Quantifying production, processing and post-slaughter effects on pork eating quality using random effects meta-regression[1]

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ABSTRACT: Random effects meta-regression techniques, analyzed using a restricted maximum likelihood (REML) approach, was used to determine the influence of various factors that may be experienced or imposed on pigs, carcases and pork on pork eating quality attributes and shear force of the M. longissimus dorsi (loin). This was done to inform the development of a pathway based eating quality system for pork. Estimated means of explanatory variables were obtained for those pathway factors where sufficient published studies met the criteria for inclusion in the analysis. Due to a lack of data for interactions between factors investigated, only single factors were included as fixed terms in the REML models. This analysis identified that moisture infusion ($P < 0.001$), ageing for more than 2 d post-slaughter ($P = 0.006$) and tenderstretching ($P = 0.006$) each resulted in significant improvements in tenderness. Cooking loins to an endpoint temperature of ≥ 80°C negatively impacted both tenderness ($P = 0.022$) and juiciness ($P < 0.001$) scores compared with 70 to 74°C. It was not possible to develop algorithms to reliably estimate the effects of multiple factors on pork eating quality attributes to a cuts-based level due to limited studies reporting data for treatment interactions.

Key words: consistency, eating quality, interventions, pathways, pigs, pork

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INTRODUCTION

The ability of pork supply chains to consistently provide high quality pork products to their customers is paramount to improve consumer satisfaction and increase pork consumption frequency (Channon and Warner 2011). This presents significant challenges to industry as production, processing, post-slaughter, and cooking parameters can, singularly or in combination, influence tenderness, juiciness and flavor of pork. This has been extensively described in qualitative reviews, both in Australia and overseas (including D'Souza and Mullan, 2001; Taverner, 2001; Rosenvold and Andersen, 2003; Ngapo and Gariepy, 2008; Channon and Warner, 2011). In the development of any eating quality system, it must be flexible and non-prescriptive in its approach to support its adoption by industry and allow individual supply chains to select those factors that will enable eating quality targets to be met.

Meta-analyses methodology has been applied to obtain single summarized effect sizes of various experimental treatments applied to pigs, carcases and/or pork, primarily for technological quality attributes (Dunshea et al., 2005; Salmi et al., 2010; Trefan et al., 2011; Trefan, 2011; Pauly et al., 2012; Salmi et al., 2012). It has enabled generalized treatment effects to be obtained when outcomes from independent, randomized studies are conflicting (Thompson and Higgins, 2002). The use of porcine somatotropin, ractopamine and conjugated linoleic acid (Dunshea et al., 2005) and gender (Trefan et al., 2013) on tenderness and juiciness has also been quantified using a meta-analysis approach. These analyses generated very useful outcomes on the size of individual factors on pork eating quality. However, an approach to quantitatively review, and analyze, the size of different treatment effects within a factor on pork eating quality attributes to establish a cuts-based predic-

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tive model for pork eating quality has not been reported. A multivariate model, incorporating all key significant factors, is needed to underpin an eating quality system to deliver consistently high quality pork to consumers, in both export and domestic markets.

A Monte Carlo simulation approach, an alternative to meta-analysis, was used as an attempt to quantify the size, effect and variability in pork eating quality traits in response to different factors imposed from production to consumption (Channon et al., 2016b). This approach utilized data primarily sourced from published, refereed studies to estimate the proportional shift in treatments imposed (termed 'comparative variates'), within a factor, on means for tenderness, juiciness, flavor, and shear force against a suite of 'standard' variates (e.g., female pigs, White genotype, etc.). However, it was not possible to determine whether the proportional shifts in the probability distributions of correction factors for tenderness, juiciness, flavor, or shear force in response to the different treatments imposed were statistically significant, or not.

Therefore, the hypothesis of this study was that random effects meta-regression statistical techniques could be applied to i) estimate means and effect size between means for tenderness, juiciness, flavor, and/or shear force of pork in response to different experimental factors experienced by pigs, carcasses, and/or pork, and ii) enable significant differences between means for eating quality traits due to various factors imposed to be statistically determined. The objective of this study was, therefore, to identify those pathway factors that improve pork eating quality traits and shortlist these for potential inclusion in an eating quality predictive model for pork.

MATERIALS AND METHODS

As all data were obtained from existing publications, Animal Care and Use Committee approval was notrequired for this study.

Database Compilation

This study utilized the extensive database generated and previously described by Channon et al. (2016b). The studies used are provided in Appendix 1 (Supplementary Material). Briefly, this database was comprised a total of 294 studies, published in English between 1968 and 2016. Studies were identified from systematic literature searches of digital databases that investigated the effects of various factors on sensory tenderness, juiciness and flavor and objective tenderness of pork, as assessed by Warner-Bratzler shear force (WBSF) produced from finisher pigs of 60 to 130 kg liveweight. Only studies that included experimental treatments for each specific factor(s) of interest and provided means for sensory attributes (flavor, tenderness, juiciness) and/or shear force were used. In contrast to Channon et al. (2016b), studies that did not present measures of variation were excluded.

Details of each study were included in the data file. Cells were left blank if details of experimental treatment were not reported. All sensory data was adjusted to a 0 (very dry, very tough, dislike extremely/bland flavor) to 100 scale (very juicy, very tender, like extremely/strong flavor) with WBSF data adjusted to Newtons. As described by Channon et al. (2016b), flavor strength/intensity scores were only used when overall acceptability scores inferred the effect of an experimental treatment on flavor intensity, where flavor, tenderness and/or juiciness scores were positively correlated and when increases in flavor scores were reported as favorable. Trained panelist scores for 'abnormal flavor' were not assessed.

Groupings were made to assist with the analysis for genotype, feeding level, housing type, electrical stimulation, ageing period, chilling, cooking treatment and final internal temperature (Table 1). For example, genotype was grouped as White (including: Landrace, Large White/Yorkshire, Swedish Landrace, Danish Landrace, Yorkshire), > 50% Duroc (including 50% and 100% Duroc), Pietrain, Hampshire and Berkshire. The Duroc percentage level of 50% was used as improved eating quality of Duroc-sired progeny compared to White genotypes has been reported (e.g., Blanchard et al., 1999).

Statistical Analysis

All measures of variation provided in studies that met the study criterion were first converted to standard errors, unless reported. Only studies that reported quantitative data for the particular factor being investigated were included in the analysis. Residual between-study variance resulting from differences in study designs and accuracy was first determined using random effects maximum likelihood (REML) model, without fixed model terms or predictors included, using Genstat 16 (VSN International Ltd., Hemel Hempstead, UK), with 'experiment' included as a random variable.

The residual between-study variance for tenderness, juiciness, flavor, and shear force was then included in a weighting term to account for both within-study variances of treatment effects and the residual between study heterogeneity. Experiment was a random term in this model. In the random effects model, the variance was determined as the variance within studies (s_i^2) plus the variation between studies (τ^2). The weighting for each study for each attribute being assessed (i.e., tenderness, juiciness, flavor, shear force) was therefore equal to the

Table 1. Pathway parameters and explanatory variables evaluated using a random effects meta-regression approach

Pathway parameter	Explanatory variables
Sex	Female;
	Surgical castrate;
	Entire male;
	Immunocastrated male
Genotype - grouped	Berkshire;
	Duroc;
	Hampshire;
	Pietrain;
	White; > 50%
Halothane gene	Homozygous dominant (NN);
	Heterozygous (Nn);
	Homozygous recessive (nn)
Hampshire gene	rn+;
	RN-
Feeding level – during grower (G) and finisher (F) phase	*Ad libitum* GF;
	Ad libitum G Restricted F
	Restricted G Ad libitum F
	Restricted GF
Metabolic modifiers	
pST	No pST;
	pST
Ractopamine	No ractopamine;
	Ractopamine (≤ 10 mg/kg)
Housing	Indoor/conventional;
	Barn/Ecoshelter;
	Outdoor/free range
Stunning method	Carbon dioxide;
	Electrical
Electrical stimulation	No electrical stimulation;
	Electrical stimulation (Constant current – 4 min post-slaughter; High voltage – 20 in post-slaughter; low voltage– 3 to 5 min post-slaughter)
Hanging method	Achilles;
	Aitchbone/tenderstretching
Chilling	0 to 4°C for 18 to 24 h post slaughter (conventional);
	Rapid chill then conventional;
	Warm/ambient chill then conventional
Ageing period	≤ 2 d;
	3 to 7 d;
	> 7 d
Moisture infusion	No moisture infusion;
	Moisture infusion
Endpoint temperature	65 to 69°C;
	70 to 74°C;
	75 to 79°C;
	> 80°C
Cut type	Chop/steak;
	Roast

inverse of the sum of the within-trial variance for that study and the residual between-trial variance. This accounted for larger studies having more influence than smaller studies, as they were weighted by the precision of their respective effect estimate.

As a random effects model was used to statistically explain between-study variability, the I^2 statistic to understand the proportion of total study variance that could be attributable to between-study variation was not determined. Data was then analyzed as a random effects meta-regression model using linear mixed models with a REML algorithm using Genstat 16. As the data set was unbalanced, both the random weights variate (either tenderness, flavor, juiciness, or shear force weighting) together with explanatory variables (within each factor) as fixed terms were included in the model.

$$y_i = \beta_1 + \beta_2 x_i + \ldots + \eta_j + e \quad (1)$$

where y is the effect size in study i, $\beta = \Sigma w_i \times (y_i - \bar{y})$ $(x_i - x) / \Sigma w_i \times (x_i - x)^2$, $w_i^* = 1 / (\tau^2 + s_i^2)$, x_i $(i = 1 \ldots k)$ reflect the different explanatory variables, η_j is the between study variance (η_i independent and $\eta_j \sim N(0,\tau^2)$ e_j is the residual error (e_i independent and $e_i \sim N(0, \sigma_i^2)$.

As few studies have evaluated the effects of pathway factors on eating quality attributes on other muscles in addition to the loin, this analysis was restricted to include only those studies reporting sensory and/or shear force data on the loin muscle. Table 2 shows the total number of studies presenting eating quality data involving different muscles when various factors were applied. The use of random effects meta-regression with additive and multiple fixed terms for different factors was explored in the analysis but was constrained by a lack of eligible studies.

RESULTS AND DISCUSSION

The total number of studies selected within each factor for each eating quality trait, the number of total

Table 2. Number of eligible studies investigating effects of various pathway parameters on sensory tenderness, juiciness, and flavor and/or shear force

Muscle	No. of studies meeting study criterion
M. longissimus dorsi	294
M. semimembranosus	27
M. biceps femoris	21
M. psoas major	9
M. gluteus medius	8
M. semitendinosus	6
M. triceps brachii	4
M. infraspinatus	1

comparisons, estimated means and standard error of differences of these means for the loin muscle are presented in Table 3. Generalized size effects, estimated means for eating quality traits resulting from various factors imposed and determination of statistical significance resulting from this study extend outcomes of Channon et al. (2016b; where correction factors were estimated) and previously reported qualitative and quantitative reviews.

Sex

In this study, estimated means for sensory tenderness, juiciness and flavor, as well as shear force of pork, did not significantly differ between females, immunocastrated males, physically castrated males and entire males. Trefan et al. (2013) also showed that while average tenderness and juiciness scores of pork from immunocastrated males were 3 to 5 sensory units higher than pork from entire male and physically castrated pigs, no differences due to sex were found. The inconsistent effects of sex on pork quality traits reported in the literature (D'Souza and Mullan, 2002; Hennessy et al., 2006; Font i Furnols et al., 2008; Jeong et al., 2008a, 2008b; Silveira et al., 2008; Font i Furnols et al., 2009; Pauly et al., 2010; Pauly et al., 2012; Moore et al., 2017), together with the small number of studies involving immunocastrated males, may assist with explaining the lack of a significant effect due to sex identified in this study. Higher intramuscular fat levels were found in pork from physically castrated males compared with pork from entire males and females, with immunocastrated males intermediate (Trefan et al., 2013). For those studies involving immunocastrated males, only 9 met the eligibility criteria for juiciness, flavor, and shear force and 7 for tenderness. Our findings highlighted that additional data is required to more reliably quantify the effect size for gender, specifically between immunocastrated males and females, in terms of eating quality.

Interestingly, when data for immunocastrated and physical castrated males were pooled in this study, a strong tendency ($P = 0.059$) for tenderness scores of loins from immunocastrated and physical castrated males to be higher was identified compared with those from entire males and females. Furthermore, shear force values of pork from entire males were higher ($P = 0.034$) than for females and pooled data for immunocastrated males and physical castrates.

These findings suggest that entire males should not be included in any eating quality system to manage potential eating quality issues, reduce product variability and minimize risks of product that does not consistently meet consumer expectations for high quality pork. In the context of the current Australian pig slaughter population, with an moving annual total hot carcase weight of 76.4 kg when pigs are slaughtered at around 20 to 22 wk of age (as at March 2017; Australian Pork Limited), the risk of boar taint is still an issue with entire male production (D'Souza et al., 2011). This is of concern particularly as physical castration of entire males is not routinely practiced in Australia.

Genotype

Genotype was analyzed based on groupings and, on average, estimated means of sensory scores for loins sourced from Berkshire or Hampshire pigs were found to be higher for tenderness ($P = 0.002$) and juiciness ($P = 0.001$) and lower ($P < 0.001$) for shear force than Pietrain, White or > 50% Duroc pigs. However, this analysis only included 9 studies that had evaluated eating quality traits for Berkshire and/or Hampshire pigs compared with other genotypes. Overall, these findings reflect those of Fjelkner-Modig and Persson (1986), Jeremiah et al. (1999) and Lundström et al. (1998) who found that pork from Hampshire pigs was more intense in flavor, juicier, and more tender than pork from Duroc and White pigs.

Pork from Berkshire pigs has been shown to be more tender and flavorsome than White pigs (Lee et al., 2012). This may be an outcome of increased fatness at a given liveweight than White or Duroc breeds (Crawford et al., 2010; Wood et al., 2004). Lee et al. (2012) also found that the cross-sectional area of Type IIB fibers was lower and the total fiber density and number of Type IIB muscle fibers were higher (as well as lower cross-sectional area of Type I fibers) in loins from Berkshire compared with Duroc and White (Landrace and Yorkshire) pigs; this was associated with initial tenderness. Higher intramuscular fat contents in loins from Berkshire and Duroc pigs compared with White pigs have also been reported (Warriss et al., 1990; Wood et al., 2004). This may reflect that selection pressure for growth efficiency and leanness has not been as intense for Berkshires as that of composite breeds. Trefan et al. (2013) found that both estimated juiciness and tenderness scores were influenced by breed, and indicated lower scores for purebred Large White and Landrace. However, neither data nor details of breed evaluated in their analysis was provided. Although some breed differences were identified from our analysis, any commercial application of these findings by the Australian pork industry to improve pork eating quality consistency via genetic selection may be challenging. This may be due to the significant variability within sire breeds (Bunter et al., 2008), the extensive use of composite genetic types for commercial pork production (comparisons of which are not in the public domain) and considerations of economic targets (including cost of production) by producers.

Table 3. Estimated means and average standard error of difference (SED) from meta-regression analysis on sensory tenderness, juiciness, and flavor scores of pork and objective shear force measures for various factors

Factor	Explanatory variable	Tenderness	Juiciness	Flavor	Shear force, kg
Sex					
n (studies)		48	45	38	53
n (comparisons)		230	219	158	197
F statistic		1.93	0.46	0.69	2.38
n.df: d.d.f		3,189.9	3,180.4	3,120.8	3,146.6
	Entire male	54.1	48.6	51.8	4.90
	Female	54.8	48.4	53.1	4.66
	Immunocastrated male	57.2	50.5	53.5	4.53
	Physical castrate	56.8	48.9	53.6	4.61
	SED	1.54	1.54	1.48	0.146
	P-value	0.13	0.71	0.56	0.072
n (studies)		48	45	38	53
n (comparisons)		230	219	158	197
F statistic		2.88	0.28	1.04	3.38
n.df: d.d.f		2,190.3	2,180.8	2,121.9	2,146.5
	Entire male	54.1	48,6	51.8	4.92
	Female	54.8	48.4	53.1	4.65
	Physical castrate + Immunocastrated male	56.9	49.2	53.6	4.61
	SED	1.14	1.16	1.18	0.111
	P-value	0.059	0.76	0.36	0.037
Genotype					
n (studies)		24	27	20	28
n (comparisons)		117	135	95	143
F statistic		3.06	4.84	1.30	12.20
n.df: d.d.f		4,94.6	4,107.8	4,71.9	4,116.2
	Berkshire	65.8	53.3	65.3	3.83
	> 50% Duroc	60.3	50.6	60.6	4.84
	Hampshire	63.7	54.7	61.8	4.02
	Pietrain	59.2	48.7	60.2	5.85
	White	57.3	48.2	59.6	5.28
	SED	3.41	2.32	2.27	0.352
	P-value	0.020	0.001	0.30	< 0.001
Halothane status					
n (studies)		13	11	8	16
n (comparisons)		41	48	27	78
F statistic		3.01	6.95	0.53	6.35
n.df: d.d.f		2,28.3	2,34.0	2,17.1	2,61.5
	Homozygous dominant	58.3	53.6	52.2	4.50
	Heterozygous	56.6	49.6	53.0	4.84
	Homozygous recessive	52.6	44.3	50.0	5.50
	SED	2.13	2.33	2.64	0.27
	P-value	0.065	0.003	0.60	0.003
Hampshire gene					
n (studies)			3	3	4
n (comparisons)			9	9	15
F statistic			32.07	0.58	4.87
n.df: d.d.f			1,5.0	1,5.0	1,10.0
	rn-		60.4	50.1	5.72
	RN+		65.8	50.7	5.16
	SED		0.96	0.86	0.252
	P-value		0.002	0.48	0.052

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Factor	Explanatory variable	Tenderness	Juiciness	Flavor	Shear force, kg
Plane of nutrition					
n (studies)		14	13	11	11
n (comparisons)		59	56	39	59
F statistic		2.66	0.65	0.44	2.56
n.df: d.d.f		3,43.3	3,40.8	3,25.1	3,46.8
	Ad libitum Grower (G) Finisher (F)	54.1	45.2	47.4	5.17
	Ad libitum G Restricted F	50.8	43.7	47.1	5.58
	Restricted G Ad lib F	53.4	43.8	47.4	5.41
	Restricted GF	51.2	44.0	46.8	5.59
	SED	2.003	1.983	1.020	0.383
	P-value	0.060	0.59	0.73	0.067
Housing type					
n (studies)		7	7	8	17
n (comparisons)		21	21	20	57
F statistic		0.38	0.24	1.43	0.23
n.df: d.d.f		2,12.3	2,10.3	2,8.1	3,37.7
	Barn/deep litter	69.2	55.7	61.6	4.55
	Indoor/conventional	66.3	57.0	62.5	4.65
	Outdoor	68.6	58.1	64.0	4.75
	Outdoor access				4.77
	SED	5.90	3.52	1.65	0.272
	P-value	0.70	0.79	0.29	0.88
Ractopamine					
n (studies)		8	8	7	15
n (comparisons)		35	35	33	50
F statistic		1.61	0.29	0.35	5.64
n.df: d.d.f		2,27.6	2,26.2	2,24.1	2,41.2
	No ractopamine	58.3	51.5	52.4	4.13
	Ractopamine – 5 mg/kg	55.7	49.9	51.6	4.65
	Ractopamine – 7.5 to 10 mg/kg	54.6	50.7	51.4	4.54
	SED	2.81	2.28	1.59	0.230
	P-value	0.22	0.75	0.71	0.013
pST					
n (studies)		8	8	6	13
n (comparisons)		74	74	50	74
F statistic		13.79	6.22	3.57	6.11
n.df: d.d.f		1,64.3	1,67.5	1,42.2	1,61.2
	No pST	61.47	54.5	61.2	4.39
	pST	55.82	50.8	58.8	4.81
	SED	1.52	1.51	1.27	0.168
	P-value	< 0.001	0.015	0.066	0.016
Stunning method					
n (studies)					5
n (comparisons)					51
F statistic					0.66
n.df: d.d.f					1,46.9
	Carbon dioxide				6.59
	Electrical				6.33
	SED				0.314
	P-value				0.42

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Factor	Explanatory variable	Tenderness	Juiciness	Flavor	Shear force, kg
Electrical stimulation					
n (studies)		5	5		7
n (comparisons)		35	26		76
F statistic		1.43	6.73		7.56
n.df: d.d.f		3,29.8	3,18.9		3,66.2
	No stimulation	50.3	50.6		5.91
	Constant current – 2 to 4 min post-slaughter	56.8	54.3		5.29
	High voltage – 20 min post-slaughter	52.7	45.9		4.90
	Low voltage – 3 to 5 min post-slaughter	50.5	49.0		4.74
	SED	5.14	3.18		0.437
	P-value	0.25	0.014		< 0.001
Hanging method					
n (studies)		3	3		3
n (comparisons)		48	48		44
F statistic		8.30	1.95		10.71
n.df: d.d.f		1,43.9	1,44.0		1,40.0
	Achilles	50.4	45.6		4.93
	Aitchbone/tenderstretch	55.5	47.2		4.26
	SED	1.75	1.13		0.203
	P-value	0.006	0.17		0.002
Chilling					
n (studies)		6	5	3	7
n (comparisons)		23	14	10	43
F statistic		4.27	0.23	0.14	8.07
n.df: d.d.f		2,16.4	2,7.6	1,6.1	2,37.2
	0 to 4°C for 18 to 24 h (conv)	54.4	49.0	57.6	6.13
	Rapid chilling then conv	53.3	49.8	58.4	4.96
	Warm chilling then conv	46.8	51.5		4.64
	SED	3.38	3.64	2.03	0.581
	P-value	0.032	0.80	0.725	0.001
Ageing period					
n (studies)		13	13	7	21
n (comparisons)		109	108	80	234
F statistic		6.56	1.04	7.11	43.13
n.df: d.d.f		2,100.3	2,99.6	2,74.8	2,215.8
	≤ 2 d	55.2	52.5	60.2	5.40
	3 to 7 d	60.0	53.5	63.3	4.44
	> 7 d	59.4	50.6	54.9	4.18
	SED	2.41	1.98	2.35	0.147
	P-value	0.002	0.36	0.001	< 0.001
Moisture infusion					
n (studies)		5	5	6	5
n (comparisons)		34	34	38	34
F statistic		27.59	20.28	4.47	7.10
n.df: d.d.f		1,28.2	1,28.7	1,31.4	1,29.7
	No moisture infusion	49.6	54.1	60.7	4.20
	Moisture infusion	63.0	64.6	65.5	3.31
	SED	2.53	2.31	2.30	0.331
	P-value	< 0.001	< 0.001	0.043	0.012

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Factor	Explanatory variable	Tenderness	Juiciness	Flavor	Shear force, kg
Endpoint temperature					
n (studies)		10	16	11	4
n (comparisons)		49	112	88	18
F statistic		4.14	31.23	4.37	1.40
n.df: d.d.f		3,39.7	3,95.1	3,74.9	1,13.4
	65 to 69°C	65.5	50.9	46.7	
	70 to 74°C	60.7	48.2	48.9	3.51
	75 to 79°C	57.9	45.0	47.7	3.70
	> 80°C	53.0	38.5	44.2	
	SED	3.48	1.63	1.80	0.152
	P-value	0.012	< 0.001	0.007	0.26
Cut type					
n (studies)		4	8	8	
n (comparisons)		30	83	70	
F statistic		27.77	2.43	0.03	
n.df: d.d.f		1,24.4	1,80.3	1,62.7	
	Chop/steak	54.5	38.7	47.6	
	Roast	64.2	35.4	47.9	
	SED	1.84	2.14	1.80	
	P-value	< 0.001	0.12	0.87	

Halothane Gene

In this study, the estimated means for juiciness was lower ($P = 0.003$) and shear force was higher ($P < 0.001$) for pork from homozygous recessive pigs compared with heterozygous and homozygous dominant pigs for the halothane gene. The impact on juiciness resulting from the mutation in the ryanodine receptor in the sarcoplasmic reticulum in muscle cells of pigs carrying the halothane gene, which leads to a hypersensitive calcium channel, can be explained by a faster post-slaughter glycolytic rate resulting in protein denaturation, paler colored pork with a lower ultimate pH, lower water holding capacity, and increased drip loss.

The incidence of pigs that are heterozygote for the halothane gene is now low in the Australian commercial slaughter pig population, as a consequence of the removal of homozygous recessive animals from nucleus herds, to overcome pork quality issues associated with pale, soft, and exudative pork (PSE; Channon and Warner 2011). While it may be that the halothane gene may no longer be playing a significant role, further understanding of the biochemical mechanisms that are contributing to the low ultimate pH of Australian pork that has more recently been observed (Jose et al., 2013; Channon et al., 2016a) is needed to optimize pork eating quality. In relation to the loin muscle, it may be that selection for growth rate, reduced subcutaneous fat content and increased lean meat deposition resulted in a higher proportion of predominantly glycolytic type IIB fibers and higher concentrations of glycogen in resting muscle (together with reduced intramuscular fat levels). This may subsequently be contributing to a faster rate of glycolysis post-mortem and lower ultimate pH. Unlike beef and lamb, neither a pH/muscle temperature window nor an ultimate pH cut-off have been established for pork. Further work may therefore also be needed to implement chilling practices to improve the management of post-slaughter temperature decline of the pig carcase, particularly those muscles that may be susceptible to a more rapid rate of pH decline and/or a lower ultimate pH, to minimize technological quality issues.

Plane of Nutrition

The effect of plane of nutrition provided to grower/finisher pigs on pork eating quality attributes did not influence estimated means for tenderness ($P = 0.060$), flavor ($P = 0.73$), and juiciness ($P = 0.73$) or objective shear force measures ($P = 0.067$). These outcomes may, in part, reflect the summary nature of this analysis; in that it was informed by different studies used for the various comparisons between explanatory variables. It was also not possible to account for variations in diet composition, including protein, lysine and energy content, sex, and genotype of pigs in this analysis due to the differences in methodologies used between studies.

Housing

Neither eating quality attributes nor shear force of pork were influenced by housing pigs in indoor/conventional, outdoor, and barn/deep litter systems. Our findings

therefore indicate that pigs raised outdoors on pasture or in deep litter systems do not produce pork of consistently higher eating quality than conventionally housed pigs. In a meta-analysis, Demori et al. (2012) also showed that juiciness, tenderness, and shear force were not affected by housing system (outdoor vs. indoor) used for pig production. No differences in sensory quality were found between conventionally raised and outdoor raised pigs (Morrison et al., 2007). This was despite pigs raised in deep litter, large group systems spending more time interacting with their environment, standing and moving, exhibiting more exploratory behavior and produced pork with lower ultimate pH and higher drip loss compared with indoor/conventionally housed animals.

Metabolic Modifiers

Ractopamine. Ractopamine, a β-adrenergic agonist that acts as a repartitioning agent, is typically included in the diet of Australian finisher pigs at a level of 5 mg/kg for a period of 28 d prior to slaughter (where permitted by customers) to increase protein deposition and muscle cell hypertrophy, with variable effects on fat accretion (Dunshea et al., 1993; Dunshea et al., 2016). This study showed that the estimated mean for shear force was reduced ($P = 0.013$) when pigs were supplemented with 5 to 10 mg/kg ractopamine compared with pork from pigs that had not been supplemented. However, no significant effects of ractopamine supplementation was found for sensory tenderness, juiciness or flavor. The inclusion of 10 mg/kg ractopamine in finisher pig diets has been shown to increase shear force (Aalhus et al., 1990) and lower tenderness scores (Carr et al., 2005a) compared with control pigs. These effects may due to a slower protein degradation rate in response to increased calpastatin activity and reduced calpain I levels in porcine muscle, however, these effects have not been consistent (e.g., Smith et al., 1995; Stoller et al., 2003; Fernandez-Duenas et al., 2008; Moore et al., 2009; Rincker et al., 2009). Our findings highlight the usefulness of the meta-regression analysis approach used here to summarize the variable outcomes that have been reported by the many randomized studies that have determined the effects on pork eating quality resulting from ractopamine supplementation to finisher pigs.

Although this study was constrained to estimating differences between different explanatory variables within a single factor, outcomes from individual studies indicate that there may be opportunities for other factors to be imposed to improve eating quality. For example, Xiong et al. (2006) concluded that ageing of vacuum packaged pork loin for 10 d from pigs supplemented with 20 mg/kg ractopamine for 28 to 30 d prior to slaughter was effective in achieving comparable shear force values to pork from control animals after ageing for 10 d. While these supplementation levels are higher than those typically used for Australian pigs, these findings suggest that extended ageing may be a useful intervention to apply to overcome slower proteolytic rates of muscles of ractopamine-fed pigs. In a study involving ractopamine supplementation at 0, 10, and 20 mg/kg to surgical castrates for 25 to 41 d prior to slaughter, Carr et al. (2005b) showed that moisture infusion negated the effect of ractopamine on tenderness, with no differences in average tenderness scores (or shear force) of loin from 10 mg/kg ractopamine-fed pigs and those from control animals.

Porcine Somatotropin (pST). The administration of pST to finisher pigs was found to negatively influence the sensory traits of tenderness ($P < 0.001$), juiciness ($P = 0.015$) and shear force ($P = 0.016$) compared with pigs not treated with pST. This was determined using a data set comprising of 8 studies for tenderness and juiciness, 6 studies for flavor and 13 for shear force. Tenderness, flavor and/or juiciness has been shown to be negatively affected by the use of pST (Prusa et al., 1989; Goodband et al., 1990; Boles et al., 1991, 1992; Hagen et al., 1991; Goodband et al., 1993; D'Souza and Mullan, 2002) while others have reported no effect on sensory quality (Nieuwhof et al., 1991; Prusa et al., 1993; Moore et al., 2012). The administration of pST to pigs can reduce subcutaneous and intramuscular fat levels and increases lean meat yield (Beermann et al., 1990; Boles et al., 1991; Prusa et al., 1993; Oksbjerg et al., 1995), as it has been shown to increase protein deposition and decrease both adipose tissue growth and the rate of fatty acid synthesis. Boles et al. (1992) suggested that pST may negatively affect myofibrillar degradation post-slaughter as initial tenderness was influenced by pST treatment (reflective of myofibrillar protein contribution), but not sustained tenderness (reflective of connective tissue component). Although the effect of pST administration on muscle fiber type distribution in myofibers of the loin has not been consistently reported, pST has been widely reported to increase cross-sectional area of muscle fiber types. In relation to the potential inclusion of metabolic modifiers into a pathway based system for pork eating quality, it must also be noted that some retailer groups do not allow producers to utilize ractopamine or pST as part of their supply agreements (Dunshea et al., 2016).

With a view to building a pathway based eating quality model, significant interactions between pST administration and other pathway factors have been reported. An interaction between pST treatment (4 mg/d or none), halothane sensitivity (NN, Nn, or nn) of broiled loin chops cooked to an endpoint temperature of 71°C) were found for initial and sustained juiciness but not for initial or sustained tenderness (Boles et al., 1992). While Aalhus et al. (1997) did not observe any

significant interactions between halothane genotype (NN, Nn, or nn) and pST supplementation (3 mg/d or none), sex and muscle specific (M. semimembranosus and M. psoas major) effects of pST on quality traits were observed. These were considered to reflect differences in muscle fiber composition between muscles and/or stage of maturity of individual muscles from pigs of different sexes when pST was administered.

Post-Slaughter Factors

Stunning Method. The effect of stunning method on pork eating quality could not be evaluated in this study due to a lack of studies investigating effects of stunning method on pork eating quality attributes. Only 5 studies presented shear force data. No effect of stunning method (electrically stunning or CO_2 stunning) on shear force of loins was found. Interestingly, Rees et al. (2003b) reported shorter sarcomere lengths of muscle fibers and higher shear force values for loin muscles from CO_2 stunned pigs at both rigor and 4 d post-slaughter compared with those stunned using head to heart (1.3 amps for 4 s) electrical stunning tongs. This suggests that electrical stunning may result in a mild stimulation effect on muscle pH decline.

Electrical Stimulation. This analysis identified differences between various types of electrical stimulation systems used on pork eating quality. Pork from pigs stimulated with constant current at 2 min post-exsanguination was found to be juicier ($P = 0.003$) than pork from electrically stimulated using constant voltage, with non-stimulated carcasses intermediate. As noted by Bowker et al. (2000), differences in the electrical current or voltage applied, timing of the electrical stimulation treatment and its duration of application may explain differences in eating quality identified from this analysis. Drip loss issues and higher PSE incidence in pig carcasses stimulated with constant voltage systems has previously been reported (Taylor and Martoccia, 1995; Taylor et al., 1995; Bowker et al., 2000). As constant current electrical stimulation using 150 mA applied at 2 min has been shown to improve eating quality without resulting in increased drip loss (Channon et al., 2003), this may hold some promise as a potential intervention that can be further validated in a commercial pork establishment. Interestingly, average tenderness scores of 2 d aged loin steaks from stimulated carcases were shown to be similar to those for 7 d aged, non-stimulated pork loins. It is noteworthy that both the sheep-meat (Thompson et al., 2005) and the beef (Thompson, 2002) Meat Standards Australia models have incorporated electrical stimulation as a voluntary intervention option.

Hanging Method. The suspension of carcasses from the aitchbone, applied immediately post-dressing, stretches the loin as well as the topside (M. semimembranosus), silverside (M. biceps femoris) and rump (M. gluteus medius) that may shorten on Achilles-hung carcasses (Harris and Shorthose, 1988). This analysis identified that tenderstretching or hanging carcasses from the aitchbone on entry into the chiller increased estimated means for tenderness ($P = 0.006$) by 5 sensory units and reduced those for shear force ($P = 0.002$) by 6.5 N compared with carcasses hung from the Achilles tendon. Hanging pork carcasses from the aitchbone has been shown to increase sarcomere length in the loin (Taylor et al., 1995; Møller and Vestergaard, 1986; Rees et al., 2003a). The increase in sarcomere length in loin muscles resulting from low voltage electrical stimulation has been shown to be comparable to that achieved by tenderstretching (Taylor et al., 1995; Rees et al., 2003a). Furthermore, Dransfield et al. (1991) concluded that tenderstretching of pig carcasses from the aitchbone improved objective tenderness (as assessed by Volodkevich shear test) to a similar extent than high voltage (700 V, 12.5 Hz for 90 s) electrical stimulation applied at 20 min post-slaughter.

Ageing Period. Proteolytic degradation of myofibrillar structure by endogenous enzymes is responsible for tenderization effects resulting from ageing of pork post-slaughter. Estimated means for tenderness ($P = 0.002$) and flavor ($P = 0.001$) were increased, and shear force values decreased ($P < 0.001$), by ageing pork loin for more than 2 d post-slaughter. These outcomes indicate that ageing period could be an effective pathway intervention to include in a pork eating quality predictive model. However, few studies have directly compared the effect of varying ageing periods on different pork primals and/or cut types. Channon et al. (2014) showed that while ageing period for 7 d and hanging carcasses from the aitchbone (compared with ageing for 2 d and Achilles hung carcasses) each improved sensory tenderness, flavor, overall liking, and quality grade scores of loin steaks cooked to an endpoint temperature of 75°C, no interactions were significant. Sensory evaluations of loin steaks and topside roasts were also conducted in separate panels. Additional eating quality data, on both a primal and cut-type basis, is needed to reliably utilize ageing period to improve eating quality consistency.

Moisture Infusion (or Enhancement). Moisture infusion, involving the injection of brines at a rate of 6 to 15% of green weight, can improve pork eating quality compared with non-infused pork as a result of swelling of the myofibrillar lattice to increase water content due to the functionality of various brine ingredients used (Detienne et al., 2003; Jensen et al., 2003; Sheard and Tali, 2004; Carr et al., 2005a; Sheard et al., 2005; Hayes et al., 2006; Moore et al., 2012). In this study, moisture infusion increased estimated means for tenderness (*P*

< 0.001), flavor ($P = 0.043$), and juiciness ($P < 0.001$) and reduced ($P = 0.012$) shear force compared to non-infused pork. Compared to all other production and processing factors investigated in this study, the largest effect size on eating quality traits resulted from moisture infusion. Improvements of more than 10 sensory units (on a 0 to 100 scale) for both tenderness and juiciness were found for moisture infused loin cuts.

Although moisture infusion may be regarded as the most reliable method available to processors and wholesalers to improve pork eating quality, appropriate systems to manage raw material selection should be implemented to optimize processing yields, color and drip loss. As moisture infusion is currently only applied to approximately 10 to 15% of Australian pork sold domestically, alternate interventions (and combinations of these) need to be identified that will enable improvements in pork eating quality that are at least equivalent to those achieved by moisture infusion. This presents significant challenges to industry as similar improvements in eating quality have not been replicated when factors other than moisture infusion have been imposed. Furthermore, no interaction between ageing period (1 or 7 d post-slaughter) and moisture infusion (none, 10% brine) was observed for any sensory traits by Moore et al. (2012).

Endpoint Temperature. While pig producers and processors may strive to do all they can to ensure that fresh pork produced is of as high as quality as possible, this may all be undone when consumers prepare pork meals in the home (Ngapo and Gariepy, 2008). Estimated means for tenderness ($P = 0.012$), juiciness ($P < 0.001$), and flavor ($P = 0.007$) decreased with increasing endpoint temperature to ≥ 80°C. The impact of overcooking of pork on eating quality therefore remains an important issue to address. Estimated means for tenderness of pork cooked as a steak/chop were also lower ($P < 0.001$) compared with roasting (either as a steak or whole piece). Notably, few published studies have directly compared the effect of cooking method and endpoint temperature of loins in addition to other muscles and cuts. This has presented difficulties in our attempts to utilize published data to develop a cuts-based predictive model for pork eating quality. These identified gaps are starting to be filled to inform a pathway based eating quality model through the conduct of multi-factorial studies (Jose et al., 2013; Channon et al., 2016b; Moore et al., 2017; Channon et al., 2017).

However, Wood et al. (1995) summed up the importance of endpoint temperature on pork eating quality by stating that greater improvements in tenderness were obtained by reducing endpoint temperature from 80 to 65°C than those resulting from the inclusion of > 50% Duroc genotype and ad libitum feeding. These eating quality improvements due to endpoint temperature were also considered by Wood et al. (1995) to be comparable to those arising from ageing period and either hanging method or electrical stimulation.

Implications

Overall, studies that have investigated various combinations of on-farm, processing and post-slaughter factors, including ageing period, electrical stimulation, tenderstretching, moisture infusion, and endpoint temperature, on their effects on pork eating quality attributes have largely concluded that these factors are additive, rather than multiplicative, as interaction terms were generally not significant.

This study highlighted the challenges, and complexities, that are being faced as attempts to build a pork eating quality system continue. The use of meta-regression statistical techniques to estimate means for key factors and to determine statistical differences between the various explanatory variables within a factor was useful, but was limited by the number of studies that met the selection criterion as well as differences in study designs and methodologies used. Unfortunately, it was also not possible to establish a rating system to summarize the effect of different explanatory variables on pork sensory attributes and shear force as the estimated means for each factor were determined using data from different studies.

Conclusion

To develop a predictive model, studies that have investigated similar combinations of pathway parameters are needed to enable more than one pathway parameter to be investigated and develop predictive models with either additive and/or multiplicative terms. This study has provided insight of current data and knowledge gaps and informed future experimental designs of multi-factorial studies to both fill these gaps and enable relationships between different pathway factors to be determined. Based on our outcomes, this should include extending the literature with eating quality data for immunocastrated males (to more reliably estimate sex effects on eating quality) as well as obtain additional data on different muscles, different cuts, cooking methods, and endpoint temperatures. This is needed to ensure that the model can be reliably applied to muscles from different anatomical locations of the carcass in addition to the loin. Those key factors that were found to positively influence eating quality traits in this study now need to be commercially validated. In conclusion, alternative model structures are needed to enable more reliable estimations of the effects of different pathway factors on pork eating quality to be predicted.

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