



THREONINE IN BROILER DIETS: AN UPDATED REVIEW

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Abstract

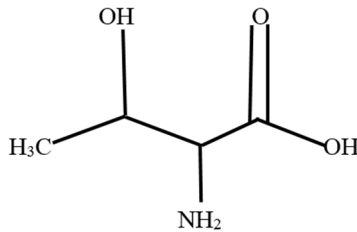
Threonine (Thr) is the third limiting essential amino acid after methionine and lysine in corn-soybean based diets of broilers. Dietary imbalance of Thr, therefore, results in a poor growth performance in broilers. This review summarizes literature data on the known effects of dietary levels of Thr on growth performance, gut morphology, immunity and carcass characteristics in broilers. Due to continuous improvement in genetic potential and management practices for poultry production, dietary Thr requirements are changing. A number of studies have shown that supplementation of Thr in broiler diet at a higher level than the current NRC recommendation (0.74–0.81%), increases body weight gain, feed conversion ratio, and improves gut morphology, carcass quality and immune status, mainly by enhancing the functional capability of digestive system and immune organs (spleen, bursa, and thymus). According to the literature data discussed in this review, the minimal and maximal total dietary Thr levels for healthy birds reared in normal conditions were 0.67 and 0.90% for growth performance, 0.77 and 1.1% for a better gut health, 0.60 and 1.02% for immunity and 0.62 and 0.97% for better carcass characteristics. This background provides impetus to further investigate the exact level of Thr and its effects on growth performance, gut morphology, immunity and carcass characteristics in broilers.

Key words: threonine, broiler, growth performance, gut health, immunity

Nutrition is the backbone of profitable broiler production and on an average it accounts for about 80–90% of the total cost of production (NAFIS, 2017). Modern broilers can potentially attain 2 kg of body weight by consuming 3 kg of feed within 5 weeks (Choct, 2009). Genetic selection and a nutritionally balanced diet are the main drivers of the faster growth of broilers (Havenstein et al., 2003). This higher growth rate, however, requires highly digestible energy and protein concentrated diets, which makes broiler feed very expensive. To minimize feed cost and increase its efficiency, modern broiler diets are formulated on digestible amino acids basis. There are about 500 types of essential molecules known as amino acids in any living organ-

isms, and only 20 of them are genetically encoded (Walsh et al., 2013). Out of these 20 amino acids, 10 are classified as essential, meaning that they cannot be synthesized at all or rapidly enough to meet metabolic requirement, and must be supplied in the diets for a maximum growth performance. Out of these 10 essential amino acids, methionine and lysine (Lys) are considered as the first two limiting amino acids for broilers (Corzo et al., 2007), whereas threonine (Thr) is ranked the third limiting amino acid (Kidd and Kerr, 1996). Threonine is very important for synthesis and maintenance of the protein in the body and contains 11.7% nitrogen (Kidd and Kerr, 1996). Threonine requirement of broilers depends on various factors including age of the birds, dietary crude protein (CP) level and core ingredients in the diet (Barkley and Wallis, 2001). National Research Council (1994) recommended values for total dietary Thr were 0.80% for starter (0 to 21 days), 0.74% for grower (22 to 42 days), and 0.68% for finisher (43 to 56 days) periods, respectively. In ideal protein concept, however, all the dietary essential amino acids need to be expressed as the percentages of Lys in the diets (NRC, 1994). In broiler diets, these recommendations for Lys and Thr ratio range from 1.10 to 1.00% and 0.80 to 0.74% for starter (0 to 3 wk) and grower (3 to 6 wk) phase, respectively.

Threonine contains both alpha as well as beta carbons in its structure and, therefore, has 4 isomers (D, L, D-allo, and L-allo). In broilers, only L-form of Thr is utilized, and transamination of alpha-keto as well as D-isomers to L-form does not occur. Catabolism of Thr results in production of pyruvate, glycine and acetyl-CoA that play a vital role in animal's body metabolism (Baker, 1985) including ketogenesis as well as gluconeogenesis. The structure of Thr is shown below:



Structure of Threonine

Threonine has a major role in intestinal development and well-functioning (Stoll, 2006), because intestinal mucin is mainly made of Thr (Faure et al., 2005). Dietary total Thr level between 0.70 and 0.93% can support optimum gut morphology (Schaart et al., 2005; Zaefarian et al., 2008). Mucin is a glycoprotein in nature, which plays a vital role in protecting the intestine from acidic chyme, pathogens and digestive enzymes as well as maintains the intestinal integrity (Horn et al., 2009). In gastrointestinal tract, mucin acts as a filtering agent for nutrients and affects their

digestion and absorption (Smirnov et al., 2006). Threonine is “furthermore” involved in different metabolic processes such as protein synthesis and uric acid formation (Eftekhari et al., 2015). Diets deficient in Thr may compromise immunoglobulin production because Thr is an integral part of immunoglobulin in broilers (Azzam and El-Gogary, 2015). Threonine is considered as the second limiting amino acid for breast meat yield (Estalkhzir et al., 2103) and, therefore, its supplementation is assumed to result in improved carcass characteristics. It is believed that Thr supplementation enhances feed intake, body weight gain and ultimately carcass weight (Estalkhzir et al., 2103; Khan et al., 2006). The present review describes the response of modern-day broilers to dietary Thr levels, and its effects on growth performance, gut morphology, immunity and carcass characteristics.

Influence of dietary threonine

Effects on growth performance

Growth performance is an important parameter to evaluate the effectiveness of feed offered to the broilers. Different environmental conditions also influence growth performance of broilers depending on the dietary Thr level (Kidd et al., 2003). These later authors reported that female broilers showed best growth performance at 0.60 to 0.67% of total dietary Thr, whereas for male broilers it was 0.63 to 0.68% during finishing (42 to 45 days) period. Corzo et al. (2003) found that ideal total dietary Thr level was 0.69% for growth performance and 0.71% for feed conversion ratio (FCR) during finisher period (30 to 42 days) in broilers. In contrast, Kidd et al. (2004) reported that total dietary Thr requirement was 0.74% for body weight gain and 0.71% for breast meat yield during grower and finisher (21 to 42 days) period. Li (2000) estimated that the requirement of the total Thr was 0.66% from 3 to 6 weeks of age. Threonine requirements for feed intake were 0.79% during starter and grower phase and 0.72% for finisher phase (Samadi and Liebert, 2006). Ciftci and Ceylan (2004) reported that ideal total dietary Thr levels for growth performance were 0.68 to 0.75% for starter (0 to 21 days) and 0.65 to 0.68% for grower (22 to 42 days) periods. This decrease in Thr requirement with increase in age may be due to varying dietary crude protein level (Rangel-Lugo et al., 1994). In hygienic environmental conditions Thr requirements might be under NRC recommended level (Azzam and El-Gogary, 2015). Recently, Najafi et al. (2017) executed a trial to investigate the effects of dietary Thr on growth performance in broilers from 1 to 14 days of age by feeding 0.89, 0.93 and 0.97% Thr, along with a control diet containing 0.65% Thr. The later authors found that the broilers fed diets containing 0.97% of Thr showed 5.1% higher feed intake, 6.4% higher body weight gain and 1.4% better FCR compared with the birds fed control diet. Similarly, Kheiri and Alibeyghi (2017) reported that the broilers fed diets containing 0.90% Thr had 1.1% increased FI, 3.2% higher body weight gain and 1.7% better FCR compared with those birds fed control diets. Min et al. (2017) conducted a trial to investigate the effects of Thr on broilers growth performance from 1 to 42 days of age by using 0.75, 0.94 and 1.12% total Thr of the diet, along with a control diet containing total 0.62% Thr. The results revealed that broilers fed diet containing 0.75% Thr had 1.6% higher FI, 6% higher average daily gain and 4.2% better FCR compared with the birds fed control diet. Chen et

al. (2017), similarly, used 0.88 and 1.08% of total dietary Thr, along with a control diet containing 0.77% Thr to evaluate its effect on growth performance of broilers. Birds fed with diet containing 0.88% dietary Thr had 0.6% higher body weight gain, 2.8% better FCR, and 1.0% reduced feed intake compared to those fed with the control diet. Valizade et al. (2016) investigated the effect of 0.675 and 0.843% of total dietary Thr on growth performance of broilers in comparison with a control diet containing 0.641% Thr. Diet containing 0.843% dietary Thr supported better growth performance in broilers, with a 4.7% better FCR and 1.0% increase in body weight gain, compared with those fed control diet. The improved growth performance with higher level of Thr may be due to provision of higher level of Thr required for an ideal growth performance. Comparing the effect of 0.74, 0.81, 0.88 and 0.96% total dietary Thr on growth performance, Eftekhari et al. (2015) reported that diet containing 0.81% total dietary Thr supported a 5.1% better FCR compared with the birds fed NRC recommended (0.74% total dietary) Thr content. In another study, Shirzadegan et al. (2015) evaluated the effect of 0.74, 0.79, 0.81 and 0.84% of total dietary Thr on growth performance of broilers. The study concluded that broilers fed diets with 0.84% total Thr performed better and resulted in a 3.1% increase in feed intake, 10.5% higher body weight gain and 4.7% better FCR compared with those fed diets containing 0.74% total dietary Thr. Ospina-Rojas et al. (2013) investigated the effect of 0.70% (control) and 0.77% total dietary Thr levels on broiler performance. The results indicated that broilers fed with 0.77% dietary Thr had good performance with 1.3% higher body weight gain and 1.4% better FCR and a 0.2% reduction in feed intake compared with those receiving control diet. Corzo et al. (2007) evaluated the influence of feeding six levels, namely, 0.51 (control), 0.58, 0.65, 0.72, 0.79 and 0.86% of total dietary Thr on broiler growth performance. This dose response study indicated that 0.86% dietary Thr was ideal for broilers growth performance that resulted in 103% higher body weight gain, 133.7% better FCR and 13.3% lower feed intake during 22–42 d compared with those fed the control diet. Ciftci and Ceylan (2004) evaluated the effect of four levels (0.54 (control), 0.60, 0.66 and 0.72%) of total dietary Thr on growth performance of broilers. The 0.72% dietary Thr supported 23.2% increase in FI, 27.1% increases in body weight and 3% better FCR compared with the control diet. Data on the influence of different levels of dietary Thr on growth performance of broilers are summarized in Table 1. This literature review highlights that broiler diets are usually deficient in Thr and supplementation of dietary Thr, above NRC recommendations, in most of the studies, resulted in an improved growth performance.

Effects on gut health

The positive effects of Thr supplementation on growth performance in broilers may be due to the involvement of Thr in the development of intestinal mucosa as well as in digestive enzymes function (Dozier et al., 2001). A healthy gut plays a key role in an ideal growth performance of broilers because it supports a better digestion and absorption of nutrients. A healthy gut is, therefore, necessary for profitable poultry production. Villus height (the distance from the apex of the villus to the junction of the villus and crypt) and crypt depth (the distance from the villus junction to the basement membrane of the epithelial cells at the bottom of the crypt) are impor-

tant indices for gut health measurement. Longer villi and shorter crypts are usually considered as markers of a healthy and well functioning gut (Qaisrani et al., 2015). Threonine is reported to have a key role in maintaining gut health in broilers. For example, Chen et al. (2017) evaluated the effect of two levels (0.88 and 1.08%) of total dietary Thr on broiler (Arbor Acres plus) performance in a comparison with the strain recommended level (0.77%). Feeding higher level of Thr (1.08%) increased villus height (VH) by 18.3%, villus height to crypt depth ratio (VCR) by 33%, and reduced crypt depth (CD) by 12.5% in the jejunum compared with birds fed the strain recommended level of Thr. Similarly, Zhang et al. (2016) studied the effect of two levels (0.49 and 0.90%) of dietary Thr on broiler (Ross-308) gut health. It was observed that feeding diets containing higher level (0.90%) of Thr resulted in an increased VH by 57.6%, VCR 35.7% and a 18.9% decrease in jejunal CD compared with those fed 0.49% of Thr. Najafi et al. (2017) conducted a trial by feeding 0.89, 0.93 and 0.97% Thr of diet along with a control diet containing 0.65% of Thr. The duodenum of broilers fed 0.89% of Thr showed increased VH by 15%, VCR by 1.8% and reduced CD by 2% compared with those fed control diet. Improvement in gut morphology may be related to the involvement of Thr in mucin synthesis. Mucin protects intestinal epithelium from acids, digestive enzymes, and works as a filtering fence against outside pathogens (Kim and Ho, 2010). Mucin is made up of Thr, serine and proline, with Thr being the major (28 to 40%) component of mucin (Carlstedt et al., 1993). Eftekhari et al. (2015) fed an NRC recommended (0.74% total dietary Thr) level and three higher levels (0.81, 0.88 and 0.96%) of total Thr to evaluate its effect on gut morphology in broilers. It was observed that the broilers fed with 0.88% total dietary Thr had better gut health with a 0.08% greater VH, 0.4% increased VCR, and a 0.3% reduction in the jejunal CD compared with the recommended level. Shirzadegan et al. (2015) evaluated the effect of three levels (0.79, 0.81 and 0.84%) of total dietary Thr on broiler gut health in comparison with a control diet containing 0.74% total dietary Thr. The finding revealed that broilers fed diets with 0.84% total dietary Thr had a 7.4% greater VH, 7.2% deeper crypts and 0.2% increased VCR in the jejunum compared with control group. Abbasi et al. (2014) investigated the effect of Thr on gut morphology in broilers using three levels, namely, 0.71% (control), 0.77%, and 0.84% total dietary Thr. Ideal gut health was observed with 0.77% total dietary Thr, with 6.1% increase in VH, 12.6% improved VCR, and a 9% reduction in the jejunal CD compared to birds fed with recommended level (0.71%). Zaefarian et al. (2008) fed diets containing 0.4 (control), 0.5, 0.6, 0.7, 0.8, 0.9, 1.0 and 1.1% total dietary Thr to broilers and evaluated its influence on gut morphology. Diet containing 1.1% total Thr had better developed gut with a 12.4% increase in VH and 28.1% shorter crypts in the ileum compared with those fed control diet containing 0.4% total dietary Thr. In another study, Chee et al. (2010) formulated balanced broiler diets with 0.55% (control), 0.8% and 1.05% total Thr and investigated its effects on gut morphology of broilers. These findings revealed that broilers fed 0.8% Thr had 33.5% greater villus, 17% deeper crypts and 20.5% greater VCR in the ileum compared to the birds fed with control diet. Data on effect of different levels of dietary Thr on broilers gut morphology are summarized in Table 2.

Table 1. Summary of literature data on the effects of different dietary levels of threonine on growth performance in broilers

Threonine inclusion level (% of the diet)	Lysine level	Age (d)	Optimal (%)	Growth performance ¹ (%)			References
				FI	BWG	FCR	
0.65 ^c , 0.89* ^c , 0.93, 0.97	1.31	1-14	0.97	+5.4	+6.4	+1.4	Najafi et al. (2017)
0.75 ^e , 0.82, 0.90	1.03	0-42	0.90	+1.1	+3.2	+1.7	Kheiri and Alibeyghi (2017)
0.3 ^m , 0.6, 0.9	1.20	1-35	0.90	+2.8	+5.5	+2.5	Al-Hayani (2017)
0.77 ^c , 0.88, 1.08	1.11	0-21	0.88	-1.0 ²	+0.6 ²	+2.8	Chen et al. (2017)
0.62 ^{cn} , 0.75, 0.94, 1.12	1.11	0-42	0.75	+1.3	+6.8	+5.4	Min et al. (2017)
0.49 ^m , 0.90	0.72	13-21	0.90	+28.0	+71.2	+33.0	Zhang et al. (2016)
0.641 ^c , 0.68*, 0.843	0.994	25-42	0.843	-4.1	+2.2	+5.2	Valizade et al. (2017)
0.69 ^c , 0.71, 0.74*, 0.76, 0.79	1.03	0-42	0.69	-1.4	+3.8	+4.9	Azzam and El-Gogary (2015)
0.74 ^e , 0.81, 0.89, 0.97	1.08	0-42	0.81	-7.0	-1.5	+5.1	Eftekhari et al. (2015)
0.74 ^e , 0.79, 0.81, 0.84	1.20	0-42	0.84	+3.1	+10.5	+4.7	Shirzadegan et al. (2015)
0.857 ^m , 0.956	1.24	0-10	0.956	ND	+4.2	ND	de Filho et al. (2015)
0.74 ^e , 0.78	1.12	1-42	0.78	-1.6	+0.1	+0.6	Rezaei pour and Gazami (2014)
0.71 ^m , 0.77, 0.84	1.03	1-42	0.77	+3.1	+5.1	+2.0	Abbasi et al. (2014)
0.81 ^m , 0.89	1.19	21-35	0.77	-0.2	+1.3	+1.4	Rojas et al. (2013)
0.6 ^c , 0.67, 0.74*, 0.81, 0.88, 0.95	0.99	21-42	0.81	+5.6	+12.2	+12.7	Roudbaneh et al. (2013)
0.72 ^c , 0.74*, 0.77, 0.79	1.0	1-42	0.79	+2.5	+5.0	+3.4	Moghaddam et al. (2011)
0.63 ^m , 0.67	1.06	9-37	0.67	+4.76	+6.8	+1.9	Star et al. (2012)
0.80 ^e , 0.87, 0.94, 1.01	1.10	1-14	0.87	+0.6	+1.1	+0.4	Moghaddam et al. (2011)
0.55 ^c , 0.80*, 1.05	1.30	1-21	0.80	+3.0	+68.1	+9.5	Chee et al. (2012)
0.4 ^c , 0.5, 0.6, 0.7, 0.8*, 0.90, 1, 1.1	1.28	0-21	0.90	+107.2	+256.5	+78.3	Zaefarian et al. (2008)
0.51 ^m , 0.58, 0.65, 0.72, 0.79, 0.86	1.16	21-42	0.86	-13.3	+103.1	+133.7	Corzo et al. (2007)
0.52 ^m , 0.59, 0.66, 0.73, 0.80, 0.87	1.08	21-42	0.80	+10.3	+51.6	+41.1	Kidd et al. (2004)
0.54 ^m , 0.60, 0.72	1.15	22-42	0.72	+23.2	+27.1	+3.0	Ciftci and Ceylan (2004)
0.6 ^c , 0.7, 0.80*	1.28	5-15	0.80	+23.0	+54.8	+26.2	Kidd et al. (2001)
0.50 ^c , 0.56, 0.62, 0.68, 0.74, 0.80*	0.92	0-56	0.80	-2.4	+11.8	+2.1	Dozier et al. (2000)

¹FI = Feed intake, BWG = Body weight gain, FCR = Feed conversion ratio.

²These values indicates the difference between control and optimal.

^cControl diet.

^mNRC Recommended level.

ⁿNRC Recommendation not provided.

ND = Not determined.

Table 2. Summary of literature studies on the effects of different dietary levels of threonine on gut morphology in broilers

Threonine inclusion level (% of the diet)	Lysine level	Age (d)	Optimal (%)	Gut morphology ^{1,2} (%)				References
				VH	VW	CD	VCR	
0.77 ^m , 0.88, 1.08	1.11	0-21	1.08(J) ³	+18.3	ND ⁴	-12.5	+33.0	Chen et al. (2017)
0.77 ^m , 0.88, 1.08	1.11	0-21	1.08(I) ⁵	+21.5	ND	-8.0	+31.8	Chen et al. (2017)
0.65 ^c , 0.89 ^a , 0.93, 0.97	1.31	1-14	0.89(D) ⁶	+15.0	-5.2	-2.0	+1.81	Najafi et al. (2017)
0.49 ^m , 0.90	0.72	13-21	0.90(J)	+57.6	ND	+18.9	+35.7	Zhang et al. (2016)
0.74 ^a , 0.81, 0.89, 0.97	1.08	0-42	0.89 (J)	+0.08	+1.9	-0.3	+0.4	Eftekhari et al. (2015)
0.74 ^a , 0.79, 0.81, 0.84	1.20	0-42	0.81 (D)	+4.2	-0.8	-0.2	+4.3	Shirzadegan et al. (2015)
0.74 ^a , 0.79, 0.81, 0.84	1.20	0-42	0.84 (J)	+7.4	+1.8	+7.2	+0.2	Shirzadegan et al. (2015)
0.74 ^a , 0.79, 0.81, 0.84	1.20	0-42	0.84 (I)	+19.1	-1.2	-3.7	+23.4	Shirzadegan et al. (2015)
0.71 ^m , 0.77, 0.84	1.03	1-42	0.77(J)	+6.1	ND	-9.0	+12.6	Abbasi et al. (2014)
0.72 ^c , 0.74 ^a , 0.77, 0.79	1.00	1-42	0.79(J)	+7.5	+3.5	+10.4	ND	Rezaei pour et al. (2012)
0.72 ^c , 0.74 ^a , 0.77, 0.79	1.00	1-42	0.79(I)	+12.2	+4.1	+13.7	ND	Rezaei pour et al. (2012)
0.80 ^a , 0.87, 0.94, 1.01	1.10	1-14	0.87(J)	+2.5	ND	+1.4	NE ⁷	Moghaddam et al. (2011)
0.55 ^m , 0.82, 1.06	1.30	1-21	0.82(I)	+33.5	ND	+17.0	+20.5	Chee et al. (2012)
0.4 ^c , 0.5, 0.6, 0.7, 0.8 ^a , 0.9, 1, 1.1	1.28	0-21	1.1(D)	+12.4	ND	+2.0	ND	Zaefarian et al. (2008)
0.4 ^c , 0.5, 0.6, 0.7, 0.8 ^a , 0.9, 1, 1.1	1.28	0-21	1.1(I)	+6.4	ND	+13.7	ND	Zaefarian et al. (2008)
0.4 ^c , 0.5, 0.6, 0.7, 0.8 ^a , 0.9, 1, 1.1	1.28	0-21	1.1(I)	+12.4	ND	+28.1	ND	Zaefarian et al. (2008)

^cControl diet.¹VH = Villus height, VW = Villus width, CD = Crypt depth, VCR = Villus height to crypt depth ratio.²± These value indicates the difference between control and optimal.³(J) = Jejunum.⁴ND = Not determined.⁵(I) = Ileum.⁶(D) = Duodenum.⁷NE = No effect.^{*}NRC Recommended level.[†]NRC Recommendation not provided.

Table 3. Summary of literature studies on the effects of different dietary levels of threonine on weight of immune organs and immunity status in broilers

Threonine inclusion level (% of the diet)	Lysine level	Age (d)	Optimal (%)	Weight of immune organ (%)			Antibody titer (%)	Disease	References
				Spleen	Bursa	Thymus			
0.77 ^m , 0.88, 1.08	1.11	1-21	0.88	+66	+4.1	-1.5	ND ²	ND	Chen et al. (2017)
0.641 ^m , 0.675, 0.843	0.994	25-42	0.675	ND	ND	ND	+7.0	IBD ³	Valizade et al. (2016)
0.74 ^e , 0.81, 0.89, 0.97	1.08	1-42	0.89	NE ⁴	+25	ND	+14.3	NDV ⁵	Eftekhari et al. (2015)
0.69 ^z , 0.71, 0.74 ^z , 0.76, 0.79	1.03	0-42	0.69	+12	-24	ND	ND	ND	Azzam and El-Gogary (2015)
0.641 ^m , 0.675, 0.843	0.994	1-35	0.843	+20	+2.5	ND	ND	ND	Valizade et al. (2014)
0.71 ^m , 0.77, 0.84	1.03	1-42	0.84	ND	ND	ND	+3.3	NDV	Abbasi et al. (2014)
0.67 ^m , 0.70, 0.73, 0.77	1.02	1-42	0.77	ND	+217	ND	ND	ND	Estalkhizir et al. (2013)
0.6 ^z , 0.67, 0.74 ^z , 0.81, 0.88, 0.95	0.99	21-42	0.95	+42	ND	ND	+39.0	IBD	Roubaneh et al. (2013)
0.70 ^m , 0.77, 0.83, 0.88	1.10	1-35	0.88	+66	-25	NA	NA	ND	Zhang and Kim (2014)
0 ^m , 0.25, 0.70, 0.75	1.00	1-42	0.75	ND	ND	ND	+71.0	NDV	Rezaei pour et al. (2012)
0.64 ^m , 0.67, 0.7, 0.73, 0.76	1.00	1-41	0.67	ND	ND	ND	+21.0	NDV	Rao et al. (2011)
0.51 ^m , 0.72	1.16	1-42	0.51	+16	+15	-21.0	ND	ND	Corzo et al. (2007)
0.96 ^m , 1.02, 1.12	1.30	1-35	1.02	+17	+17	+3.0	ND	ND	Mandal et al. (2006)
0.60 ^z , 0.70, 0.80 ^z	1.28	1-15	0.6	+4	+15	+12.0	ND	ND	Dozier et al. (2000)
0.68 ^z , 0.74, 0.80 ^z , 0.86	1.10	1-18	0.80	ND	ND	ND	+6.0	NDV	Kidd et al. (1997)
0.3 ^m , 0.5, 0.7, 0.9, 1.1	1.4	1-18	1.1	ND	ND	ND	+77.0	NDV	Bhargava et al. (1971)

¹± These values indicates the difference between control and optimal.

²Not determined.

³Infectious bursal disease.

⁴NE = No effect.

⁵Newcastle disease virus.

^zControl diet.

^mNRC = Recommended level.

ⁿNRC Recommendation not provided.

ND = Not determined.

Table 4. Summary of literature studies on the effects of different dietary levels of threonine on carcass characteristics in broilers

Threonine inclusion level (% of the diet)	Lysine level	Age (d)	Optimal (%)	Carcass characteristics ¹ (%)							References
				Carcass	DS ²	Breast	Thigh	Pancreas	Liver		
0.3 ^{en} , 0.6, 0.9	1.20	1-35	0.90	+3.66	ND	+2.3	-1.1	ND	-16.3	Al-Hayani (2017)	
0.75 ^e , 0.82, 0.90	1.03	0-42	0.90	+5.3	ND	+1.6	+1.0	ND	+1.3	Kheiri and Alibeyghi (2017)	
0.77 ^{en} , 0.87	1.26	0-35	0.87	ND	+1.6	+26	-9.6	ND	ND	El-Faham et al. (2017)	
0.69 ^e , 0.71, 0.74 ^e , 0.76, 0.79	1.03	0-42	0.71	ND ³	ND	ND	ND	+5.1	+15.4	Azzam and El-Gogary (2015)	
0.74 ^e , 0.79, 0.81, 0.84	1.20	0-42	0.79	ND	ND	+1.0	-0.5	+8.0	+8.0	Shirzadegan et al. (2015)	
0.74 ^e , 0.81, 0.89, 0.97	1.08	0-42	0.97	ND	ND	+8.0	-1.8	+3.6	+1.1	Eftekhari et al. (2015)	
0.74 ^e , 0.77	1.12	1-42	0.74	ND	ND	+0.4	+0.2	+5.0	+10	Rezaeipour and Gazani (2014)	
0.71 ^{en} , 0.77, 0.84	1.03	1-42	0.77	+1.6	+2.7	+5.3	ND	N.E. ⁴	ND	Abbasi et al. (2014)	
0.67 ^{en} , 0.70, 0.73, 0.77	1.02	1-42	0.77	+11.5	ND	+29.8	2.0	ND	+3.5	Estalkhizir et al. (2013)	
0.72 ^e , 0.74 ^e , 0.77, 0.79	1.00	1-42	0.79	ND	ND	+6.3	-1.4	ND	-5.0	Rezaeipour et al. (2012)	
0.51 ^{en} , 0.58, 0.65, 0.72, 0.79, 0.86	1.16	21-42	0.86	+57.5	ND	+77.0	ND	ND	ND	Corzo et al. (2007)	
0.52 ^{en} , 0.59, 0.66, 0.73, 0.80, 0.87	1.08	21-42	0.87	+31.7	ND	+43.0	ND	ND	ND	Kidd et al. (2004)	
0.54 ^{en} , 0.60, 0.66, 0.72	1.15	22-42	0.72	+8.0	+4.4	+8.0	-5.6	+8.4	-26.5	Ciftci and Ceylan (2004)	
0.68 ^e , 0.81, 0.95, 1.08, 1.22	0.97	1-48	1.08	-0.2	ND	+0.6	ND	ND	ND	Kidd et al. (2002)	
0.50 ^e , 0.56, 0.62, 0.68 ^e , 0.74, 0.80	0.92	42-56	0.62	ND	+6.5	ND	+9.3	ND	ND	Kidd et al. (1997)	

^cControl diet.¹±These values indicates the difference between control and optimal.²DS = Drumstick.³ND = Not determined.⁴NE = No effect.^{*}NRC Recommended level.ⁿNRC Recommendation not provided.

After absorption, Thr is used for gut protein synthesis, which groups together the inner side of the gut as mucin, and protects gut from anti-nutritional factors and pathogens (Lee et al., 2007). This may explain the better developed gut in broilers fed higher than recommended levels of dietary Thr. Synthesis of *de novo* mucin and mucosal protein is faster in the presence of Thr in the lumen, which indicates the importance of Thr for proper functioning of the gut (Nichols and Bertolo, 2008). In portal drained viscera, protein synthesis requires higher amount of Thr, compared with other amino acids, that further highlights the importance of Thr for broilers (Schaart et al., 2005). Geyra et al. (2001) reported that development of CD was important to speed up gut maturation as well as for renewal of intestinal cells. The increased intestinal mucin secretion (as a result of developed crypt, which contains mucin secreting goblet cell) as well as proliferation of enterocytes (as a result of Thr) also increases absorption of nutrients (Ospina-Rojas et al., 2013). The small intestine uses about 30 to 50% of Thr along with other amino acids (arginine, proline, isoleucine, valine, leucine, methionine, Lys, phenylalanine, glycine and serine) and does not spare it for other extra-intestinal tissues (Wu, 1998). For rapid turnover of intestinal tissues, and to increase surface area for nutrients absorption, therefore, higher than NRC recommended level of Thr is required for a better gut health.

Effects on immunity

Immunity plays a major role in achieving maximum growth performance of broilers. The immune status of the broilers improves, with an improvement in the function of immune organs (Corzo et al., 2007; Zhang et al., 2016). For example, a greater activity and an increase in weight of immune organs including thymus, bursa, and spleen can result in more antibodies production. The NRC recommendations for Thr are normally established for healthy birds reared in ideal management conditions, whereas in commercial production systems birds are generally exposed to various types of stresses (Roudbaneh et al., 2013). Under unhygienic environmental conditions, dietary Thr requirements are increased to sustain the maintenance necessities in the gut mucosa (Corzo et al., 2003) and to enhance immunity (Bhargava et al., 1971). Roudbaneh et al. (2013) reported that Thr requirement under stress and unhygienic environmental conditions were increased to 0.81% to maintain growth performance including FCR. Corzo et al. (2007), similarly, described that total dietary Thr requirements to improve growth performance were 0.71 to 74% on new litter, whereas for used litter these were 0.73 to 0.78%. During finisher phase due to old litter and poor management conditions, immunity of birds is compromised. It has been reported that dietary Thr requirements increase in birds suffering from disease including clostridial infection (Star et al., 2012). Increasing Thr concentration in diet might enhance immune organs growth, stimulate the synthesis of antibodies and relieve immune stress caused by *Escherichia coli* challenge or Newcastle disease (ND) virus (Azzam et al., 2012; Trevisi et al., 2015). Chen et al. (2017) conducted a trial to observe the effect of Thr on broilers immunity by feeding 0.77 (control), 0.88 and 1.08% of total dietary Thr. It was observed that broilers fed with diet containing 0.88% Thr had 66% greater spleen and 4.1% greater bursa weights, whereas a numerical (1.5%) reduction in thymus weight was observed compared with those fed the control diet. Valizade et al. (2016) fed 0.641 (control), 0.675 and 0.683% of

total dietary Thr to broilers and observed its effect on their immunity. It was found that the broilers fed diet containing 0.675% Thr had 7% greater antibody titer against infectious bursal disease (IBD) compared with those fed 0.641% of Thr. Eftekhari et al. (2015) used a NRC recommended (0.74%) and three higher levels (0.81, 0.89 and 0.97%) of total dietary Thr in broiler rations to investigate its effect on bird's immunity. Broilers fed diet containing 0.89% Thr had 25% greater bursa weight, 14.3% greater ND antibodies titer, whereas spleen weight remained unchanged compared with birds receiving NRC recommended level of Thr in the feed. In another study, Azzam and El-Gogary (2015) used 0.71, 0.74, 0.76 and 0.79% of total dietary Thr, along with a control diet containing 0.69% Thr to evaluate its effect on immunity of broilers. It was observed that broilers fed diet containing 0.69% of total dietary Thr had a numerically (3%) higher spleen weight, and 24% lower bursa weight compared with those fed 0.74% of Thr. Valizade et al. (2014) fed 0.675 and 0.843% of total dietary Thr, along with a control diet containing 0.641% Thr, in broilers to evaluate its effect on immunity. It was reported that broilers fed diet having 0.675% total dietary Thr had 20% greater spleen and 2.5% greater bursa weights compared with those fed control diet. In another study, Zhang and Kim (2014) used 0.70, 0.77, 0.83 and 0.88% of total dietary Thr to observe its influence on immunity in broilers. The later authors concluded that broilers fed diet containing 0.80% Thr had 66% greater spleen weight and 25% smaller bursa weight compared with those fed the control diet containing 0.70% Thr. Estalkhzir et al. (2103) evaluated the effect of higher levels i.e. 0.70, 0.73, and 0.77% of dietary Thr on broiler immunity in comparison with a control (0.67%). The study found that broilers fed with 0.77% total dietary Thr had 217% greater bursa weight compared to those fed with recommended level of Thr. Rao et al. (2011) studied the effect of 0.64, 0.67 (control), 0.70, 0.73, and 0.76% of total dietary Thr on broiler immunity. It was reported that broilers fed diet containing 0.67% total Thr had 21% increase in ND antibody titer compared with those fed control diet. Data on the effect of different levels of dietary Thr on broilers immunity is summarized in Table 3. Mandal et al. (2006) compared the effect of feeding 0.96% (control), 1.02% and 1.12% total dietary Thr on broilers immunity. It was observed that the broilers fed with the 1.02% total dietary Thr had 16% greater spleen weight, 17% greater bursa weight and 7% greater thymus weight compared with those fed control diet containing 0.96% total Thr.

From the above cited literature, it is evident that immunity in broilers is enhanced by providing Thr higher than NRC recommendation, because extra dietary Thr promotes the growth of immune organs, especially bursa and spleen that stimulates the synthesis of immunoglobulin resulting in an improved antibody titer against various diseases including ND and *Escherichia coli*.

Effects on carcass characteristics

After slaughtering, except blood and feathers, body of eviscerated bird is called carcass. In broilers, Thr requirement for carcass yield is variable, depending upon age, strain, sex of broilers, CP content of feed, type and proportion of dietary ingredients used (Barkley and Wallis, 2001). The improved carcass characteristics may be due to increased amount of essential amino acids (Thr) in diet (Estalkhzir et al., 2013). Al-Hayani (2017) conducted a trial by feeding three different levels (0.3, 0.6

and 0.9%) of Thr in broilers diet. The later authors concluded that the broilers fed diet containing 0.90% of Thr resulted in an enhanced carcass weight by 3.7%, breast weight by 2.3%, whereas thigh weight was reduced by 1.1%, respectively. Similarly, El-Faham et al. (2017) executed a trial using two levels (0.77 and 0.87%) of Thr in broiler diet. The results revealed that the broilers fed diets containing 0.87% of Thr showed 28% higher breast weight, 1.6% increased drumstick weight, whereas 9.6% reduced thigh weight compared with those fed diet containing 0.77% of Thr. Shirzadegan et al. (2015) executed a trial to evaluate effect of different levels (0.74, 0.79, 0.81 and 0.84%) of total dietary Thr on broilers carcass characteristics. The results indicated that broilers fed diets containing 0.79% total dietary Thr had higher meat by 1%, liver weight by 8% and pancreas weight by 8%, and reduced thigh weight by 0.5% compared with the control diet containing 0.74% total dietary Thr. In another study, Abbasi et al. (2014) used 0.77 and 0.84% of dietary Thr, along with the strain (Ross-308) recommended (0.71%) total Thr level during finisher phase, to evaluate its influence on carcass characteristics in broilers. Broilers fed diet containing 0.77% total dietary Thr increased carcass weight by 1.6%, drumstick weight by 2.7%, and breast meat by 5.3% compared with those fed recommended level of Thr. The improvement in carcass weight and quality characteristics may be related to the role of Thr on digestive enzymes function and intestinal mucosa development. Rezaei-pour and Gazani (2014) compared the effect of 0.74 and 0.77% total dietary Thr on carcass characteristics in broilers. It was observed that broilers fed 0.77% total dietary Thr showed increased breast meat by 0.4%, pancreas weight by 5%, and liver weight by 10% compared with those fed control diet containing 0.74% total dietary Thr. Estalkhzir et al. (2013) evaluated the effect of higher levels of total dietary Thr, i.e. 0.70, 0.73 and 0.77% in comparison with a control diet containing 0.67% Thr. Broilers fed diet having 0.77% of Thr showed best results, with an increase of 11.5% carcass weight, 29.8% breast meat, and a numerically higher (2%) thigh and (3.5%) liver weights, compared with those fed control diet. Kidd et al. (2004) conducted an experiment by using 0.59, 0.66, 0.73, 0.80 and 0.87% of total dietary Thr, along with a control diet having 0.52% Thr, to observe its effect on carcass characteristics in broilers. The later authors found that broilers fed diet containing 0.87% of Thr had a 31.7% higher carcass weight and 43% higher breast meat compared with those fed control diet. It is evident from these studies that for a better growth and improved carcass characteristics, a higher than the currently recommended level of Thr in broiler diet is necessary. Threonine is involved in building muscle mass along with serine and improves gut morphology resulting in better absorption of nutrients, and ultimately improves carcass characteristics of broilers. The effects of different levels of dietary Thr on carcass characteristics in broilers are summarized in Table 4.

Conclusions

Finding of literature data on the influence of dietary Thr on growth performance, gut morphology, immune status and carcass characteristics of broilers is summarized in this review. From a number of direct comparisons, and indirect comparison across the studies, it is clear that inclusion of dietary Thr, higher than NRC recommenda-

tion (0.74–0.81%), enhances the growth performance and improves gut morphology, immunity and carcass characteristics in broilers. According to the literature data discussed in this review, the minimal and maximal total dietary Thr for optimum growth performance, in normal conditions for healthy birds, during starter (0 to 21 d) phase were 0.80 and 0.90%, in grower (22 to 42 d) phase 0.72 and 0.86%, and 0.67 to 0.84% during the whole (0 to 42 d) period. For a healthy gut during normal conditions in healthy birds, the minimum and maximum total dietary Thr requirements in starter (0 to 21 d) phase were 0.82 and 1.1%, whereas during the whole (0 to 42 d) phase these were 0.77 and 0.89%. For a better immune response, the minimum and maximum total dietary Thr requirements, during the starter (0 to 21 d) phase were 0.6 and 1.1%, and during the grower (22 to 42 d) phase were 0.675 and 0.95%, whereas these were 0.67 and 0.89% during the whole (0 to 42 d) period. The minimum and maximum levels of total dietary Thr in healthy birds' feed for a better carcass characteristics during grower (21–42 d) phase were 0.71 and 1.08%, whereas during the whole (0–42 d) phase these requirements were 0.72 to 0.87%. These dietary Thr requirements were, however, increased during disease, stress and in unhygienic rearing conditions. Dietary Thr level is, therefore, still contradictory in literature, because there are no defined levels of Thr, which could be fed to broilers for the best growth performance. The data regarding influence of dietary Thr on ileal and total tract digestibility of protein in broilers is still inadequate. In future, there is a dire need to conduct further studies to better elaborate the significance of dietary Thr on ileal and total tract protein digestibility in broilers.

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Received: 18 XII 2017

Accepted: 11 IV 2018