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# Algae beta-glucan inhibits African Swine (ASF) virus replication in macrophages

| Key Conclusions |
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| **Algae beta-1,3-glucan (BG) reduced ASF viral replication in porcine alveolar macrophage (PAM) cells.** |

## INTRODUCTION

African swine fever (ASF), a highly contagious disease of swine has brought a big socioeconomic impact on the swine industry worldwide. The virus that causes this disease mainly infects macrophages and monocytes but is also known to infect other cells. Studying ASFv *in vitro* usually involves the use of porcine monocytes and macrophages to mimic the natural ASFv infection, in which all the virus isolates readily grow. At this moment, other than biosecurity there is no preventive measure available nor a treatment.

There have been several studies done on beta-1,3-glucan, derived from algae, *Euglena gracilis*, showing that it triggers immunomodulatory properties because it is recognized by cell surface receptors such as Dectin-1 on vertebrate immune cells as a pathogen-associated molecular pattern. Binding algae beta-1,3-glucan to macrophages and dendritic cells initiates an immunomodulatory effect: a cascade of signals that results in increased rates of phagocytosis and antigen presentation, production of reactive oxygen species, and secretion of cytokines and chemokines. Macrophages being the primary replication site of African Swine Fever virus (ASFv) and the immune modulating effects of algae beta-1,3-glucan initiated this study to determine if algae beta-1,3-glucan will be able to help decrease propagation of the virus in porcine alveolar macrophage (PAM) cells. A first study to investigate whether, the commercially available supplement for pigs, Aleta, an algae beta-1,3-glucan, could impact the pathogenesis of ASF in pigs.

## KEYWORDS

Aleta™, African Swine Fever, macrophages, viral replication

## MATERIAL AND METHODS

*African Swine Fever Virus Strain*

Ha Nam isolate was used in the study. This isolate is genotype II, serotype 8 and IGR variant II. Accession numbers: MN199633 for p72 gene and MN199634 for CD2v gene. Virus stock: 106 HAD50/ml. OIE protocol was used for Virus isolation and HAD (haemadsorption) test.

*Preparation of algae beta-1,3-glucan*

0.1 g of algae beta-1,3-glucan was dissolved in 10 ml of 1N potassium hydroxide (KOH) solution. 1N KOH solution was made by dissolving 5.611 grams of KOH in 100ml of distilled water.

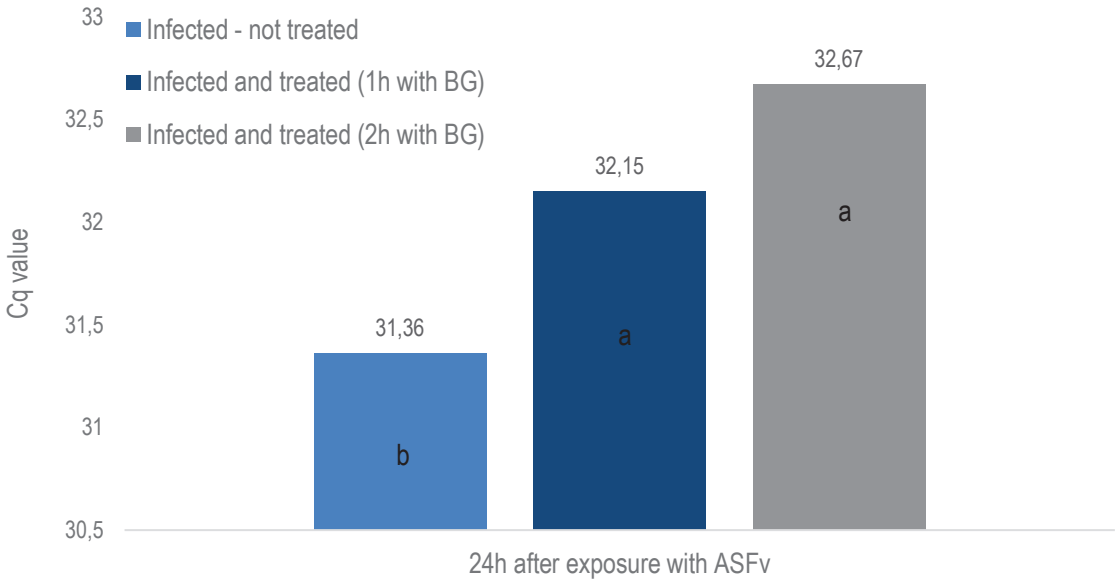
*Effect of algae beta-1,3-glucan on ASFv propagation by Realtime PCR*

Porcine alveolar macrophages (PAM) cells were treated with 50 micrograms/ml of algae beta-1,3 glucan and were incubated at different times: 60 mins and 120 mins. Treated PAM cells with algae beta-1,3- glucan were inoculated with ASF virus exposure at 103 $HAD_{50}$/ml. Three samples were then collected at 24 h post virus exposure for Real-time PCR for each dried algae beta-1,3 glucan exposure time. Real-time PCR used the OIE protocol for the quantification of ASF virus. Positive controls were also included in the trial represented by PAM cells not treated with dried algae beta-1,3 glucan and exposed and to ASF virus. Quantification of viral load through PCR runs in cycles, and results are expressed in quantitation cycle (Cq) value. This is the term for the cycle in which a positive signal (presence of the pathogen) can be detected. The higher the presence of the pathogen, the earlier it will be detected, the shorter the quantitation cycle, Cq-value.

## RESULTS AND DISCUSSION

The study shows that algae beta-1,3 glucan (BG) has potential effects to inhibit/reduce ASFv replication in PAM cells. A 50 microgram/ml for prior 2 h incubation of the macrophage with BG showed a good protection against ASFV infection. At the dose of 50 micrograms/ml, a significant increase of Cq value was noted at 24 hours after ASF virus exposure in groups of 1 h and 2 h incubation with BG. Graph 1 shows a significant lower Cq value for the control group compared to the BG incubated groups, meaning, a shorter PCR cycle is needed to detect virus, indicating a higher presence of ASFv compared to the BG groups.

Graph. 1. Effect of algae beta-1,3-glucan (BG) at 50 micrograms/ml on ASFv propagation in macrophages (MF) by Real-Time PCR




Although further study is needed to understand the mode of action of algae beta-1,3-glucan on ASF virus propagation, a hypothesis can be that the ability of algal beta-1,3-glucans to enhance infection defense mechanisms and simultaneously increase secretion of IL-10 is responsible for the algae beta-1,3-glucan to inhibit the propagation of ASF virus in the PAM cells. When ASF virus infects monocytes, it stimulates the secretion of proinflammatory cytokines and down-regulates the expression of IL-10 an anti-inflammatory cytokine. This cytokine secretion of monocytes when infected with the ASF virus plays an important role in the pathogenesis of the disease and the action of algae beta-1,3 glucan on these cytokines can help decrease the propagation of the ASFv in the PAM cells.

## CONCLUSION

In conclusion, the results show a significant effect of algae beta-glucan on viral replication of ASFv in porcine macrophages. This study is a first indication that supplementing Aleta, algae beta-glucan, to pigs, could be a tool to inhibit the pathogenesis of this disease *in vivo*.

## ACKNOWLEDGEMENTS

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## REFERENCES

1. TL-21-19718