Measuring thirst in broiler chickens

M Sprenger1, C vangestel2 and FAM Tuyttens*†

1 Institute for Agricultural and Fisheries Research (ILVO), Animal Sciences, Animal Husbandry & Welfare, Scheldeweg 68, 9090 Melle, Belgium
2 Department of Biology, Terrestrial Ecology Unit, University of Ghent, KL Ledeganckstraat 35, B-9000 Ghent, Belgium
* Contact for correspondence and requests for reprints: frank.tuyttens@ilvo.vlaanderen.be

Abstract

Freedom from thirst has been long considered of paramount importance for animal welfare, however a feasible and sensitive animal-based indicator to assess thirst, on-farm or at-slaughter, is not available. In this study, voluntary water consumption over time was investigated and validated as a non-invasive behavioural parameter for assessing thirst in broiler chickens. Thirty-two groups of four broilers were used in this study and the effect of three factors on water consumption from a test drinker was investigated: duration of water deprivation (0, 6, 12, or 24 h); familiarity with the test drinker, and age (35 or 37 days). Water consumption was measured after 30, 60, 90 and 120 min following the deprivation period. Water consumption increased with the length of the deprivation period and was greater in birds that had been habituated to the test drinker. The effect of familiarity was smaller for 24-h deprived groups compared to 6- and 12-h deprived groups. When birds were habituated to the drinker, they started to drink sooner than when the drinker was new. These findings illustrate the potential of simple, animal-based measures, such as water consumption over time to assess thirst in chickens and this behavioural test may form the basis of an on-farm test that could be included in integrated animal welfare assessment schemes.

Keywords: animal welfare, broilers, dehydration, thirst, water consumption, welfare assessment

Introduction

Thirst is a subjective perception that motivates an animal to drink. It is a component of the regulatory mechanism that maintains body fluid homeostasis and is activated by several factors related to water loss or dehydration, such as a decrease in volume of body fluids, increase of osmolality of the blood, and activity of specific dipsogenic hormones (McKinley & Johnson 2004). Dehydration can be assessed by physiological parameters related to blood volume and composition, but as physiological mechanisms buffer fluctuations in these blood parameters with the aim of keeping them at constant level for as long as possible, these parameters are more suited for indicating severe dehydration. To assess dehydration at an earlier stage, measures of drinking motivation are probably more suitable. Spontaneous water intake is a promising animal-based indicator of thirst as it has been shown to be related to duration of water deprivation in quail (Takei et al 1988).

Since dehydration is extremely damaging to the production performance of farm animals, it is often assumed that it is unlikely to be a common problem in intensive livestock production. It is presumed that this explains the paucity of research on this subject and the lack of validated measures of thirst, despite it long being recognised that freedom from (chronic) thirst is a crucial aspect of animal welfare (Brambell 1965). For example, out of 72 aspects related to farm animal welfare, Belgian farmers and citizens alike allocated the highest importance to water availability (Vanhonacker et al 2008). However, when asked to score the same aspects according to whether or not it was a problem in current Belgian livestock production systems, water availability was given one of the lowest scores.

In our opinion, whether or not thirst is a concern in current livestock production cannot be properly documented until an animal-based, on-farm indicator of thirst is developed and validated. If assessed at all, freedom from (chronic) thirst is mostly evaluated in existing welfare assessment schemes by a design-based (sometimes also called resource-based) indicator, such as the number of animals per drinker, checking possible leaks in the drinker line and by checking drinker alarms. Although such resource-based measures can often be assessed quickly and have good inter- and intra-observer reliability, they may not be very sensitive and accurate measures of thirst. Animal-based parameters can show the outcome of integrated resource and management factors in the experience of the animal itself (Main et al 2003) and can therefore be a more valid measure of welfare (Keeling & Veissier 2005). Despite the common perception that thirst is not a problem in current livestock production, there might be certain factors causing thirst that cannot be properly assessed by these simple, resource-based indicators. In large groups, in
particular, individual animals that are severely thirsty because they are unable, for some reason, to reach or operate the drinker, may pass unnoticed. In intensive broiler chicken production systems, for example, water is often provided by nipple drinkers (May et al. 1997). Nipple lines are heightened with the age of the chickens, so that they need to stretch their necks, raise their heads and then trigger the nipple to drink water. Small birds might be unable to reach these high nipples. In addition, drinking from high nipples is unnatural for birds and individual chickens differ in the strategy they use to drink from the nipples. Thus, some birds’ water intake may be constrained as a result of inefficient nipple use (SCAHAW 2000). Houldcroft et al. (2008) showed that chickens prefer to drink from lower drinkers compared to higher drinkers, irrespective of the drinker type (bell or nipple). It was suggested that it is easier for broiler chickens to keep a stable position during drinking from a lowered nipple drinker compared to a nipple of standard height. When nipple height was lowered many birds started drinking “as if they were somewhat dehydrated” (Houldcroft et al. 2008). May et al. (1997) observed that chickens probably have difficulty drinking from elevated nipples during high temperatures (while panting), as water consumption from high nipples decreased with increasing temperature. This effect was reduced when nipples were lowered, and reversed with open drinkers. Other factors possibly increasing the risk of thirst include, locomotion problems, high stocking density and disease. Animals with locomotion problems — one of the major welfare issues in broilers — reach the drinkers less easily and possibly suffer from thirst (SCAHAW 2000) or even severe dehydration (Butterworth et al. 2002). High stocking densities and inappropriate design of the (water provisions inside the) barn can exacerbate the difficulties animals have in gaining access to drinkers. Small or very weak animals are at risk of becoming severely water constrained since they are unable to reach the drinkers and are likely to die (SCAHAW 2000).

The aim of this study was to investigate the potential of voluntary water uptake as a non-invasive, animal-based indicator of thirst in broiler chickens. This indicator was validated in an experimental setting by testing whether water uptake increases proportionally to the duration of the deprivation period. As the ultimate goal is for this indicator to be the basis of a method for assessing thirst as part of an on-farm welfare assessment scheme, it ought to be feasible to perform the test during a short farm visit (maximum one day) without requiring much labour or expensive equipment. Moreover, the indicator should be appropriate for comparing farms and therefore require as few restrictions as possible regarding drinking system used on the farm or timing of the farm visit. Mostly, such visits are planned when the chickens reach slaughter-age, but it is logistically very difficult to co-ordinate each visit to each farm so that all chickens have exactly the same age or weight. Therefore, it was also tested whether voluntary water uptake after various water deprivation periods was affected by a two-day age difference of the chickens. To assess whether or not familiarity with the drinker used during testing influenced water consumption after deprivation half of the birds in the experiment were made familiar with the test drinker, while the other were only used to commercial nipples. If water consumption from the test drinker is dependent on whether or not the chickens are familiar with this type of drinker, the test drinker should be designed so that it is equally unfamiliar to all commonly-used drinker types in commercial units.

In this experiment, we aimed to measure levels of thirst by measuring drinking behaviour. Animals were subjected to different levels of water deprivation and we assume they were subjected to different levels of thirst. However, we have to point out that different levels of deprivation might not be linearly correlated to different levels of thirst (dependent on other factors, such as temperature, internal state, feeding level, diet, time since last meal). These factors have been controlled for as much as possible in this experiment.

**Materials and methods**

**Animals and housing**

Test animals were sampled from a population of 200 Ross 308 broiler chickens. From day one until day 33, the animals were housed in single sex groups (two male and two female) of approximately 50 animals in four home pens (2 × 1.5 × 0.5 m; length × width × height) at a density of 16.7 chickens per m². The floor was covered with wood shavings. Food and water were offered ad libitum with water offered in bell drinkers for the first week, and nipple drinkers thereafter (7 chickens per nipple). In the first week, extra heating lamps were provided with ambient temperature kept initially at 29°C, before being decreased by 1°C every three days until 21°C was reached. The lighting schedule was lightdark, 18 h:6 h. In two of the home pens, one female and one male pen, the mobile test drinker (Figure 1) was included in the final week before testing to allow the birds to become accustomed to it.

**Experimental design**

The experimental set-up consisted of 16 wooden pens (1 × 1 × 0.5 m) with wood shavings on the floor. On day 33, a total of 32 animals from the pens that were accustomed to the drinker (‘familiar’) were placed in eight of the experimental pens in groups of four, balanced for sex. Thirty-two chickens from the groups that were unfamiliar with the drinker were placed in the other eight pens. Groups of four chickens were weighed and marked before transfer to the experimental pens. After a two-day habituation period, the groups in the experimental pens were subjected to a water deprivation period of 0, 6, 12 or 24 h using a balanced design. Temperature and ventilation were kept equal to the home pens. Light was provided at a constant level of 30-40 lux. Food was offered ad libitum until two hours prior to testing to reduce differences in drinking motivation due to a different feeding level. Water was offered in drink nipples (two per pen). From the onset of the deprivation period,
these nipples were closed with plastic tubes. All deprivation periods ended between 1200 and 1500h at day 35, in order to reduce the effect of time of day on water consumption. After the deprivation period, water was offered to all groups in a mobile test drinker (Figure 1) for two hours. The test drinker consisted of a wooden framework measuring 31 × 20 × 22 cm, which contained a Tupperware box filled with 1 L of water. After 30, 60, 90 and 120 min, the box with the remaining water was weighed, refilled up to 1 L and placed back in the pen. After 120 min, all chickens were placed back in the home pens. Immediately thereafter, a total of 32 new chickens from the familiar home pens and 32 new chickens from the unfamiliar home pens were weighed in groups of four, balanced for sex, and placed in the experimental pens. The test was performed with these chickens on day 37, using the same procedure as described previously.

The experiment consisted of two replicates. In each replicate, a total of 64 35-day-old birds and 64 37-day-old birds were tested, both divided into 16 groups of 4 chickens. The first replicate was performed at the end of February 2008, the second at the end of May 2008. For each treatment (deprivation period × familiarity with drinker × age), 4 groups of 4 chickens were used in total. This experiment was approved by the Ethical Commission of the Institute of Agricultural and Fisheries Research (ILVO).

Statistical analysis
A group of four chickens was the experimental unit used for analysis. Water consumption was measured at group level. Group means expressed in ml per chicken were used for the analysis. The differences in water consumption between treatments were analysed using a general linear mixed model. Deprivation period, familiarity with the test drinker, age and interactions were included as fixed variables and replicate was included as a random factor to account for dependency between repetitions. To test whether cumulative water consumption between 30 and 120 min differed between treatments, time was included as a continuous variable and the residual correlation was modelled using an autoregressive covariance structure.

To assess the point of time where the mean response between different treatments (deprivation period) differed significantly from each other within ‘familiar’ and ‘unfamiliar’ groups, post hoc comparisons of treatment means were inspected at 30, 60, 90 and 120 min. To test whether the effect of age was due to increased weight of the chickens, water consumption was also expressed in ml kg⁻¹ and data were re-analysed using the same model. Weights on test days were estimated using the difference in average weight on day 32 and 35. Daily growth was 71.9 and 81.9 g day⁻¹ for replicates 1 and 2, respectively. Ad libitum growth can be described as linear between day 32 and 42 (Cangar et al 2006). Therefore, growth between day 35 and 37 can be predicted by the growth rate between day 32 and 35.

When significant effects or interactions were present, exact location of the effects were determined with post hoc contrast statements (t-tests) and Bonferroni corrections for multiple tests were applied. Fixed effects were tested using traditional F-tests and degrees of freedom were estimated using Satterthwaite formulas (Littell et al 1996). All statistical procedures were performed using SAS 9.1.

Results
Average water consumption
Figure 2 shows the effects of deprivation period and familiarity with the test drinker on average water intake during 120 min. There was a significant interaction of familiarity by deprivation period ($F_{3,55} = 6.68, P < 0.001$). The effect of familiarity was significantly smaller in the 24-h deprivation treatment compared to the 12- ($F_{1,55} = 15.81, P < 0.001$) and 6-h deprivation periods ($F_{1,55} = 8.51, P < 0.01$). In the 0–12-h deprivation treatments, chickens that were familiar with the drinker consumed more than those unfamiliar. In the 24-h deprived chickens, the effect of familiarity was not significant.

In general, chickens consumed more water when deprived for a longer period. Within the ‘familiar’ treatment, water intake differed significantly between all deprivation periods. For the ‘unfamiliar’ treatment, water intake also differed significantly between deprivation treatments with the exception of the 6-h deprivation treatment which did not differ significantly from the 0- or 12-h deprivation treatments (Table 1).
Figure 3(a) shows the difference in responses of 35- and 37-day-old chickens. There was a significant interaction effect of age by deprivation period ($F_{3, 55.6} = 7.14$, $P < 0.001$). With the exception of the 6-h deprived groups, 35-day-old chickens drank less water than 37-day-old ones. This age difference, however, was statistically significant only for the 12-h deprived groups ($t_{55.6} = 3.7$, $P < 0.01$; Figure 3a).

To test whether these age effects were related to increased bodyweight at day 37, water consumption was expressed in ml kg$^{-1}$ and the data were re-analysed. The differences between 35- and 37-day-old chickens were smaller (see Figure 3b), but the reversed effect of age after 6 h of deprivation compared to the other groups remained (age × deprivation period interaction: $F_{3,54.6} = 5.41$, $P < 0.01$). Within deprivation period, effect of age was never significant.

Cumulative water consumption over time

Figures 4(a) and (b) show the average cumulative water consumption over time and the effects of deprivation period and familiarity with the drinker. Most water was consumed within the first 30 min after deprivation, with the exception of birds that were not familiar with the test drinker and had been deprived for a maximum of 12 h. From 30 to 120 min after deprivation, there was an interaction of time by deprivation period ($F_{3,239} = 6.29$, $P < 0.001$) and of time by familiarity ($F_{1,239} = 5.74$, $P < 0.05$). Post hoc tests showed that cumulative water consumption increased significantly more for 24- and 12-h deprived groups compared to 0-h deprived groups ($t_{239} = 3.52$, $P > 0.05$, and $t_{239} = 3.62$, $P > 0.05$, respectively).

Water consumption of unfamiliar groups increased significantly more over time than that of familiar groups. For chickens familiar with the test drinker, water consumption after 30 min already differed significantly between all deprivation treatments (all $P < 0.05$), with the exception of between the 6- and 12-h deprived groups. The difference between the latter treatments was significant from 90 min onwards ($P < 0.01$).

For chickens unfamiliar with the test drinker, water consumption of 24-h deprived groups differed significantly from the other groups after only 30 min of testing ($P < 0.0001$). The difference in water consumption between control groups and 12-h deprived groups approached significance after 60 min ($P = 0.05$) and was highly significant after 90 min of testing.

Table 1  Mean (± SEM) water consumption (ml chicken$^{-1}$) of familiar and unfamiliar groups after various deprivation periods.

<table>
<thead>
<tr>
<th>Deprivation period (h)</th>
<th>Unfamiliar</th>
<th>Familiar</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>1.1 (± 6.3)$^a$</td>
<td>37.5 (± 6.3)$^a$</td>
</tr>
<tr>
<td>6</td>
<td>18.3 (± 6.3)$^a$</td>
<td>80.6 (± 6.3)$^b$</td>
</tr>
<tr>
<td>12</td>
<td>35.8 (± 6.3)$^a$</td>
<td>111.6 (± 6.3)$^c$</td>
</tr>
<tr>
<td>24</td>
<td>122.2 (± 6.3)$^a$</td>
<td>147.8 (± 6.3)$^d$</td>
</tr>
</tbody>
</table>

$^a,b,c,d$ Means within familiarity with no common superscript differ significantly ($P < 0.05$).
Voluntary water intake appears to be a valid indicator of thirst. As hypothesised, it becomes greater with increasing periods of water deprivation (which, in turn, presumably reflect increasing levels of thirst). Three potentially confounding factors that ought either to be standardised, taken into account or corrected for, have been highlighted in this experiment. These are whether or not the chickens were familiar with the test drinker, the duration that water intake is recorded, and the age of the chickens.

Chickens familiar with the test drinker drank more than those unfamiliar with it, unless they had been severely deprived (24-h treatment). Perhaps the shorter the deprivation period the greater the fear of the novelty of the drinker, relative to the motivation to drink. It has been reported that high motivational states such as hunger and thirst can decrease fear levels. Savory et al (1993a) and Hocking et al.

(Figure 3)

Mean (± SEM) water consumption during 120 min in (a) ml chicken⁻¹ and (b) ml kg⁻¹ after 0, 6, 12, and 24 h of water deprivation for 35- and 37-day-old chickens, (n = 8). Significant effects of age within deprivation period are indicated as * P < 0.05. Data for familiar and unfamiliar groups were averaged.

(P < 0.01). Water consumption of 6-h deprived groups never differed significantly from control and 12-h deprived groups.

**Discussion**

Voluntary water intake appears to be a valid indicator of thirst. As hypothesised, it becomes greater with increasing periods of water deprivation (which, in turn, presumably reflect increasing levels of thirst). Three potentially confounding factors that ought either to be standardised, taken into account or corrected for, have been highlighted in this experiment. These are whether or not the chickens were familiar with the test drinker, the duration that water intake is recorded, and the age of the chickens.

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Figure 4

Mean (± SEM) cumulative water consumption over time after 0, 6, 12 and 24 h of deprivation, for groups (a) familiar and (b) unfamiliar with the test drinker. Data of 35- and 37-day-old chickens were averaged.

(1996) found a reduced level of arousal and fear in food-restricted broiler breeders. Severely water-deprived birds may also have been more active in exploring the novel object or in actively searching for drink-related objects, as water deprivation is known to increase motor activity in novel environments (Bolles 1975; Fowler 1965 cited in Maren & Fanselow 1998). The water in the test drinker may not have been easy for the birds to detect. Water level was at approximate breast height and the birds may not have noticed the water unless they stood close to the drinker. An increase in activity and exploratory behaviour is likely to have resulted in a quicker recognition of the novel drinkers.
Also, high physical needs will draw the attention towards needs-specific stimuli, a state which is called perceptual readiness (Aarts et al. 2001). In this experiment, video recordings during the test were taken and differences in exploration and general activity will be reported elsewhere. Chickens familiar with the drinker drank most in the first 30 min of the test. Thereafter, total water consumption did not increase by much. Chickens unfamiliar with the drinker started drinking later and kept on drinking throughout the entire 2-h test period. As already suggested, ‘unfamiliar’ chickens probably needed more time to explore the drinker and to detect the water inside. The practical implications are that a shorter testing time (30 vs 90 min) is needed to distinguish 24- and 12-h deprived groups from 0-h deprived groups, when a familiar instead of an unfamiliar drinker is used. Slightly thirsty chickens (6-h deprivation) could be detected with the familiar drinker recording water consumption after 90 min, whereas with the unfamiliar drinker they could not be differentiated from non-deprived birds.

When using this test to compare thirst levels between flocks, water consumption should preferably be expressed in ml kg⁻¹ in order to reduce the effects of bodyweight on water intake. Ideally, birds should be weighed shortly before testing if this does not disturb normal drinking behaviour. If water consumption is expressed in ml chicken⁻¹ then bodyweight (and/or age) ought to be standardised or adjusted for as much as possible. If this behavioural test of thirst is to be included in an on-farm welfare assessment scheme, it should not require a lot of labour, equipment or time. Moreover, the outcome of the test should be independent of drinking system used on the farm and the timing of the visit.

A feasible way of using this indicator, on-farm, is to separate small groups of chickens on several locations in the house, offer them water and measure water consumption after approximately 1–1.5 h, dependent on the familiarity of the drinker. Using a familiar drinker reduces recording time needed, but may imply that only chickens on that particular drinker system can be compared with each other. If, however, it is important to compare thirst across farms using different types of drinkers, we advise the use of a test drinker that is different from all currently-used drinker systems. Recording time will have to be longer to maintain equal sensitivity. The use of an unfamiliar drinker may also reduce the risk of false positives. In this experiment, 0-h deprived chickens that were familiar with the drinker consumed more water than 0-h deprived birds unfamiliar with the drinker. An explanation for this could be that despite the fact that they were not deprived, they drank a fair amount of water from the open-test drinker as chickens prefer an open drinker compared to high nipple drinkers (Houldcroft et al. 2008). The unfamiliarity or fear of a novel drinker more effectively prevents non-thirsty animals from drinking. Indeed, in the present experiment, the animals needed to be seriously deprived (12 h) in order to be detected with an unfamiliar drinker type.

One limitation of this behavioural thirst test is that severely lame or sick animals will not be able to walk to a test drinker and will probably be missed. Thirst could be assessed ‘individually’ in these chickens (regardless of how time consuming) by bringing the water to them or by measuring levels of dehydration, using physiological parameters such as plasma osmolality (Butterworth et al. 2002). It is important, nevertheless, to be able to assess thirst in healthy animals using a valid indicator. Previous research has shown that healthy birds can also have trouble drinking from high nipples, especially during high temperatures, and may suffer from chronic thirst (May et al. 1997; Houldcroft et al. 2008). This cannot be assessed properly by design-based indicators that are used in present assessment schemes. Another aspect in need of further investigation is the reaction of birds to chronic water restriction instead of acute water deprivation. Savory et al. (1993b) studied differences in eating motivation between chronically restricted-broiler breeders and 72-h deprived, ad libidum-fed broiler breeders. Restricted-fed broilers were 1.9–3.6 times more motivated to eat than 72-h deprived, ad libidum-fed birds. Commercial units may also be subjected to a degree of chronic water restriction due to inefficient use of nipples or troubles with drinking from them (SCAHAW 2000; Houldcroft et al. 2008).

**Conclusion and animal welfare implications**

The behavioural thirst test developed and validated in this experimental setting, may form the basis of an animal-based indicator of thirst that could be included in on-farm, integrated animal welfare assessment schemes. As the validity and sensitivity of currently-used, resource-based indicators of thirst are questionable, this could be an important addition given the fact that freedom from thirst is being considered as one of the most important elements of animal welfare.

**Acknowledgements**

This research was supported financially by the Federal Governmental Agency of Health, Food Chain Safety and Environment, Belgium. The useful discussions with the members of the WELBROIL steering group and user group were very much appreciated, as was the input of Sue Haslam (Department of Clinical Veterinary Science, University of Bristol, UK) and Linda Keeling (Department of Animal Environment and Health, Swedish University of Agricultural Sciences, Sweden). The authors greatly acknowledge the technical assistants and animal caretakers of the Institute for Agricultural and Fisheries Research (ILVO), Animal Sciences, for their help during the experiments.

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