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Transfer of α-tocopherol stereoisomers in laying hens from feed to egg yolk and tissues of hatched chicken

Transfer von α-Tocopherol-Isomeren vom Legehennen-Futter zum Dotter und zu den Geweben von geschlüpften Küken

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Introduction

Vitamin E status in egg yolk is closely related to dietary level and sources of vitamin E. Commercially available vitamin E products are stable esters of α-tocopherol with various acids, e.g. acetic acid, forming α-tocopheryl acetate. Two major sources are largely commercially available, i.e. synthetic vitamin E in the form of all-rac-α-tocopheryl acetate and natural source vitamin E in the form of RRR-α-tocopheryl acetate. Different from any other vitamin, the chemically synthesized form differs from the naturally occurring form in terms of bio-availability based on retention of α-tocopherol in plasma and tissues. This is due to different optical stereoisomerism in the two products. All-rac-α-tocopheryl acetate is an equimolar mixture of 8 stereoisomers. Vitamin E derived from natural ingredients contains 100% of the RRR-α-tocopherol stereoisomer. Thus synthetic vitamin E contains only 12.5% of the naturally occurring form of vitamin E. The other seven isomers have different stereo-configurations and lower bio-availability. In general, 2R forms (RRR, RRS, RSR, RSS, i.e. R-configuration at position 2 of the side chain) are more bio-available, whereas, 2S forms (SSS, SSR, SRS, SRR, i.e. S-configuration at position 2 of the side chain) are basically not bio-available in humans and various animal species. However, bio-availability of different α-tocopherol stereoisomers differs among animal species (Jensen and Lauridsen, 2007).

In ruminants and swine, higher bio-availability of natural source vitamin E has been established. RRR-α-tocopherol is the dominating stereoisomer retained in plasma and milk in cows, accounting for 96 and 86% of the total α-tocopherol in plasma and milk, respectively. The 2S forms amounted to less than 1% of the total α-tocopherol in plasma and milk (Jensen and Lauridsen, 2003; Meglia et al., 2006). In sows, RRR-α-tocopherol stereoisomer was preferentially transferred and retained into plasma and milk, with 31% and 35% of the total α-tocopherol in plasma and milk, respectively (Lauridsen and Jensen, 2005). Similar results as for sows and piglets were observed in veal and rearing calves (Dersjant-Li et al., 2009). Information on α-tocopherol stereoisomer transfer from feed to egg is limited. An earlier study showed that the isomer pair of RRR/SSS is preferentially transferred from feed to egg yolk (Piironen et al., 1991). A preliminary study with commercial eggs showed, that RRR and RRS isomers (59 and 20%) had been preferentially retained in egg yolk, while 2S and RSR, RSS isomers show less than 5% retention per isomer (Dersjant-Li and Peisker, 2010). The objective of this study was to determine the transfer and distribution of α-tocopherol stereoisomers from feed to egg yolk in laying hens and tissues of newly hatched chickens.

Materials and Methods

The trial was carried out at the Institute of Animal Nutrition of Friedrich-Loeffler-Institute (FLI), Federal Research Institute for Animal Health, Braunschweig, Germany.

Dietary treatments

Five diets were used in the trial. The control diet consisted of mainly corn and soybean meal (Table 1). Test diets were formulated by addition of 30 or 60 mg/kg RRR-α-tocopheryl acetate and 30 or 60 mg/kg all-rac-α-tocopheryl acetate to the control (basal) diet. Treatment code, vitamin E sources and inclusion levels are shown in Table 2.

Table 1. Composition and analysed values of basal diet

Zusammensetzung und analysierte Nährstoffgehalte der Basisration

Ingredient, as is	g/kg
Corn	719.90
SBM, 44%CP	164.07
Soy oil	11.20
Calcium carbonate	76.98
DCP 40	12.49
NaCl	3.31
Premix (no Vitamin E)	10.00
DL-Methionine	1.06
L-Lys-HCl	0.99
DM	882.5
CP	160
Crude fat	26.6
Crude fiber	28.9
Crude ash	124.5
Starch	427.3
ME, MJ/kg	11.0

Table 2. Dietary treatments, vitamin E sources and inclusion levels

Behandlungen, Vitamin E-Quellen und Zulagehöhen

Treatment code	Treatments
Pre-feeding	Commercial vitamin E supplement
11	Basal diet, no supplementvitamin E (control)
12	Basal + 30ppm RRR-α-tocopheryl acetate*
13	Basal + 60 ppm RRR-α-tocopheryl acetate*
14	Basal + 30 ppm all-rac-α-tocopheryl acetate
15	Basal + 60ppm all-rac-α-tocopheryl acetate

* Natural-Source Vitamin E 250 (Adsorbate) ADM SID (Europe) B.V.

Animals and feeding

In total, 210 laying hens (Lohmann LB) aged 22 weeks were used in the study (42 per treatment). They were fed a commercial layer diet (containing 20 mg/kg vitamin E via premix) for 4 weeks (23 to 27 weeks of age, 1st laying month), followed by a diet (basal) without any vitamin E addition from 28 to 35 weeks of age. The treatment diets were fed in meal form from 35 weeks of age onwards. Hens were singly caged and water was freely available.

Sampling and measurements

Egg samples were taken on day 0, 7, 14, 28 and 84, respectively, after feeding the basal and experimental diets. At each sampling point, 2 egg yolks were collected from 7 hens, and pooled as one sample. Thus 5 pooled samples (14 egg yolks per sample) were collected for each treatment at each sampling point. Samples were freeze-dried for analysis of total α-tocopherol and stereoisomer composition. Samples from feed and premix were taken during the experiment. All laying hens were artificially inseminated and eggs were hatched in laying month 7 and 11. Plasma and tissue samples (heart, breast, liver, brain and yolk sac) were taken from newly hatched birds (10 birds per treatment) and analyzed for total α-tocopherol and stereoisomer composition.

Laboratory analyses

Total α-tocopherol and stereoisomers composition were analyzed at Aarhus University, Department of Animal Science, by using HPLC method as described by JENSEN and LAURIDSEN (2007). Tissue samples of newly hatched chicks were pooled in order to get enough sample material for analyses. Therefore, statistics on stereoisomer distribution in tissues of newly hatched chicks could not be run.

Statistical analysis

For statistical analysis of data, the MIXED procedure of SAS software package (SAS Inst., Inc., Cary, NC) was applied.

Results

Vitamin E content in basal diet and premix

The total α-tocopherol content was analyzed in the basal diet and premix and the results are presented in Table 3. Premix contained about 300 mg/kg α-tocopherol from all-rac-α-tocopheryl acetate. The stereoisomer distribution in premix showed 50% 2S and 12.5% each of the 2R forms. The diet used in the pre-feeding period (week 23–27) contained 25 mg/kg α-tocopherol. Based on analysis of the tocopherol containing ingredients corn, soybean meal and soy oil, the basal diet was calculated containing 11 mg/kg α-tocopherol.

Table 3. Total α-tocopherol (mg/kg) in premix and basal/control diet

Gesamtgehalt an α-Tocopherol (mg/kg) in der Vormischung und in der Basisration (Kontrolle)

	Pre-Feeding	Premix	Basal*
Mean	24.8	302	11
SD	1.8	1.4	0.2 – 0.8

* Based on α-tocopherol analysis of ingredients

Vitamin E content in feed and egg yolk

Total α-tocopherol concentration in feed and treatment groups is presented in Table 4. The results for the treatment were combined and are shown as average values, since there was no significant difference after two and twelve week's trial period within the same treatment.

Table 4. Average α-tocopherol concentration in feed (mg/kg) and egg yolk (mg/kg)

Durchschnittliche α-Tocopherol-Konzentrationen im Futter (mg/kg) und im Dotter (mg/kg)

Treatment	Feed		Avg values Egg Yolk	Sx
	mg/kg		mg/kg	
11	11	Basal	125^{e}	8
12	41	B + 30 RRR	214^{d}	18
13	95	B + 60 RRR	320^{b}	30
14	45	B + 30 all-rac	251^{c}	8
15	95	B + 60 all-rac	434^{a}	26
		P-Value	< 0.001	

Supplementation of both sources of vitamin E increased α-tocopherol concentration in egg yolk compared to basal diet significantly. With the same level of dietary addition, significantly more α-tocopherol was deposited with all-rac than with RRR α-tocopherol (treatment 12 vs. 14 and 13 vs. 15, resp.).

α-Tocopherol stereoisomer distribution in feed and egg yolks

Distributions of stereoisomers as % of total α-tocopherol in egg yolk was significantly influenced by dietary treatments (Table 5). Feeding natural source vitamin E increased the proportion of RRR-α-tocopherol stereoisomer and reduced synthetic 2R and 2S forms in egg yolk, compared to control group and baseline. This is more pronounced within the 60 mg/kg RRR-α-tocopheryl acetate supplementation group. Here the RRR α-tocopherol stereoisomer contributed up to 74% of total α-tocopherol. In treatments with synthetic vitamin E supplementation, the RRR-α-tocopherol stereoisomer contributed to 20–22% of total α-tocopherol. Comparison of feed and egg yolk stereoisomer distribution in the all-rac treatments shows that 2S isomer percentage reduces and RRS isomer increases in yolk at all sampling points. This resulted in an increased 2R/2S ratio in egg yolk compared to feed. The distributions pattern of stereoisomers maintained basically the same after the first week until week 12 of the experiment within each treatment group. Therefore, statistical analyses was carried out based on results of sampling points on days 14 and 28 combined (Table 6).

Table 5. Distributions of stereoisomer in feed and egg yolk (% of total α-tocopherol)

Verteilung der Stereoisomere im Futter und im Dotter (% des Gesamtgehalts an α-Tocopherol)

Treatments		Days	2S	RSS	RRS	RRR	RSR	2R/2S
Baseline/ Pre-feeding	feed		42.6	5.7	10.0	27.9	13.7	1.35
	yolk	0	29.4	11.1	22.7	27.2	9.5	2.40
11 Control	feed		34.9	6.5	10.4	32.3	16.0	1.87
11	yolk	7	28.9	11.2	21.9	26.5	11.5	2.46
11		14	28.2	10.7	22.9	27.9	10.3	2.55
11		28	28.8	10.7	22.3	27.7	10.5	2.47
11		84	28.4	10.7	21.2	27.9	11.7	2.52
12 + 30 RRR	feed		23.5	3.2	5.7	59.6	8.0	3.26
12	yolk	7	14.9	6.1	13.2	59.7	6.1	5.71
12		14	16.3	6.0	15.0	57.8	4.9	5.14
12		28	16.1	6.0	15.3	58.0	4.7	5.19
12		84	20.8	7.6	16.6	46.8	8.1	3.80
13 + 60 RRR	feed		8.5	1.2	2.1	85.3	3.0	10.78
13	yolk	7	16.3	3.6	11.5	63.3	5.3	5.13
13		14	11.4	4.1	11.4	69.6	3.5	7.78
13		28	11.4	4.4	11.4	69.7	3.4	7.82
13		84	10.2	2.9	10.2	73.5	3.1	8.79
14 + 30 all-rac	feed		46.6	7.3	10.9	21.6	13.6	1.15
14	yolk	7	34.4	13.5	19.5	19.8	12.9	1.91
14		14	34.3	13.0	19.2	21.5	11.9	1.92
14		28	33.2	13.0	19.8	22.0	12.0	2.01
14		84	30.5	12.4	20.5	24.6	12.1	2.28
15 + 60 all-rac	feed		48.8	11.3	11.5	17.2	11.2	1.05
15	yolk	7	34.9	14.7	18.2	18.4	13.6	1.15
15		14	36.2	13.5	17.9	19.3	13.1	1.76
15		28	37.8	9.7	19.3	19.5	13.7	1.64
15		84	34.4	12.6	19.0	21.3	12.7	1.91

Table 6. Distribution of stereoisomers in egg yolk (% of total α-tocopherol; average of days 14 and 28)

Verteilung der Stereoisomere im Dotter (% des Gesamtgehalts an α-Tocopherol; Durchschnitt der Tage 14 und 28)

Treatments		2S	RSS	RRS	RRR	RSR	2R/2S
Pre-feeding	yolk	29.4	11.1	22.7	27.2	9.5	2.40
11	yolk	28.5^{c}	10.7^{b}	22.6^{a}	27.8^{c}	10.4^{c}	2.51^{c}
12	yolk	16.2^{d}	6.0^{c}	15.1^{c}	57.9^{b}	4.8^{d}	5.17^{b}
13	yolk	11.4^{e}	4.2^{d}	11.4^{d}	69.7^{a}	3.5^{e}	7.81^{a}
14	yolk	33.8^{b}	13.0^{a}	19.5^{b}	21.8^{d}	12.0^{b}	1.96^{d}
15	yolk	37.0^{a}	11.6^{a}	18.6^{b}	19.4^{e}	13.4^{a}	1.70^{e}

Different superscripts within a column indicate $P < 0.05$

All-rac-α-tocopheryl acetate supplementation increased the percentage of 2S and decreased the percentage of 2R isomers in egg yolk. The 2R/2S ratios increased when feeding RRR and decreased when feeding all-rac α-tocopherol compared to the control group.

α-Tocopherol stereoisomer distribution in tissues of newly hatched chicks

Total α-tocopherol concentration increased in plasma and tissues of newly hatched chickens with supplementation of vitamin E from both sources and dietary levels (Table 7). There were only small differences in total α-tocopherol concentration in tissues between vitamin E sources, except that 30 mg/kg RRR-α-tocopheryl acetate (treatment 12) had higher level in yolk sac, plasma, heart and breast when compared to 30 mg/kg all-rac-α-tocopheryl acetate (treatment 14). In general, tissue distribution pattern of stereoisomers mirrors that in egg yolk. However, there are tissue related differences. For example, in control group (11), less RRR- isomer was retained in the yolk sac, resulting in higher RRR proportion in other tissues, especially in the brain (100%). Also, in the all-rac-α-tocopheryl acetate treatments (14, 15), the RRR α-tocopherol stereoisomer contributed to the highest proportion in brain (30and 24%). In the RRR-α-tocopheryl acetate supplementation groups, RRR-α-tocopherol stereoisomer contributed to 86–99% of total α-tocopherol, with higher proportions observed for breast and brain.

Table 7. Distribution of stereoisomers in tissues of hatched chicks (% of total α-tocopherol)

Verteilung der Stereoisomere im in den Geweben der geschlüpften Küken (% des Gesamtgehalts an α-Tocopherol)

Treatment (mg/kg)		α-toc	2S	RSS	RRS	RRR	RSR	2R/2S
		µg/g	%	%	%	%	%	
Yolk sac								
11	Control	35.6	11.1	2.9	17.2	70.1	1.7	2.57
12	30 RRR	125.0	2.8	0.6	7.8	88.4	0.4	34.71
13	60 RRR	133.6	1.5	0.2	6.4	91.7	0.1	65.60
14	30 all-rac	86.6	35.0	13.5	18.1	12.6	20.8	1.86
15	60 all-rac	116.5	36.6	14.7	17.8	17.5	13.3	1.73
Plasma								
11	Control	17.1	3.5	1.0	15.5	77.3	1.4	27.20
12	30 RRR	32.9	1.4	0.3	7.8	86.8	0.5	68.14
13	60 RRR	34.7	1.2	0.3	7.2	90.2	0.4	81.75
14	30 all-rac	26.6	29.4	12.4	22.3	23.7	12.2	2.40
15	60 all-rac	52.1	33.0	14.3	20.1	19.3	13.4	2.03
Liver								
11	Control	83.1	12.4	3.0	6.5	71.1	5.1	6.91
12	30 RRR	309.4	2.3	1.2	7.6	88.3	0.6	42.48
13	60 RRR	406.0	2.4	0.9	1.4	93.8	1.5	40.67
14	30 all-rac	289.1	32.9	12.9	19.3	22.2	12.7	2.04
15	60 all-rac	440.6	33.3	15.3	18.5	18.7	14.2	2.00
Heart								
11	Control	8.6	6.8	5.2	30.8	51.1	6.1	13.71
12	30 RRR	21.9	1.3	0.5	10.5	86.1	0.7	75.23
13	60 RRR	27.4	0.7	0.5	9.3	86.4	1.2	139.14
14	30 all-rac	15.9	30.6	12.8	23.1	22.8	10.7	2.27
15	60 all-rac	27.6	30.9	14.5	21.2	19.7	13.6	2.23
Brain								
11	Control	1.6	n.d.	n.d.	n.d.	100.0	n.d.	n.d.
12	30 RRR	3.1	2.2	0.6	6.4	90.6	0.2	44.45
13	60 RRR	4.8	1.0	0.4	5.6	92.3	0.7	99.00
14	30 all-rac	3.2	25.0	9.9	21.9	30.4	12.9	3.00
15	60 all-rac	4.0	25.2	14.1	22.7	23.6	14.3	2.96
Breast								
11	Control	4.7	7.3	3.2	6.4	70.3	6.4	11.82
12	30 RRR	15.4	0.9	n.d.	n.d.	99.1	n.d.	110.11
13	60 RRR	17.8	1.4	0.3	2.3	93.5	1.2	69.50
14	30 all-rac	11.7	27.6	12.5	24.3	24.6	11.0	2.62
15	60 all-rac	17.3	30.5	15.2	22.3	20.3	11.7	2.28

Figure 1, 2 and 3 show the amounts (µg/g) of total α-tocopherol and RRR-α-tocopherol in yolk sac and selected tissues (brain, breast) of newly hatched chicken when feeding increasing levels of dietary vitamin E, either as RRR-α-tocopherol or as all-rac-α-tocopherol. It is noticed that for lower supplementation levels of vitamin E to diets the total amount of α-tocopherol is higher (except for brain tissue) for the RRR- than for the all-rac-α-tocopherol source. For the high level of supplementation no differences were observed. The amount of RRR -α-tocopherol shows differences in the assessed tissues and yolk sac. With synthetic vitamin E as dietary source there was no increase of RRR-α-tocopherol, whereas in the RRR treatment groups the analysed amounts are almost identical to the total α-tocopherol content (Figure 1, 2 and 3).

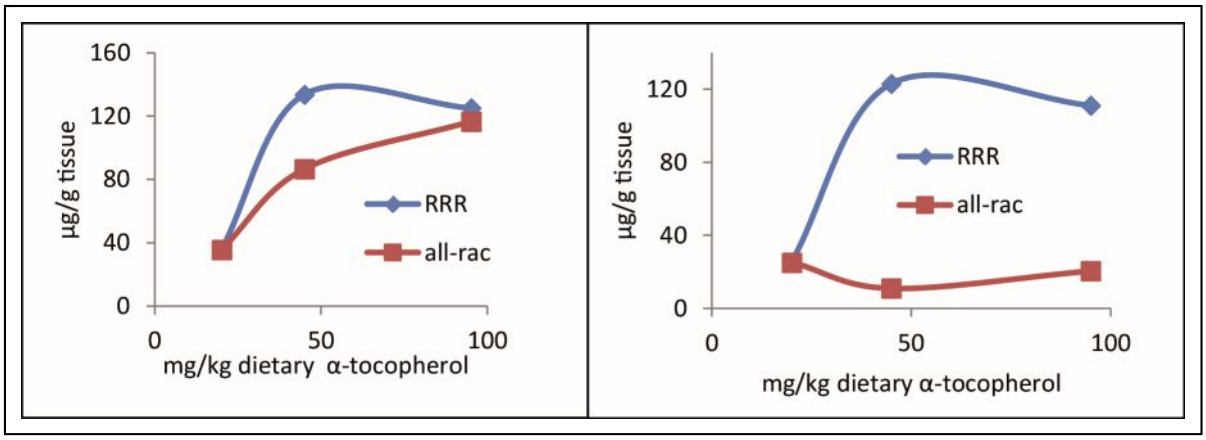

Figure 1. Total (left) and RRR α-tocopherol content (right) in chicken yolk sac

Gesamt- (links) und RRR α-Tocopherol-Gehalt (rechts) im Dottersack der Küken

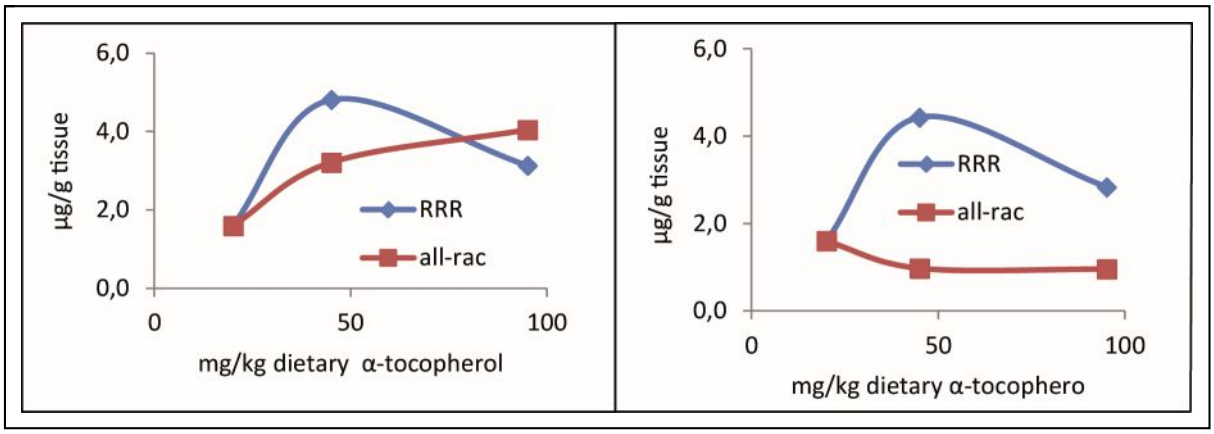

Figure 2. Total (left) and RRR-α-tocopherol (right) in chicken brain tissue

Gesamt- (links) und RRR α-Tocopherol-Gehalt (rechts) im Hirngewebe der Küken

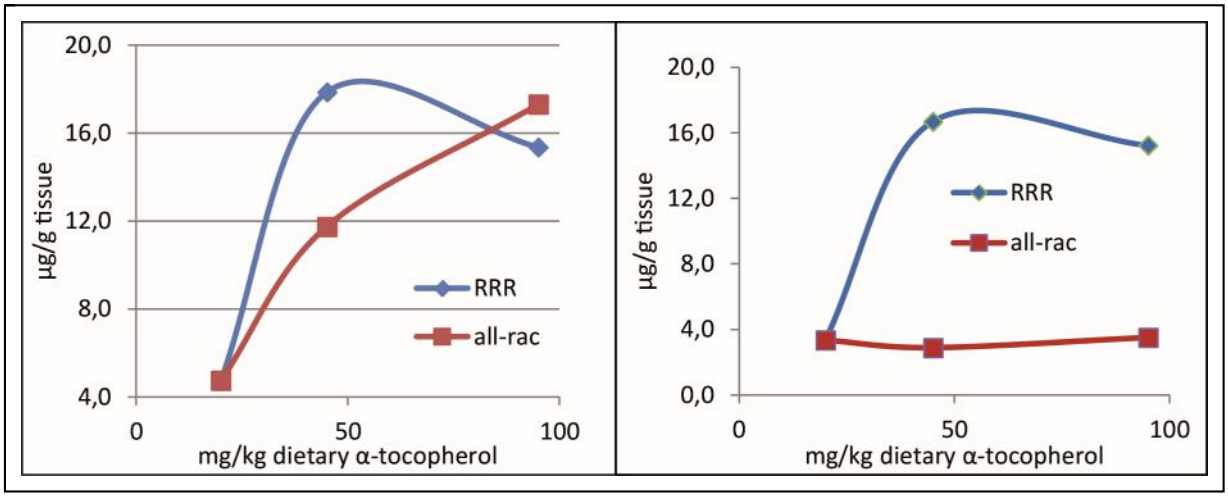

Figure 3. Total (left) and RRR-α-tocopherol (right) in chicken breast tissue

Gesamt- (links) und RRR α-Tocopherol-Gehalt (rechts) im Brustmuskel der Küken

Discussion

Deposition of feed-borne tocopherol after luminal absorption into cells of different tissues is facilitated by several mechanisms (MARDONES and RIGOTTI, 2004). Lipid transfer proteins and lipases, receptor mediated lipoprotein endocytosis and selective Scavenger Receptor BI (SRBI) uptake models are discussed in literature (KOSTNER et al., 1995; NAKAMURA et al., 1998; GOTI et al., 2000; GOTI et al., 2002; KAEMPF-ROTZOLL et al., 2003). Chylomicrons as carriers of tocopherol from the gut are relieved from their tocopherol by lipoprotein lipase-mediated remodelling in muscle and adipose tissue, yielding remnant chylomicrons. Remnant chylomicrons are ultimately transported to the liver, where the associated tocopherol is selectively incorporated into very low density lipoproteins (VLDL) by the α-tocopherol transfer protein (α-TTP) (SATO et al., 1991; ARAI et al., 1993; KAEMPF-ROTZOLL et al., 2003). However, exchange of α-tocopherol is possible between different classes of lipoproteins from VLDL to LDL (low density lipoprotein) to HDL (high density lipoprotein) (TRABER et al., 1993; KOSTNER et al., 1995). The α-TTP was first described by SATO et al. (1991). α-TTP not only sorts out the α-form of all tocopherols but also has a preference for 2R stereoisomers (BRIGELIUS-FLOHE and TRABER, 1999). A plasma factor of 2 in favour of the RRR form has been observed in humans and this largely exceeds the accepted ratio of biological activity of 1.36 (ACUFF et al., 1994; TRABER et al., 1994; KIYOSE et al., 1997; ACUFF et al., 1998; BURTON et al., 1998; TRABER et al., 1998). Humans with defective α-TTP genes are vitamin E deficient (TRABER and ARAI, 1999). Significant differences in metabolism of individual tocopherols have to be taken into account when assessing vitamin E effects (BRIGELIUS-FLOHE and TRABER, 1999). Presence or absence of a specific α-TTP presumably is one of the metabolic factors to be considered.

TRABER (1998) suggested that every tissue must have its own tocopherol transferring protein. MARDONES and RIGOTTI (2004) hypothesized a novel role for lipoprotein lipase for the delivery of α-tocopherol across the blood-brain barrier into the central nervous system. KAEMPF-ROTZOLL et al. (2003) proposed specific transfer proteins for brain and uterus/placenta but also tissues without specific α-TTP.

Egg yolk

Most studies are done in rats and humans or other mammals. Very little is known about avian species. When looking at the tocopherol content in egg yolk (Table 4) it is obvious that at equal dose level more α-tocopherol was deposited when the feed was supplemented with all-rac α-tocopherol. The reason for this difference remains unclear, however, it can be assumed that α-tocopherol transfer most likely occurs directly from chylomicrons to egg yolk. The results suggest that the laying hen has no hepatic α-TTP for selective incorporation of the RRR isomer of α-tocopherol into lipoproteins.

The stereoisomer distribution in egg yolk is reflecting their distribution in feed. However, the proportion of 2S-isomers in the all-rac treatments is lower in egg yolk compared to feed (Table 5).The percentage of RRR-isomers in the all-rac treatments is similar to the feed, whereas, the percentage of the RRS-isomer almost doubles. The 2S/2R ratio in the all-rac-α-tocopheryl acetate supplementation groups 14 and 15 is about 1.1 in feed, while it is about 1.7 – 2.0 in egg yolk (Table 6) and increases with trial time (Table 5). In the treatments with natural source vitamin E (12 and 13) the RRR isomer is the dominating form.

Compared to control (treatment 11) the 2R/2S isomer ratio increases significantly in the RRR treatments 12 and 13 with increasing dietary vitamin E supplementation and decreases significantly in treatments 14 and 15 (Table 6). However, the percentage of RRR isomer does not increase in the all-rac treatments compared to control, which would be expected if a specific α-TTP would selectively retrieve the RRR-α-tocopherol from remnant chylomicrons.

Yolk sac/Tissues

The amount of α-tocopherol in yolk sac at similar dietary dose level is numerically higher with natural than with synthetic vitamin E. This contrasts the findings displayed in Table 4. Also for plasma and the other tissues except the brain, the absolute amounts of α-tocopherol are numerically higher with the lower dose level of natural vitamin E (treatments 12 vs. 14). The cause of this remains unclear. When looking at the fate of the different isomers, there are indications, that in the all-rac treatments the 2R isomers are enriched in plasma and tissues (except yolk sac). They are higher than the expected level of 50%, and in particular the RRR and the RRS isomers exceed the expected level (12.5%) as analysed in the premix (Table 7).

In yolk sac the percentage of 2S isomers is about equal to egg yolk in the all-rac treatments (30 – 37%), whereas, it is reduced in the RRR treatments from 12–16% to only 1.5–2.8%. Stereoisomer distribution in other tissues of newly hatched chicken show the RRR isomer prevailing in the RRR treatments. In the all-rac treatments the sum of

RRS + RRR exceeds the sum of 2S isomers at both dose levels. This supports the previous findings of Dersjant-Li and Peisker (2010). In brain, the RRR makes up 30% and RRR + RRS make up 52% of total α-tocopherol at the lower dose level. There is a specific preference of these two isomers from the all-rac α-tocopherol for uptakes by the brain and, to a lesser extend also breast muscle and heart tissue.

Piironen et al. (1991) presented results showing a specific selectivity for α-tocopherol isomers. They separated the eight stereoisomers from all-rac-α-tocopherol to 4 pairs of enantiomers. Hens receiving diets based on barley (A), barley/oats (B) and oats (C). All three feeds were supplemented with 16 mg/kg all-rac-α-tocopheryl acetate. Total α-tocopherol content in feed was 18.3, 19.0 and 26.5 mg/kg for feeds A, B and C, respectively. This indicates that feed A contained relatively low amounts of naturally occurring vitamin E, while feed C contained almost 10.5 mg/kg natural source vitamin E from oats. However, regardless of the different natural source vitamin E content in the diets, it was observed that in all feed vs. egg comparisons, the eggs contained significantly more of the enantiomeric pairs of RRR + SSS than did the feed. The proportion of the other pairs was lower in the egg than in the feeds.

The results of the presented study are in agreement with Cortinas et al. (2004), who determined the effect of supplementation with different levels of all-rac-α-tocopheryl acetate on the deposition of α-tocopherol stereoisomers in liver and thigh meat in broilers. All-rac-α-tocopheryl acetate was supplemented at 100, 200 and 400 mg/kg. However, α-tocopherol stereoisomer proportion in liver and thigh was not affected by vitamin E inclusion levels. It was observed that both tissues preferentially accumulated 2R stereoisomers (main isomers are RRR and RRS). The 2R/2S ratio determined in this study was 2.3 and 3.5 in liver and thigh, respectively, while the 2R/2S ratio was 1.1 in feed.

In humans, a hepatic tocopherol transfer protein was identified, which selectively transfers the RRR and other 2R forms into VLDL, which transfers them to other lipoproteins and eventually to different tissues. In avian species such a protein has not yet been identified. However, there might be other mechanisms for selective transfer and incorporation of α-tocopherol isomers into avian tissues as the results for brain and breast muscle indicate. The transfer from feed to egg yolk seems to be executed none-selectively by chylomicron transportation. From yolk sac to tissues, in particular the brain, discrimination between isomers occurs. The mechanisms responsible for this selection needs to be investigated in further studies.

Conclusions

The results of this study show that the α-tocopherol stereoisomer composition of egg yolk and various tissues of newly hatched chicken are closely related to dietary vitamin E sources. When all-rac-α-tocopheryl acetate is supplied to feed, 2R forms are preferentially transferred to egg yolk and tissues as reflected by an increase in the 2R/2S ratio. This discrimination of different isomers is related to dose level and target tissues. Brain turned out to be the most discriminating tissue amongst tissues tested in this study. Low dose levels of all-rac α-tocopherol result in relatively higher RRR isomer retention in tissues. The degree of stereoisomer discrimination is less pronounced in laying hens compared to pigs and ruminants. However, a tissue specific discriminatory mechanism for selecting different α-tocopherol isomers appears to exist also in layers and needs to be addressed in further research.

Summary

Alpha-tocopherol (vitamin E) derived from natural source consists solely of a single stereoisomer (i.e. RRR-α-tocopherol) while synthetic vitamin E consists of a mixture of eight stereoisomers (all-rac-α-tocopherol). These stereoisomers are not equally retained in the body and their efficiency of utilization differs amongst animal species. A study with laying hens was carried out to determine the transfer of different α-tocopherol stereoisomers from feed to egg yolk and various tissues of newly hatched chicken. The results of this study show that the α-tocopherol stereoisomer composition of egg yolk and various tissues of newly hatched chicken are closely related to dietary vitamin E sources. When all-rac-α-tocopheryl acetate is supplied to feed, 2R forms are preferentially transferred to egg yolk and tissues as reflected by an increase in the 2R/2S ratio. Discrimination of different isomers is related to dose level and target tissues. Brain turned out to be the most discriminating tissue amongst tissues tested in this study. Low dose levels of all-rac α-tocopherol result in relatively higher RRR isomer retention in tissues. The degree of stereoisomer discrimination is less pronounced in laying hens compared to pigs and ruminants. However, a tissue specific discriminatory mechanism for selecting different α-tocopherol isomers appears to exist also in layers.

Key words

Layers, nutrition, eggs, vitamin E, α-tocopherol stereoisomer, transfer, deposition

Zusammenfassung

Transfer von α-Tocopherol-Isomeren vom Legehennen-Futter zum Dotter und zu den Geweben von geschlüpften Küken

Alpha-Tocopherol (Vitamin E) aus natürlichen Quellen besteht ausschließlich aus einem einzelnen Stereoisomer (d.h. RRR-α-Tocopherol), während synthetisches Vitamin eine Mischung aus acht Stereoisomeren (All-rac-α-Tocopherol) ist. Diese Stereoisomere werden nicht gleichmäßig im Körper gespeichert und die Aufnahmeeffizienz variiert zwischen den Tierarten. Das Ziel der Studie war daher die Bestimmung des Transfers von verschiedenen α-Tocopherol-Stereoisomeren vom Futter in den Eidotter und in verschiedene Gewebe von frisch geschlüpften Küken. Die Ergebnisse zeigen, dass die Verteilung der α-Tocopherol-Stereoisomere im Dotter und in den Geweben der Küken eng von dem im Futter verwendeten Vitamin E-Quellen abhängt. Beim Einsatz von All-Rac-α-Tocopherol-Acetat im Futter werden in erster Linie die 2R-Formen zum Dotter und den Geweben transferiert, wie die Erhöhung des 2R/2S-Verhältnisses belegt. Die Unterscheidung zwischen den verschiedenen Isomeren hängt von der Zulagehöhe zum Futter und vom Zielgewebe ab. Dies war beim Gehirn am deutlichsten ausgeprägt. Eine geringe Zulagenhöhe an All-Rac-α-Tocopherol bewirkte eine relativ höhere Retention des RRR-Isomers in den Geweben. Die Unterscheidung zwischen den Stereoisomeren ist bei der Legehennen weniger stark ausgeprägt als bei Schweinen oder Wiederkäuern. Allerdings scheint auch bei der Legehenne ein spezifischer Unterscheidungsmechanismus für die Auswahl von unterschiedlichen α-Tocopherol-Isomeren zu existieren.

Stichworte

Legehenne, Fütterung, Eier, Vitamin E, α-Tocopherol-Isomere, Transfer, Deposition

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