°C and 90°C within 10 to 30 min.

**Table 20.** Survival of PEDV in spray dried plasma at four different temperatures (60°C, 70°C, 80°C, 90°C) and at 70% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	5.0x10 <sup>4</sup>	*NA
	5	1.7x10 <sup>4</sup>	66.00
	10	1.9x10 <sup>3</sup>	96.20
	15	2.4x10 <sup>3</sup>	95.20
	30	3.2x10 <sup>2</sup>	99.36
70°C	0	3.2x10 <sup>4</sup>	NA
	5	3.6x10 <sup>3</sup>	88.75
	10	3.2x10 <sup>2</sup>	99.00
	15	1.1x10 <sup>2</sup>	99.66
	30	6.8x10 <sup>1</sup>	99.79
80°C	0	5.0x10 <sup>4</sup>	NA
	5	1.8x10 <sup>4</sup>	64.00
	10	1.9x10 <sup>2</sup>	99.62
	15	1.1x10 <sup>2</sup>	99.78
	30	6.8x10 <sup>1</sup>	99.86
90°C	0	5.0x10 <sup>4</sup>	NA
	5	1.6x10 <sup>4</sup>	68.00
	10	1.9x10 <sup>2</sup>	99.62
	15	1.1x10 <sup>2</sup>	99.78
	30	6.8x10 <sup>1</sup>	99.86

\*NA: Not Applicable.

**Conclusion:** At 70% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in spray dried plasma within 10 to 30 min. None of the temperatures studied was able to kill 4 log<sub>10</sub> of the virus within 30 min.

**Table 21.** Survival of PEDV in meat meal at four different temperatures (60°C, 70°C, 80°C, 90°C) and at 70% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	3.2x10 <sup>4</sup>	*NA
	5	1.6x10 <sup>4</sup>	50.00
	10	1.8x10 <sup>3</sup>	94.38
	15	2.4x10 <sup>2</sup>	99.25
	30	1.9x10 <sup>2</sup>	99.41
70°C	0	5.0x10 <sup>4</sup>	NA
	5	3.6x10 <sup>3</sup>	92.80
	10	1.8x10 <sup>3</sup>	96.40
	15	1.1x10 <sup>2</sup>	99.78
	30	6.8x10 <sup>1</sup>	99.86
80°C	0	3.2x10 <sup>4</sup>	NA
	5	1.8x10 <sup>3</sup>	94.38
	10	2.4x10 <sup>2</sup>	99.25
	15	1.1x10 <sup>2</sup>	99.66
	30	6.8x10 <sup>1</sup>	99.79
90°C	0	2.4x10 <sup>4</sup>	NA
	5	3.6 x10 <sup>3</sup>	99.94
	10	≤1	≥99.99
	15	≤1	≥99.99
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 70% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in meat meal within 5 to 15 min. More than 4 log<sub>10</sub> was inactivated at 90°C within 10 min.

**Table 22.** Survival of PEDV in bone meal at four different temperatures (60°C, 70°C, 80°C, 90°C) and at 70% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	5.0x10 <sup>4</sup>	*NA
	5	1.7x10 <sup>4</sup>	66.00
	10	2.4x10 <sup>3</sup>	95.20
	15	9.1x10 <sup>2</sup>	98.18
	30	1.1x10 <sup>2</sup>	99.78
70°C	0	5.0x10 <sup>4</sup>	NA
	5	3.6x10 <sup>3</sup>	92.80
	10	4.2x10 <sup>2</sup>	99.16
	15	6.8x10 <sup>1</sup>	99.86
	30	≤1	≥99.99
80°C	0	2.4x10 <sup>4</sup>	NA
	5	1.9x10 <sup>3</sup>	92.08
	10	1.9x10 <sup>2</sup>	99.21
	15	≤1	≥99.99
	30	≤1	≥99.99
90°C	0	3.2x10 <sup>4</sup>	NA
	5	1.8x10 <sup>2</sup>	99.44
	10	≤1	≥99.99
	15	≤1	≥99.99
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 70% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in meat and bone meal within 5 to 30 min. More than 4 log<sub>10</sub> was inactivated at 80°C and 90°C within 10 to 15 min.

**Table 23.** Survival of PEDV in blood meal at four different temperatures (60<sup>0</sup>C, 70<sup>0</sup>C, 80<sup>0</sup>C, 90<sup>0</sup>C) and at 70% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	*NA
	5	3.8x10 <sup>3</sup>	92.40
	10	2.4x10 <sup>3</sup>	95.20
	15	5.0x10 <sup>2</sup>	99.00
	30	2.4x10 <sup>2</sup>	99.52
70 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	1.6x10 <sup>4</sup>	68.00
	10	1.8x10 <sup>3</sup>	96.40
	15	3.2x10 <sup>2</sup>	99.36
	30	1.9x10 <sup>1</sup>	99.96
80 <sup>0</sup> C	0	3.2x10 <sup>4</sup>	NA
	5	7.6x10 <sup>3</sup>	76.25
	10	2.0x10 <sup>2</sup>	99.94
	15	3.2x10 <sup>2</sup>	99.00
	30	6.8x10 <sup>1</sup>	99.79
90 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	9.1x10 <sup>2</sup>	98.18
	10	6.8x10 <sup>1</sup>	99.86
	15	≤1	≥99.99
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 70% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in blood meal within 10 to 30 min. More than 4 log<sub>10</sub> was inactivated at 90<sup>0</sup>C within 15 min.

**Table 24.** Survival of PEDV in Grow-Finish premix at four different temperatures (60°C, 70°C, 80°C, 90°C) and at 70% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	3.2x10 <sup>4</sup>	*NA
	5	1.1x10 <sup>4</sup>	65.63
	10	2.4x10 <sup>3</sup>	92.50
	15	1.8x10 <sup>3</sup>	94.38
	30	2.4x10 <sup>2</sup>	99.25
70°C	0	5.0x10 <sup>4</sup>	NA
	5	1.6x10 <sup>4</sup>	68.00
	10	1.8x10 <sup>3</sup>	96.40
	15	3.2x10 <sup>2</sup>	99.36
	30	2.4x10 <sup>2</sup>	99.52
80°C	0	3.2x10 <sup>4</sup>	NA
	5	1.6x10 <sup>4</sup>	50.00
	10	2.4x10 <sup>2</sup>	99.25
	15	1.5x10 <sup>2</sup>	99.53
	30	6.8x10 <sup>1</sup>	99.79
90°C	0	5.0x10 <sup>4</sup>	NA
	5	7.7x10 <sup>3</sup>	84.60
	10	2.4x10 <sup>2</sup>	99.25
	15	6.8x10 <sup>1</sup>	99.79
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 70% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in Grow-Finish premix within 10 to 30 min. More than 4 log<sub>10</sub> was inactivated at 90°C within 30 min.

**Table 25.** Survival of PEDV in corn at four different temperatures (60°C, 70°C, 80°C, 90°C) and at 70% relative humidity

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
60°C	0	5.0x10 <sup>4</sup>	*NA
	5	5.0x10 <sup>3</sup>	90.0
	10	1.8x10 <sup>3</sup>	96.40
	15	3.2x10 <sup>2</sup>	99.36
	30	3.2x10 <sup>2</sup>	99.36
70°C	0	5.0x10 <sup>4</sup>	NA
	5	5.0x10 <sup>3</sup>	90.0
	10	5.0x10 <sup>2</sup>	99.00
	15	3.2x10 <sup>2</sup>	99.36
	30	3.2x10 <sup>2</sup>	99.36
80°C	0	5.0x10 <sup>4</sup>	NA
	5	5.0x10 <sup>3</sup>	90.0
	10	5.0x10 <sup>2</sup>	99.00
	15	1.1x10 <sup>2</sup>	99.78
	30	5x10 <sup>1</sup>	≥99.99
90°C	0	5.0x10 <sup>4</sup>	NA
	5	5.0x10 <sup>2</sup>	99.00
	10	2.0x10 <sup>2</sup>	99.60
	15	≤1	≥99.99
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 70% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in corn within 10 to 15 min. More than 4 log<sub>10</sub> was inactivated at 80°C and 90°C within 15 to 30 min.

**Table 26.** Survival of PEDV in Soybean meal at four different temperatures (60°C, 70°C, 80°C, 90°C) and at 70% relative humidity

Temperature	Incubation period (in min)	Average Virus titer TCID <sub>50</sub> /mL	Average Percent virus reduction
60°C	0	5.0x10 <sup>4</sup>	*NA
	5	16.8x10 <sup>4</sup>	NA
	10	1.5x10 <sup>3</sup>	97.00
	15	9.1x10 <sup>2</sup>	98.18
	30	1.1x10 <sup>2</sup>	99.78
70°C	0	5.0x10 <sup>4</sup>	NA
	5	1.8x10 <sup>3</sup>	96.40
	10	3.2x10 <sup>2</sup>	99.36
	15	3.2x10 <sup>2</sup>	99.36
	30	2.4x10 <sup>2</sup>	99.52
80°C	0	3.2x10 <sup>4</sup>	NA
	5	9.1x10 <sup>2</sup>	97.16
	10	1.5x10 <sup>2</sup>	99.53
	15	≤1	≥99.99
	30	≤1	≥99.99
90°C	0	3.2x10 <sup>4</sup>	NA
	5	8.3x10 <sup>2</sup>	97.41
	10	≤1	≥99.99
	15	≤1	≥99.99
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 70% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in Soybean meal within 10 to 30 min. More than 4 log<sub>10</sub> was inactivated at 80°C and 90°C within 10 to 15 min.



**Table 27.** Survival of PEDV in DDGS at four different temperatures (60<sup>0</sup>C, 70<sup>0</sup>C, 80<sup>0</sup>C, 90<sup>0</sup>C) and at 70% relative humidity

Temperature	Incubation period (in min)	Average Virus titer TCID <sub>50</sub> /mL	Average Percent virus reduction
60 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	*NA
	5	5.0x10 <sup>3</sup>	90.00
	10	9.1x10 <sup>2</sup>	98.18
	15	1.1x10 <sup>1</sup>	99.78
	30	1.1x10 <sup>1</sup>	99.78
70 <sup>0</sup> C	0	23.5x10 <sup>3</sup>	NA
	5	1.8x10 <sup>3</sup>	92.50
	10	2.4x10 <sup>2</sup>	99.00
	15	≤1	≥99.9
	30	≤1	≥99.9
80 <sup>0</sup> C	0	5.0x10 <sup>4</sup>	NA
	5	1.8x10 <sup>3</sup>	96.40
	10	6.8x10 <sup>1</sup>	99.86
	15	≤1	≥99.99
	30	≤1	≥99.99
90 <sup>0</sup> C	0	3.2x10 <sup>4</sup>	NA
	5	5.0x10 <sup>2</sup>	98.44
	10	2.0x10 <sup>2</sup>	99.38
	15	≤1	≥99.99
	30	≤1	≥99.99

\*NA: Not Applicable.

**Conclusion:** At 70% RH, more than 2 log<sub>10</sub> of PEDV was inactivated in DDGS within 10 to 15 min. More than 4 log<sub>10</sub> was inactivated at the three high temperatures within 15 min.

**Table 28.** Time required (in minutes) to kill  $\geq 99.99$  of PEDV in nine feed ingredients at four different temperatures and at three humidity levels<sup>A</sup>

Feed ingredient	60°C			70°C			80°C			90°C		
	30%	50%	70%	30%	50%	70%	30%	50%	70%	30%	50%	70%
Complete feed	NA <sup>B</sup>	NA	NA	NA	NA	NA	NA	NA	30	30	NA	10
Spray dried plasma	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA
Meat meal	NA	NA	NA	NA	30	NA	NA	30	NA	30	30	10
Meat and bone meal	NA	NA	NA	30	NA	30	30	NA	15	15	NA	10
Blood meal	NA	NA	NA	NA	NA	NA	NA	NA	NA	NA	15	15
Grow-Finish premix	NA	NA	NA	NA	NA	NA	NA	NA	NA	30	NA	30
Corn	NA	NA	NA	NA	NA	NA	NA	30	30	NA	30	15
Soybean meal	NA	NA	NA	NA	NA	NA	NA	NA	15	NA	NA	10
DDGS	NA	NA	NA	NA	NA	15	NA	NA	15	30	NA	15

<sup>A</sup> The results in this Table are a composite of results from Tables 1 through 27.

<sup>B</sup> NA=99.99% inactivation not achieved

**Table 29.** The effect of various irradiation doses on PEDV survival in complete feed.

<b>Irradiation dose (kGy) used</b>	<b>Actual Dose (kGy)</b>	<b>Virus titer TCID<sub>50</sub>/mL</b>	<b>Percent virus reduction</b>
Control	No irradiation	3.2x10 <sup>3</sup>	NA
10	9.28	3.2x10 <sup>2</sup>	90.00
20	22.39	1.5x10 <sup>2</sup>	95.30
30	30.65	6.8 x10 <sup>1</sup>	97.90
50	48.13	≤ 1	≥99.97

**Conclusions:** Three log<sub>10</sub> of PEDV was inactivated by the 50 kGy dose of irradiation. As the dose increased, the amount of virus inactivation also increased.

**Table 30.** Survival of PEDV in complete feed when heated to high temperatures

Temperature	Incubation period (in min)	Average virus titer TCID <sub>50</sub> /mL	Average percent virus reduction
120°C	0	6.8x10 <sup>3</sup>	*NA
	5	6.8x10 <sup>2</sup>	90.00
	10	3.2 x10 <sup>2</sup>	95.29
	15	2.4 x10 <sup>2</sup>	96.47
	20	1.1 x10 <sup>2</sup>	98.38
	25	≤1	≥99.99
	30	≤1	≥99.99
130°C	0	6.8x10 <sup>3</sup>	*NA
	5	3.2 x10 <sup>2</sup>	95.29
	10	1.5 x10 <sup>2</sup>	97.79
	15	≤1	≥99.99
	20	≤1	≥99.99
	25	≤1	≥99.99
	30	≤1	≥99.99
140°C	0	6.8x10 <sup>3</sup>	*NA
	5	2.4 x10 <sup>2</sup>	96.47
	10	1.5 x10 <sup>2</sup>	97.79
	15	≤1	≥99.99
	20	≤1	≥99.99
	25	≤1	≥99.99
	30	≤1	≥99.99
145°C	0	5.0 x10 <sup>3</sup>	*NA
	5	3.2 x10 <sup>2</sup>	93.60
	10	≤1	≥99.99
	15	≤1	≥99.99
	20	≤1	≥99.99
	25	≤1	≥99.99
	30	≤1	≥99.99

NA\*: not applicable.

**Conclusion:** Greater than 4 log<sub>10</sub> of PEDV are inactivated various temperatures within 10 to 25 minutes.

**Table 31.** Survival of PEDV in feed with various additives

Additive	Incubation period (in days)	Control Virus titer TCID <sub>50</sub> /mL <sup>a</sup>	Virus titer TCID <sub>50</sub> /mL	Av. % virus reduction
Nutriad (Ultrap)	0	5.0 x10 <sup>3</sup>	5.0 x10 <sup>3</sup>	0
	1	3.6 x10 <sup>3</sup>	6.8 x10 <sup>3</sup>	0
	3	4.4 x10 <sup>3</sup>	6.8 x10 <sup>2</sup>	84.5
	5	9.8 x10 <sup>3</sup>	6.8 x10 <sup>2</sup>	93.1
	7	4.7 x10 <sup>3</sup>	6.8 x10 <sup>2</sup>	85.5
	14	8.6 x10 <sup>2</sup>	3.2 x10 <sup>2</sup>	62.8
	21	2.5 x10 <sup>2</sup>	6.8 x10 <sup>1</sup>	72.8
Novus International Inc. (ADA)	0	5.0 x10 <sup>3</sup>	6.8 x10 <sup>3</sup>	0
	1	3.6 x10 <sup>3</sup>	1.5 x10 <sup>3</sup>	58.3
	3	4.4 x10 <sup>3</sup>	1.5x10 <sup>2</sup>	96.6
	5	9.8 x10 <sup>3</sup>	6.8 x10 <sup>2</sup>	93.1
	7	4.7 x10 <sup>3</sup>	6.8 x10 <sup>1</sup>	98.6
	14	8.6 x10 <sup>2</sup>	6.8 x10 <sup>1</sup>	92.1
	21	2.5 x10 <sup>2</sup>	6.8 x10 <sup>1</sup>	72.8
Agri Nutrition: (AB)	0	5.0 x10 <sup>3</sup>	5.0 x10 <sup>3</sup>	0
	1	3.6 x10 <sup>3</sup>	1.5x10 <sup>3</sup>	58.3
	3	4.4 x10 <sup>3</sup>	6.8 x10 <sup>2</sup>	84.5
	5	9.8 x10 <sup>3</sup>	3.2 x10 <sup>2</sup>	96.7
	7	4.7 x10 <sup>3</sup>	6.8 x10 <sup>2</sup>	85.5
	14	8.6 x10 <sup>2</sup>	6.8 x10 <sup>1</sup>	92.1
	21	2.5 x10 <sup>2</sup>	6.8 x10 <sup>1</sup>	72.8
Kemin Agrifoods North America (Kem)	0	5.0 x10 <sup>3</sup>	5.0 x10 <sup>3</sup>	0
	1	3.6 x10 <sup>3</sup>	6.8 x10 <sup>3</sup>	0
	3	4.4 x10 <sup>3</sup>	6.8 x10 <sup>3</sup>	0
	5	9.8 x10 <sup>3</sup>	3.2 x10 <sup>2</sup>	96.7
	7	4.7 x10 <sup>3</sup>	6.8 x10 <sup>1</sup>	98.6
	14	8.6 x10 <sup>2</sup>	6.8 x10 <sup>1</sup>	92.1
	21	2.5 x10 <sup>2</sup>	≤ 1	≥99.6
Sugar (sugar)	0	5.0 x10 <sup>3</sup>	6.8 x10 <sup>3</sup>	0
	1	3.6 x10 <sup>3</sup>	6.8 x10 <sup>3</sup>	0
	3	4.4 x10 <sup>3</sup>	1.5 x10 <sup>3</sup>	65.9
	5	9.8 x10 <sup>3</sup>	3.2 x10 <sup>2</sup>	96.7
	7	4.7 x10 <sup>3</sup>	1.5x10 <sup>2</sup>	96.8
	14	8.6 x10 <sup>2</sup>	3.2 x10 <sup>2</sup>	62.8
	21	2.5 x10 <sup>2</sup>	≤ 1	≥99.6
Salt	0	5.0 x10 <sup>3</sup>	5.0 x10 <sup>3</sup>	0
	1	3.6 x10 <sup>3</sup>	6.8 x10 <sup>3</sup>	0
	3	4.4 x10 <sup>3</sup>	3.2 x10 <sup>3</sup>	27.3
	5	9.8 x10 <sup>3</sup>	3.2 x10 <sup>3</sup>	67.3
	7	4.7 x10 <sup>3</sup>	6.8 x10 <sup>1</sup>	98.6
	14	8.6 x10 <sup>2</sup>	3.2 x10 <sup>2</sup>	62.8
	21	2.5 x10 <sup>2</sup>	≤ 1	≥99.6

**Conclusion:** Only 3 of 6 additives (Kemin, sugar, and salt) were able to inactivate more than 2 log<sub>10</sub> of PEDV and that too after 21 days.

**Table 32.** Comparison of different wipes for PEDV recovery from stainless steel surface

Wipe Used	Experiment No.	Virus titer TCID <sub>50</sub> /mL	Total Virus recovered	Percent virus recovery
Swiffer	1	3.2x10 <sup>3</sup>	1.6x10 <sup>4</sup>	47.1%
	2	3.2x10 <sup>3</sup>	1.6x10 <sup>4</sup>	47.1%
	3	3.2x10 <sup>2</sup>	1.6x10 <sup>3</sup>	4.7%
	Average	2.2x10 <sup>3</sup>	1.1x10 <sup>4</sup>	<b>16.2%</b>
Gauze	1	3.2x10 <sup>3</sup>	1.6x10 <sup>4</sup>	47.1%
	2	3.2x10 <sup>3</sup>	1.6x10 <sup>4</sup>	47.1%
	3	1.5x10 <sup>3</sup>	7.5x10 <sup>3</sup>	11.0%
	Average	2.6x10 <sup>3</sup>	1.3x10 <sup>4</sup>	<b>19.1%</b>
1-MDS filter	1	6.8x10 <sup>2</sup>	3.4x10 <sup>3</sup>	10.0%
	2	1.5x10 <sup>3</sup>	7.5x10 <sup>3</sup>	22.1%
	3	3.2x10 <sup>2</sup>	1.6x10 <sup>3</sup>	4.7%
	Average	8.3x10 <sup>2</sup>	4.2x10 <sup>3</sup>	<b>6.1%</b>

**Conclusion:** Although virus recovery was only 16-19%, the Swiffer and gauze appeared to be better than 1-MDS for recovering PEDV from stainless steel surfaces.

**Table 33:** MID50 of PEDV grown in Vero cells

<b>Virus dilution</b>	<b>TCID<sub>50</sub></b>	<b>Initial cT value</b>	<b>Extent of diarrhea</b>	<b>cT value in mucosal samples from inoculated piglets</b>
Undiluted (p13)	6.8 x 10 <sup>4</sup>	22.52	3	16.60
10 <sup>-1</sup>	6.8 x 10 <sup>3</sup>	25.13	ND*	ND
10 <sup>-2</sup>	6.8 x 10 <sup>2</sup>	27.09	ND	ND
10 <sup>-3</sup>	6.8 x 10 <sup>1</sup>	30.20	ND	ND
10 <sup>-4</sup>	6.8 x 10 <sup>0</sup>	33.10	ND	ND
10 <sup>-5</sup>	<6.8	35.50	1	29.04
10 <sup>-6</sup>	<0.68	39.60	0	-
10 <sup>-7</sup>	<0.068	-	-	-
10 <sup>-8</sup>	<0.0068	-	-	-
10 <sup>-9</sup>	<0.00068	-	-	-
10 <sup>-10</sup>	<0.000068	-	-	-

\*ND= not done.

**Conclusion:** A 10<sup>-5</sup> dilution of Vero cell grown PEDV (<6.8 TCID<sub>50</sub> of the virus) was able to infect piglets resulting in diarrhea. The mucosal samples from piglets inoculated with this dilution were also positive for PEDV by RT-PCR.