

## Evaluation of the acute phase protein haptoglobin as an indicator of herd health in slaughter pigs

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### Abstract

Health is an important aspect of animal welfare, which is difficult to assess at herd level. Clinical examination of individual animals is time-consuming, and most measures of clinical herd health depend significantly on the examiner. Acute phase proteins are produced during inflammatory processes, and could therefore be used as general markers of infection and injury. Our objective was to evaluate whether haptoglobin could be used to monitor the health status of herds at slaughter.

107 groups of 20 slaughter pigs each were examined at two large abattoirs. Clinical signs of tail biting, lameness, injuries, abscesses, pneumonia, pleurisy, hepatitis and gastritis were recorded. The association between elevated haptoglobin concentration in meat juice samples and clinical signs was assessed via multiple logistic regression.

Pigs from groups in which tail biting, lameness, othaematomas, abscesses, pneumonia, pleurisy, pericarditis or condemnations of livers, lungs or carcasses occurred, showed higher levels of haptoglobin than pigs from slaughter groups without any of these clinical signs. In the multiple regression model, only the variables of lameness and tail biting were statistically significant. The haptoglobin test classified 66% of all slaughter groups correctly according to the presence of these clinical signs. These results demonstrate the potential for haptoglobin to be used as a screening test to identify problem herds at slaughter.

**Keywords:** animal health, animal welfare, haptoglobin, immunology, monitoring, pig

### Introduction

Good health is a requirement for a good welfare status of farm animals. Traditionally, health is assessed through examination of clinical symptoms or diagnostic testing for specific pathogens. The results of clinical health examinations depend greatly on the person examining the animals (Baadsgaard & Jorgensen 2003; Petersen *et al* 2004). In addition, short-lasting clinical conditions will often remain undetected, because considerable effort is required to examine the animals repeatedly.

Physiological parameters have been widely used in the assessment of animal welfare. Since most diseases trigger an immune reaction, physiological parameters might also be useful as indicators of impaired health status. Acute phase proteins such as haptoglobin are produced during inflammatory processes, and could therefore be used as general markers of infection and injury. An increase in serum haptoglobin concentration has been demonstrated in several diseases of swine, eg infection with *Actinobacillus pleuropneumoniae* (Hall *et al* 1992), *Streptococcus suis* (Knura-Deszczka *et al* 2002), atrophic rhinitis (Francisco *et al* 1996), and postweaning multisystemic wasting syndrome (Segales *et al* 2004). In addition, epidemiological studies have demonstrated an association with haptoglobin concen-

tration and the prevalence of clinical symptoms, hygiene status, and management of pig herds (Petersen *et al* 2002b; Geers *et al* 2003).

Our objective was to evaluate whether haptoglobin could be used as a screening test to monitor the health status of herds at slaughter.

### Materials and methods

Finishing pigs from 107 randomly selected slaughter batches were examined at two large abattoirs in Switzerland. Only batches consisting of at least 20 pigs from the same finishing farm were included in the study. A clinical examination was performed on arrival at the abattoir, and the prevalence of shortened and bleeding tails, peritarsitis, lameness, lordosis, othaematomas, abscesses and wounds were recorded. From each slaughter batch, 20 animals were inspected at post mortem examination, where lungs were examined for pneumonia and pleurisy, livers for milk spots, and hearts for pericarditis via the modified protocol of Pointon *et al* (1992). Carcasses were examined for muscle and spinal abscesses. Stomachs were separated from the slaughter line and examined for the clinical signs of gastritis using the protocol of Hessing *et al* (1992). The number and type of condemnations per slaughter batch were obtained from slaughterhouse records.

**Table 1** Prevalence of clinical symptoms and haptoglobin concentration in 2192 pigs from 107 slaughter batches.

Clinical symptom	% prevalence in individual animals	% positive slaughter batches	Mean ( $\pm$ SD) haptoglobin concentration (mg ml <sup>-1</sup> )	
			Batches without clinical symptoms	Batches with symptoms
Shortened tail	16	82	0.14 (0.20)	0.15 (0.26)
Bleeding tail	2	34	0.14 (0.22)	0.18 (0.29)
Peritarsitis	15	83	0.16 (0.25)	0.15 (0.25)
Lameness	2	36	0.13 (0.22)	0.18 (0.29)
Lordosis	1	14	0.15 (0.24)	0.18 (0.28)
Othematomas	0.2	7	0.15 (0.24)	0.21 (0.31)
Milk spots	54	100	-	0.15 (0.25)
Pneumonia	11	64	0.13 (0.22)	0.16 (0.26)
Pleurisy	4	44	0.14 (0.22)	0.17 (0.27)
Pericarditis	2	30	0.14 (0.22)	0.18 (0.30)
Abscesses	2	27	0.14 (0.23)	0.19 (0.30)
Gastritis	36	84	0.15 (0.22)	0.15 (0.25)
<i>Condemnations</i>				
Livers	8	40	0.15 (0.26)	0.16 (0.24)
Lungs	1	15	0.15 (0.25)	0.17 (0.24)
Carcass parts	1	23	0.14 (0.22)	0.20 (0.31)

Meat juice from *m. brachiocephalicus* was obtained by freezing and thawing of samples in Salmostore® Containers. Haptoglobin analyses in meat juice samples were carried out with RIDASCREEN® Haptoglobin ELISA (R-Biopharm AG, Darmstadt, Germany), a competitive enzyme immunoassay. With this test, the correlation between haptoglobin concentration measured in serum and meat juice was high ( $r = 0.8$ ), with approximately 10-fold lower concentrations in meat juice than in serum (Hiss *et al* 2003).

Herds were classified according to the presence or absence of each of the recorded clinical signs. The different clinical signs were initially screened for significant association with an increased prevalence of haptoglobin positive animals with the Mann-Whitney *U* test. Variables significantly ( $P < 0.05$ ) associated with increased haptoglobin levels were entered into a multiple logistic regression model. For this analysis, slaughter batches were classified as having an increased haptoglobin level if at least 10% of the animals had a haptoglobin concentration above 0.4 mg ml<sup>-1</sup> in meat juice. With a stepwise backward variable selection procedure, only variables significant at  $P < 0.05$  were retained in the model. Agreement between clinical examination results and elevated haptoglobin levels was evaluated by means of the classification table of the regression model.

## Results

The distribution of the haptoglobin concentrations of all 2192 samples showed a mean value of 0.15 mg ml<sup>-1</sup> and a median of 0.06 mg ml<sup>-1</sup> (25th percentile; 0.03, 75th

percentile; 0.16). Thirty-one percent of all slaughter batches were classified as batches with elevated haptoglobin concentration.

The screening for associations between the presence of clinical signs or lesions and an increase in haptoglobin concentration revealed that pigs from herds classified as positive for bleeding tails, lameness, lordosis, othaematomas, pneumonia, pleurisy, pericarditis, abscesses, liver and lung condemnations and partial condemnations of the carcass showed significantly ( $P < 0.05$ ) higher haptoglobin concentrations than pigs from herds where no clinical signs were observed (Table 1). There was no significant correlation between haptoglobin concentration and *Salmonella* antibodies in meat juice.

The results of the logistic regression analysis are presented in Table 2. If lameness or bleeding tails were observed in the clinical examination before slaughter, slaughter batches were also significantly more likely to have an elevated haptoglobin concentration in at least 10% of the examined pigs. Pneumonia recorded at the slaughter examination showed a weak association with elevated haptoglobin concentration after correcting for the effect of lameness and tail biting ( $OR = 2.1$ ,  $P = 0.13$ ). According to the classification table of the regression model, 66% of the slaughter batches were classified correctly by the haptoglobin test. Of the healthy slaughter batches, 88% were classified correctly, compared to only 47% of the batches with lameness and/or bleeding tails.

**Table 2 Risk factors for a slaughter batch being 'haptoglobin positive' in the final multiple logistic regression model. A slaughter batch was classified as positive if at least 10% of the pigs had a haptoglobin concentration of at least 0.4 mg ml<sup>-1</sup> meat juice.**

Risk factor	Odds ratio	95% confidence interval odds ratio	Wald P-value
Lameness	2.8	1.1 - 6.8	0.02
Bleeding tails	2.7	1.1 - 7.0	0.02

## Discussion

Most of the recorded health parameters were associated with an elevated haptoglobin concentration in meat juice. Because our study reported a correlation between health status and haptoglobin concentrations in meat juice at herd level, it was difficult to compare the results with other studies, where the correlation has been demonstrated on individual animals using concentrations of serum haptoglobin. Nevertheless, the health parameters lameness, pneumonia and tail biting, which have been reported to be associated with elevated haptoglobin at the individual animal level (Lipperheide *et al* 1998; Petersen *et al* 2002a, b), also showed an association at herd level in our study. Despite the associations between health parameters and haptoglobin, a large amount of the variation in haptoglobin concentration among slaughter groups remained unexplained. Acute infections do not always result in clinical signs that can be observed at slaughter. In addition, a poor hygiene standard on a farm could be the reason for constantly elevated haptoglobin concentration in the absence of clinical signs (Geers *et al* 2003). On the other hand, certain clinical signs can remain visible after the acute phase response has terminated and the haptoglobin concentration has decreased. This could be an explanation for the missing association between an increased haptoglobin concentration and antibodies to *Salmonella*, or the prevalence of milk spots in the liver.

## Animal welfare implications

The acute phase protein haptoglobin might be used as a generalised measure for acute infections and chronic inflammatory conditions such as lameness and tail biting. However, the classification of the health status of a herd according to its haptoglobin status was imprecise. Measuring haptoglobin cannot therefore be used to replace a clinical examination, but to supplement other measures of health. Even though the present results are promising, further studies are needed before haptoglobin can be used as a screening test to detect herds with a poor health status.

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