

# Success of fishmeal replacement through poultry by-product meal in aquaculture feed formulations: a meta-analysis

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## Abstract

Poultry by-product meal (PBM) is a popular animal-based protein source tested in aquaculture feed formulations for replacing fishmeal (FM), mainly due to its high protein content, relatively cheap price and broad availability. However, due to the well-documented variability in success of PBM inclusions, a meta-analysis approach was utilized to summarize the efficacy and success of PBM inclusion in aquaculture diet formulations substituting FM. Hedges'g effect sizes were calculated by quantitatively comparing final weight and feed conversion ratio (FCR) between animals fed control (100% FM) and PBM-supplemented FM diets using data from 47 published articles targeting 33 different species (141 and 96 effect size calculations for final weight and FCR, respectively). In addition, several analyses were performed to determine the effect of different moderators as well as variation across species. Meta-regression was conducted to evaluate the effect of varying PBM proportions of the diet on the response variables. Overall, a non-significant final weight but a significant higher FCR was detected with aquaculture species fed PBM diets. Similar trends in final weight were observed for subgroups. However, FCR for crustaceans and marine fish fed PBM diets was not significantly different than those fed control diets. In both freshwater and marine fish, 'species' was a significant moderator effect on responses, while the 'level of FM replacement' was significant only in marine fish. Higher between-study heterogeneity was detected for fish, which may be due to the influence of certain factors, such as variability in nutritional quality and palatability and digestibility of PBM or due to variability on the fish meal used. In contrast, low between-study variation was observed in crustaceans indicating consistent success in PBM-supplemented diets for shrimp compared with fish.

**Key words:** feed conversion ratio, feeds, fishmeal, growth, poultry by-product meal.

## Introduction

The aquaculture industry has been expanding rapidly over time; exceeding the annual growth rates of poultry, pork, dairy and beef industries (Troell *et al.* 2014), aquaculture now provides roughly half of the fish consumed globally. Although aquaculture contributes significantly to the global fish and shrimp production, its future growth is heavily dependent on the effectiveness of sustainable feed formulations due to trophic transfer inefficiencies. As a result, feed production has grown at an average annual rate of 10.3% per year since 2000 and is expected to grow to  $6.54 \times 10^{10}$  kg by 2020 and  $8.71 \times 10^{10}$  kg by 2025 (Tacon

& Metian 2015). In general, commercial fish and shrimp feeds contain 25–50% crude protein, which is the dominant and most expensive component in these feeds (Lim & Dominy 1990; Mente *et al.* 2002; Dawson *et al.* 2018) and one of the major nutrients required for maintenance and growth of animals (Shiau 1998; Davis & Arnold 2000).

Fishmeal (FM) has traditionally been the main protein source used in aquaculture feed accounting for ~68% of global fishmeal production (Mallison 2013; Tacon & Metian 2015). This is not only due to its excellent amino acid profile, palatability and digestibility, but also because fishmeal is a source of nucleotides, essential fatty acids, phospholipids, minerals and fat and water soluble vitamins

(Tacon *et al.* 2009). Considering the upsurge in usage, Hardy (2010) argued that the demand will soon exceed the world production of fishmeal based on the expected growth rates of aquaculture production and fishmeal utilization in feeds. However, a steady decline of fishmeal inclusion levels in aquaculture feeds has been observed in recent years and may be in response to static supply, increasing price or ethical considerations (Oliva-Teles *et al.* 2015; Han *et al.* 2018). As an alternative to fishmeal, several animal protein sources have been tested, including rendered by-products, such as meat and bone meal and poultry by-product meal (PBM), due to their high crude protein (45–65%) content, good amino acid profile (Davis & Arnold 2000), consistent availability and relative low cost. However, some of the rendered animal protein meals, such as blood meal, hydrolysed feather meal or meat and bone meal, often have deficiencies or excesses in essential amino acids resulting in considerable variability in performances in fish especially when used alone as the main source of protein in diets (Davies *et al.* 1989; Hegedüs *et al.* 1990; Bureau *et al.* 2000; Abdel-Warith *et al.* 2001; Fasakin *et al.* 2005).

Poultry by-product meal, one of the most common poultry-based ingredients used in feed formulations, consists of ground rendered clean parts of the carcasses of slaughtered poultry, such as head, neck, feet and undeveloped eggs, exclusive of feathers and intestines (Dong *et al.* 1993; Cruz-Suárez *et al.* 2007). PBM emerged as one of the most promising alternative ingredients for fishmeal due to its high protein content, essential fatty acids, vitamins, minerals, palatability and protein quality (Cruz-Suárez *et al.* 2007; Gunben *et al.* 2014). Furthermore, PBM was identified as a relatively cheap source of protein compared with FM and readily available in large quantities throughout the year, especially in poultry-producing regions such as Asia (Abdul-Halim *et al.* 2014). However, like other animal-based proteins, variation in compositional quality is common, largely due to the variability in raw material composition, quality and processing specifications, such as temperature, time and pressure. Such variation results in deficiencies in certain essential amino acids, higher ash content and variability in digestibility (Davis & Arnold 2000; Robinson *et al.* 2001; Tacon *et al.* 2006; Garza De Yta *et al.* 2012; Dawson *et al.* 2018). Nevertheless, advancements in processing technologies in modern rendering facilities counteract these challenges through computerized process control in time and temperature of the cooking process that is critical in determining the final product quality (Cruz-Suárez *et al.* 2007; Najafabadi *et al.* 2007). In addition, laws and regulations in the selection of raw materials (such as prohibiting renderers from accepting and processing animals infected with avian influenza), sanitary transportation and handling play a vital role in safeguarding the product quality to stimulate its use in aquaculture

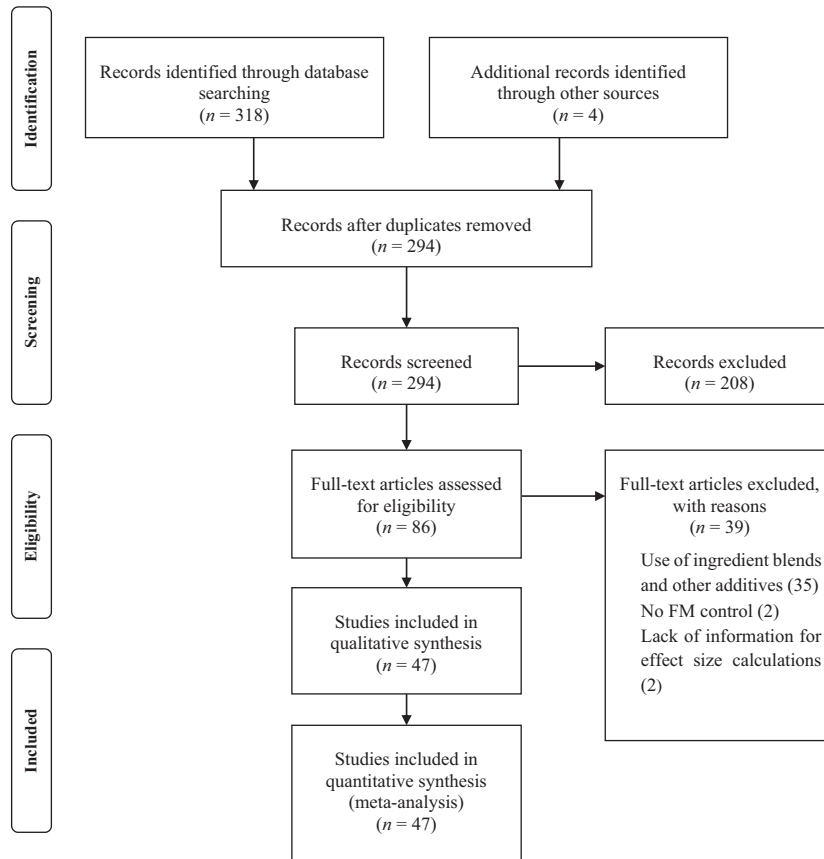
feeds (Bureau *et al.* 1999; Cruz-Suárez *et al.* 2007; Garza De Yta *et al.* 2012). As a result, PBM has been tested with varying success to replace FM in different dietary inclusion levels for numerous finfish and shellfish species. Hence, the current study uses meta-analysis to quantitatively synthesize the efficacy and success of PBM inclusion in aquaculture diet formulations as an alternative for FM across a wide range of studies, species and environmental conditions.

## Materials and methods

### Literature search and inclusion criteria

To evaluate growth performances (final weight and feed conversion ratio (FCR)) of fish and shrimp in response to different levels of FM replacements with PBM, inclusion criteria were set prior to the database search to minimize publication selection bias. Therefore, the following inclusion criteria were considered: (i) use of pure PBM in the feed formulations without any supplementation; thus, supplementation with any additional products, such as animal by-products and other attractants to yield a unique taste and improve the nutritional content of diet, was not considered; (ii) availability of isonitrogenous dietary information; (iii) reduction levels of FM as the inclusion effect of PBM in dietary formulation; and (iv) the assessment of organismal growth performance providing sufficient details for effect size calculations, such as sample size, mean response and some standard measurement of error. Studies fulfilling the above criteria were considered eligible to be included in this study. Following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines (Liberati *et al.* 2009), a comprehensive literature search was conducted in Web of Science to identify scientific articles investigating the growth responses of fish or shrimp over different dietary inclusion levels of PBM that replaced FM as the main protein source (Fig. 1). Literature searches used multiple combinations of the following terms: 'poultry-by-product meal', 'fishmeal', 'fishmeal replacement and growth'. In addition, several papers and data sets were added to the study based on previous experience with this field but that were missed by earlier searches as well as from citations within the papers found during the original search.

As of 10 March 2019, Web of Science generated 318 articles in response to the keyword combinations of 'poultry by-product meal and fishmeal', while 'poultry by-product meal and fishmeal replacement' resulted in 140 articles. After careful assessment of the titles and abstracts of these articles, 86 papers were selected to be screened for further processing through the inclusion criteria. Thirty-nine articles were excluded due to issues with the diet formulation (use of ingredient blends and other additives ( $n = 35$ )),



**Figure 1** Flow chart for search results and selection details based on Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA).

lack of FM control ( $n = 2$ ) or lack of data for effect size calculations ( $n = 2$ ). A total of 47 articles were selected for data extraction (Fig. 1). A total of 237 comparisons (141 for final weight and 96 for FCR) across different levels of PBM substitution among 33 different aquatic species were included in this study (Tables 1, 2).

### Data collection and analysis

All effect size calculations were conducted using the *metafor* package (Viechtbauer 2010) in the statistical software program *R* (R Core Team 2013). Hedges'  $g$  was chosen as the effect size due to its ease of interpretation, strong statistical properties and ability to correct for differences in sampling efforts between studies while accounting for small sample sizes (Cooper *et al.* 2009; Rosenberg *et al.* 2013). In simple terms, Hedges'  $g$  quantifies the magnitude of difference between the mean of the treatment population ( $\mu_1$ ; PBM replacement diet) to the mean of the control population ( $\mu_2$ ; FM diet) scaled by the pooled weighted standard deviation ( $SD_p$ ) (Hedges 1992; Preisser *et al.* 2005; Nakagawa *et al.* 2017).  $N$  is equal to the number of sample sizes of the

two populations or groups ( $N = n_1 + n_2$ ) which is used for the bias correction in small sample sizes.

$$g = \frac{\mu_1 - \mu_2}{SD_p} \times \frac{N - 3}{N - 2.25} \times \sqrt{\frac{N - 2}{N}}$$

A Hedges'  $g$  estimate of zero indicates no statistical difference in final weights or FCR between FM control and PBM replacement treatment diets. Hedges'  $g$  estimates  $> 0$  (lower 95% confidence interval is greater than zero) indicate a higher final weight or FCR due to PBM replacement compared with the FM control. Additional available information that could influence effects sizes, such as experiment duration, type of species, culture system and salinity, was also collected and used in moderator analyses. Some of the studies only provided pooled standard error (PSE) as the variance term. In such scenarios, PSE was used in the effect size calculation given that PSE is approximately the average SE of all treatment groups. SE (or PSE) values were converted to standard deviation (SD) using the formula:  $SD = SE \times \sqrt{n}$ ; where  $n$  = number of experimental replicates.

**Table 1** Effect size calculation outcomes for final weight comparisons through random-effect model and mixed-effect model with FM replacement level and species as moderators

Final weight	Effect size (random-effects model)						Effect of moderators (mixed-effect model)								
	FM replace level			Species			FM replace level			Species					
	<i>k</i>	$\bar{f}^2$	Hedges' <i>g</i> value	SE	C.L.	<i>P</i> -value	$\bar{f}^2$	<i>R</i> <sup>2</sup>	<i>P</i> -value	$\bar{f}^2$	<i>R</i> <sup>2</sup>	<i>P</i> -value	$\bar{f}^2$	<i>R</i> <sup>2</sup>	<i>P</i> -value
All species	134	63.48	-0.265	0.139	-0.538 to 0.008	0.057	62.88	2.32	0.055	53.02	31.74	<0.0001	63.05	1.46	0.111
Subgroups															
Species category															
All fish species	106	79.24	-0.424	0.221	0.858 to 0.009	0.055	79.19	0	0.090	64.5	48.61	<0.0001	79.61	0	0.223
All crustaceans	28	0.98	-0.076	0.149	-0.368 to 0.217	0.612	4.74	0	0.458	7.87	0	0.525	7.65	0	0.682
Habitat															
Freshwater species	62	85.05	-0.421	0.347	-1.100 to 0.258	0.224	86.04	0	0.838	73.98	44.71	<0.0001	85.66	0	0.100
Brackish water species	20	19.06	-0.048	0.192	-0.425 to 0.328	0.802	22.14	0	0.434	17.51	9.53	0.281	21.97	0	0.474
Marine species	52	69	-0.402	0.243	-0.878 to 0.075	0.099	66.74	9.53	0.006	34.29	75.26	<0.0001	61.82	26.81	<0.0001
Fish + habitat															
Freshwater fish	56	90.04	-0.601	0.477	-1.535 to 0.333	0.207	90.53	0	0.862	77.57	58.51	<0.0001	89.37	8.57	0.027
Marine fish	52	69	-0.402	0.243	-0.878 to 0.075	0.099	66.74	9.53	0.006	34.29	75.26	<0.0001	61.82	26.81	<0.0001

C.L., confidence limits (lower and upper); FM, fishmeal;  $\bar{f}^2$ , percentage variation across studies due to heterogeneity; *k*, sample size (no. of comparisons); *R*<sup>2</sup>, amount of heterogeneity accounted for particular moderator or moderators; SE, standard error.

**Table 2** Effect size calculation outcomes for food conversion ratio (FCR) comparisons through random-effect model and mixed-effect model with FM replacement level and species as moderators

FCR	Effect size (random-effects model)						Effect of moderators (mixed-effect model)								
	FM replace level			Species			FM replace level			Species					
	<i>k</i>	$\bar{f}^2$	Hedges' <i>g</i> value	SE	C.L.	<i>P</i> -value	$\bar{f}^2$	<i>R</i> <sup>2</sup>	<i>P</i> -value	$\bar{f}^2$	<i>R</i> <sup>2</sup>	<i>P</i> -value	$\bar{f}^2$	<i>R</i> <sup>2</sup>	<i>P</i> -value
All species	94	46.41	0.389	0.134	0.127-0.652	0.004	42.61	14.02	0.0026	8.75	88.44	<0.0001	44.19	8.11	0.006
Subgroups															
Species category															
All fish species	77	69.16	0.631	0.210	0.219-1.043	0.003	63.75	21.28	0.0013	36.37	72.79	<0.0001	64.64	17.89	0.003
All crustaceans	17	0	0.042	0.180	-0.311 to 0.395	0.815	0	N/A	0.387	0	N/A	0.6329	0	N/A	0.571
Habitat															
Freshwater species	38	46.58	0.75	0.221	0.317-1.183	0.001	50.57	0	0.2351	25.28	57.49	<0.0001	40.29	21.21	0.017
Brackish water species	11	0	-0.037	0.217	-0.461 to 0.389	0.865	0	N/A	0.5462	0	N/A	0.481	0	N/A	0.636
Marine species	45	68.98	0.314	0.269	-0.213 to 0.841	0.243	58.21	37.09	0.0013	30.83	79.12	<0.0001	36.18	74.24	<0.0001
Fish + habitat															
Freshwater fish	32	68.99	1.111	0.336	0.453 to 1.770	0.001	71.02	0	0.2421	42.45	63.17	<0.0001	51.48	50.75	0.009
Marine fish	45	68.98	0.314	0.269	-0.213 to 0.841	0.243	58.21	37.09	0.0013	30.83	79.12	<0.0001	51.48	50.75	0.009

C.L., confidence limits (lower and upper); FM, fishmeal;  $\bar{f}^2$ , percentage variation across studies due to heterogeneity; *k*, sample size (no. of comparisons); *R*<sup>2</sup>, amount of heterogeneity accounted for particular moderator or moderators; SE, standard error.

To understand variation in effect sizes between potentially important subgroups, the studies were categorized within a variety of groups, including 'taxa-group' (i.e. fish, crustaceans), 'habitat' (i.e. freshwater, brackish, marine) and 'fish-habitat' (i.e. freshwater or marine fish excluding all crustaceans) groupings prior to meta-analyses. Random-effects models were used to calculate Hedges'  $g$  for final weight and FCR, because this model assumes that each study has a unique Hedges'  $g$  estimate due to biological and environmental variation across studies (Hedges 1992; Nakagawa & Santos 2012). To determine whether heterogeneity was due to the influence of moderators, a mixed-effects model was used with 'species' and 'percentage FM replacement through PBM' as primary moderators. Sensitivity analysis was done using the leave-one-out study analysis, which allows for the comparisons of between-study heterogeneity ( $I^2$ ; Higgins & Thompson 2002) with and without individual studies to determine their influence on effect sizes (Nakagawa *et al.* 2017). Random-effects meta-regression analysis (Borenstein *et al.* 2009) was conducted using the percentage FM replacement through PBM as the continuous moderator to detect its influence on Hedges'  $g$  effect sizes for final weight and FCR.

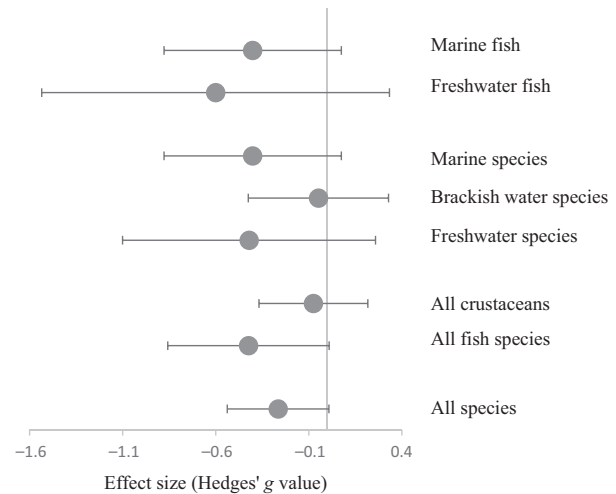
To determine whether there was publication bias, funnel plots, Egger's regression test (Egger *et al.* 1997) and Begg's rank correlation test (Begg & Mazumdar 1994) were applied to the data set. Funnel plots are commonly used to detect publication bias as they approximately resemble a symmetrical (inverted) funnel in the absence of bias (Harbord *et al.* 2009). However, this has been used as an informal technique, in which skewness in the graph is identified visually. Therefore, two additional common statistical tests, Egger's regression test and Begg's rank correlation tests, were used to quantify funnel plot asymmetry (Harbord *et al.* 2009).

## Results

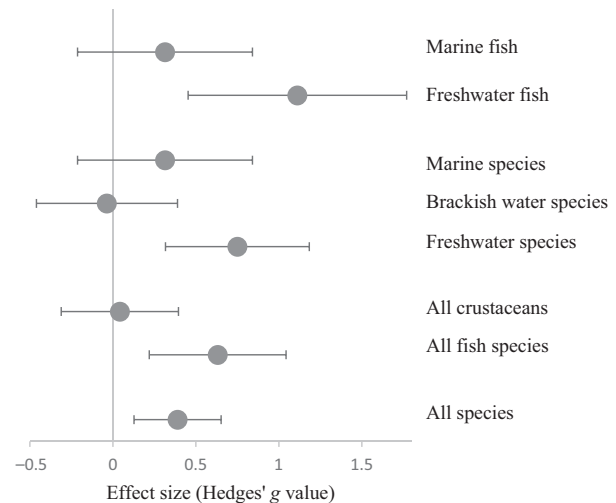
### Effect size comparisons between control and treatment groups

Comparisons between 100% FM (control) and PBM substitutions (treatments containing any concentration of PBM) yielded an overall non-significant effect of  $-0.27$  ( $P = 0.057$ ) for final weight and highly significant effect of  $0.39$  ( $P = 0.004$ ) for FCR (Figs 2, 3). Significantly higher FCR was detected during the subgroup analysis for freshwater fish ( $1.11$ ;  $P = 0.001$ ), all freshwater species ( $0.75$ ;  $P = 0.001$ ) and all fish species ( $0.63$ ;  $P = 0.003$ ) fed PBM diets. However, FCR of crustaceans ( $0.04$ ;  $P = 0.82$ ) and marine fish ( $0.31$ ;  $P = 0.24$ ) was not significantly different between control and PBM diets.

Measures of heterogeneity ( $I^2$ ) indicated considerable between-study variation for both response variables in fish



**Figure 2** Hedges'  $g$  comparisons for final weight (mean  $\pm$  95% CI); subgroup analysis (random-effects model).



**Figure 3** Hedges'  $g$  comparisons for food conversion ratio (mean  $\pm$  95% CI); subgroup analysis (random-effects model)

for final weight (79%) and FCR (69%; Tables 1, 2). Such levels of heterogeneity suggested further moderator analyses were needed to explain the factors mediating the variation in effect sizes across studies. In contrast, crustaceans had extremely low  $I^2$  (<1% for both final weight and FCR comparisons) suggesting that most of the variability in a particular subgroup was likely due to sampling error within individual experiments (Tables 1, 2).

In terms of publication bias, both Egger's regression test and Begg's rank correlation test were significant ( $P < 0.005$ ) using the complete data set or when only fish studies were included, which were supported by asymmetrical funnel plots. However, no significant publication bias was detected for crustaceans ( $P > 0.005$ ). Significant funnel

plot asymmetry could be attributed to reporting bias, true between-study heterogeneity and/or chance (Higgins & Green 2008; Sterne *et al.* 2011).

### Effect of moderators on Hedges' $g$

#### *Final weight*

Only 1.5% of the total amount of heterogeneity was accounted for by including FM replacement level and species as moderators in the mixed-effect model for final weight across all studies (Table 1). The omnibus test ( $QM = 4.40$ ,  $d.f. = 2$ ,  $P = 0.11$ ) confirmed the non-significant effect of both moderators to the overall heterogeneity, while the test for residual heterogeneity was significant ( $QE = 406.32$ ,  $d.f. = 131$ ,  $P < 0.0001$ ), possibly indicating that other moderators not considered in the model are influencing the response (Viechtbauer 2010). A non-significant effect ( $QM = 0.77$ ,  $d.f. = 2$ ,  $P = 0.68$ ) of moderators on the outcomes was noted with the crustacean subgroup, while the residual heterogeneity test ( $QE = 30.92$ ,  $d.f. = 25$ ,  $P = 0.19$ ) indicated no need for further moderator analyses due to its relatively low between-study variance. However, when it comes to final weight comparisons in freshwater fish, FM replacement level and species as moderators accounted for a significant 8.6% of the heterogeneity ( $QM = 7.21$ ,  $d.f. = 2$ ,  $P = 0.03$ ). Out of the two moderators, only species had significant influence on heterogeneity ( $P = 0.01$ ), while FM replacement level ( $P = 0.45$ ) did not contribute much to the variability between studies. In addition, a significant test for residual heterogeneity ( $QE = 216.65$ ,  $d.f. = 51$ ,  $P < 0.0001$ ) suggests other moderators not considered in the model may explain additional variability between studies. Further pronounced effects of moderators were detected in marine fish, which shared 27% of the total amount of heterogeneity for final weight. Significant effects of both FM replacement and species were detected on the overall heterogeneity in final weight comparisons of marine fish ( $QM = 19.10$ ,  $d.f. = 2$ ,  $P < 0.0001$ ); however, additional unexplained heterogeneity remained for this subgroup ( $P < 0.0001$ ).

#### *FCR*

About 8.1% of the total amount of heterogeneity was accounted for by including FM replacement level and species as moderators in the mixed-effect model for FCR in the overall data set (Table 2). Though omnibus test ( $QM = 10.27$ ,  $d.f. = 2$ ,  $P = 0.01$ ) revealed a significant contribution of moderators to the overall heterogeneity, only FM replacement level ( $P = 0.004$ ) appeared significant and other unknown moderators influenced the remaining residual heterogeneity ( $QE = 406.32$ ,  $d.f. = 131$ ,  $P < 0.0001$ ). There was a non-significant effect ( $QM = 0.75$ ,  $d.f. = 2$ ,  $P = 0.39$ ) of moderators for

crustaceans, which is further supported by low residual heterogeneity ( $QE = 4.65$ ,  $d.f. = 15$ ,  $P = 0.99$ ). However, when it comes to the FCR comparisons in freshwater fish, both FM replacement level and species as moderators accounted for 51% of the heterogeneity ( $QM = 9.43$ ,  $d.f. = 2$ ,  $P = 0.01$ ). Both FM replacement level ( $P = 0.04$ ) and species ( $P = 0.01$ ) showed a significant influence on heterogeneity as moderators for the between-study variance in FCR. However, other unexplained variation is likely due to other moderators not considered in this analysis ( $QE = 82.75$ ,  $d.f. = 29$ ,  $P < 0.0001$ ). As noted in final weight, a significant moderator effect was noted in marine fish for FCR as well, which shared 74% of the total amount of heterogeneity. For marine fish, both FM replacement ( $P = 0.005$ ) and species ( $P < 0.0001$ ) were important moderators ( $QM = 36.42$ ,  $d.f. = 2$ ,  $P < 0.0001$ ) and explained a significant portion of the residual heterogeneity ( $QE = 91.90$ ,  $d.f. = 42$ ,  $P < 0.0001$ ).

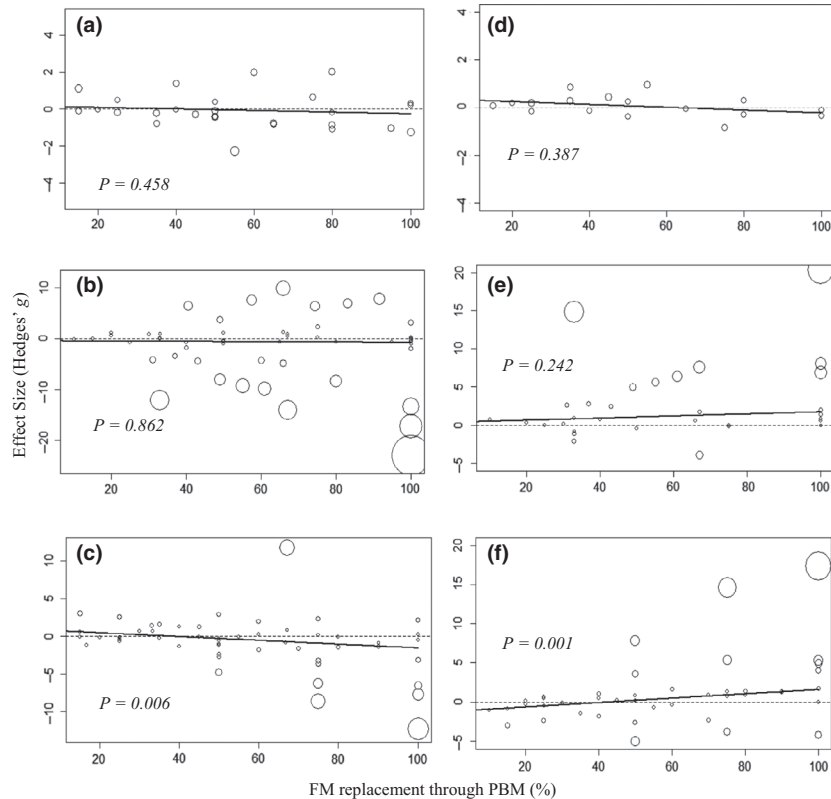
### Meta-regression analysis

Meta-regression analysis conducted using the percentage FM replacement with PBM as the continuous moderator revealed a non-significant relationship on the effect sizes of both final weight and FCR in crustaceans and freshwater fish ( $P > 0.24$ ; Fig. 4). In marine fish, a significant negative relationship was noted on the effect sizes for final weight ( $P = 0.006$ ; Fig. 4c), while a significant positive trend was noted for FCR ( $P = 0.0013$ ; Fig. 4f).

### Discussion

Ingredient characterization is a key strategy to determine the potential use of any ingredient in aquaculture feed. Chemical composition and variability due to its origin and processing specifications serves as a preliminary evaluation, while the estimation of energy and nutrient availability of ingredient and its palatability to an animal is also vital. PBM emerged as a promising alternative ingredient for FM in aquaculture feed formulations due to a variety of reasons, including high protein content, reduced ash content, good acceptance by many species based on palatability and attractability, as a good source of cholesterol and phospholipids, worldwide availability and reduced cost compared with FM (Yu 2006; Cruz-Suárez *et al.* 2007; Abdul-Halim *et al.* 2014; Gunben *et al.* 2014). As a result, numerous studies from around the world have examined a large diversity of fish and crustaceans fed diets containing various levels of PBM. However, the outcomes of these studies have varied widely, emphasizing the need to synthesize the results.

Through this quantitative synthesis, the relative difference for final weight between 100% FM (control) and PBM



**Figure 4** Fishmeal replacement through poultry by-product meal (%) as continuous moderator (random-effect model meta-regression) to detect its relationship with pooled effect sizes of final weight (a = crustaceans, b = freshwater fish and c = marine fish) and feed conversion ratio (d = crustaceans, e = freshwater fish and f = marine fish; each effect size represented by one circle, and the diameter of circle shows the confidence interval).

substitutions (treatments) was small (Hedges'  $g = -0.265$ ) and non-significant ( $P = 0.057$ ) for all species where data were available (Fig. 2). One explanation for the high between-study heterogeneity ( $I^2$ ) of freshwater and marine fish species groups could be due to high species diversity included in this study (13 freshwater fish species and 12 marine fish species). Species was shown to be a significant moderator for both freshwater and marine fish species (Tables 1, 2). In addition, FM replacement level was an important moderator for final weight in marine, but not freshwater, fish species. Numerous studies support these quantitative outcomes, revealing the possibility of total replacement of fishmeal with PBM in freshwater fish species, such as European eel (Appelbaum *et al.* 1996), Nile tilapia (*Oreochromis niloticus*) (El-Sayed 1998; Hernández *et al.* 2010), catla (*Catla catla*) (Hasan *et al.* 1993), rohu (*Labeo rohita*) (Hasan & Das 1993), sunshine bass (female White Bass *Morone chrysops* × male Striped Bass *M. saxatilis*) (Webster *et al.* 1999, 2000), hybrid striped bass (*Morone chrysops* × *Morone saxatilis*) (Rawles *et al.* 2011) and tenches (*Tinca tinca*) (Panicz *et al.* 2017). Growth performance and feed utilization of snakehead (*Channa striata*) were not adversely affected by the incremental

inclusion of PBM up to the highest protein replacement level tested (40%) by Abdul-Halim *et al.* (2014), while FM replacements with PBM did not compromise growth in rainbow trout (*Oncorhynchus mykiss*; PBM ≤ 59%) (Parés-Sierra *et al.* 2014), European eel (*Anguilla anguilla*; PBM ≤ 50%) (Gallagher & Degani 1988), African catfish (*Clarias gariepinus*; PBM ≤ 40%) (Abdel-Warith *et al.* 2001), tench (*Tinca tinca*; PBM ≤ 25%) (González-Rodríguez *et al.* 2016) and mirror carp (*Cyprinus carpio*; PBM ≤ 20%) (Emre *et al.* 2003). In summary, it appears that many freshwater fish species tolerate PBM up to 100% FM replacements, while a majority accept >50% PBM in diet without an issue.

However, broader variation in final weights in marine fish confirmed the significant effect of both moderators (species and FM replacement level) considered in this study. As a result, equivalent growth to those fed the 100% FM protein (control) was noted with PBM inclusion level up to 90% in Juvenile Black Sea Bass (*Centropristis striata*) (Dawson *et al.* 2018), up to 80% in marine Japanese Sea Bass (*Lateolabrax japonicus*) (Wang *et al.* 2015), up to 67% in Totoaba (*Totoaba macdonaldi*) (Zapata *et al.* 2016) and Florida Pompano (*Trachinotus carolinus*) (Rossi & Davis

2012; Riche 2015), up to 60% for cobia (*Rachycentron canadum*) (Zhou *et al.* 2011), up to 59% in red sea bream yearlings (*Pagrus major*) (Takagi *et al.* 2000), up to 50% in spotted rose snapper (*Lutjanus guttatus*) (Hernández *et al.* 2014), gilthead sea bream (*Sparus aurata*) (Nengas *et al.* 1999), black sea turbot (*Psetta maeoticus*) (Yigit *et al.* 2006) and Chinook salmon (*Oncorhynchus tshawytscha*) (Fowler 1991), up to 25% in silver seabream (*Rhabdosargus sarba*) (El-Sayed 1994), up to 21% in Australian snapper (*Pagrus auratus*) (Quartararo *et al.* 1998) and up to 14% in red drum (*Sciaenops ocellatus*) (Kureshy *et al.* 2000). In addition, success with 100% FM substitutions through PBM was documented with tiger grouper juveniles (*Epinephelus fuscoguttatus*) (Gunben *et al.* 2014) and humpback grouper (*Cromileptes altivelis*) (Shapawi *et al.* 2007) with no adverse effects on growth performances of fish.

In contrast to observed effects for fish, PBM inclusion in crustacean diets had negligible between-study heterogeneity ( $I^2 < 1$ ) and statistically insignificant effects on final weight ( $-0.08$ ;  $P = 0.612$ ) or FCR ( $0.04$ ;  $P = 0.815$ ). The success of PBM in crustaceans might be related to relatively few species included in our data set (i.e. two brackish water shrimp species, freshwater prawn and crayfish), minor effects of AA and FA imbalances or that the PBM diet satisfied AA and FA requirements for these taxa, or higher palatability and digestibility of PBM diets in crustaceans.

The protein content of PBM used in the studies selected for the current meta-analysis ranged from 51% to 72% due to the variation in the raw material quality and processing specifications. Several authors noted the importance of protein quality, emphasizing the lower availability of certain essential amino acids (AA) in PBM compared with fishmeal, which is assumed to have the ideal AA profile for a majority of aquaculture species (Abdel-Warith *et al.* 2001; Keramat Amirkolaie *et al.* 2014). Amino acid shortages in PBM are cited as the reason for the reduced fish growth at higher FM replacement levels relative to fishmeal, which is conditional based on the essential amino acid (EAA) requirements of the targeted species (Karapanagiotidis *et al.* 2019). In general, PBM protein is known to contain lower levels of methionine and lysine compared with FM, which is considered to be the limiting factor for growth in many species, such as Florida pompano (Riche 2015), African catfish (Abdel-Warith *et al.* 2001), humpback grouper (Shapawi *et al.* 2007), rainbow trout (Steffens 1994; Milla-mena 2002; El-Haroun *et al.* 2009; Keramat Amirkolaie *et al.* 2014), hybrid striped bass (Gaylord & Rawles 2005) and gilthead seabream (Nengas *et al.* 1999) at higher PBM inclusions in the diet. However, Yigit *et al.* (2006) suggested that the negative growth observed with black sea turbot to lysine alone, while methionine was identified as the limiting factor for hybrid striped bass (Gaylord & Rawles 2005), gilthead sea bream (*Sparus aurata*)

(Karapanagiotidis *et al.* 2019), juvenile tench (González-Rodríguez *et al.* 2016) and cobia (Zhou *et al.* 2011) at higher PBM inclusions. In a study with Sobaity sea bream, limited methionine and taurine contents were noted as the reasons for suppressed fish growth ( $>60\%$  PBM) (Hekmatpour *et al.* 2018), while lower levels of histidine and lysine negatively affected growth in sea bream (Nengas *et al.* 1999), and limited histidine, methionine, isoleucine, lysine and phenylalanine reduced growth in spotted rose snapper were identified to cause growth reductions with high inclusions of PBM (Cowey *et al.* 1985; Nengas *et al.* 1999; Hernández *et al.* 2014). In conclusion, the absence or imbalance of certain AAs in PBM may play a key role in determining the success of their used as FM replacements in aquaculture feed formulations for both freshwater and marine fish and crustaceans.

Generally,  $n - 3$  polyunsaturated fatty acids (PUFA) are indispensable for optimum growth and survival of many fish species, specifically for marine fish, with requirements vary based on target species, water temperature and natural feeding habits (Watanabe 1982; Nengas *et al.* 1999). Fish oil is considered to be the key lipid source in aquaculture feeds in part because of the high proportion of  $n - 3$  long chain highly unsaturated fatty acids (LC-HUFA), such as eicosapentaenoic acid (EPA,  $20:5n - 3$ ) and docosahexaenoic acid (DHA,  $22:6n - 3$ ) (Asdari *et al.* 2011; Zhou *et al.* 2014) which are available, typically in low abundance, in FM. In contrast, PBM and poultry oil are rich in monounsaturated fatty acids (MUFA) (particularly oleic acid) and  $n - 6$  PUFA, but with lower levels of essential fatty acids (EFA) such as  $n - 3$  LC-PUFA, EPA and DHA (Higgs *et al.* 2006; Parés-Sierra *et al.* 2014; Zapata *et al.* 2016; Panicz *et al.* 2017). This was highlighted and identified as one of the reasons for the reduced growth in certain species, such as Totoaba (Zapata *et al.* 2016), early life stages of catfish (García-Pérez *et al.* 2018), black sea turbot (Yigit *et al.* 2006) and gilthead sea bream (Nengas *et al.* 1999). In contrast, certain freshwater fish species, including rainbow trout, seem to use MUFA from PBM efficiently as they have low requirements for HUFA (Sargent *et al.* 2003; Parés-Sierra *et al.* 2014). Beyond growth in most fish species, FA content of a diet can have a strong influence on FA content of flesh and other tissues (Bell *et al.* 2002; Fonseca-Madrigal *et al.* 2005; Grant *et al.* 2008; Parés-Sierra *et al.* 2014). This was observed through accumulated lipids in liver and flesh in African catfish (Abdel-Warith *et al.* 2001), juvenile tench (González-Rodríguez *et al.* 2016) and Chinook salmon (Fowler 1991) with higher PBM inclusions in diet. In this study, the lipid content of PBM averaged 14% (ranged from 6% to 23%) compared with the average lipid content of FM (8%; range from 3% to 12%). Therefore, higher lipid content and deficiency in LC-PUFA in PBM seems to have a measurable effect on fish growth that



might explain some of the observed between-study heterogeneity observed ( $I^2$ ; Tables 1, 2).

Another possible reason for the variation in responses of fish and crustaceans fed PBM containing diets could be due to differences in digestibility of the different PBM categories (Dong *et al.* 1993). Lower digestibility values for the diets with higher inclusion levels of PBM were noted with rainbow trout (Alexis *et al.* 1985; Keramat Amirkolaie *et al.* 2014), red drum (Gaylord & Gatlin 1996), tilapia (Hanley 1987), African catfish (Abdel-Warith *et al.* 2001), humpback grouper (Shapawi *et al.* 2007) and spotted rose snapper (Hasan *et al.* 1997) compared with the diets with FM. However, Badillo *et al.* (2014) and Yan *et al.* (2014) revealed higher dry matter and protein digestibility of PMB than FM in rainbow trout and Korean rockfish, respectively. When it comes to shrimp, the majority of the studies confirmed the high digestibility of PBM in Pacific white shrimp (Davis & Arnold 2000; Cruz-Suárez *et al.* 2007) and giant tiger prawn, *P. monodon* (Luo *et al.* 2012) explaining the potential for complete replacement of FM with PBM without affecting performances (Cruz-Suárez *et al.* 2007).

It was noted by several authors (Davis & Arnold 2000; Cruz-Suárez *et al.* 2007) that PBM substitutions for FM could affect the physical properties of diets, such as reduced water stability, increased water absorption capacity and less pellet hardness, due to higher lipid and fibre content of PBM. Such changes may negatively affect consumption and growth in some fish and crustaceans (Davis & Arnold 2000; Kureshy *et al.* 2000; Abdel-Warith *et al.* 2001; Yigit *et al.* 2006; Cruz-Suárez *et al.* 2007; Shapawi *et al.* 2007; Hernández *et al.* 2014). Given that commercial feed can be found with high levels of poultry meal, this is likely an artefact of laboratory processing of feed. In any case, each ingredient has an effect on processing which should be considered. Other considerations would include palatability of the meals which can influence feed intake. At last, it is required to mention that the quality of FM (or any ingredient used to replace another) also can vary considerably depending on the freshness of the fish from which it was produced, type of fish, processing conditions, storage conditions etc. Similar to the variability in PBM used between studies, FM also has this variability which could contribute to the final result. This was not captured through this analysis, since the nutritional data of both ingredients (except for protein and lipid level) were rarely analysed or presented in research publications.

## Conclusions

Overall, no significant effect on growth was detected with PBM-supplemented diets compared with control diets with 100% FM. Similar trends were observed for all subgroups (mainly freshwater fish, marine water fish and crustaceans)

for final weight. Significantly higher FCR was detected in fish compared with crustaceans and in freshwater fish compared with marine fish. However, FCR for crustaceans and marine fish was not significantly different from control FM diets. Heterogeneity for both response variables in fish was high, emphasizing the necessity of moderators to be included to the study, while extremely low between-study variation was observed in crustaceans, might be due to relatively few species included in the data set, minor effects of AA and FA imbalances of PBM or that the PMB diet satisfied AA and FA requirements for these taxa. In both freshwater and marine fish, effects varied across different species, while level of FM replacement only significantly influenced effect sizes for marine fish. Relatively high unexplained heterogeneity suggests that other factors, such as nutritional quality (specifically amino acids or fatty acids), palatability and digestibility of PBM, could also mediate the influence of PBM supplements on the growth and FCR of fish.

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## Supporting Information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

**Table S1.** Outcomes of Egger's regression test and Begg's rank correlation test to evaluate publication bias during the study for final weight comparisons.

**Table S2.** Outcomes of Egger's regression test and Beggs's rank correlation test to evaluate publication bias during the study for FCR comparisons.