Monitoring the Health of Brown Layers Using the Transmission Colour Value of the Eggs Defined by VIS/NIR Spectroscopy

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Summary

1. Stress or diseases will quickly and possibly strongly influence the deposition of eggshell pigmentation during egg formation. Therefore defining the shell colour of the daily produced eggs by means of fast VIS/NIR transmission spectra could be a representative method for monitoring flock health. 2. Two experimental flocks of laying hens were monitored during a complete laying period: in the first flock stress situations were induced (red mite infestation, heat stress) while in the second flock spontaneous problems (Infectious Bronchitis) occurred. Daily the Transmission Colour Value (TCV) of all daily produced eggs was defined by the ratio of T643 nm, the most important absorbance peak of the pigmentation molecules (mainly protoporphyrin-IX), and T610 nm, transmission at a reference wavelength. Deviating values of the TCV were detected by means of an intelligent CuSum monitoring scheme (control chart). 3. In both monitored flocks problem situations were quickly reflected by an increased TCV-value: more transmission due to reduced pigmentation and hence less light absorbance at the pigment wavelength. Furthermore the TCV-value changed faster than other quality aspects like average egg weight or technical aspects like feed and water

consumption. 4. Measuring the TCV of all produced eggs on a daily basis provides relevant information on the health of a flock of brown layers. This could be used for an early detection of stress situations or emerging diseases, even before important quality and health damage can occur.

Introduction

The eggshell colour of brown eggs is a quality aspect which is mainly important towards the consumer's perception. In general a more homogeneous brown colour is preferred over spotty or pale eggs. This aspect makes the shell colour an important economic quality parameter (Wei and Bitgood, 1989). The brown colour of eggs as perceived by consumers is mainly caused by the pigments protoporphyrin-IX (PPIX) and to a lesser extent by biliverdin-IX. Most of these pigments are located in the cuticle of the egg, yet they can also be found in the shell (Kennedy and Vevers, 1973; Lang and Wells, 1987). Different measurement techniques for defining eggshell colour have been developed. The most known and used is reflectometry described by Wei and Bitgood (1989), and recently Karoui *et al.* (2006a) used front-face fluorescence spectroscopy. The reflectometry measurement method is being used in most selection centres of layer lines, yet despite the economic importance of shell colour few large scale grading machines are equipped with automated colour grading techniques.

Combined results of several publications have suggested that the eggshell colour might be a promising tool for estimating the birds' health. Work by Siefferman *et al.* (2006), Moreno *et al.* (2006) and Martinez de la Puente *et al.* (2007) indicate that eggshell colour and spottiness are indicators for stress and the general health condition of birds. It is indeed known that stress or diseases will quickly and possibly strongly influence the deposition of eggshell pigmentation during egg formation (Whittow, 1999). In commercial laying hens, for instance viral diseases infecting the reproductive tract will cause pale eggs. Different kinds of stress (environmental, social,...) might also induce a higher degree of spottiness (Butcher and Miles, 2003). Mills *et al.* (1987) already suggested the use of reflectometry as a measure of stress in commercial layers. Yet independent of stress or diseases, shell colour of commercial eggs also changes with age. Odabasi *et al.* (2007) stated that older hens lay lighter coloured eggs as a result of the increased egg

size since they found no proportionate change in the quantity of pigment deposited over the shell surface. Furthermore, there are considerable differences in colour between the genetic lines (Solomon, 1987).
The new suggested use of the shell colour might be an interesting parameter for using in a monitoring scheme of the production process of consumption eggs. For this purpose, it would be desirable to asses the colour of every single egg. Candidate technologies and sensors should be fast, suitable for automation and reliable (De Ketelaere *et al.*, 2004). Kemps (2006) found that the observed changes in the spectra of consumption eggs were related to changes in shell pigments and therefore stated that the use of VIS/NIR transmission spectroscopy could prove useful in defining and monitoring shell colour.

In the presented work it was investigated whether the nondestructive method of VIS/NIR transmission spectroscopy could be used for defining the eggshell colour in a fast and objective way. A new parameter for measuring shell colour will be defined. Furthermore, it was examined if this parameter could be used for indirect monitoring of the health status of a flock of brown laying hens.

Materials and methods

In this research eggs of two experimental flocks were used. Based on the daily VIS/NIR transmission spectra of the eggs a new colour parameter was defined and this measure was inserted into an intelligent monitoring scheme.

Layer flocks

Two experimental flocks of laying hens were monitored. The first flock consisted of 72 Isa Brown ® laying hens housed per 2 in a 3 tier battery cage with 36 cages. The cage was located in a climate controlled room. These hens were subjected to three heat stress challenges of a constant (24/24h) temperature of 32°C. The first and the second challenge lasted for 8 days while the third challenge lasted for 4 days. Besides the heat stress challenge on the chickens, there was a serious natural challenge of red mite (*Dermanyssus gallinae*) infestation, especially in the beginning of the laying period.

The second flock consisted of 500 Isa Warren ® laying hens housed in an experimental aviary system. This group of layers was followed during 295 days of lay. Each day all the eggs were collected and the average weight was registered. The TCV was defined on a sample of 120 eggs. Furthermore, a log was kept of all events.

The Transmission Colour Value

To quantify the eggshell colour, a new parameter was developed using the daily VIS/NIR transmission spectra of eggs: the *Transmission Colour Value* (TCV). The raw VIS/NIR transmission spectra were defined using the method presented and discussed by Bamelis *et al.* (2002), Bamelis (2003), Kemps *et al.* (2006) and Kemps (2006). To define the TCV, first the raw transmission spectra were smoothed by a Savitzky-Golay smoothing filter with a bandwidth of 10 nm (Kemps, 2006; Kemps *et al.*, 2006). Next, the TCV was calculated by taking the ratio $TCV = T_{643}/T_{610}$ with T_{643} the relative transmission at 643 nm, most important absorbance peak of PPIX (Kadish *et al.*, 1999), and T_{610} the relative transmission through the egg at the reference wavelength 610 nm (to exclude egg specific effects like shell thickness and egg size). Higher TCV values correspond to a higher transmission rate of light at the PPIX wavelength and hence less pigment is present in the shell.

Intelligent monitoring algorithm

In this investigation we would like to look at the course of the *CV* in time and if changes could be related to possible health problems of the flock. In order to do so, a cusum control chart was constructed to detect deviating TCV values caused by possible problems like stress or disease. The algorithm used for the construction of this control chart was presented by Mertens *et al.* (2008, 2009).

Results and discussion

Figures 1 and 2 show the control chart for the TCV in both flocks. From figure 1 it can be seen that for all applied heat stress challenges the TCV of the eggs increased and resulted in postive alarms indicating that the eggs became paler. As a result of the heat stress the egg weight dropped as well (data not presented). Furthermore, the influence of the red mite infestation causes paler eggs as well (first alarm). The alarm around 150 days of lay was caused by a problem

with the quality of the experimental feed. Figure 2 shows the resulting control chart for the second flock. Three alarms can be discerned. The first alarm (at day of lay 51) was the result of an infectious bronchitis (IB) infection. Pale egg shells are a typical phenomenon of IB. An investigation of the average egg weight revealed that the IB infection also caused a drop in the average egg weight and that the decrease in egg weight started later (day of lay 54) than the alarmed increase of TCV. The second alarm (days of lay 112 to 114) was the result of an increased TCV following an abrupt transition to phase 2 feed. As a result, the feed intake decreased, hen's health was affected which resulted in the production of paler eggs. The alarms at days of lay 195 might have been caused by measurement error, since no clear explanation was found in the log. At the end of the laying period, there is a sudden drop in the TCV meaning that the eggs become darker again. Although this drop was larger than expected hence generating a signal, this is actually a good sign for the layer manager.

Figure 1: Cusum control chart for TCV of the first experimental flock. Upper graph: raw TCV data with mathematical trend detection. Lower graph:Cusum chart.

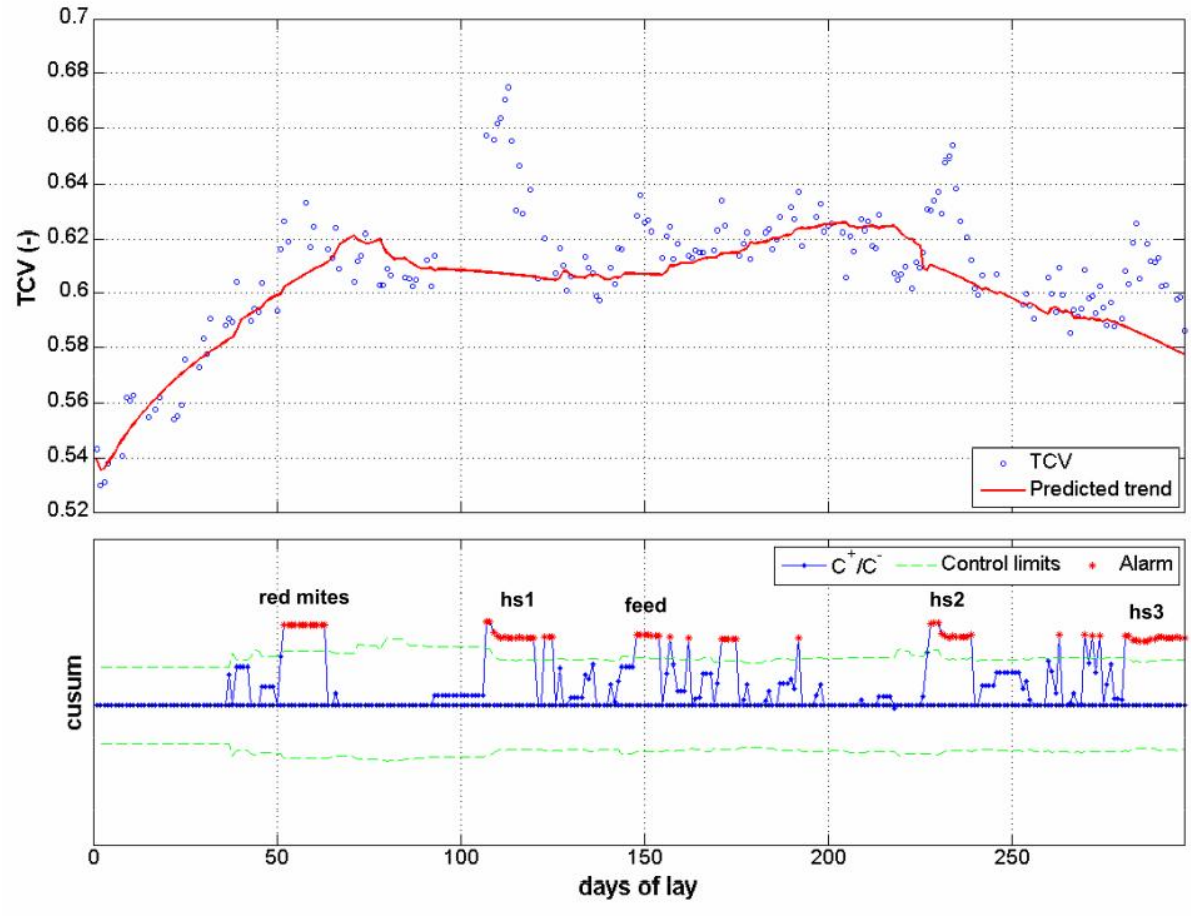
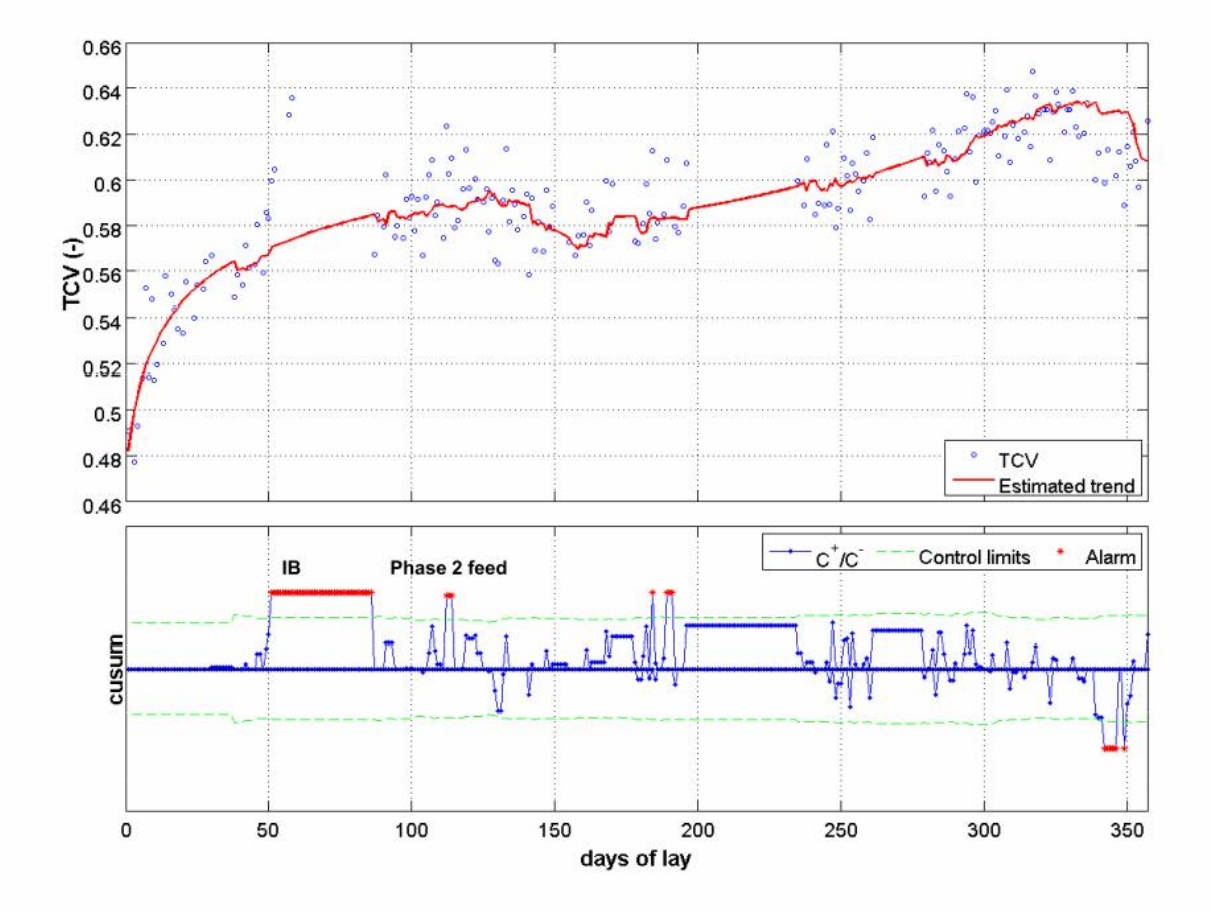


Figure 2: Cusum control chart for TCV of the second experimental flock. Upper graph: raw TCV data with mathematical trend detection. Lower graph:Cusum chart.



From these results it can be concluded that it is useful and practically relevant to use the novel quality measurement, TCV, in a monitoring scheme as an indicator for flock health, as suggested by Kemps (2006), Siefferman *et al.* (2006), Moreno *et al.* (2006) and Martinez-de la Puente *et al.* (2007). Since unhealthy hens will produce fewer eggs, low quality eggs and/or no eggs at all (Whittow, 1999), it is essential to guard the health of the chickens. Environmental stress and diseases are important factors that can quickly affect the health of the birds (Butcher and Miles, 2003). In current common practice the farm manager tries to avoid stress in the henhouse and aims at visually checking his hens as often and as good as possible in order to note emerging health problems before economical damage is caused. The success rate of this visual controlling procedure for disease detection greatly depends on the clinical symptoms of the possible emerging diseases and the management skills of the farmer to interpret the symptoms. With the new availability of the TCV it should become possible to improve the success rate of early disease detection.

Usage of TCV is of double economic significance. First of all TCV determines the shell colour as perceived by the consumer (Wei and Bitgood, 1989), and second the flock health measure is directly related to flock productivity. Guarding optimal flock health by using TCV will improve productivity and even egg quality.

Conclusions

In conclusion, measuring eggshell colour defined by the Transmission Colour Value of all produced eggs on a daily basis provides relevant information on evolution of the health of a flock of brown layers. Therefore TCV could be used as part of a monitoring scheme for an early detection of stress situations and/or emerging diseases, even before important quality and health damage and in the end economic damage can occur.

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