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Basic nutritional investigation

Free-range farming: A natural alternative to produce vitamin D-enriched eggs

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ABSTRACT

Objective: Food-based strategies need to be developed to improve the vitamin D status of individuals. Recent studies identified ultraviolet B irradiation as an efficient method to enrich mushrooms and eggs with vitamin D. The aim of this study was to determine whether free-range farming of hens could provide a valuable method to produce vitamin D-enriched eggs.
Methods: Laying hens were randomly assigned to three groups of 33 to 34 animals each, and were kept either indoors (indoor group), outdoors (outdoor group), or with an indoor/outdoor option (indoor/outdoor group) over 4 wk.
Results: The study shows that the vitamin D_3 content of egg yolk was three- to fourfold higher in the groups that were exposed to sunlight (outdoor and indoor/outdoor groups) compared with the indoor group ($P < 0.001$). Egg yolk from the outdoor group revealed the highest vitamin D_3 content, which averaged 14.3 μg/100 g dry matter (DM), followed by that from the indoor/outdoor group (11.3 μg/100 g DM). Yolk from indoor eggs contained only 3.8 μg vitamin D/100 g DM. The 25-hydroxyvitamin D (25[OH]D_3) content of egg yolk was also influenced by sunlight exposure, although less pronounced than the vitamin D content ($P < 0.05$). In contrast, free-range eggs randomly acquired from supermarkets had relatively low vitamin D contents.
Conclusion: Free-range farming offers an efficient alternative to fortify eggs with vitamin D, provided that farming conditions are sufficiently attractive for hens to range outside.

Introduction

The main source of vitamin D in humans is the conversion of 7-dehydrocholesterol (7-DHC) in the epidermis to previtamin D_3, which isomerizes to form vitamin D_3. Seasonal and local variations of ultraviolet (UV) radiation, increasing indoor activities, but also public awareness of the hazardous effects of sun exposure that lead to sun avoidance strategies or the use of sunscreens limit the endogenous synthesis of vitamin D_3. As a consequence, the need for vitamin D from food sources is on the rise.

In contrast to vitamin D from sunlight, dietary vitamin D is available during any season and offers a reliable source of the vitamin for individuals who do not have access to sunlight exposure. Oily fish is the most important dietary source of vitamin D [1], whereas most other foods from animal origin contain very low amounts and do not significantly contribute to an improvement of vitamin D status. A recent study was able to ascertain UVB exposure of hens as a highly effective approach to increase the vitamin D content in eggs [2]. Data from that study showed that UVB radiation increased the vitamin D content much stronger than feeding the maximum permissible dosages of dietary vitamin D_3.

Currently, three farming systems for laying hens are frequently implemented in practice: Free-range farming, floor management, and small-group systems, whereby mixed forms of these housing systems are also common. In free-range systems, hens have the possibility to go outside and become exposed to natural sunlight, whereas floor management and small-group systems operate with UV-free light regimens. Therefore, we hypothesized that eggs from free-range hens might contain higher vitamin D contents than barn eggs. The present study aimed to investigate the vitamin D content of eggs in response to different housing conditions.

GIS, JK, and AS conceived and designed the experiment. JK, AS, and HK performed the experiment. JK and FH analyzed the data. JK, GIS, and AS wrote the article.

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Materials and methods

Animals and treatment

The study was conducted with 101 25-wk-old laying hens (Lohmann silver). The hens were randomly assigned to three groups. Group 1 (n = 34, indoor group) was kept indoors without any exposure to natural sun light over the entire study period; group 2 (n = 34, outdoor group) was placed outside for 9 h/d from 0700 h to 1600 h. Group 3 (n = 33, indoor/outdoor group) could choose freely between inside and outside (outside option from 0700 h to 1600 h). The free-range area was partly covered by UV-permeable acrylic glass to provide rain protection. All hens received a standard feed with 2500 IU vitamin D/kg (Deuka, Könnern, Germany). Feed and water were offered ad libitum. The indoor group was fed inside the barn; the outdoor and indoor/outdoor groups were provided with feed and water, both indoors and outdoors.

Study protocol

The study was performed from August 22 to September 19, 2012 at the experimental facility of the Martin-Luther-University Halle-Wittenberg (51° latitude). Before the experiment, all hens were housed in a barn system for 2 wk. The experimental period lasted 4 wk. Eggs were collected at the beginning of the study (basal) and after the fourth week (final) of the experiment. To study the choice behavior of the indoor/outdoor group, hens were monitored on three randomly selected days during the experimental period.

Additionally, free-range and barn eggs were collected randomly from supermarkets and analyzed for their vitamin D content. To ensure that the commercial free-range eggs were produced under similar weather and sunlight conditions as the experimental eggs, we acquired eggs from two different hen farms located near the experimental facility. The eggs were collected in September and January to determine out seasonal influence on the vitamin D.

Sun light exposure conditions and sunshine duration

The outdoor enclosure conditions (location, incidence of sun light) were arranged to ensure an optimal UVB exposure from 1000 h to 1600 h. The UV permeability of the coverage was >90% (manufacturer's specification). Sunshine duration, defined as the time during which the solar irradiance exceeds 120 W/m^2, was recorded daily.

Analysis of eggshell thickness and stability

Eggshell stability was determined by an electronically controlled breaking strength tester (Messtechnik Gutsch, Nauendorf, Germany). Eggshell thickness was measured using a micrometer screw with an accuracy of 10.0 μm. Shell fragments were collected from the equatorial area and mechanically purified from organic materials.

Analysis of vitamin D_3, 25-hydroxyvitamin D_3, and 7-DHC contents in egg yolk

Pooled egg yolk samples (of three yolks each) of each group were used for the analysis of vitamin D metabolites. Commercial eggs were analyzed individually. Vitamin D_3, 25-hydroxyvitamin D_3 (25[OH]D_3) and 7-DHC were determined by liquid chromatography-tandem mass spectrometry, as described previously [2].

Statistical analysis

Statistical analyzes were performed using SPSS Statistics version 20 (IBM, Armonk, NY, USA). Differences in the vitamin D content of eggs from basal to final were analyzed by a paired *t* test. Differences between the three groups of hens were analyzed by one-way analysis of variances (ANOVA). Vitamin D content of the purchased eggs was compared by two-way ANOVA with the factors "farming" and "season," and the interaction between these two factors. When two-way ANOVA revealed significance for one of these factors, a post hoc comparison was performed. In case of variance homogeneity, means of the four groups were compared by Tukey's test, or in case of variance heterogeneity, by Games-Howell test. Means were considered significantly different at $P < 0.05$.

Results

Sunshine duration and behavior of hens

The average weekly daily sunshine durations were 6.6, 5.5, 5.9, and 7 h, respectively (Fig. 1). Observations from three randomly selected days showed that hens from the indoor/outdoor group spent most of their time outside the barn. Thus, we assumed that UVB exposure of this group was comparable to that of the outdoor group.

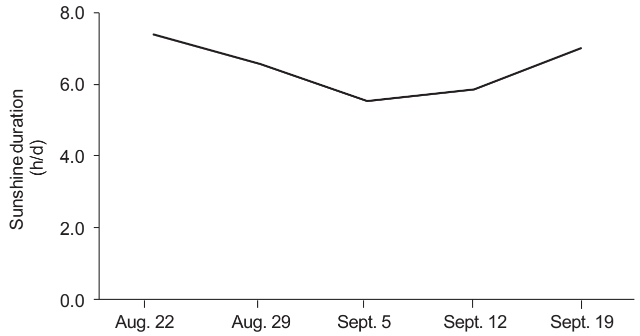

Fig. 1. Sunshine duration at the trial site (data averaged to weekly sunshine duration).

Egg weight and eggshell quality

Egg weight, eggshell thickness, and stability did not differ between the three groups of hens at basal (Table 1). In all groups of hens, egg weights significantly increased from basal to final ($P < 0.05$). The housing systems did not reveal any effect on final egg weights and eggshell stability (Table 1). The eggshell thickness increased from basal to final in the outdoor and indoor/outdoor groups ($P < 0.05$), but not in the indoor group (Table 1). Final eggshell thickness was highest in the outdoor group, lowest in the indoor group, and showed intermediate values in the indoor/outdoor group.

Vitamin D content of egg yolk in response to the housing system

Analysis of vitamin D metabolites in pooled egg yolks from the treatment groups was performed at basal and after 4 wk of the experiment (Fig. 2). Egg yolk data did not reveal any differences in basal contents of vitamin D_3, 25(OH)D_3, and 7-DHC between the three groups of hens. At the end of the experiment, the vitamin D_3 content of egg yolk was three- to fourfold higher in the sunlight-exposed groups (outdoor group and indoor/outdoor group) than in the indoor group ($P < 0.001$; Fig. 2A). Egg yolks from the outdoor group revealed the highest vitamin D_3 content, which averaged 14.3 μg/100 g dry matter (DM), followed by yolk from the indoor/outdoor group with 11.3 μg/100 g DM. Egg yolk

Table 1
Egg weights and eggshell quality in response to the housing system

	Indoor group	Outdoor group	Indoor/outdoor group
Egg weight (g)			
Basal	54.3 ± 3.1	52.7 ± 2.4	52.9 ± 1.9
Final	62.2 ± 4.8*	59.8 ± 1.9*	59.3 ± 2.6*
Eggshell thickness (μm)			
Basal	352 ± 17	337 ± 20	333 ± 14
Final	362 ± 14^{b}	380 ± 16^{*,a}	368 ± 13^{*,a,b}
Eggshell stability (N)			
Basal	39.3 ± 7.7	36.8 ± 5.1	38.1 ± 4.4
Final	46.5 ± 7.2	49.6 ± 7.5*	49.3 ± 9.1*

Values are presented as mean ± SD (n = 7 to 11)
Means not sharing the same superscripts (a,b) are significantly different ($P < 0.05$, Tukey's test).
* Means are significantly different compared with basal ($P < 0.05$, paired *t* test).

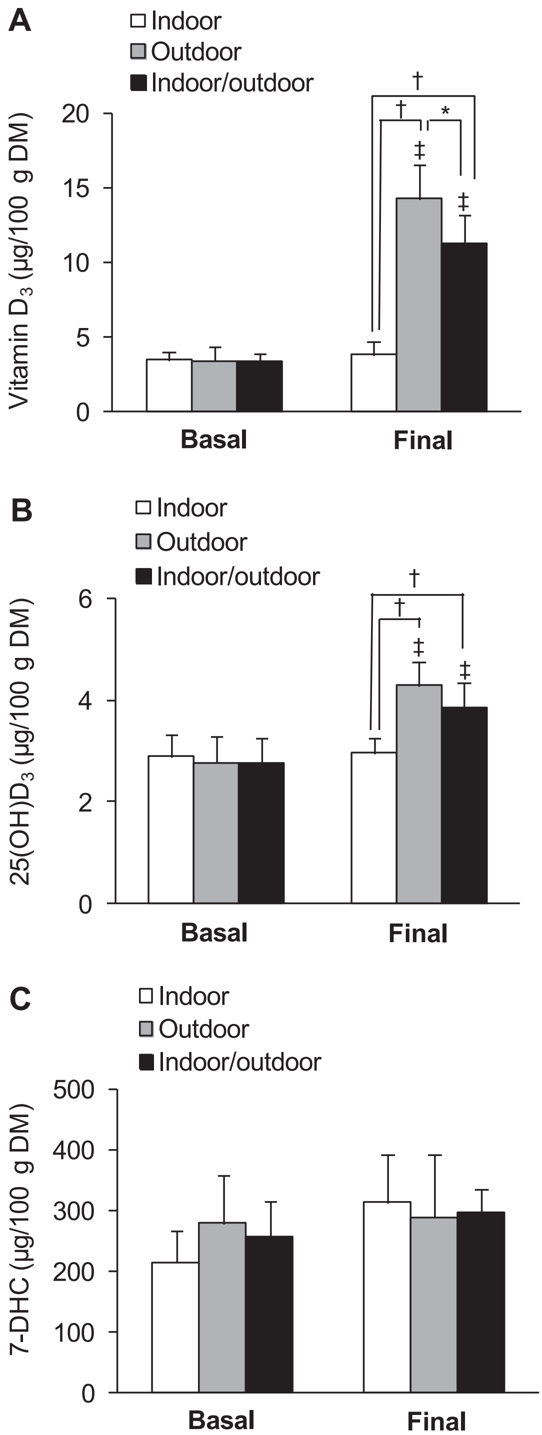

Fig. 2. Content of (A) vitamin D_3, (B) $25(OH)D_3$, and (C) 7-dehydrocholesterol (7-DHC) in dry matter (DM) of pooled egg yolks from three hens each (n = 7 to 11, resulting from inconsistent numbers of laid eggs). Values are expressed as mean ± SD. Data were analyzed by one-way ANOVA using multiple comparison procedures to analyze means of the groups (Tukey's test or Games-Howell test). *$P < 0.05$, †$P < 0.001$, ‡significantly different from basal ($P < 0.05$, paired t test).

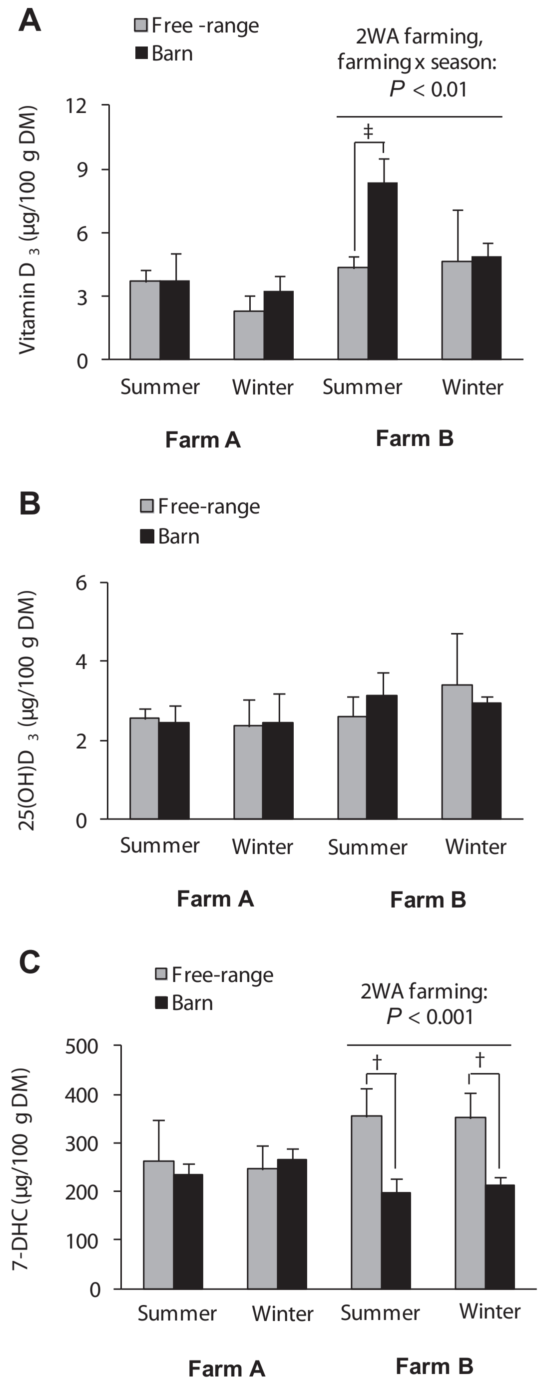

Fig. 3. Content of (A) vitamin D_3, (B) $25(OH)D_3$, and (C) 7-dehydrocholesterol (7-DHC) in dry matter (DM) of yolks from commercial eggs (n = 6 per set). Values are expressed as mean ± SD. Data were analyzed by two-way ANOVA (2 WA) with the factors "farming" and "season" and the interaction between these factors, respectively. Tukey's test and Games-Howell test were applied to compare means: †$P < 0.001$, ‡$P < 0.01$.

of the indoor group revealed the lowest vitamin D levels. The content of $25(OH)D_3$ in egg yolk also increased in response to sunlight exposure, although less pronounced than the vitamin D_3 content ($P < 0.05$; Fig. 2B). The 7-DHC contents of eggs were not influenced by the housing system (Fig. 2C).

Vitamin D content of commercial eggs

Vitamin D data did not show a consistent picture of free-range and barn eggs. Regardless of the housing system and the season, the vitamin D_3 content of commercial eggs from farm A was low and ranged between 2 and 4 µg/100 g DM (Fig. 3A). Also, free-range eggs from farm B were not characterized by higher vitamin D content compared with barn eggs (Fig. 3A). The yolk content of 25(OH)D_3 did not differ between the commercial eggs analyzed (Fig. 3B). The 7-DHC content was not different between eggs from farm A, but was higher in free-range eggs than in barn eggs from farm B ($P < 0.05$; Fig. 3C).

Discussion

UV radiation is essential for endogenous vitamin D synthesis in humans and animals [3]. Recent data show that eggs from hens exposed for 3 h to artificial UVB radiation contained sixfold higher amounts of vitamin D_3 than eggs from non-exposed animals [2]. Here, we show that free-range versus barn-rearing systems for laying hens could offer an appropriate and cheap alternative to fortify eggs with vitamin D. Eggs from free-range hens that spent 9 h/d outdoors contained on average fourfold higher amounts of vitamin D_3 and 45% higher amounts of 25(OH) D_3 than barn eggs. However, we need to acknowledge that the findings are specific to the season (August 22 to September 19) and latitude (51°, Central Germany) under which the experiment was conducted. It is to be expected that the efficacy of free-range systems to fortify animal-based foods such as eggs, largely depend on weather, season, and the latitude of the area where the animals are housed [4,5]. We further found that eggs from chickens with the inside/outside option contained markedly more vitamin D_3 than the indoor group, although the vitamin D_3 contents of the outdoor group were not fully reached. Although such eggs may not cover the daily vitamin D recommendation, the idea of vitamin D bio-addition by sunlight could be used for other animal housing systems to improve the vitamin D content of meat or milk. The fact that the vitamin D content of milk could be increased by sunlight exposure of cows [6,7] indicates the effectiveness of the free-range system in production of other vitamin D-enriched food.

Owing to the efficient bio-addition of eggs with vitamin D by natural sunlight exposure of hens, we hypothesized that commercial free-range eggs produced in summer should contain higher amounts of vitamin D than barn eggs or free-range eggs from winter. However, the vitamin D analysis did not support our hypothesis. After visiting the farms, we assume that the free-range housing conditions are crucial for the choice behavior of the chickens. An important aspect that could influence the movement of chickens in and out of the hen house is the offered space for feeding [8]. We observed that chickens in a free-range husbandry avoided moving outside if they received their feed exclusively inside the barn. Besides the feeding points, habitat preferences of free-range chickens are also important. One study observed that the number of chickens found to be ranging outside was positively correlated with the amount of tree cover the range area contained [9]. Thus, the efficiency of sunlight exposure to produce vitamin D-enriched eggs also depends on the habitat conditions.

Another interesting finding from this study was an increase in the stability of eggshells from basal to final in the sunlight-exposed groups, but not in the indoor group. Eggshell quality could be affected by diverse factors [10]. Most of the studies did not found any effect of the housing system on eggshell quality [11–13], but it is still a subject of controversy whether UVB radiation could improve the eggshell quality.

Conclusion

Data show that the free-range farming system could be an appropriate method to fortify eggs with vitamin D, provided free-range farming conditions are sufficiently attractive for the hens to range outside.

References

[1] Mattila PH, Piironen VI, Uusi-Rauva EJ, Koivistoinen PE. Contents of cholecalciferol, ergocalciferol, and their 25-hydroxylated metabolites in milk products and raw meat and liver as determined by HPLC. J Agric Food Chem 1995;43:2394–9.
[2] Schutkowski A, Krämer J, Kluge H, Hirche F, Krombholz A, Theumer T, et al. UVB exposure of farm animals: Study on a food-based strategy to bridge the gap between current vitamin D intakes and dietary targets. PLoS ONE 2013;8:e69418.
[3] Webb AR. Who, what, where and when-influences on cutaneous vitamin D synthesis. Prog Biophys Mol Biol 2006;92:17–25.
[4] Webb AR, Kline L, Holick MF. Influence of season and latitude on the cutaneous synthesis of vitamin D3: Exposure to winter sunlight in Boston and Edmonton will not promote vitamin D3 synthesis in human skin. J Clin Endocrinol Metab 1988;67:373–8.
[5] Sherman SS, Hollis BW, Tobin JD. Vitamin D status and related parameters in a healthy population: The effects of age, sex, and season. J Clin Endocrinol Metab 1990;71:405–13.
[6] Kurmann A, Indyk H. The endogenous vitamin D content of bovine milk: Influence of season. Food Chem 1994;50:75–81.
[7] Hymoller L, Jensen SK. Vitamin D(3) synthesis in the entire skin surface of dairy cows despite hair coverage. J Dairy Sci 2010;93:2025–9.
[8] Bubier NE. Movement of flocks of laying hens in and out of the hen house in four free range systems. Br Poult Sci 1998;39:5–6.
[9] Stamp Dawkins M, Cook PA, Whittingham MJ, Mansell KA, Harper AE. What makes free-range broiler chickens range? In situ measurement of habitat preference. Animal Behaviour 2002;66:151–60.
[10] Roberts JR. Factors affecting egg internal quality and egg shell quality in laying hens. J Poult Sci 2004;41:161–77.
[11] Minelli G, Sirri F, Folegatti E, Meluzzi A, Franchini A. Egg quality traits of laying hens reared in organic and conventional systems. Ital J Anim Sci 2007;6:728–30.
[12] Van Den Brand H, Parmentier HK, Kemp B. Effects of housing system (outdoor vs cages) and age of laying hens on egg characteristics. Br Poult Sci 2004;45:745–52.
[13] Mugnai C, dal Bosco A, Castellini C. Effect of rearing system and season on the performance and egg characteristics of ancona laying hens. Ital J Anim Sci 2009;8:175–88.