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EXIGÊNCIA DE TREONINA PARA SUÍNOS EM CRESCIMENTO: UMA ABORDAGEM META-ANALÍTICA

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EXIGÊNCIA DE TREONINA PARA SUÍNOS EM CRESCIMENTO: UMA ABORDAGEM META-ANALÍTICA

Dissertação apresentada como requisito para obtenção do Grau de Mestre em Zootecnia, na Faculdade de Agronomia, da Universidade Federal do Rio Grande do Sul.

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2020

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EXIGÊNCIA DE TREONINA PARA SUÍNOS EM CRESCIMENTO: UMA ABORDAGEM META-ANALÍTICA[1]

Autora: Carolina Haubert Franceschi
Orientadora: Ines Andretta

Este trabalho teve como objetivo realizar uma extensa revisão sistemática, seguida de meta-análise, acerca da exigência de treonina (Thr) para suínos em crescimento. Os estudos foram levantados em plataformas de pesquisa indexadoras como PubMeb, Web of Science e Scopus. A base de dados foi composta por 38 estudos de dose-resposta publicados entre os anos de 1982 a 2019. A base de dados foi dividida em inicial (5 – 25 kg PV), crescimento (25 – 75 kg PV) e terminação (75 – 135 kg PV). Os estudos selecionados apresentavam concentração de Lisina e outros aminoácidos que atendiam as exigências em 90% e 100%, respectivamente, das recomendações do NRC. Devido às diferenças entre os estudos, a concentração de treonina e os critérios de resposta foram padronizados. A variável resposta para ganho de peso médio diário (GMD%) foi expressa relativa à melhor resposta presente no estudo (ex: maior ganho de peso médio diário considerado 100%). A variável independente (concentração de treonina) foi expressa em relação ao manual de exigências do NRC (2012) (ex: exigências do NRC foram consideradas 100%). Regressões não lineares foram ajustadas utilizando o procedimento NLIN do programa estatístico SAS. Os dados médios observados nos estudos foram ajustados utilizando o modelo linear-plateau (LRP) e quadrático linear-plateau (QP). O ponto de quebra do LRP foi interpretado como a concentração relativa de Thr necessária para maximizar o GMD%. Considerando os estudos individualmente, a concentração relativa de Thr para maximizar o GMD% variou de 32 a 117% de acordo com as exigências previstas pelo NRC (2012). Em 10 experimentos de dose-resposta, os animais não responderam ao aumento da concentração relativa de Thr na dieta. Ambos resultados (variação de exigência e falta de resposta à Thr) indicam uma grande variação nas exigências de treonina que compõem a base de dados. O requerimento ideal de treonina para a fases inicial e de crescimento foi de 0,019 e 0,015 g/g de ganho de peso, respectivamente de acordo com o modelo QP. Para a fase final os resultados foram ajustados pelo modelo QR e apresentaram 0,015 g de treonina por cada g de

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[1] Dissertação de mestrado em Zootecnia – Produção Animal, Faculdade de Agronomia, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brasil (49p) Março, 2020.

ganho de peso. Considerando a análise de todo banco de dados, a concentração relativa de treonina que maximizou o GMD% foi 14,25% maior na fase inicial, 11,36% maior na fase de crescimento, e 11,98% maior na fase de terminação quando comparadas às exigências previstas no NRC (2012). Esses resultados representam uma subestimação das exigências de treonina para suínos durante todas as fases de criação dos suínos.

Palavras-chave: Aminoácidos; Meta-análise; Nutrição de precisão; Revisão

THREONINE REQUIREMENTS FOR GROWING PIGS: A META-ANALITYCAL APPROACH[2]

Author: Carolina Haubert Franceschi
Supervisor: Ines Andretta

This study aimed to perform an extensive systematic review, followed by a meta-analysis, on the requirement of threonine (Thr) for growing pigs. The studies were search in electronic databases PubMed, Web of Science, and Scopus. The database contained 38 dose-response studies published between 1982 and 2019. The database was divided in weaning (5 - 25 kg BW), growing (25 – 75 kg BW) and finishing (75 – 135 kg BW). The selected studies had a concentration of Lysine and other amino acids that met the requirements in 90% and 100%, respectively, of the NRC requirements. Due to differences between studies, threonine concentration and response criteria were standardized. The response variable for average daily gain (ADG%) was expressed in relation to the greatest response present in the study (i.e greatest ADG was considered 100%) and in kg per day (ADG). The independent variable (threonine concentration) was expressed in relation to NRC (2012) requirements manual (i.e NRC requirements were considered 100%). Results of Thr intake were presented in grams per day. Non-linear regressions were fitted by NLIN procedure of SAS for each dose-response study using the linear-plateau (LRP) and quadratic linear-plateau (QP) model. The breaking point in the LRP was interpreted as the relative SID Thr concentration, required to maximize ADG%. Considering the studies individually, SID Thr concentration to maximize ADG% varied from 32 to 117% of the NRC (2012) recommendation. In 10 experiments, pigs did not respond to relative SID Thr concentration. Both results (range of requirements and lack of response to Thr) indicated a large variance in the Thr requirements the among studies that composed the database. The ideal threonine requirements for weaning and growing was 0.019 and 0.015 g/g of weight gain, respectively according to the QP model. For the finishing phase the results were adjusted for the LRP model and presented 0.015 g of threonine by each g of weight gain. Considering the entire database, the results of the QP model showed that relative SID Thr concentration that maximizes ADG% were 14.25% higher in weaning phase, 11.36% higher in growing phase, and 11.98 higher in finishing phase

[2] Master of Science dissertation in Animal Science, Faculdade de Agronomia, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil. (49p) March, 2020.

than the recommendations of NRC (2012), which represents an underestimation of Thr requirements for pigs during all phases.

Keywords: Amino acids; Meta-analysis; Precision feeding; Review

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RELAÇÃO DE ABREVIATURAS E SÍMBOLOS

AA - Amino acid

AIC – Akaike information criterion

ADFI – Average daily feed intake

ADG – Average daily gain

BSAS – The British society of animal science

BW – Body weight

GMD – Ganho médio diário

GRR – Gauge repeatability and reproducibility

GRU – Gated recurrent unit

INRA - Institut national del la recherche agronomique

Lys – Lysine

LRP – Linear Plateau

NLIN – Nonlinear

NRC – National Research Council

NSNG – National swine nutritional guide

PV – Peso vivo

QP – Quadratic Plateau

SeqA – Sequence A

SID – Standardized ileal digestibility

S2e – Standard error

Thr – Threonine

USA – United States of America

VSP – Danish agriculture & food council

CAPÍTULO I

INTRODUÇÃO

A produção de carne suína alcançou destaque no cenário nacional nos últimos anos. O Brasil produziu cerca de 3,9 milhões de toneladas em 2018, o que representou um aumento de cerca de 6% na produção em relação ao ano de 2017 (ABPA, 2019). No quesito exportação, o Brasil ocupa a quarta colocação, atrás apenas da União Europeia, Estados Unidos e Canadá (FAO, 2019).

A nutrição é um dos pilares da suinocultura industrial e a alimentação dos animais chega a representar cerca de 60 a 70% dos custos totais de produção dos suínos. Considerando isto, conhecer as exigências nutricionais dos animais se torna um fator importantíssimo para a sustentabilidade econômica da produção de suínos.

As exigências nutricionais podem ser definidas como a quantidade de um nutriente ou substância necessária para atingir determinado objetivo de produção, como por exemplo maximizar ganho de peso ou melhorar a conversão alimentar (WU, 2017). Além disso, as exigências nutricionais não são fixas e podem variar entre suínos de uma mesma população (POMAR, 2007) e, também, de acordo com o status sanitário (JAYARAMAN, HTOO E NYACHOTI., 2015) e ambiental (FERGUSON et al., 2000) em que esse animal se encontra.

A treonina é um aminoácido essencial na dieta de suínos, e sua exigência diária pode ser encontrada em diversos manuais disponíveis, tais como as tabelas brasileiras de aves e suínos (ROSTAGNO, 2017) e as tabelas publicadas pelo National Research Council (NRC, 2012). Entretanto, os manuais apresentam valores médios de uma população, sendo insuficientes ou pouco flexíveis para garantir máximo desempenho dos animais. Alimentar os animais sem conhecer suas exigências exatas pode aumentar a excreção de nutrientes no ambiente e aumentar os custos de produção significativamente (POMAR et al., 2014).

Considerando estes fatores, ferramentas como revisão sistemática e meta-análise podem se tornar aliadas para estabelecer níveis nutricionais ajustados aos diferentes aspectos que influenciam nas respostas dos animais. Assim compilando inúmeras informações acerca de um único assunto para, no fim, gerar dados que sejam coerentes e condizentes com a realidade observada. Desta forma, este trabalho tem como objetivo avaliar as respostas dos animais a diferentes níveis de treonina para suínos através de uma revisão sistemática e meta-análise.

REVISÃO BIBLIOGRÁFICA

1.1 Exigências nutricionais

A exigência nutricional pode ser definida como a quantidade de um nutriente necessário para atingir um objetivo específico da criação de animais, seja ganho de peso, produção de leite ou conversão alimentar. Exigências nutricionais não são consideradas quantidades fixas, uma vez que o animal pode ser condicionado a inúmeras circunstâncias que podem alterar a percepção de exigências (FULLER, 2004). Uma característica fundamental da produção animal moderna é fornecer nutrientes de maneira apropriada para que o desempenho e a eficiência dos animais possam ser otimizados.

As exigências nutricionais podem ser estimadas utilizando dois métodos: empírico e fatorial. Ambos os métodos utilizam uma população como base do estudo de exigências. O método empírico, que é comumente mais utilizado, estima as exigências para maximizar ou minimizar determinada característica (ex: ganho de peso ou deposição de proteína) através de experimentos de dose-resposta. No método dose-resposta, as exigências de um nutriente são estimadas avaliando a resposta de grupo de animais submetidos a diferentes concentrações de um nutriente na dieta durante um determinado período. Essa resposta é de natureza curvilínea, pois considera as diferenças entre os animais para mantença e potencial de crescimento (Figura 1.1).

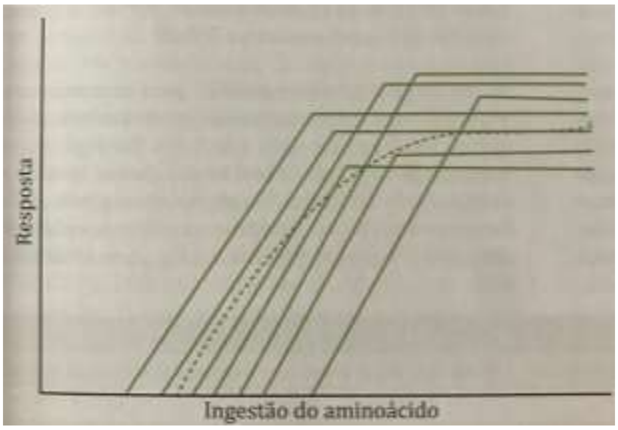

Figura 1.1 Relação entre respostas e aumento da ingestão de aminoácidos de diferentes indivíduos (linhas contínuas) e média da população (linha tracejada).

Fonte: SAKOMURA et al., (2014)

Modelos matemáticos são constantemente utilizados na nutrição animal e, principalmente, aplicados em experimentos de dose-resposta. Para isto, a resposta de um nutriente, como por exemplo um aminoácido, deve ser assumida como linear entre o aumento da ingestão do aminoácido e a resposta esperada a esse aumento, até o momento de cessamento da resposta em relação ao nutriente (DUMAS, DIJKSTRA, E FRANCE, 2008). Assim, modelos como o *broken line*, com uma resposta-platô podem ser representativos de cada indivíduo ou, com alguns ajustes, da média da população (SAKOMURA et al.,2013).

Por sua vez, o método fatorial associa a exigência necessária para mantença e para produção, como ganho de peso. Esse método permite explicar as modificações do metabolismo no tempo, como taxas de crescimento e peso corporal que se alteram com o avançar da idade dos animais (SAKOMURA et al., 2014). Neste método, o indivíduo médio da população é utilizado como referência para estimar as exigências do restante. Por este fato, populações heterogêneas devem usar com cautela as exigências nutricionais previstas por este método.

A exigência de proteína é indispensável para animais em crescimento, porém são os aminoácidos que possuem um papel importante nas diversas vias metabólicas presentes no organismo animal. Então, conhecer a exigência destes aminoácidos torna-se imprescindível.

1.2 Exigências de aminoácidos

Os aminoácidos são unidades estruturais das proteínas formados por dois grupos funcionais diferentes: uma amina e uma carboxila. Durante o processo de digestão, as proteínas são quebradas em aminoácidos e peptídeos, estes que posteriormente serão absorvidos pelo organismo e utilizados para formar novas proteínas, por exemplo, em forma de músculo (NELSON E COX, 2018). Em dietas balanceadas, os níveis de aminoácidos são ofertados de acordo com a exigência dos animais, especialmente para os aminoácidos essenciais.

Suínos durante todos os estágios do seu ciclo de vida necessitam de aminoácidos que facilitem seu crescimento. As exigências de aminoácidos podem ser determinadas assumindo que todos aminoácidos sejam fornecidos em quantidades suficientes e sem excessos, baseando-se no conceito de proteína ideal (SAKOMURA, 2014). Porém, na prática, podemos observar que dietas formuladas para atingir um menor custo podem apresentar deficiência ou excesso de alguns aminoácidos. Na Tabela 1.1 podemos observar aminoácidos essenciais, suas propriedades químicas e sua ordem de limitação.

Tabela 1.1 Aminoácidos essenciais de acordo com a geral ordem de limitação em dietas práticas comuns para suínos em crescimento.

Aminoácido essencial	Propriedade Química	Ordem de limitação
Lisina	Aminoácido base	Primária
Treonina	Aminoácido hidroxi	
Metionina	Aminoácido sulfurado	
Metionina + cistina	Aminoácido sulfurado	
Triptofano	Aminoácido indol	
Isoleucina	Aminoácido de cadeia ramificada	Secundária
Leucina	Aminoácido de cadeia ramificada	
Valina	Aminoácido de cadeia ramificada	
Histidina	Aminoácido imidazólico	
Fenilalanina	Aminoácido aromático	
Fenilalanina + tirosina	Aminoácido aromático	

Em dietas para suínos a base de milho e farelo de soja, o primeiro aminoácido limitante é a lisina, mas outros aminoácidos também podem se tornar limitantes em razão da formulação e dos ingredientes utilizados. Para estimar a exigência dos outros aminoácidos, a lisina é utilizada como padrão em virtude de ser destinada basicamente para síntese proteica, sendo o componente principal do tecido magro de suínos. Assim, todos os outros aminoácidos são estimados a partir da exigência de lisina (D'MELLO, 2003).

A ingestão de quantidades desproporcionais de aminoácidos pode resultar em efeitos adversos como deficiência, toxicidade e antagonismo (D'MELLO, 2003). Outro fator importante é o aumento do custo de produção e do impacto ambiental causado por esse desbalanço de nutrientes fornecidos aos animais (POMAR et al., 2014). Assim, conhecer e entender as exigências dos animais é essencial para uma produção que, além de ser sustentável, seja coerente com o uso dos nutrientes disponíveis.

Diversas tabelas e manuais estão disponíveis como ferramentas de apoio para que os nutricionistas possam obter uma formulação de ração que forneça a quantidade ideal de nutrientes para os animais. As tabelas publicadas pelo *National*

Research Council (NRC, 2012) são uma das referências mais utilizadas a nível mundial. Nessas tabelas, as exigências de aminoácidos são apresentadas em relação à lisina e são alteradas de acordo com a idade dos animais (peso) (Tabela 1.2).

Tabela 1.2 Padrões de proteína ideal (%) para suínos em crescimento, segundo o NRC (2012).

Aminoácido	Faixa de peso (kg)		
	20-50	50-80	80-120
Lisina	100	100	100
Treonina	62,5	64,5	67,2
Metionina	28,9	28,8	28,8
Metionina + cistina	57,0	57,8	58,9
Triptofano	17,4	17,7	18,2
Arginina	45,9	46,0	46,1
Valina	65,8	66,6	67,7
Isoleucina	50,8	51,3	52,0
Leucina	101,1	101,5	102,0
Histidina	34,3	34,3	34,3
Fenilalanina	60,2	60,7	61,3
Fenilalanina + tirosina	94,7	95,5	96,6

1.3 Fatores influentes na exigência de aminoácidos

Fatores como idade, genótipo, sexo e ambiente podem influenciar na exigência de aminoácidos para suínos em crescimento, uma vez que estes fatores interferem na expressão do potencial máximo de desempenho dos animais (SAKOMURA, 2014). Ao longo da vida, os animais apresentam diferentes taxas de crescimento em seus muitos tecidos corporais. Animais jovens depositam uma proporção maior de tecido magro, o que se altera progressivamente à medida que os animais atingem seu peso de maturidade. As alterações nas taxas de crescimento repercutem em alterações nas exigências nutricionais, sobretudo de aminoácidos.

As genéticas suínas também apresentam exigências variadas para aminoácidos, uma vez que podem expressar diferentes taxas de crescimento e composição corporal. Nas tabelas brasileiras apresentadas por ROSTAGNO et al. (2017), as recomendações nutricionais para suínos são divididas por sexo e por rebanhos de diferentes desempenhos esperados (ex: regular-médio e médio-superior). Em suínos machos castrados o desempenho esperado irá afetar na exigência de aminoácidos, ou seja, animais com médio-superior desempenho serão mais exigentes (Tabela 1.3). Além disso, diversas empresas focadas no desenvolvimento de material genético apresentam suas próprias recomendações nutricionais, tendo em vista as curvas de crescimento características dos seus animais.

Tabela 1.3 Exigência nutricional de treonina digestível (%) de acordo com o desempenho de suínos machos castrados em diferentes faixas de peso[1].

Faixa de peso (kg)	Desempenho	
	Regular-médio	Médio-superior
15 – 30	0,72	0,81
30 – 50	0,62	0,69
50 – 70	0,54	0,60
70 – 100	0,43	0,52
100 – 125	0,37	0,45

[1] Exigências estimadas de acordo com as tabelas brasileiras para aves e suínos (ROSTAGNO, 2017)

Características do ambiente onde o animal é mantido também são importantes no contexto das exigências nutricionais. A temperatura ambiental e o desafio imunológico são alguns dos fatores ambientais que influenciam a exigência de nutrientes dos animais. Usualmente as exigências de aminoácidos são obtidas em condições ambientais controladas e existem poucos estudos que avaliam as exigências de aminoácidos em temperaturas extremas. Em situações de desafio sanitário e imunológico, o consumo de alimento é afetado e, com isso, a concentração de nutrientes nas rações poderia ser alterada para atingir a exigência mínima (STAHLY, WILLIAMS E ZIMMERMAN, 1994). Além disso, animais desafiados apresentam uma maior demanda de nutrientes para tarefas do sistema imunológico, assim comprometendo a deposição de proteína em casos de limitação da oferta (NOGUEIRA et al., 2001).

Como apresentado nos parágrafos anteriores, as exigências de aminoácidos podem ser variáveis de acordo com as características dos animais e do ambiente onde eles são criados. Conhecer essa heterogeneidade é fundamental para garantir que os animais sejam alimentados de maneira mais precisa, sobretudo para assegurar uma produção mais sustentável e econômica.

1.4 Importância da treonina para suínos

A treonina é o segundo aminoácido limitante em dietas para suínos em crescimento. Esta condição é dependente dos ingredientes que compõem a dieta desses animais, mas tende a ser frequente na maioria dos padrões utilizados na suinocultura industrial. Apesar de ser essencial para suínos, a treonina não é sintetizada pelo metabolismo do animal em quantidades necessárias para suprir suas exigências. A treonina, junto com outros aminoácidos, é importante para a manutenção do sistema imune e participa ativamente de processos de reparação da mucosa intestinal dos animais (BEQUETTE, 2003). Dentre suas inúmeras funções dentro do organismo, a treonina é utilizada principalmente na produção de mucina, que está relacionada a proteção no intestino dos animais (WU, 1998).

Além de estar presente nos ingredientes, este aminoácido pode ser utilizado na sua forma sintética nas dietas. Os animais não são capazes de transformar o isômero D- em L-treonina, por não haver transaminases no tecido animal capazes de transaminar a molécula (D'MELLO, 2003). Em consequência disso, a produção industrial de treonina é realizada através de processos fermentativos, nos quais somente o isômero L-treonina é gerado.

As moléculas de treonina são encontradas dentro das cadeias de polipeptídios das proteínas e, dentre os aminoácidos, foi a última molécula a ser descoberta (WU, 2013). O catabolismo da treonina, além de inúmeras funções metabólicas, pode ser utilizado como precursor da síntese de outros aminoácidos, como a isoleucina (BENDER, 2012).

1.5 Exigências de treonina para suínos

As exigências de treonina para suínos são influenciadas por diferentes fatores, especialmente com a fase produtiva em que esses animais se encontram. Conforme mencionado anteriormente, as exigências para treonina são estabelecidas em função das exigências de lisina. Entretanto, essa relação pode variar de acordo com a referência utilizada (Tabela 1.4).

Tabela 1.4 Relação ideal de treonina digestível comparada a lisina (100%) conforme diversas recomendações disponíveis.

Aminoácido	NRC[1] (2012)	BSAS[2] (2003)	INRA[3] (2013)	VSP[4] (2013)	Ajinomoto[5] (2013)	Rostagno[6] (2017)
Treonina, %	59	65	65	61	65	65

[1] National Research Council
[2] The British Society of Animal Science
[3] Institut National de la recherche agronomique
[4] Danish Agriculture & Food Council.
[5] Ajinomoto Eurolysine
[6] Tabelas Brasileiras para aves e suínos.

Diversos estudos foram realizados para encontrar o nível ideal de treonina para suínos, porém os resultados ainda são controversos (ADEOLA, LAWRENCE E CLINE, 1994; BERENDE et al., 1983; CHANG et al., 2000). Além dos diversos fatores influentes já mencionados, as exigências também serão dependentes das variáveis utilizadas no processo de estimação, como ganho de peso, a conversão alimentar e a deposição de proteína (MA et al., 2015). As exigências de treonina ainda podem ser dependentes do nível de fibra da dieta (MATHAI, 2016) e do nível sanitário dos animais (BEQUETTE et al., 2003).

O NRC é um dos principais manuais de referência de exigências para animais de produção e companhia. Para suínos, sua primeira edição foi publicada em 1994 e a última, e mais recente, foi publicada em 2012, sendo sua décima primeira edição. A Tabela 1.5 apresenta as exigências presentes nas tabelas do NRC recentemente publicados.

Tabela 1.5. Exigências de treonina digestível (%) para suínos de acordo com o peso corporal segundo o NRC.

	Variação do peso corporal (kg)						
Treonina digestível (%)	3 – 5	5 – 10	10-20	20-50	50 – 80	80 - 120	
NRC 1998	0,75	0,66	0,56	0,46	0,37	0,30	
	Variação do peso corporal (kg)						
Treonina digestível (%)	5 – 7	7 – 11	11 – 25	25 – 50	50 – 75	75 – 100	100 – 135

NRC 2012	0,88	0,79	0,73	0,59	0,52	0,46	0,40

Como apresentado anteriormente, os estudos em exigências de treonina para suínos são muito variáveis, sobretudo em suas características experimentais. A tabela 1.6 apresenta algumas diferenças metodológicas que foram encontradas na literatura acerca da utilização de treonina na dieta de suínos em diferentes fases de crescimento. Além disso, outras condições, como o potencial genético e a abordagem analítica (modelo escolhido para a estimação das exigências), também não são constantes entre os estudos disponíveis na literatura.

A variabilidade encontrada na literatura abre espaço para revisões sistemáticas e estudos meta-analíticos que, com cautela, podem avaliar os diferentes resultados encontrados em cada um dos estudos avaliados.

Tabela 1.6. Características experimentais de diferentes estudos acerca da exigência de treonina para suínos.

Artigo	Ano	País	Peso Inicial (kg)	Peso final (kg)	Tabela utilizada	Níveis de treonina digestível na dieta (%)
Berto[1]	2002	Brasil	7,20	12,71	NRC (1998)	0,65; 0,72; 0,78; 0,84
Berto[2]			12,60	23,79		0,55; 0,74; 0,80; 0,85
Berende	1983	Holanda	14,80	38,92	NA*	0,32; 0,4; 0,48; 0,56; 0,64; 0,63
Bisinoto	2007	Brasil	6,04	11,60	ROSTAGNO et al., (2000)	0,72; 0,88; 1,03; 1,19
Borg[1]	1987	EUA	7,70	19,71	NRC (1979)	0,45; 0,5; 0,55; 0,6; 0,65; 0,63
Borg[2]			9,70	21,13		0,44; 0,51; 0,57; 0,65; 0,72; 0,65
Buraczewska	2006	Polônia	25,35	51,9	NA	0,47; 0,53; 0,57; 0,61
De lange	2001	Canadá	39,10	76,65	NRC (1998)	0,7; 0,68; 0,79; 0,69; 0,71; 0,71
Defa	1999	China	17,45	29,42	NRC (1998)	0,47; 0,55; 0,65; 0,74
Ettle[1]	2003	Alemanha	32,80	67,86	NA	0,42; 0,46; 0,51; 0,54; 0,50; 0,53; 0,59; 0,65;
Ettle[2]			67,80	109,57		0,32; 0,35; 0,38; 0,44; 0,32; 0,37; 0,39; 0,43
Friesen	1992	EUA	31,0	51,94	NA	0,33; 0,4; 0,5; 0,61; 0,71
Goodband	1991	EUA	59,42	104,32	NA	0,35; 0,45; 0,55; 0,65
Jayaranan	2015	Canadá	7,31	17,51	NRC (2012)	0,65; 0,7; 0,75; 0,79; 0,84
Kovar	1993	EUA	10,90	19,39	NRC (1988)	0,34; 0,39; 0,44; 0,49; 0,54; 0,59
Leibholz	1988	Austrália	4,80	13,85	NA	0,55; 0,59; 0,66; 0,67; 0,71
Leibholz			6,40	16,21		0,43; 0,48; 0,53; 0,58; 0,63
Lewis	1986	EUA	6,40	14,38	NRC (1979)	0,45; 0,49; 0,54; 0,6; 0,67; 0,75
Li	1998	China	17,45	29,42	NA	0,35; 0,44; 0,54; 0,64

Lopez	2010	México	24,02	54,70	NRC (1998)	0,42; 0,47; 0,52; 0,57; 0,62
Ma	2015	China	90,0	116,96	NRC (2012)	0,27; 0,30; 0,33; 0,36; 0,39
Mathai[1]	2016	EUA	26,39	49,10	NRC (2012)	0,39; 0,48; 0,57; 0,65; 0,73; 0,81
Mathai[2]			26,60	51,51		0,39; 0,48; 0,57; 0,65; 0,73; 0,81
Mauricio	2014	Brasil	7,45	13,88	ROSTAGNO et al., (2012)	0,66; 0,75; 0,85; 0,94; 1,04
Paiano	2008	Brasil	15,15	30,50	NRC (1998)	0,55; 0,59; 0,64; 0,69; 0,74; 0,55; 0,59; 0,64; 0,69; 0,74
Pinheiro	2014	Brasil	5,31	14,82	ROSTAGNO et al., (2005)	0,75; 0,85; 0,95; 1,05; 1,15
Plitzner	2007	Áustria	66,5	113,13	NRC (1998)	0,38; 0,41; 0,45; 0,49; 0,51; 0,55
Pozza	2000	Brasil	15,46	33,36	ROSTAGNO et al., (1983)	0,41; 0,46; 0,51; 0,56; 0,61
Remus	2019	Canadá	26,02	41,87	INRA (2004)†	0,35; 0,42; 0,50; 0,57; 0,65
Remus	2018	Canadá	109,42	133,4	INRA (2004) †	0,35; 0,42; 0,50; 0,57; 0,65
Rossoni	2008	Brasil	59,0	95,22	ROSTAGNO et al., (2000)	0,48; 0,50; 0,52; 0,55; 0,57
Rodrigues	2001	Brasil	5,8	15,08	ROSTAGNO et al., (2000)	0,58; 0,63; 0,68; 0,73; 0,78
Rodrigues	2001b	Brasil	29,2	60,28	NRC (1988)	0,51; 0,56; 0,61; 0,66; 0,71
Saldana[1]	1994	EUA	6,5	16,48	NRC (1988)	0,46; 0,5; 0,54; 0,58; 0,62; 0,64
Saldana[2]			58,0	93,21		0,23; 0,28; 0,33; 0,38; 0,43; 0,40
Santos	2010	Brasil	95,0	127,15	ROSTAGNO et al., (2000)	0,47; 0,49; 0,52; 0,55; 0,58
Saraiva	2006	Brasil	14,9	30,23	ROSTAGNO et al., (2000)	0,53; 0,57; 0,61; 0,65; 0,68
Saraiva	2007	Brasil	15,1	29,95	ROSTAGNO et al., (2000)	0,53; 0,57; 0,61; 0,65; 0,68

Saraiva	2007b	Brasil	30,0	61,09	ROSTAGNO et al., (2000)	0,50; 0,53; 0,56; 0,59; 0,63
Schutte[1]	2010	Holanda	50,0	61,93	NA	0,38; 0,42; 0,46; 0,50
Schutte[2]			50,0	61,41		0,44; 0,48; 0,52; 0,56
Schutte[3]			50,0	95,79		0,38; 0,42; 0,46; 0,50
Schutte[4]			50,0	93,60		0,44; 0,48; 0,52; 0,56
Taylor	1982	Inglaterra	25,0	53,77	NA	0,39; 0,43; 0,45; 0,47; 0,49; 0,51; 0,53; 0,57
Wang	2006	China	9,85	23,35	NRC (1988)	0,53; 0,58; 0,65; 0,75; 0,85
Xie	2013	China	72,5	95,06	NSNG (2010)	0,34; 0,37; 0,41; 0,44; 0,47
Zhang	2013	China	22,0	49,72	NRC (1998)	0,5; 0,54; 0,59; 0,63; 0,68

[1]Experimento 1; [2] Experimento 2; [3] Experimento 3; [4] Experimento 4.

[*]Não apresenta as tabelas utilizadas para a formulação de rações.

[†] SAUVANT, PEREZ e GILLES (2004)

1.6 Revisão sistemática e meta-análise como ferramentas na nutrição animal

A meta-análise é uma ferramenta muito utilizada em diversas áreas do conhecimento, incluindo ciências da saúde, economia, ciências humanas e biológicas. Na agricultura não poderia ser diferente, visando o grande número de dados que hoje são trabalhados nesta área. Essa ferramenta sintetiza ou combina informações de múltiplos estudos para obter informações que possam ser aplicadas à grandes populações (BORENSTEIN, 2009). Na figura 1.2 observa-se o aumento dos estudos de revisão sistemática e meta-análise ao longo dos anos, o que demonstra a relevância desta técnica na pesquisa científica.

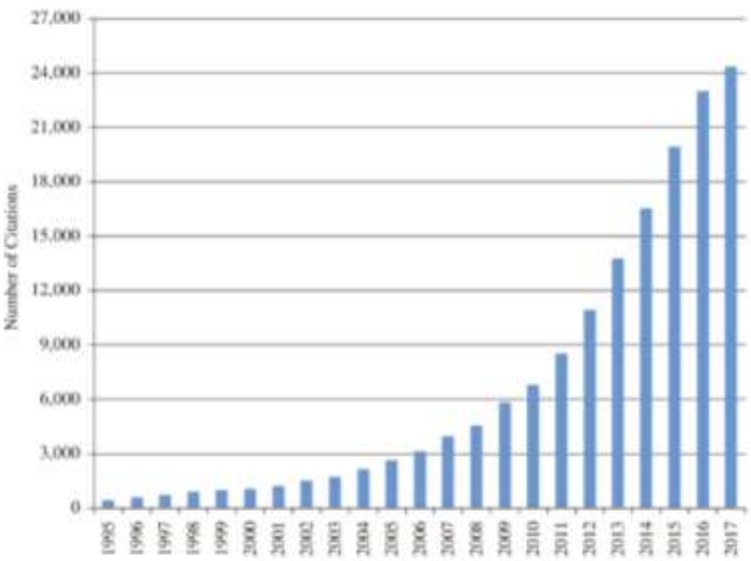

Figura 1.2 Citações a artigos incluindo os termos *Research synthesis*, *Systematic Review*, ou *Meta-Analysis* nos seus títulos.
Fonte: COOPER, HEDGES E VALENTINE (2019)

Uma das grandes vantagens da meta-análise na produção animal, ou em qualquer outra grande área, é o aumento no tamanho da amostra (pela combinação de vários estudos), o que diminui o grau de incertezas que existem nos resultados (LOVATTO et al., 2007). Outro ponto positivo desta técnica, é o poder de extrapolar os dados obtidos de um estudo meta-analítico em inúmeras situações. Isto se diferencia de resultados de um experimento clássico, onde os resultados são específicos para aquelas condições experimentais (SAUVANT et al., 2008).

O número de estudos na área de ciência animal tem crescido consideravelmente na última década. Esta é uma condição comum em várias subáreas, mas é particularmente importante em nutrição. O aumento do número de pesquisas é positivo. Porém, muitas vezes os estudos se tornam demasiadamente repetitivos e apresentam resultados antagônicos. Neste contexto, ferramentas que podem auxiliar na busca de um resultado comum para um assunto de interesse são bastante importantes.

No estudo de aminoácidos, a meta-análise é uma aliada para sintetizar as inúmeras informações que estão presentes na literatura. E, também, utilizar de análises estatísticas para estimar exigências ou, até mesmo, comparar com as exigências presentes em tabelas e manuais de exigências existentes.

2. OBJETIVO

Este estudo foi desenvolvido para fornecer um resumo completo da literatura disponível sobre exigências de treonina para suínos em crescimento e avaliar as recomendações de treonina digestível ileal estandardizada.

CAPÍTULO II[1]

[1]Artigo escrito nas normas da revista *Journal of Animal Science*

Threonine requirements of growing pigs: a systematic review and meta-analysis

Abstract

Precise estimations of standardized ileal digestible (SID) threonine (Thr) requirements can maximize nitrogen efficiency of utilization in growing pigs. Threonine is considered the second limiting amino acid in corn-soybean meal diets for pigs. Although several studies have been conducted to study SID Thr requirements, still there is no consensus regarding the ideal recommendation. Therefore, this study aimed to verify SID Thr requirements for pig populations. The database contained 38 dose-response studies published between 1982 and 2019. The database was divided in weaning (5 - 25 kg BW), growing (25 – 75 kg BW) and finishing (75 – 135 kg BW). The selected studies had a concentration of Lysine and other amino acids that met the requirements in 90% and 100%, respectively, of the NRC requirements. Due to differences between studies, threonine concentration and response criteria were standardized. The response variable for average daily gain (ADG%) was expressed in relation to the greatest response present in the study (i.e greatest ADG was considered 100%) and in kg per day (ADG). The independent variable (threonine concentration) was expressed in relation to NRC (2012) requirements manual (i.e NRC requirements were considered 100%). Results of Thr intake were presented in grams per day. Non-linear regressions were fitted by NLIN procedure of SAS for each dose-response study using the linear-plateau (LRP) and quadratic linear-plateau (QP) model. The breaking point in the LRP was interpreted as the relative SID Thr concentration, required to maximize ADG%. Considering the studies individually, SID Thr concentration to maximize ADG% varied from 32 to 117% of the NRC (2012) recommendation. In 10 experiments, pigs did not respond to relative SID Thr concentration. Both results (range of requirements and lack of response to Thr) indicated a large variance in the Thr requirements the among studies that composed the database. The ideal threonine requirements for weaning and growing was 0.019 and 0.015 g/g of weight gain, respectively according to the QP model. For the finishing phase the results were adjusted for the LRP model and presented 0.015 g of threonine by each g of weight gain. Considering the entire database, the results of the QP model showed that relative SID Thr concentration that maximizes ADG% were 14.25% higher in weaning phase, 11.36% higher in growing phase, and 11.98 higher in finishing phase than the recommendations of NRC (2012), which represents an underestimation of Thr requirements for pigs during all phases.

Keywords: Amino acid, nutrition, nutritional requirements, precision feeding, swine.

Introduction

Threonine (Thr) is usually the second limiting amino acid in pig diets formulated based on corn and soybean meal. This amino acid is essential for several metabolic pathways and can not be synthesized by the animal from nitrogen sources such α-keto acids to meet its requirements. The Thr, together with glutamate, arginine, and cysteine are involved in many maintenance functions, particularly those which are part of immune system and gut mucosa repair processes (Bequette, 2003). The Thr also plays an important role on defence mechanism once the intestine is a barrier against bacterial translocation and Thr is used in the mucin production.

Usually, requirements for Thr are established in function of lysine. However, several ratios were proposed (NRC, 2012; Rostagno et al., 2017) and the previous studies present controversial results (De Lange et al., 2001; Pedersen et al., 2003; Mathai et al., 2016). Likely because Thr requirements will vary according to the sanitary level, the fibre level in the diet, and the presence of microbes and parasites in the intestinal tract (Bequette, 2003). Also, the variation may be assigned by the difference between the models (broken-line, curvilinear) used to determine the requirement (Pomar et al., 2003) and also by the difference between the criteria responses that had been used to estimate the ideal requirement (Ma et al., 2015). Although several studies have been conducted to study Thr requirements, still there is no consensus regarding the ideal recommendation. This study was developed to provide a complete summary of the literature available on Thr requirements for growing pigs and to evaluate an important recommendation for standardized ileal digestible (SID) Thr requirements.

Material and methods

Search strategy, selection of the articles, and database construction

A critical systematic review was performed to select the peer-reviewed studies for the construction of the database. Records were identified from the electronic databases of PubMed, Scopus, and Web of Science using the search terms 'threonine' or 'threonine requirements', in combination with 'pig', 'pigs' or 'swine', in addition to 'performance', 'weight gain' or 'feed intake'. The literature search was performed in November 2019.

The studies obtained in the search were then checked for their compliance with different criteria in order to determine their inclusion in the database. The criteria used for including the studies were: a) at least four different Thr levels in the diets (e.g. a basal diet plus at least other three levels of Thr); b) composition of the experimental diets available in the paper, c) lysine was provided at levels lower than its requirement (90%) and all other amino acids (AA) were provided to meet at least 100% of their optimal levels; and d) presentation of performance results.

All references obtained in each database were exported to EndNote software, where the title and the abstract of each record was revised independently by two researchers in order to select the papers that were fully evaluated in the next step. The full version of the selected papers was critically evaluated as to their quality and relevance considering the systematic review objectives. To complete the database, all citations in the reference lists of selected publications were revised in order to search for additional studies that could meet the criteria to be included in the databases. If found, these scientific publications (papers, thesis, or abstracts) were evaluated following the same criteria used in the previous step.

After the selection of the scientific communications, the data were transferred to an electronic spreadsheet. Each row of the spreadsheet represented a treatment and each column represented a study variable. Information related to the objective of the study (animal performance) and other variables (genetic strain, sex, dietary nutritional composition, and rearing phase) were used to provide a descriptive analysis of the studies included in the database.

Two moderating codes were applied: a) general code (study effect), where each dose-response study received a sequential number and b) inter-code, consisting of the general code number plus a sequential number representing the treatment (e.g. article 1, treatment 01 – 1+01 = 101). A categorical code was used to analyse data from weaning (1) 5 – 25 kg BW, growing (2) 25 – 75 kg BW and finishing (3) 75 – 135 kg BW phases. The ingredient composition of the diets was computed into a spreadsheet and the nutrient composition was recalculated using the software EvaPig (version 1.3.1.4, INRA, Saint-Gilles, France). This method was used in order to have the complete composition of the diets and to minimize the variation among experiments.

The AA were always evaluated as SID values. The Thr concentration was adopted as the independent variable and expressed in grams per day and as 'relative Thr concentration'. The relative Thr concentration was expressed as relative percentage to the SID Thr requirement recommended by the National Research Council (NRC, 2012). In this case, the 100% value represented the Thr requirements estimated by the NRC model. Similar procedure was already applied in a previous study (van Milgen et al., 2012).

The ADG% results were also used as relativized information. For this approach, the responses of each treatment were relativized to the treatment with the maximum response in the trial and expressed as the variation (percentage) between the results (i.e. treatment with the greatest ADG in each study was considered 100%). This procedure was adopted to reduce the variability among studies in the database (inter-study effect). The results of ADG were also presented in kilogram per day.

Meta-analysis

Exploratory analyses were performed to better understand the database. First, graphical analyses were used to observe the biological coherence of the data and to obtain a general view of the consistency and heterogeneity of the data. This procedure was also used to observe data distribution in terms of dietary Thr levels. Non-linear models were applied to fit the individual dose-response curves and to check the adjustment of current recommendations for SID Thr requirements. The analyzes were divided into two-step procedure. First, non-linear regressions were fitted by N-LIN procedure individually for each dose-response trial included in the database. The Linear Response Plateau (LRP) was used, by the equation:

$$Y = U * (X < R) * (R - X) + L$$

Where, U is the slope coefficient when X<R, L is the break point (plateau) in the Y-axis, and R represents the break point in the X-axis beyond which there is no change in the dependent variable. The R variable was interpreted as the relative Thr level required to maximize the ADG.

In the second step, random effects were fitted by NLMIXED procedure. This step was performed to combine the models generated for each trial and to obtain a model that considered the variability among studies. Therefore, a new database containing the estimated (L, U, and R) as well the standard error (S2e) of each one of the trials that composed the database was generated and subdivided in the three groups previously defined (weaning, growing, and finishing phases). The average values of all parameters in each group were obtained to be considered in subsequent steps.

So, four models were fitted in sequential analyzes (SeqA) where the effects of parameters U, R, and L were gradually transformed from fixed to random effects (St-Pierre, 2001). Each model was dependent on the previous one. For this, average values of the parameters U, R, L, and S2e estimated in the first step were used as initial parameters, where the parameter S2e was considered as random effect. After that, the three parameters were considered one by one as random by including its variance and covariance as random effects in the model. Thus, in the first SeqA was included variance of the parameter R (Grr); in the second SeqA variance of the parameter U (Guu); in the third SeqA covariance between R and U (Gru) and fourth SeqA variance L (Gll) and the covariances between R and L (Grl) and between U and L (Gul). In the last SeqA, all parameters were randomized, considering thus the variability among studies.

Additional to the NLMIXED procedure previously described, a quadratic-plateau and quadratic regression models were also tested. Quadratic regression was conducted using MIXED procedure, using the study as a random effect. For the quadratic-plateau (QP), each of the parameters U, R and, L were one at time introduced as random effects and the best model was chosen for presenting the smallest AIC and gradient for the estimated parameters. Exploratory analyses were performed using Minitab statistical package (version 18, Minitab Inc., State College, PA). Modelling analyses were carried out using SAS statistical software (version 9.4, SAS Inst. Inc. Cary, NC, USA).

Results

Literature research results

After the literature search, 992 references were identified and imported from the electronic databases. With the assistance of the reference manager, a total of 425 duplicates were removed. After, 518 studies were unqualified through titles and abstracts assessment. Following the evaluation of the full papers, eleven references were excluded because they did not meet the selection criteria of this meta-analysis. From those, two studies presented high lysine levels (more than 90% of the NRC recommendation), six studies used less than 4 levels of Thr in the trials, two did not presented performance responses, and one did not describe the diets used in the trials. After all the evaluation steps (online databases and references of selected studies), 38 scientific publications were retained for this meta-analysis, from which 35 were full research articles, 2 abstracts published in scientific conferences and 1 chapter of a PhD thesis (Figure 2.1).

Description of the database

The studies that composed the database were published between 1982 and 2019 (median: 2006) and described 47 dose-response trials with 6,922 pigs. The final database consisted of 250 rows (treatments) and 141 columns (variables). The average composition of the diets is presented in Table 2.1. Considering the address of the main author, 34% of the studies were from Brazil, 18% from USA, 16% from China, 11% from Canada, and 21% were distributed in Europe and Asia.

The database included 12 articles for weaning phase published between 1986 and 2015. In these papers, 14 dose-response trials were developed. The average initial BW was 7.33 (±1.77) kg and the average final BW was 16.47 (±3.71) kg. The average age at weaning was 25 (±2.95) days and the average trial duration was 23.73 (±5.12) days. The ADFI was 670 (±180) g and the ADG was 398 (±70) g. Calculated SID Thr levels in the diets ranged from 0.34 to 1.19 % and Thr:Lysine ratio ranged from 0.34 to 0.86%.

For growing pigs, the database included other 20 articles published between the 1982 to 2019, which described 22 dose-response trials. The average initial BW in these studies was 25.08 (±9.77) kg and the average final BW was 47.67 (±14.68) kg. Trial average duration was 31 (±9.41) days. The ADFI for this phase was 1,540 (±320) g and the ADG was 686 (±140) g. Calculated SID Thr level varied from 0.32 to 0.81% in the diets, while Thr:Lysine ratio ranged from 0.30 to 0.96%.

The database presented also 10 articles describing 11 dose-response trials with finishing pigs. These papers were published between 1991 and 2018. The average initial BW was 71.18 (±17.97) kg and the final BW was 107.51 (±13.52) kg in these studies. The ADFI was 2,840 (±450) g and ADG was 939 (±140) g. Calculated SID Thr level presented in the diets ranged from 0.23 to 0.65%, while Thr:Lysine ratio ranged from 0.30 to 0.89% in these studies.

Exploratory analysis

In this database, it was possible to observe that Thr:Lysine does not explain the variation observed in the ADG, which was better explained by SID Thr intake (Figure 2.2). Therefore, the analysis were performed ignoring Thr:Lys, and only accounting for SID Thr intake. Additionally, in the exploratory graphical analysis it was possible to observe that weaning pigs presented a linear response to Thr intake, while growing and finishing pigs presented a quadratic response (Figure 2.3).

The average level of SID Thr in the database was close (108%) to the requirement tables of the Nutritional Research Council (NRC, 2012). However, these levels ranged widely among the studies (Figure 2.4). Relative Thr concentration required to maximized ADG% varied from 32 to 117% of the level estimated by the NRC (2012) model. Most of the experiments used animals weighting 15 to 50 kg, and there is a lack of information in the finishing phase. All trials developed with weaning piglets lasted more than 11 d and used ADG as the variable response to establish Thr requirements. Most part of the trials developed with growing and finishing pigs lasted more than 21 and 28 d, respectively. These trials also used ADG as response to establish the Thr requirements. Numbers of Thr levels tested in each trial varied among studies. Eleven experiments used 4 levels of Thr, 21 experiments used 5 levels, 11 used 6 levels, and 4 trials used 8 or more levels of Thr in the experimental design.

Assessing the current Thr requirements

For weaning pigs, when using the QP the optimal SID Thr intake was 0.019g/ g of weight gain. The LRP found that the optimal SID Thr intake was 0.013g/ g of weight gain. For growing pigs, when using the QP the optimal SID Thr intake was 0.015g/ g of weight gain. The LRP found that the optimal SID Thr intake was 0.014g/ g of weight gain. For finishing pigs, QP overestimated largely the requirements and was excluded from the analysis. The LRP found that the optimal SID Thr intake was 0.015g/ g of weight gain, showing no change from growing pigs' requirements.

The ADG% was maximized at SID Thr levels that corresponded to 114.25, 111.36, and 111.98% of the current recommendation of NRC (2012) for weaning, growing, and finishing pigs, respectively using the QP model (Table 2.2). The QP model it was chosen because presented greater AIC when compared to the RLP model. Thus, obtained values were very close to the current recommendations for all the growing phases. However, it is important to point out that results represented the midpoint of each phase, which has been shown to be appropriate in previous studies (Van Milgen et al., 2012).

Discussion

General over-view of the systematic review

As an important amino acid for the growth of the pigs, studies of Thr started in the 70's and provided the first requirement recommendation for this amino acid for the swine industry. Although the research focused in pig nutrition has developed considerably in the last decades, there is still a lack of information in some specific topics. Most of the studies identified during the systematic review were developed with weaning (12) or growing (20) pigs. Just a few articles (10) were found for the finishing phase. Van Milgen et al. (2012) studied the requirements of isoleucine for pigs and found few information on finishing phases. This condition is probably related to the cost of the trials, which are higher for finishing phase compared to the experiment with younger piglets (less feed, labour, and space are needed). Despite the difficulties in developing trials with the finishing pigs, it is important to point out that more studies are needed in this phase to increase the precision in the nutritional requirement estimation. This is a very relevant recommendation, particularly considering that nutritional requirements are influenced by several intrinsic and extrinsic factors (Hauschild et al., 2010), which may vary among the studies. In addition, finishing pigs are responsible for a large part of the feed consumption in the commercial operations, and could also be responsible for a large share of the emissions caused by diets that do not meet their requirements. Thus, increasing the precision in which the nutritional requirements are assessed could be as important way to mitigate the environmental impacts associated with the pig production (Andretta et al, 2018).

Another important topic to be addressed concerning the systematic review procedure was that several articles were excluded for missing very important information, as number of animals used in the experiment, duration of the trials, and even the dietary composition. As previously stated, the nutritional requirements are influenced by several factors, that need to be fully described in the paper in order to allow the correct interpretation of the results. The high variability found among the studies in terms of Thr recommendation is probably related to the characteristics of animals, diets, or environment used in the trials (Ferguson et al., 2000; Xie et al., 2013). Thus, the description of these features is needed not only to interpret the results (Thr recommendations), but also to better understand the heterogeneity (each Thr recommendation when assessed in the context of the entire database).

In addition to the lack of important information in the paper, other reasons were responsible for the exclusion of studies from the database. Some trials were developed with very few levels of Thr (three or less), which do not allow the LPR models (or other indicated models, as exponential or curvilinear-plateau) to be applied in the data analysis (NRC, 2012). Other limitation found in the review process was that some studies varied the levels of other

AA together with Thr, making difficult to access the individual effect of each AA. Similar limitations were already reported by Remus et al., (2015) when assessing methionine requirements of pigs, highlighting the importance of good protocols for experimentation and also for data presentation.

Lastly, studies that challenged the animals with sanitary or environmental conditions could not be used on this meta-analysis. However, the exclusion was only motivated by the scope of the current study. The evaluating of nutritional requirements of challenged animals is a highly pertinent topic in modern pig research. Particularly for Thr, these studies are needed due to its relation with immune response and other defence mechanisms. Recently, Jayaraman et al. (2015) studied the requirements of Thr:Lysine of weaned pigs challenged by uncleaned installations and found that the recommendations increase in this condition. Other study developed by Capozzalo et al. (2017) showed the effect of weaned pigs challenged with *Escherichia Coli* increased the requirements for sulphur amino acids and the relation with lysine when compared to the recommendations estimated by NRC (2012).

Thr requirements using non-linear models

The dietary Thr levels varied greatly within the database. This was already expected because a minimum range in the levels was necessary in the original studies to stablish the Thr requirements. In only one study (Li et al., 1998), all Thr levels tested were above the NRC (2012) recommendations, with a minimum of 153% and a maximum of 217%. The other studies of the database tested Thr levels under and above the NRC (2012) recommendations.

The NRC (2012) requirements were chosen to be the standard reference in this study due to its importance in pig production. The reference is also recent and widely used in feed formulation all around the world. The NRC recommendations for Thr are similar to other important references. For instance, the Thr levels suggested by the Brazilian Tables for Poultry and Pigs (Rostagno, 2017) are 9.8% lower for weaning, 6.0%, and 2.3% higher for growing, and finishing pigs comparing to the NRC (2012). The InraPorc model (van Milgen et al., 2008) suggested Thr requirements 10% higher than the NRC recommendations for growing phase. This model uses the protein deposition as a variable for the estimation of the requirements.

The Thr requirements found in this meta-analysis were higher than the recommended by the NRC (2012) model, which indicates an underestimation on the requirements of Thr for pigs. Considering these findings, the animals could receive more Thr in the diet and have greater growth. Consequently, their nutrient efficiency rate would be maximized, and more resources would be necessary during the pig production. Estimating the nutritional requirements precisely is very important for the sustainability and rentability of the production systems. Feeding pigs with diets that correctly meet their requirements is a way to save resources (Monteiro et al., 2019), mitigate emissions to the environment (Andretta et al., 2018), and reduce feeding costs (Pomar and Remus, 2019) and still maintain the weight gain.

The LRP model is commonly used for requirement estimation and have been extensively adopted in pig research (Campbell et al., 1984; Xie et al., 2013; Ma et al., 2015). It is important to point out that LRP models are limited because linearity cannot be applied in every situation. Curvilinear and sigmoid models, for example, can differentiate dose-response and factorial methods of amino acid estimation for the pig population (Hauschild et al., 2010) and represent biologically. In this study the quadratic linear-plateau model it was performed and presented greater results.

The current study was able to summarize the data presented in this research field, which helped understanding further the Thr requirements. In this study, the weight gain of the pigs was maximized at SID Thr levels that nearly correspond to the current recommendations, but that could be adjusted in precision feeding systems. Precise estimations of Thr requirements can be used to maximize the nutrient utilization efficiency, consequently benefiting the growth and sustainability of pig production systems.

Declaration of interest

The authors declare no conflict of interests.

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Table 2.1 Average diet composition used in the studies of the database[1].

Composition, %	Weaning[2]	SD[3]	Growing	SD	Finishing	SD
CP	17.73	3.15	15.95	2.35	13.44	2.37
SID Thr	0.667	0.17	0.564	0.10	0.440	0.09
SID Lys	1.671	0.14	0.874	0.10	0.734	0.20
SID Trp	0.231	0.04	0.197	0.18	0.145	0.02
SID Met	0.415	0.10	0.332	0.12	0.26	0.07
SID Met + Cys	0.631	0.09	0.556	0.09	0.457	0.09
SID Val	0.726	0.16	0.643	0.09	0.514	0.07
SID His	0.396	0.11	0.341	0.05	0.268	0.05
SID Phe	0.826	0.65	0.650	0.10	0.520	0.09
SID Leu	1.330	0.19	1.230	0.30	0.997	0.16
SID Ile	0.663	0.11	0.539	0.09	0.435	0.09
SID Arg	1.094	0.51	0.793	0.15	0.679	0.21
Total Ca	0.844	0.16	0.755	0.12	0.599	0.22
Total P	0.713	0.06	0.600	0.10	0.588	0.16
Available P	0.400	0.06	0.424	0.41	0.502	0.58
Net energy (kcal)	2,419	186	2,355	112	2,354	102

[1]Number of treatments: 73 in weaning phase, 119 in growing phase, and 57 in the finishing phase

[2] Weaning (5 – 25 kg BW), growing (25 – 75 kg BW), finishing (75 – 135 kg BW)

[3] SD, standard deviation

Table 2.2 Non-linear model parameters[1] of the independent response variable (ADG%) in function of relative Thr concentration[2], estimated with a linear-plateau and a quadratic linear-plateau model.

Model	Threonine concentration	AIC
Weaning		
Quad-Plateau with a random component for parameter R	114.25 ± 8.9	427.7
Lin-Plateau with a random component for parameter R	104.86 ± 0.0007	427.9
Growing		
Quad-Plateau with a random component for parameter R	111.36 ±3.7	709.6
Lin-Plateau with a random component for parameter U, R, L	95.192 ±0.004	718.5
Finishing		
Quad-Plateau with a random component for parameter L	111.98 ± 4.8	296.5
Lin-Plateau with a random component for parameter R	97.1629 ±2.4	297.8

[1] Parameters: U, fit intercept; R, relative Thr concentration required to reach the plateau; L, average response estimated by the model.

[2] Relative Thr concentration was expressed in relation to the recommended SID Thr requirement (NRC, 2012)

[3]Weaning (5 – 25 kg BW), growing (25 – 75 kg BW), finishing (75 – 135 kg BW)

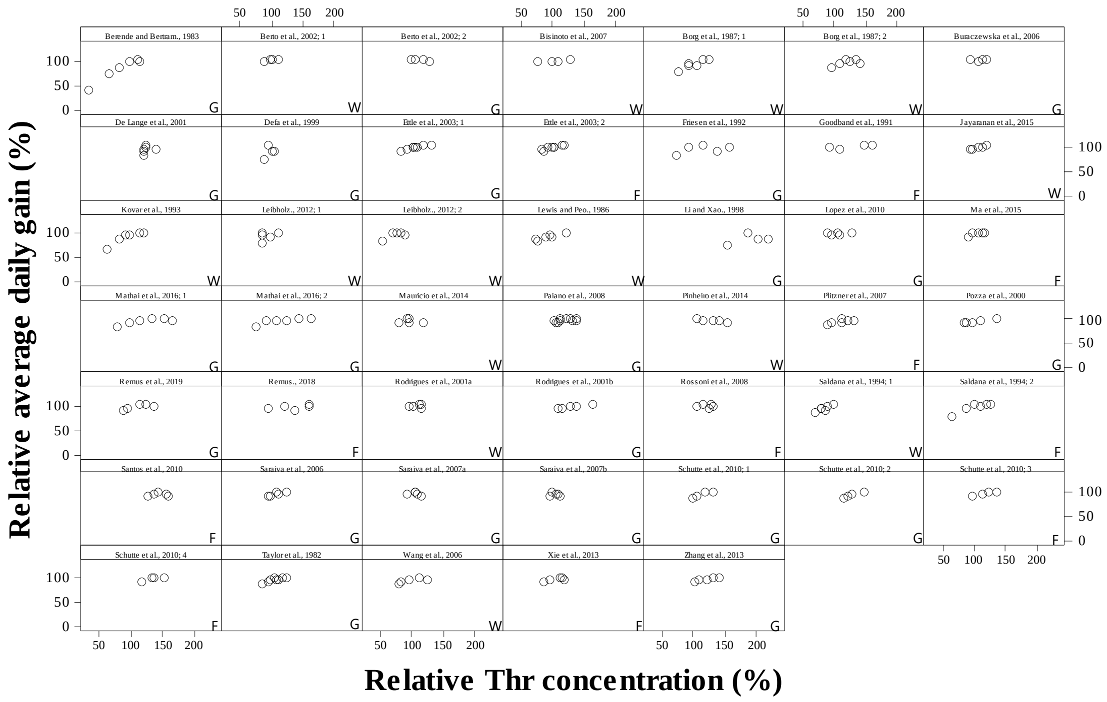

Fig. 2.1 Effect of Thr concentration[1] in the diets on the relative average daily gain[2] within each study[3] of the database.

[1] Average daily gain was relativized according to the maximum weight gain found within the study.

[2] Relative Thr concentration was expressed relative to the SID Thr requirement estimate for the treatment that maximized ADG (100%).

[3] Capital letters are references to the phases of the studies: W, weaning; G, growing; and F, finishing.

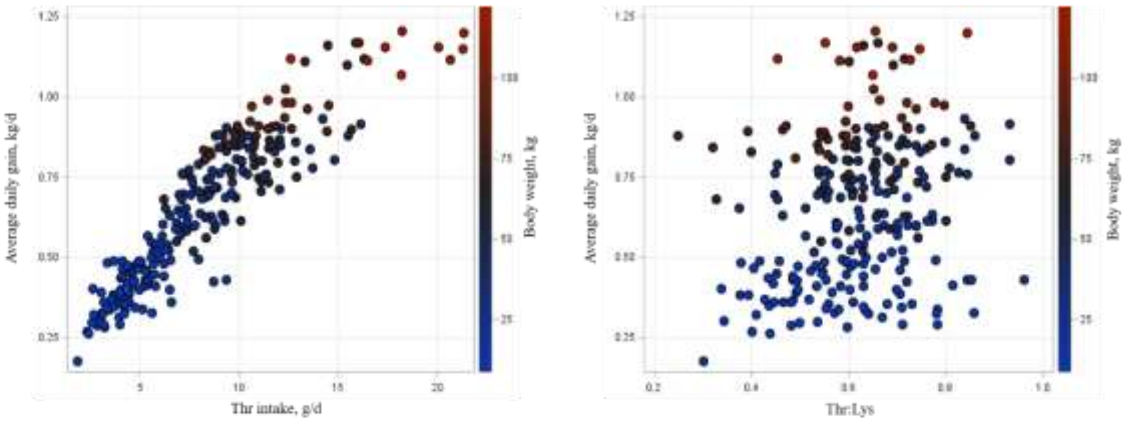

Fig 2.2. Variation between Thr intake (grams per day) and average daily gain (kg per day) according to the body weight of the animals.

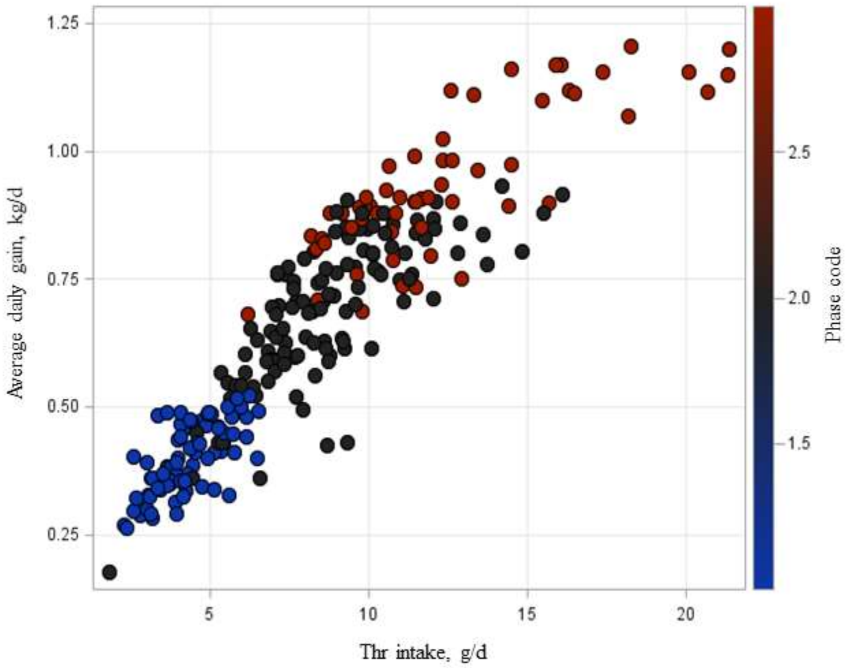

Fig. 2.3 Distribution of the Thr intake (grams per day) and average daily gain a (kilograms per day) according to the phase code[1] presented in the database.

[1] Phase code: 1 weaning, 2 growing and 3 finishing.

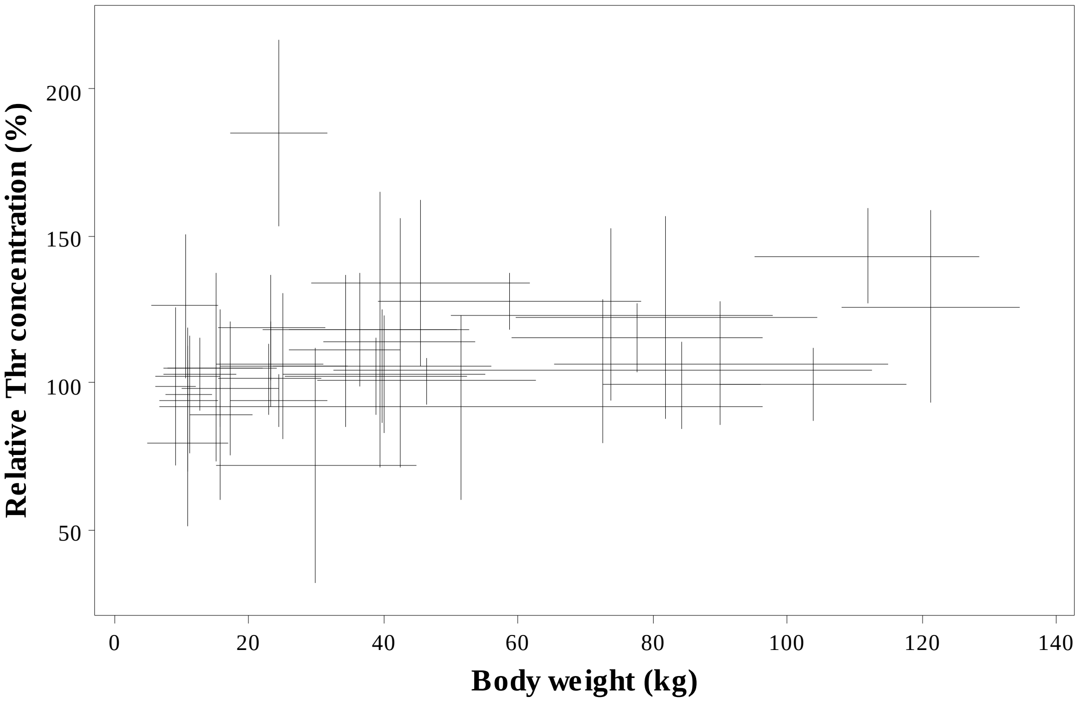

Fig. 2.4 Meta-design[1] - relation of body weight and relative Thr concentration[2].

[1] Each study is indicated by a cross, of which the horizontal line indicates the range of the body weight of the animals used in the studies, and the vertical line indicates the range of the relative Thr concentration used in each study.

[2] Relative Thr concentration was expressed in relation to the recommended SID Thr requirement (NRC, 2012).

CAPÍTULO III

CONSIDERAÇÕES FINAIS

Revisões sistemáticas e meta-análises são ferramentas úteis na nutrição animal. Uma vez que não há utilização de recursos, como animais e instalações para realizar o estudo, então torna-se economicamente viável e aceito do ponto de vista do bem-estar animal.

Esta extensa revisão sistemática apresentou inúmeros estudos de exigência de treonina para suínos e sua variação entre os autores. O ganho de peso dos animais foi maximizado nos níveis de treonina digestível, estes que estão próximos às recomendações atuais fornecidas pelos manuais disponíveis. Porém, alguns ajustes poderiam ser realizados para aumentar a utilização deste aminoácido da dieta e tornar o sistema mais produtivo.

Por fim, alimentar os animais de forma mais precisa é ambientalmente sustentável e economicamente interessante para o sistema produtivo. Mais estudos meta-analíticos em relação à exigência de aminoácidos deveriam ser abordados por pesquisadores da área de nutrição animal.

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APÊNDICE

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Required items on the page are,

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4. Institutions of the authors with location denoted with a symbol (*, †, ‡, §, #,||, and ¶) behind the author last name
5. Department, city, state or country and postal code
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Abstract

A single paragraph of no more than 2,500 keystrokes (characters plus spaces) that summarizes the results in an understandable form using statistical evidence (*P*-values). Abbreviations are defined at first use in the ABSTRACT and again in the body of the manuscript.

Key words

List up to 6 words in alphabetical order and separated by a comma. Capitalize only proper nouns. Do NOT use abbreviations. Place at the end of the ABSTRACT.

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A comprehensive list of all abbreviations used in the manuscript and their definition. An example format is MRF, myogenic regulatory factor. The list should include neither standard JAS Abbreviations nor diets. Abbreviations must be defined at first use in the manuscript text but not in tables and figures unless unique.

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Plural abbreviations do not contain a final “s” because the context of an abbreviation implies whether it is singular or plural. Use of the standard 3-letter abbreviations for amino acids (e.g., Ala) is acceptable in *JAS*. Use of the internationally recognized chemical symbols for chemical elements (e.g., P and S) is acceptable in *JAS*. Except for N (not italicized), which is the recognized abbreviation for nitrogen and newton (unit of force), chemical symbols for elements are reserved for elements (e.g., C is for carbon and never for control).

Introduction

A clear justification for conducting the research with a stated hypothesis and objective(s) is required. The rationale for the experiments should place the work into the context of existing literature. There is NO word limit on the section but brevity is encouraged.

Materials and Methods

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Journal articles

Perez, V. G., A. M. Waguespark, T. D. Bidner, L. L. Southern, T. M. Fakler, T. L. Ward, M. Steidinger, and J. E. Pettigrew. 2011. Additivity of effects from dietary copper and zinc on growth performance and fecal microbiotia of pigs after weaning. J. Anim. Sci. 89:414–425. doi:10.2527/jas.2010-2839.

Abstracts

Centon, J. R., G. E. Erickson, T. J. Klopfenstein, K. J. Vander Pol, and M. A. Greenquist. 2007. Effects of roughage source and level in finishing diets containing wet distillers grains on feedlot performance. J. Anim. Sci. 85(Suppl. 2):76. (Abstr.) doi:10.2527/jas.2006-354.

Books and chapters in books

AOAC. 1990. Official methods of analysis. 15th ed. Assoc. Off. Anal. Chem., Arlington, VA.

NRC. 2000. Nutrient requirements of beef cattle. 7th rev. ed. Natl. Acad. Press, Washington, DC.

Robinson, P. H., E. K. Okine, and J. J. Kennelly. 1992. Measurement of protein digestion in ruminants. In: S. Nissen, editor, Modern methods in protein nutrition and metabolism. Academic Press, San Diego, CA. p. 121–127.

Conference proceedings

Bailey, E. A., J. R. Jaeger, J. W. Waggoner, G. W. Preedy, L. A. Pacheco, and K. C. Olson. 2012. Effect of weaning method on welfare and performance of beef calves during receiving. Proc. West. Sec. Amer. Soc. Anim. Sci. 63:25-29.

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