Salivary IL-18 and IgA are useful non-invasive stress markers in Pigs.

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Introduction
There has been increasing interest in animal welfare with regards to the handling, housing and feeding of domestic animals and the spread of intensive agriculture and changes in environmental factors may enhance the physiological stress of animals. These stresses negatively affect immune responses and resistance to diseases, quality of meat and reproduction in a variety of animals. Recently, animal welfare guidelines for the handling, housing, and feeding of pigs have been introduced in Japan. However, non-invasive methods and biomarkers to evaluate stress in pigs have not been established, but should be established to eliminate unnecessary stress, improve animal welfare and increase productivity. Saliva sampling has the advantage of being non-invasive, easy to perform, and stress-free in comparison with blood sampling. Therefore, saliva is considered to be a good material for evaluating the stress condition, and salivary biomarkers to evaluate stress have received a great attention. Taken together, in the present study, we investigated the expression of IL-18 and IgA in pig salivary gland and saliva, and examined their dynamics during an acute immobilization stress to evaluate IL-18 and IgA as non-invasive stress markers in pigs.

Materials and Methods
1) IL-18 and IgA expression in pig salivary glands were examined by immunohistochemistry.
2) IL-18 and IgA levels in pig saliva were measured by ELISA.
3) The change of salivary IL-18 and IgA concentration during an acute immobilization stress were measured by ELISA.

Results
We investigated the expression of IL-18 and IgA in porcine salivary gland and their relationship with restraint stress in pigs. IL-18 was expressed in the salivary duct cells and IgA was expressed in plasma cells in salivary gland as confirmed by immunohistochemical staining. IL-18 and IgA was also detected in pig saliva itself by ELISA, and salivary IL-18 and IgA levels were increased by an acute immobilization stress. Moreover, there was a circadian rhythm of IL-18 and IgA over the course of a day. These results are the first evidence of IL-18 and IgA expression related to stress in the pig saliva.

Discussion
Immobilization stress sometimes occurs in pig production. For example, when blood samples are taken from pigs for diagnosis or treatment, pigs must be immobilized using a pig keeper etc. However, the saliva collection carried out in this study was easy, non-invasive, and relatively stress-free. Moreover, anyone can carry out the sampling by this method without a special apparatus or techniques in comparison with blood sampling. Therefore, stress evaluation using saliva samples are straightforward in relation to animal welfare issues. The data described herein provide the first evidence of the usefulness of salivary IL-18 and secretory IgA as a biomarker for an acute stress in pigs. These results also suggest that non-invasive measurement of salivary IL-18 and IgA may be one of a promising method for monitoring stress conditions in pigs.

Reference