PROBIOTIC-YEAST IMPROVES PERFORMANCE INDICATORS IN BROILER CHICKENS: EVIDENCE FROM META-ANALYSIS

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Abstract. Antibiotics modulate gut microbiomes and enhance broiler chicken productivity. Nevertheless, their use in chicken nutrition has been linked to the spread of resistant strains of bacteria. Studies have shown that probiotic-yeast improves the productivity of broiler chickens. However, agreement has not been reached among investigators as to whether yeast improves the production indices of broiler chickens. The objective of this meta-analysis therefore was to determine the effect of yeast supplementation on feed consumption (FC), feed conversion ratio (FCR) and body weight gain (BWG) in broiler chicken using meta-analysis. The study focused on published primary studies comparing broiler chickens fed diet supplemented with yeast versus without yeast. Sixteen published primary studies were included in the meta-analysis. Results of pooled effects estimate revealed that yeast supplementation improved FCR ($p < 0.001$), BWG ($p < 0.001$) and reduced FC ($p < 0.001$) in broiler chickens. The results of sub-analysis indicated that broiler chickens that received yeast at < 10 g/kg diet had better performance than those that received yeast at 10 g/kg feed and > 10 g/kg feed. The chosen moderators were predictors of study effect observed in the meta-analysis. It is concluded that yeast can be used as a performance enhancer in broiler chickens instead of antibiotics.
Keyword: *beneficial microbes, meat-typed chickens, productivity, meta-analysis, meta-regression*

Introduction

Broiler chicken farming plays a vital part in supporting the livelihoods of most households in many parts of the world, because of its role as a source of animal protein. Incidentally, several households in developing nations, still regard poultry products as luxury (Adene and Oguntade, 2006). This has been attributed to the high cost of poultry products driven mainly by the elevated feed price, which led poultry nutritionist to search for feed additives with the potential to increase feed digestion and nutrient uptakes (Gadde et al., 2017). Antibiotics, one of such feed additives has been reported to modulate gut microbiomes, enhance feed efficiency, boost growth rate as well as deter the emergence of diseases in chickens (Costa et al., 2017). Regardless of the established positive actions of in feed antibiotics in boosting growth performance in chickens, their use has been tied to the proliferation of resilient strains of bacteria and meat with remnants of antibiotics (Kabir et al., 2004; Chen et al., 2017; Gadde et al., 2017) which is a threat to public health (Kabir et al., 2004; Piątkowska et al., 2012). Although, views are mixed on whether the use of antibiotics in poultry feed can transfer resistance genes from animals to humans (Gadde et al., 2017), consumers of poultry meat were increasingly concerned about the risk of continued use of antibiotics in animal feeds (Kabir et al., 2004; Piątkowska et al.,

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2012). This ugly development called for an investigation into the use of probiotics as an alternative to antibiotics in broiler nutrition (Ahmed et al., 2015; Gao et al., 2017; Ogbuewu et al., 2019).

Probiotics are living organisms that elicit positive health effects in animal and humans when included in the diet at the right proportions (FAO, 2016). The beneficial health property of probiotics can be ascribed to either its ability to modulate gut microbiomes or their direct nutritional effect (Saulnier, 2007; Shareef and Al-Dabbagh, 2009). Yeast (*Saccharomyces cerevisiae*), one of the commonly used probiotics in both human and animal food is a single-celled organism that measures about 3 to 4 microns in size. Yeast is rich in protein and amino acids (Gomes et al., 2014; Ogbuewu et al., 2019) as well as vitamin B complex and minerals (USDA, 2018). Chand et al. (2014) reported that yeast contains 93.00% dry matter, 1.00% ether extract, 44.40% crude protein, 2.70% crude fibre, 0.12% calcium and 1.40% phosphorus. Indisputably, the results of the production parameters of broiler chickens fed yeast supplemented diets in literature as reported by several authors were disaggregated and conflicting, hence calling for evidence synthesis. Evidently, there are several studies on the actions of yeast on broiler performance (Paryard and Mahmoudi, 2008; Ezema, 2013; Ahmed et al., 2015; Chen et al., 2017), and many have undoubtably shown that yeast enhance broiler chicken productivity (Ezema and Ugwu, 2014; Ogbuewu et al., 2019). Earlier reviews conducted to ascertain the beneficial effect of yeast on broiler chicken performance, however, were narrative, which is bias-friendly due to non-repeatability of the results and lacked the ability to manage large datasets. For example, Ezema and Ugwu (2014) did a narrative review of published studies of the effect of probiotic-yeast on broiler productivity and discovered that yeast improve growth rate and nutrient digestibility in chickens. Incongruity, a recent narrative review by Ogbuewu et al. (2019) found disparity on production data of broiler chickens fed yeast supplemented diets. The current study aims to use meta-analytical technique to identify and quantify several variables that affect the outcomes of the effect of probiotic yeast on the growth performance indices of broiler chickens.

Materials and methods

Selection guidelines and ethical approval

The main criteria for selecting the primary studies included in this meta-analysis were: randomized and controlled tests (RCTs) in diseased-free broiler chickens fed yeast supplemented diets. Studies were included if peer-reviewed and published in English. Study reported at least one of these growth performance indices (FC, FCR and BWG) and a dispersion metric such standard error (SE), standard deviation (SD) or 95% confidence interval (CI) for each effect size. This study was conducted at the University of South Africa during the months of June to September 2019. Literature search and data analysis were conducted in line with the guidelines of the University of South Africa Ethics Committee.

Data sources, extraction and data integrity

The investigators independently searched for articles in PubMed, Scopus and Google scholar databases from 1999 to 2017. Studies were panel-selected to guarantee the reliability of outcome of the meta-analysis, which relies on the validity of papers that

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make up the database. Yeast* and broiler chickens* were included in the search conditions. Articles were evaluated, and those that met the selection guidelines were selected. Information on the surname of the first author, the year the study was published, broiler breed, yeast supplementation level, duration of yeast supplementation, outcomes of interest (FC, FCR, BWG) and measures of variance (SE, SD or p-value) from the papers included in the meta-analysis are presented in the supplementary file. Most of the articles assessed did not report the yeast type used and the failure of the authors to obtain extra information from the corresponding author led to the removal of yeast type from the a priori selection criteria that an article must meet in order to be included in the meta-analysis. Furthermore, where the SD was not supplied, but can be estimated from SE, where it is reported using the method (Higgins and Deeks (2011).

Statistical analysis

Data analysis was performed in Open Meta-analyst for Ecology and Evolution (OpenMEE) software and forest plots were produced (Wallace et al., 2016). Continuous variable results (FC, BWG and FCR) were evaluated as the difference between the groups in yeast treatment and control with 95% CI. Q-statistic (DerSimonian and Laird, 1986) and I^2 – statistic; (Higgins et al., 2003) were used to calculate heterogeneity. Pooled effects estimate of the responses of broiler chickens to yeast supplementation were calculated using the DerSimonian and Laird (1986) random-effects model (REM). The REM selection is based on the premise that information used in the current study were not the same, therefore, the variance must be split into variance within and variance between studies plus sampling error (Borenstein et al., 2009).

For robustness of our results, we used sensitivity analysis which according to Lean et al. (2009) is used to assess the impacts of studies judged to be deviant or to have an undue influence on the analysis. This was accomplished by leaving one study each time the analysis was conducted. The influence graph shows the global impact without each study. The magnitude of the effect of yeast supplementation on broiler chicken productivity may be affected by several explanatory variables (moderators). The moderators considered in this study matched the factors we assumed to predict the association between yeast supplementation and broiler performance. In the subgroup analysis, the data were stratified using the following modifiers (study continent, study country, supplementation level, duration of supplementation and broiler breed used) thought to influence the physiological traits of broiler chickens fed probiotic yeast. Subgroup analysis was not performed when there are comparatively few studies (< 3 comparisons) in an individual stratum. The same variables used in the subgroup analysis were included as the modifier variables in the meta-regression (Dohoo et al., 2003). Meta-regression was used to examine whether the selected predictors (study continent, study country, yeast supplementation level, duration of yeast supplementation and breed of broiler used) explained any of the sources of heterogeneity.

Rosenberg's fail-safe number (Nfs) and funnel plots were used in this study to assess the existence of publication bias. Studies are said to have no publication bias when the funnel is inverted and symmetrical. Evidently, Jennions et al. (2013) have shown the robustness of meta-analysis results in the presence of publication bias regarded Nfs is greater than "5 (n = number of effect sizes) + 10". Forest plots (DerSimonian and Laird, 1986; IntHout et al., 2014) displayed the outcome of yeast intervention on broiler chicken productivity. Points to the left of the no effect line show a decline in the results

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of our parameters of interest, and the opposite demonstrates the reverse. The effect size is represented by individual square in the forest plot, while the upper and lower 95% CI for the effect size are the line that joined the square. According to Leah et al. (2009), the inverse of the effect size variance is shown by the weight of each study, whereas the square box size is equivalent to the inverse variance of the estimates with larger square boxes suggesting greater weight. The pooled effects estimate depicts the dotted line with the diamond at the bottom making up 95% CI. The pooled estimate was not significant when the diamond at the bottom is in contact with the no effect line (Koricheva et al., 2013). Effect sizes of approximately 0.2 and 0.5 are designated low and medium respectively, and large when they are more than 0.8.

Results

Overview of the articles included in the meta-analysis

The literature search in PubMed, Scopus and Google scholar produced 64 articles evaluating the productive indices of broiler chickens fed yeast supplemented diets (*Fig. 1*). A total of 25 articles were excluded from the analysis because they were reported in animal species other than broiler chickens. Six papers were removed from the study because they were narrative reviews, while five papers were excluded because they reported only the abstract. Five articles were excluded from the meta-analysis because of they were not randomized, and the control group was missing. Seven articles were not considered suitable for the analysis because they did not report any of the outcomes of interest. For the analysis of FC, 16 studies (with 4874 birds and 62 comparisons) representing 10 study countries drawn from three study continents met the eligibility criteria (*Table 1*). The publications used in the meta-analysis to assess the effect of yeast supplementation on FC in broiler chickens span 18 years with first study published in 1999 and the most current published in 2017 (*Table 1*). The FCR analysis included 15 studies containing 59 comparisons and 4874 birds conducted in 10 study countries drawn from 3 study continents (*Table 1*), while 13 studies comprising 49 comparisons and 3930 birds were included for the assessment of the effect of yeast on BWG in broiler chickens (*Table 1*). Of the all articles used in the meta-analysis to evaluate the effect of yeast on production indices of broiler chickens, the oldest study was published in 1999 and the most recent published in 2017. Most of the studies used Ross followed with Arbor acres. Yeast was added via the feed in the present meta-analysis.

Table 1. *Studies used to evaluate the effect of yeast-based diets on FC (g/bird), FCR and BWG (g/bird) of broiler chickens*

Study	Year	Sources of variation					NOB	Outcomes
		Study country	Study continent	SL (g/kg)	Broiler breeds	DS (d)		
Oyedeji et al.	2008	Nigeria	Africa	0, 0.2	Ross	28	80	FC, FCR, BWG
Al Mansour et al.	2011	Saudi Arabia	Asia	0, 1	Ross	21	120	FC, FCR
Al Mansour et al.	2011	Saudi Arabia	Asia	0, 1.25	Ross	21	120	FC, FCR
Al Mansour et al.	2011	Saudi Arabia	Asia	0, 1.5	Ross	21	120	FC, FCR, BWG
Al Mansour et al.	2011	Saudi Arabia	Asia	0, 1	Ross	42	120	FC, FCR
Al Mansour et al.	2011	Saudi Arabia	Asia	0, 1.25	Ross	42	120	FC, FCR
Al Mansour et al.	2011	Saudi Arabia	Asia	0, 1.5	Ross	42	120	FC, FCR
Njeru	2013	Kenya	Africa	0, 1.25	Arbor Acres	21	80	FC, FCR, BWG
Njeru	2013	Kenya	Africa	0, 0.63	Arbor Acres	42	80	FC, FCR, BWG

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El Fatah	1999	Sudan	Africa	0, 25	Ross	42	60	FC, FCR, BWG
El Fatah	1999	Sudan	Africa	0, 50	Ross	42	60	FC, FCR, BWG
El Fatah	1999	Sudan	Africa	0, 75	Ross	42	60	BWG
El Fatah	1999	Sudan	Africa	0, 100	Ross	42	60	FC, FCR, BWG
Mohamed et al.	2015	Sudan	Africa	0, 10	Hubbard	21	80	FC, FCR, BWG
Mohamed et al.	2015	Sudan	Africa	0, 20	Hubbard	21	80	FC, FCR, BWG
Mohamed et al.	2015	Sudan	Africa	0, 30	Hubbard	21	80	FC, FCR, BWG
Mohamed et al.	2015	Sudan	Africa	0, 10	Hubbard	42	80	FC, FCR, BWG
Mohamed et al.	2015	Sudan	Africa	0, 20	Hubbard	42	80	FC, FCR, BWG
Mohamed et al.	2015	Sudan	Africa	0, 30	Hubbard	42	80	FC, FCR, BWG
Chen et al.	2017	Taiwan	Asia	0, 2.5	Arbor Acres	21	120	FC, BWG
Chen et al.	2017	Taiwan	Asia	0, 2.5	Arbor Acres	35	120	FC, FCR, BWG
Aluwong et al.	2012	Nigeria	Africa	0, 15	Marshall	42	100	FC, FCR
Aluwong et al.	2012	Nigeria	Africa	0, 20	Marshall	42	100	FC, FCR
Osman	2006	Sudan	Africa	0, 0.2	Lohman	28	100	FC, FCR, BWG
Osman	2006	Sudan	Africa	0, 0.4	Lohman	28	100	FC, FCR, BWG
Osman	2006	Sudan	Africa	0, 0.6	Lohman	28	100	FC, FCR, BWG
Shareef and Al-Dabbagh	2009	Iraq	Asia	0, 5	Faobrow CD	21	40	FC, FCR, BWG
Shareef and Al-Dabbagh	2009	Iraq	Asia	0, 10	Faobrow CD	21	40	FC, FCR, BWG
Shareef and Al-Dabbagh	2009	Iraq	Asia	0, 15	Faobrow CD	21	40	FC, FCR, BWG
Shareef and Al-Dabbagh	2009	Iraq	Asia	0, 20	Faobrow CD	21	40	FC, FCR, BWG
Chand et al.	2014	Pakistan	Asia	0, 3.5	Hubbard	35	60	FC, FCR
Chand et al.	2014	Pakistan	Asia	0, 7	Hubbard	35	60	FC, FCR
Chand et al.	2014	Pakistan	Asia	0, 10.5	Hubbard	35	60	FC, FCR
Manal	2012	Egypt	Africa	0, 3	Arbor Acres	21	60	FC, FCR, BWG
Manal	2012	Egypt	Africa	0, 5	Arbor Acres	21	60	FC, FCR, BWG
Manal	2012	Egypt	Africa	0, 7	Arbor Acres	21	60	FC, FCR, BWG
Manal	2012	Egypt	Africa	0, 3	Arbor Acres	42	60	FC, FCR, BWG
Manal	2012	Egypt	Africa	0, 5	Arbor Acres	42	60	FC, FCR, BWG
Manal	2012	Egypt	Africa	0, 7	Arbor Acres	42	60	FC, FCR, BWG
Buba et al.	2016	Nigeria	Africa	0, 5	White Rose	28	102	FC, FCR, BWG
Buba et al.	2016	Nigeria	Africa	0, 10	White Rose	28	102	FC, FCR, BWG
Buba et al.	2016	Nigeria	Africa	0, 15	White Rose	28	102	FC, FCR, BWG
Buba et al.	2016	Nigeria	Africa	0, 20	White Rose	28	102	FC, FCR, BWG
Buba et al.	2016	Nigeria	Africa	0, 5	White Rose	56	102	FC, FCR, BWG
Buba et al.	2016	Nigeria	Africa	0, 10	White Rose	56	102	FC, FCR
Buba et al.	2016	Nigeria	Africa	0, 15	White Rose	56	102	FC, FCR, BWG
Buba et al.	2016	Nigeria	Africa	0, 20	White Rose	56	102	FC, FCR, BWG
Atul Shankar et al.	2017	India	Asia	0, 1	Cobb	42	144	FC, FCR, BWG
Atul Shankar et al.	2017	India	Asia	0, 1.5	Cobb	42	144	FC, FCR, BWG
Atul Shankar et al.	2017	India	Asia	0, 2	Cobb	42	144	FC, FCR, BWG
Yalçin et al.	2013	Turkey	Europe	0, 1	Ross	21	70	FC, FCR, BWG
Yalçin et al.	2013	Turkey	Europe	0, 2	Ross	21	70	FC, FCR, BWG
Yalçin et al.	2013	Turkey	Europe	0, 3	Ross	21	70	FC, FCR, BWG
Yalçin et al.	2013	Turkey	Europe	0, 4	Ross	21	70	FC, FCR, BWG
Yalçin et al.	2013	Turkey	Europe	0, 1	Ross	42	70	FC, FCR, BWG
Yalçin et al.	2013	Turkey	Europe	0, 2	Ross	42	70	FC, FCR, BWG
Yalçin et al.	2013	Turkey	Europe	0, 3	Ross	42	70	FC, FCR, BWG
Yalçin et al.	2013	Turkey	Europe	0, 4	Ross	42	70	FC, FCR, BWG
Mohamed et al.	2016	Sudan	Africa	0, 2.5	Ross	42	42	FC, FCR, BWG
Mohamed et al.	2016	Sudan	Africa	0, 5	Ross	42	42	FC, FCR, BWG
Mohamed et al.	2016	Sudan	Africa	0, 10	Ross	42	42	FC, FCR
Priya and Buba	2013	India	Asia	0, 5	Ross	36	100	FC
Priya and Buba	2013	India	Asia	0, 10	Ross	36	100	FC

DS – duration of supplementation; SL – supplementation level; NOB – number of birds

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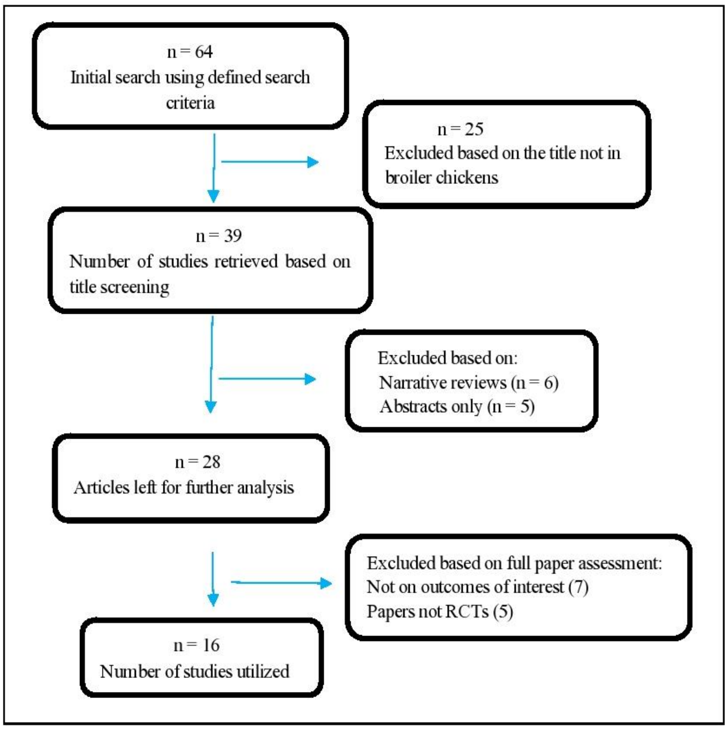

Figure 1. *Flow chart of articles used for the meta-analysis*

Feed consumption (FC)

The results of the overall effects estimate of yeast supplementation revealed that the incorporation of yeast in broiler diets reduced FC relative to broilers on control diet (d = -0.429, 95% CI: -0.606 to -0.253, I^2 = 89.59%, $p < 0.001$; *Fig. 2*). The temporal trends of the outcome of yeast supplementation on FC of broiler chickens are presented in *Figure 3*. The results of stratified subgroup analysis of the relationship between modifier variables and FC in broiler chickens are presented in *Table 2*. When the analysis was disaggregated, the results of the grand mean indicated that FC was significantly influenced by the explanatory variables (study continent, study country, duration of supplementation, supplementation level, broiler breed used; $p < 0.001$) following the removal of subgroup with one or two effect sizes. Disaggregation based on study continent, revealed that FC was not significantly reduced for studies performed in Africa, whereas studies conducted in Asia ($p < 0.001$) and Europe ($p < 0.001$) had significantly reduced FC. Results of the disaggregated studies of the effect of yeast on FC in broiler chickens based on study country showed that studies conducted in Pakistan, Egypt, India and Turkey were significantly different from zero ($p < 0.001$). Broiler chickens fed diet supplemented with yeast at < 10 g/kg feed had significantly reduced FC ($p < 0.001$). However, there was no association between FC and supplementation level (> 10 g/kg feed, $p = 0.054$;10 g/kg feed, $p = 0.113$, respectively). Stratified subgroup analysis results showed that FC was significantly reduced in broiler chickens fed yeast supplemented diet for 21 days ($p = 0.001$), 35 days ($p = 0.038$) and 42 days ($p < 0.001$), while FC in studies where broiler chickens were placed on yeast supplemented diets for 28 days ($p = 0.340$) and 56 days ($p = 0.665$) were not significant. Ross ($p < 0.001$), Arbor acres ($p < 0.001$) and Cobb ($p < 0.001$) had significantly decreased FC, whilst other breeds were not significant ($p > 0.05$).

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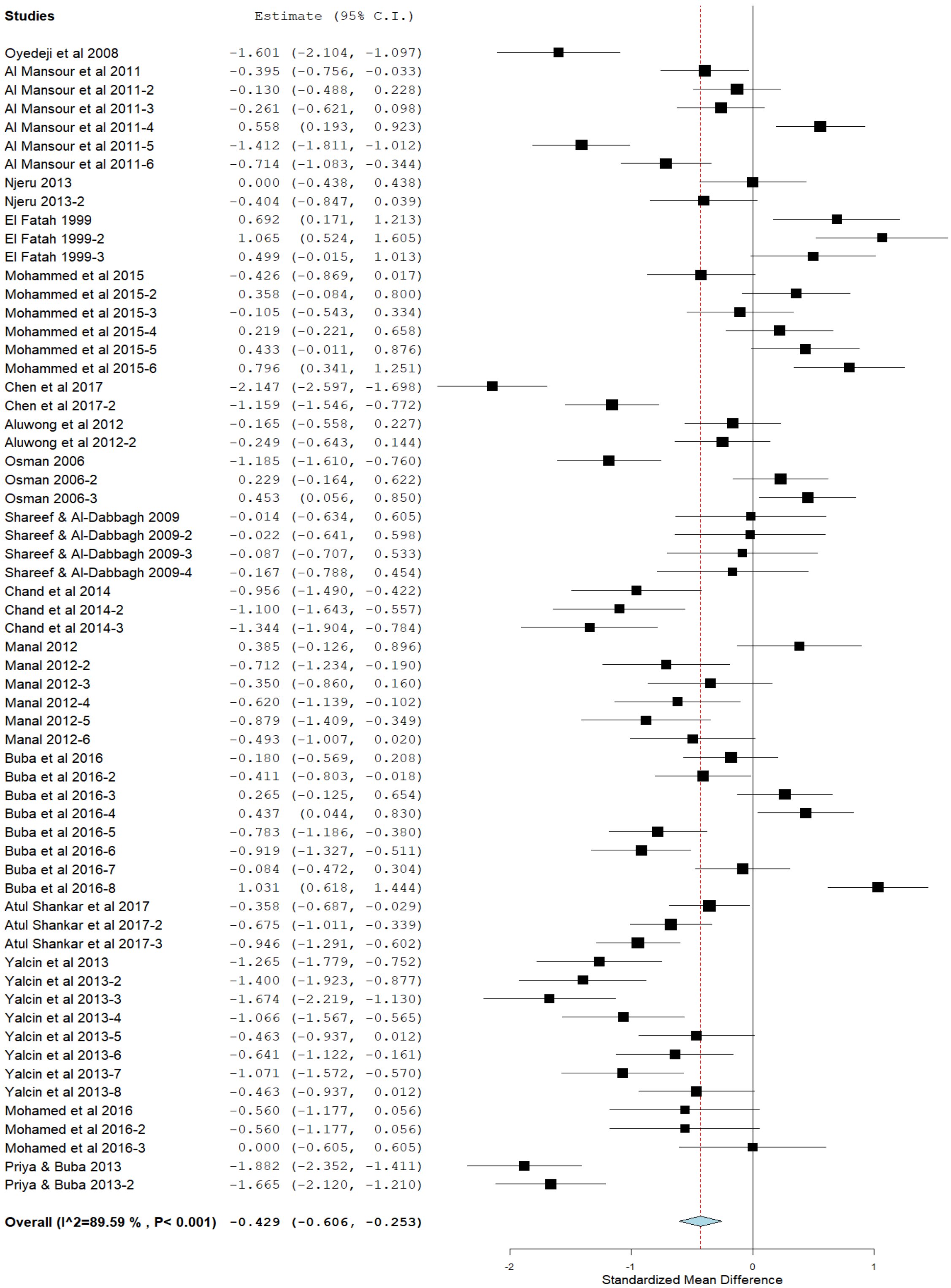

***Figure 2.** Forest plot of the effect of yeast supplementation on feed consumption in broiler chickens. The points to the right of the no effect line shows an increase in the outcome of interest, and the opposite depicts the reverse. The effect size is represented by individual square in the forest plot, while the upper and lower 95% CI for the effect size are the line that joined the square*

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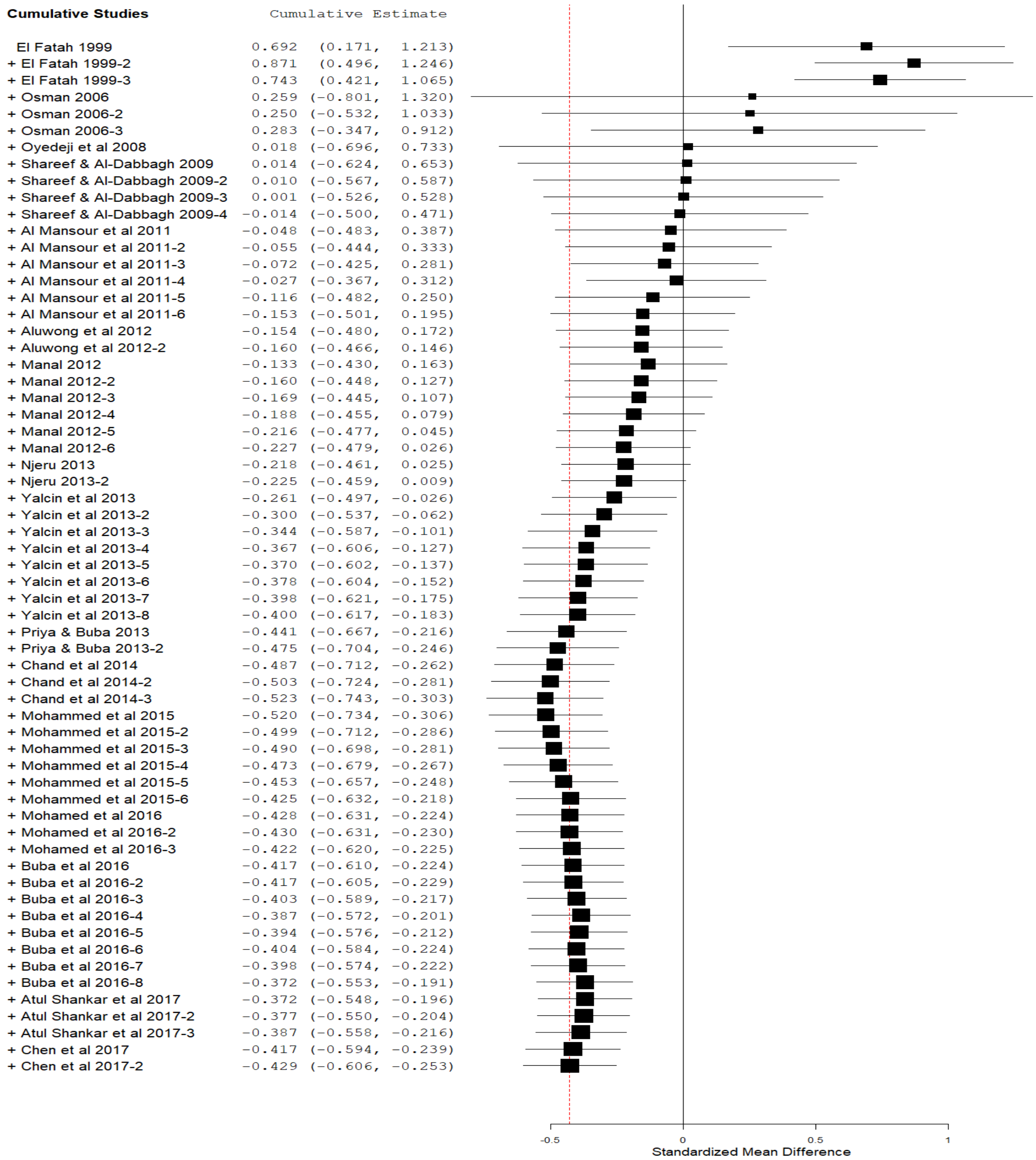

***Figure 3**. Trends of FC in broilers fed yeast supplemented diets*

Feed conversion ratio (FCR)

The results of pooled effects estimation found a positive association between yeast supplementation and FCR in broiler chickens (d = -0.354, 95% CI: -0.517 to -0.190, $I^2 = 87.27\%$, $p < 0.001$; *Fig. 4*). The temporal trends on the effect patterns of yeast supplementation on FCR in broiler chickens are shown in *Figure 5*. The results of the stratified analysis found an association between modifier variables and FCR in broiler chickens on yeast diet as presented in *Table 3*. Results of the stratified subgroup analysis revealed that FCR was positively influenced by the explanatory variables (study continent, $p < 0.001$; study country, $p < 0.0001$; duration of supplementation, $p < 0.001$; supplementation level, $p < 0.001$; broiler breed used, $p < 0.001$) when subgroup with one or two effect size(s) were removed from the analysis. Birds from

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studies undertaken in Europe ($p < 0.001$) and Asia ($p < 0.001$) had superior FCR compared to chickens from studies performed in Africa ($p = 0.296$). Stratified analysis results indicate that the FCR for studies performed in Pakistan, Egypt, Indian and Turkey differed significantly ($p < 0.001$). There was a correlation between FCR and duration of supplementation (21 days, $p = 0.002$; 35 days, $p < 0.001$; 42 days, $p = 0.038$), whereas no correlation exists between the FCR and the duration of supplementation (28 days, $p = 0.340$; 56 days, $p = 0.665$). Broiler chickens fed yeast supplemented diet at a level of > 10 g/kg feed ($p = 0.111$) and 10 g/kg feed ($p = 0.113$) had poor FCR, whilst those that received yeast supplemented diet at a level of < 10 g/kg feed ($p < 0.001$) had improved FCR. Ross ($p = 0.001$), Arbor acres ($p = 0.003$) and Cobb ($p < 0.001$) had significantly improved FCR relative to Hubbard ($p = 0.354$), Lohman ($p = 0.742$), Faobrow CD ($p = 0.648$) and White Rose ($p = 0.718$).

Table 2. *Relationship between moderators and FC in broiler chickens on yeast-based diets*

Subgroups	n	*d*	95% CI)	I^2 (%)	*p*-value
Study continent	62	-0.429	-0.606 to -0.253	88.59	< 0.001
Africa	34	-0.109	-0.313 to 0.095	85.61	0.296
Asia	20	-0.747	-1.052 to -0.443	90.14	< 0.001
Europe	8	-0.993	-1.301 to -0.685	66.94	< 0.001
Study country	58	-0.394	-0.571 to -0.217	88.80	< 0.001
Nigeria	11	-0.235	-0.662 to 0.152	90.09	0.233
Saudi Arabia	6	-0.389	-0.894 to 0.116	91.12	0.131
Sudan	15	0.131	-0.172 to 0.434	83.64	0.395
Iraq	4	-0.072	-0.382 to 0.238	0.00	0.648
Pakistan	3	-1.127	-1.442 to -0.812	0.00	< 0.001
Egypt	6	-0.442	-0.800 to -0.084	65.12	< 0.001
India	5	-1.086	-1.618 to -0.554	89.88	< 0.001
Turkey	8	-0.993	-1.301 to -0.685	88.80	< 0.001
DS (days)	60	-0.384	-0.555 to -0.213	88.25	< 0.001
21	19	-0.499	-0.808 to -0.191	87.41	< 0.001
28	8	-0.239	-0.731 to 0.253	91.43	0.340
35	4	-1.140	-1.384 to -0.896	0.00	0.038
42	25	-0.260	-0.506 to -0.015	86.78	< 0.001
56	4	-0.189	-1.047 to 0.669	94.49	0.665
SL (g/kg feed)	62	-0.429	-0.606 to -0.253	89.59	< 0.001
< 10	39	-0.684	-0.882 to -0.485	87.17	< 0.001
> 10	16	0.218	-0.051 to 0.487	81.94	0.054
10	7	-0.473	-0.955 to 0.008	86.56	0.113
Broiler breed used	60	-0.437	-0.620 to -0.254	89.90	< 0.001
Ross	23	-0.626	-0.952 to -0.300	91.20	< 0.001
Arbor Acres	10	-0.642	-1.084 to -0.201	83.32	< 0.001
Hubbard	9	-0.221	-0.689 to -0.246	88.63	0.354
Lohman	3	-0.165	-1.144 to -0.815	94.31	0.742
Faobrow CD	4	-0.072	-0.382 to 0.238	0.00	0.648
White Rose	8	-0.081	-0.519 to 0.358	89.76	0.718
Cobb	3	-0.657	-0.990 to -0.323	66.02	< 0.001

n – number of effect sizes; d – Hedges' d, CI – confidence interval, I^2 – heterogeneity

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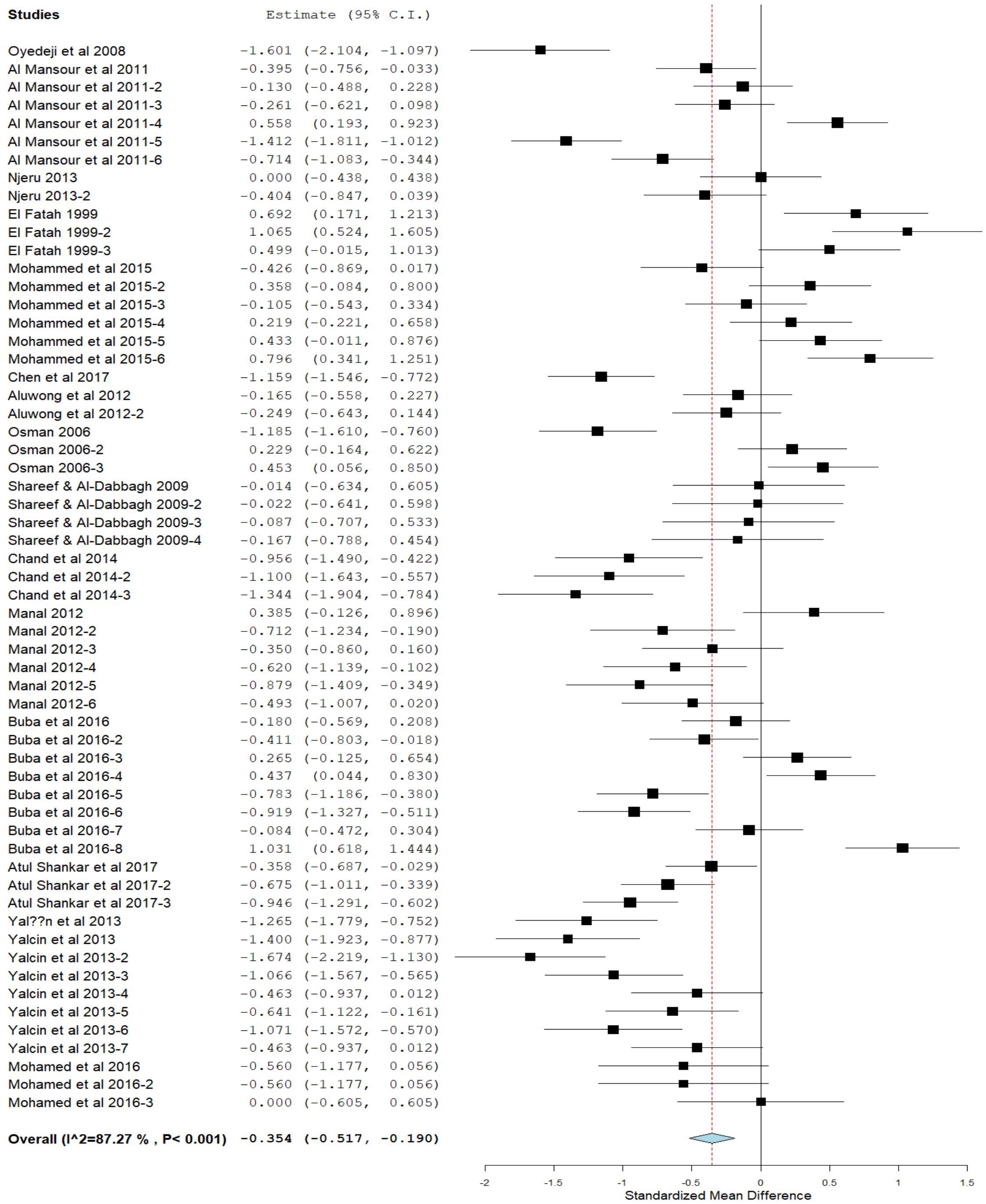

Figure 4. *Effect of probiotic-yeast supplementation on FCR of broiler chickens*

Body weight gain (BWG)

BWG was statistically increased from zero ($d = 0.310$, 95% CI: 0.130 to 0.491, $I^2 = 87.13\%$, $p < 0.001$; *Fig. 6*). The impact of yeast on BWG in broiler chickens in chronological order are shown in *Figure 7*. The results of the stratified subgroup analysis of the association between the explanatory variables and the BWG in broiler chickens are presented in *Table 4*. The mean effects estimate for the meta-analysis were 0.310, 0.325, 0.329, 0.310 and 0.310 for study continent ($p < 0.001$), study

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country (p = 0.001), duration of supplementation (p < 0.001), supplementation level (p < 0.001) and broiler breed used (*p* < 0.001), respectively, when the analysis was stratified. When the BWG analysis was disaggregated by study continent, studies conducted in Asia (p < 0.001) and Europe (p < 0.001) had improved BWG compared to those conducted in Africa. The results of BWG disaggregated by study country revealed that studies conducted in Iraq (*p* = 0.015), India (p = 0.019) and Turkey (p < 0.001) had statistically increased BWG following the removal of studies conducted in Saudi Arabia, Kenya and Taiwan that had less than 3 effect sizes. Studies conducted in Egypt, Sudan and Nigeria had similar values (Egypt: p = 0.556, Sudan: p = 0.728, Nigeria: p = 0.172). Broiler chickens from experiments that received yeast supplemented diets for 21 days (p < 0.001) had increased BWG, while those that received yeast supplemented diets for 42 days (*p* = 0.125) and 56 days (p = 0.983) were not significant. Studies that fed yeast for 35 days were removed from the analysis because they had less than 3 effect sizes. There was a significant positive association between BWG, and yeast supplementation level (< 10 g/kg feed, *p* < 0.001), whereas there was no significant association between BWG, and supplementation level (10 g/kg feed, p = 0.135; > 10 g/kg feed, p = 0.349). When BWG was disaggregated by breed of broiler used, Ross (p = 0.001), Faobrow CD (p = 0.015) and Cobb (p < 0.019) had significantly improved BWG.

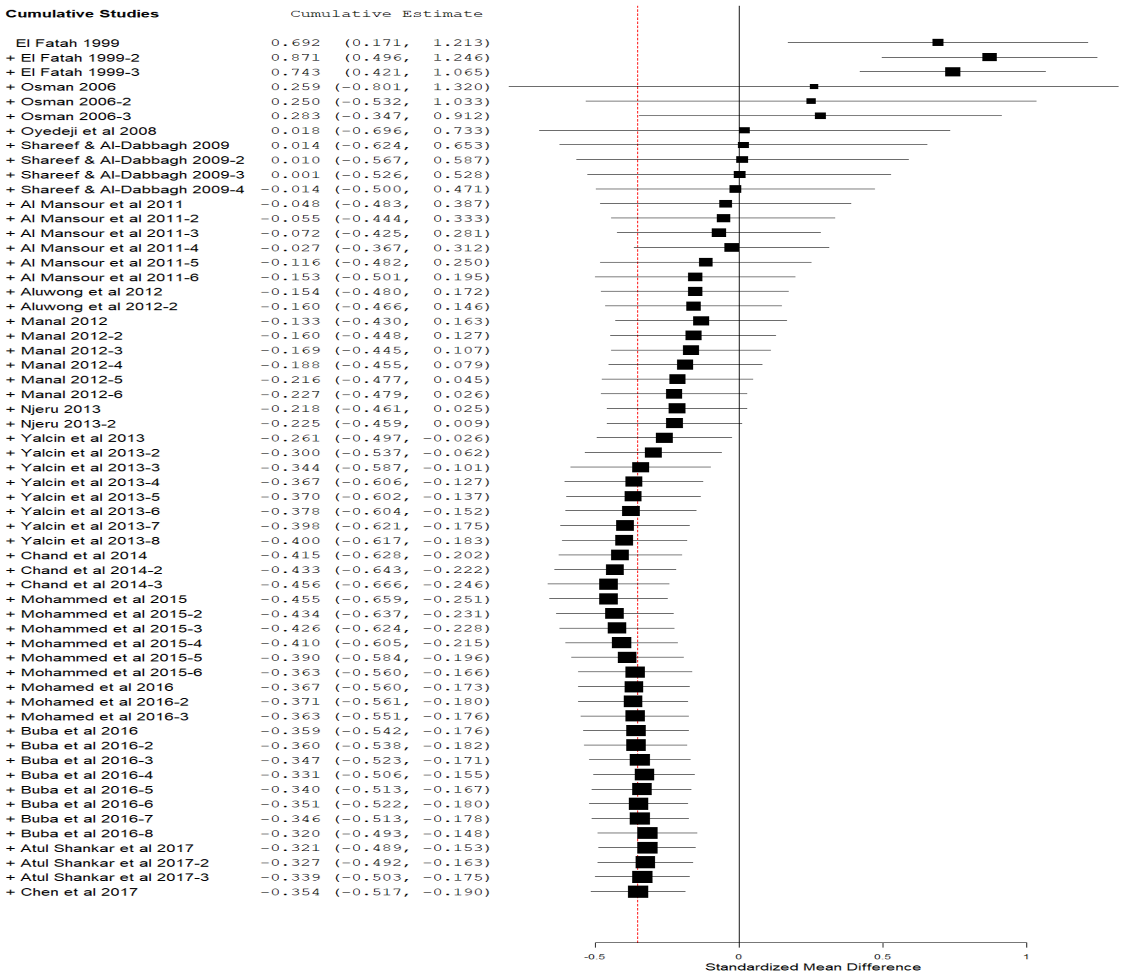

Figure 5. *Trends of FCR in broilers fed yeast supplemented diets*

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Table 3. *Association between moderators and FCR in broilers on yeast-based diets*

Subgroups	n	*d*	95% CI)	I^2 (%)	*p*-value
Study continent	59	-0.354	-0.517 to -0.190	87.27	< 0.001
Africa	34	-0.109	-0.313 to 0.095	85.61	0.296
Asia	17	-0.545	-0.806 to -0.285	84.32	< 0.001
Europe	8	-0.993	-1.301 to -0.685	66.94	< 0.001
Study country	56	-0.344	-0.513 to -0.175	87.37	< 0.001
Nigeria	11	-0.235	-0.662 to 0.152	90.09	0.233
Saudi Arabia	6	-0.389	-0.894 to 0.116	91.12	0.131
Sudan	15	0.131	-0.172 to 0.434	83.64	0.395
Iraq	4	-0.072	-0.382 to 0.238	0.00	0.648
Pakistan	3	-1.127	-1.442 to -0.812	0.00	< 0.001
Egypt	6	-0.442	-0.800 to -0.084	65.12	< 0.015
India	3	-0.657	-0.990 to -0.323	66.02	< 0.001
Turkey	8	-0.993	-1.301 to -0.685	88.80	< 0.001
DYS (days)	59	-0.354	-0.517 to -0.190	88.27	< 0.001
21	18	-0.404	-0.659 to -0.149	80.36	0.002
28	8	-0.239	-0.731 to 0.253	91.43	0.340
35	4	-1.140	-1.384 to -0.896	0.00	< 0.001
42	25	-0.260	-0.506 to -0.015	86.78	0.038
56	4	-0.189	-1.047 to 0.669	94.49	0.665
YSL (g/kg feed)	59	-0.354	-0.517 to -0.190	87.27	< 0.001
< 10	37	-0.610	-0.791 to -0.429	83.78	< 0.001
> 10	16	0.218	-0.051 to 0.487	81.94	0.111
10	6	-0.284	-0.633 to 0.065	69.57	0.113
Breeds of broiler used	57	-0.359	-0.529 to -0.189	87.68	< 0.001
Ross	21	-0.516	-0.833 to -0.199	89.77	0.01
Arbor Acres	9	-0.474	-0.790 to -0.159	74.20	0.003
Hubbard	9	-0.221	-0.689 to -0.246	88.63	0.354
Lohman	3	-0.165	-1.144 to -0.815	94.31	0.742
Faobrow CD	4	-0.072	-0.382 to 0.238	0.00	0.648
White Rose	8	-0.081	-0.519 to 0.358	89.76	0.718
Cobb	3	-0.657	-0.990 to -0.323	66.02	< 0.001

Analysis of heterogeneity and moderators

Data in *Table 5* presents the mixed effects meta-regression of the impact of yeast supplementation in broiler chicken performance. Forest plots of the 16 studies comprising 62 comparisons that evaluated the effect of yeast on FC in broilers provided evidence of significant heterogeneity (I^2-statistic = 89.59%, 95% CI: -0.606 to -0.253, p < 0.001, *Fig. 2*) and sensitivity analysis was not able to resolve the problem heterogeneity. Subgroup analysis did not remove the challenges of large heterogeneity among the studies included in the meta-analysis. Thus, the substantial heterogeneity continues, suggesting that these analyses cannot fix the problem of heterogeneity. Meta-regression, revealed that study continent (Q_B = 19.9, degree of freedom, df = 2, p < 0.001), study country (Q_B = 44.87, df = 9, p < 0.001), duration of supplementation

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(Q_B = 15.5, df = 5, p = 0.0084) and supplementation level (Q_B = 23.8, df = 2, p < 0.001) accounted for most the heterogeneity. Heterogeneity existed amongst the articles utilized in the analysis (FCR; I^2 = 87.27%, 95% CI: -0.517 to -0.190, p < 0.001, *Fig. 4* and BWG; I^2 = 87.13%, 95% CI: 0.130 to 0.491, p < 0.001, *Fig. 6*). Mixed effect meta-regression analysis revealed that study continent and yeast supplementation level were predictors of the study effect observed on FCR, whereas study continent, study country, supplementation level and broiler breed were predictors of study effects noticed on BWG results.

Table 4. *Relationship between moderators and BWG in broilers on yeast-based diets*

Subgroups	**n**	***d***	**95% CI)**	**I^2 (%)**	***p*-value**
Study continent	49	0.310	0.133 to 0.487	86.51	< 0.001
Africa	31	0.167	-0.067 to 0.401	87.63	0.163
Asia	10	0.335	0.054 to 0.615	77.12	0.019
Europe	8	0.838	0.512 to 1.164	71.35	< 0.001
Study country	44	0.325	0.130 to 0.520	71.35	0.001
Nigeria	8	0.307	-0.314 to 0.748	89.49	0.172
Sudan	15	0.068	-0.315 to 0.451	89.76	0.728
Iraq	4	0.389	0.075 to 0.703	0.00	0.015
Egypt	6	0.147	-0.343 to 0.637	81.49	0.556
India	3	0.549	0.089 to 1.009	82.33	< 0.019
Turkey	8	0.838	0.512 to 1.164	71.35	< 0.001
DS (days)	48	0.329	0.147 to 0.510	86.36	< 0.001
21	17	0.555	0.243 to 0.867	83.62	0.003
28	8	0.322	-0.176 to 0.819	91.58	0.205
42	20	0.208	-0.058 to 0.473	84.73	0.125
56	3	-0.007	-0.673 to 0.658	88.34	0.983
SL (g/kg feed)	49	0.310	0.133 to 0.487	86.51	< 0.001
< 10	31	0.470	0.271 to -0.669	83.72	< 0.001
> 10	14	-0.134	-0.413 to 0.146	80.11	0.349
10	4	0.573	-0.179 to 1.325	89.79	0.135
Broiler breed used	49	0.310	0.133 to 0.487	86.51	< 0.001
Ross	16	0.651	0.342 to 0.960	83.53	0.001
Arbor Acres	10	0.145	-0.174 to 0.464	79.08	0.374
Hubbard	6	-0.173	-0.758 to 0.412	90.07	0.563
Lohman	3	0.027	-0.933 to 0.988	94.11	0.956
Faobrow CD	4	0.389	0.075 to 0.703	0.00	0.015
White Rose	7	0.182	-0.249 to 0.612	87.93	0.408
Cobb	3	0.549	0.089 to 1.009	88.51	0.019

Analysis of publication bias

The results of the funnel plot of the impact of yeast supplementation on the productivity of broiler chickens as shown in *Figure 8A-C* revealed that the plots were asymmetrical. However, the Rosenberg fail-safe number of 3138 (FC), 1913 (FCR) and 964 (BWG) which is more than 9, 6 and 4-folds, respectively above the threshold of 320

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(5 × 62 + 10), 305 (5 × 59 + 10) and 255 (5 × 49 + 10) is required to declare the mean effect size robust. Consequently, the existence of publication bias was not a problem in this study as comparatively large number of unpublished studies would be needed to alter the statistically significant effects.

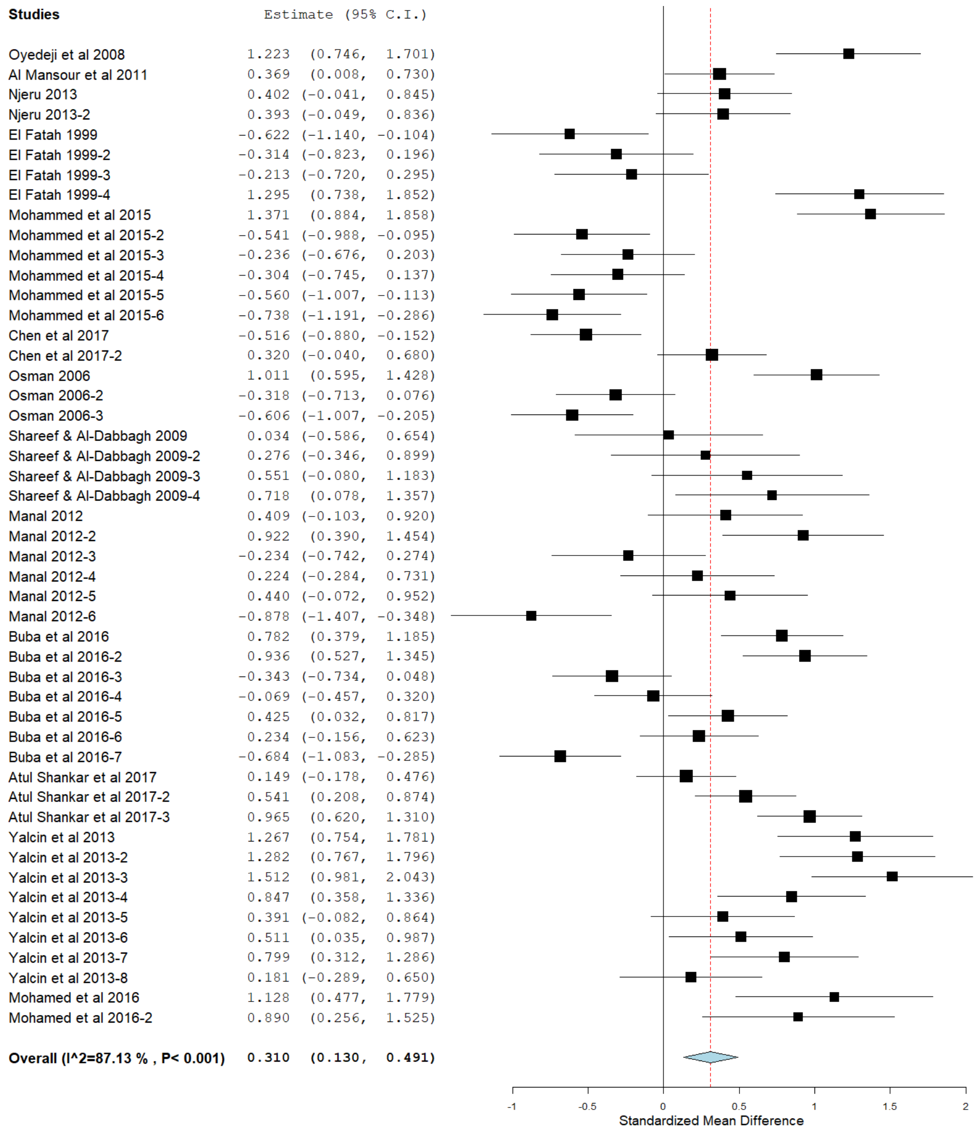

***Figure 6.** Forest plot of the effects probiotic (yeast) supplementation on BWG of broiler chickens*

Discussion

Probiotic effect

In the last couple of years, the incorporation of small amounts of yeast in animal feed as a growth enhancing agent instead of antibiotics has received considerable attention (Ahmed et al., 2015; Gao et al., 2017; Ogbuewu et al., 2019). Basically, yeast is rich in essential nutrients and contains mannan oligosaccharides (MOS) that helps in the multiplication of beneficial microbes in the guts of broilers (Spring et al., 2000). The findings from the present meta-analysis of 16 RCTs representing 10 study countries

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drawn from three study continents indicated that broilers fed yeast supplemented diets had improved productive indices. This finding is consistent with the previous results of Hooge (2004) and Frizzo et al. (2011) who reported that probiotic (lactic acid bacteria) and actigen® (second-generation, yeast (*Saccharomyces cerevisiae* var. *boulardii*) cell wall product) supplementation improved BWG and FCR in broilers and calves, respectively. The enhanced FC, FCR and BWG recorded in the current meta-analysis could be partly ascribed to the direct nutritional effect of yeast that has been documented to increase productivity in broiler chickens (Hooge, 2004; Yalcin et al., 2013; Mohamed et al., 2016) and in calves (Frizzo et al., 2011). In addition, yeast has been recorded to improve the multiplication of helpful microbes in the intestines of chickens (Schneitz, 2005; Awad et al., 2006; Apata, 2008; Musa et al., 2009). It has also been documented that yeast activates the innate immune response of broilers (Haghighi et al., 2006; Apata, 2008; Musa et al., 2009); stimulates digestive enzyme production and activity (Yoon et al., 2004); and competes with pathogens for adhesion sites in the gut, thus preventing their multiplication in the intestine (Choudhari et al., 2008). The enhanced feed efficiency and the resultant increase in body weight gain is the ultimate result of probiotic intervention as reported by Bozkurt et al. (2009) in male broiler chickens fed MOS.

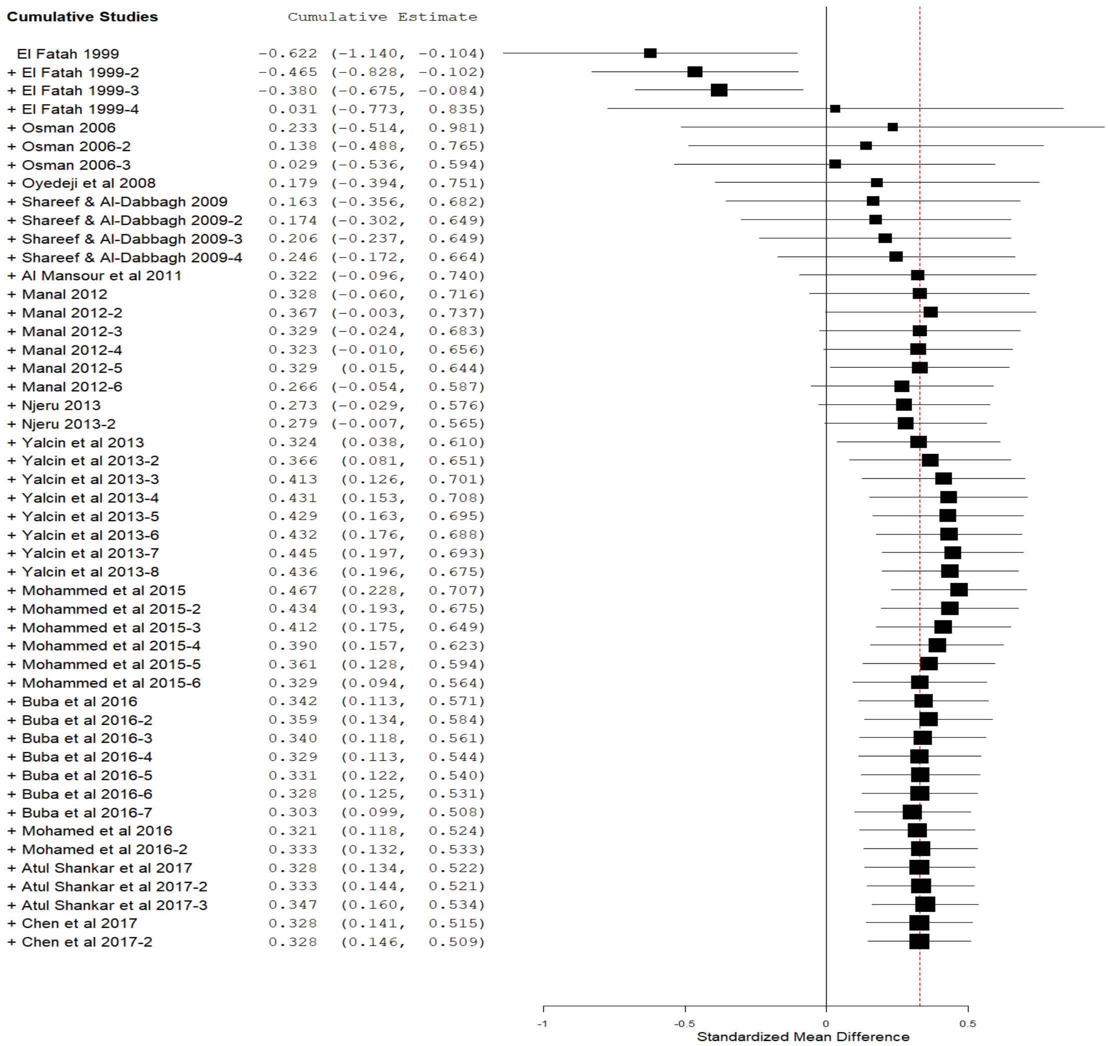

Figure 7. *Trends of BWG in broilers fed yeast supplemented diets*

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Table 5. *Summary of the random weighted meta-analysis for the explanatory variables that act as a modifier on the effects of yeast supplementation on productivity of broiler chickens*

Parameters	Moderators	QM	df	p-value	I^2 accounted
FC	Study continent	19.9	2	$p < 0.001$	25.61
	Study country	44.87	9	$p < 0.001$	41.20
	Duration of supplementation	15.5	5	$p = 0.0084$	16.42
	Supplementation level	23.8	2	$p < 0.001$	29.56
	Broiler breed used	7.04	7	$p = 0.425$	0.24
FCR	Study continent	7.80	2	$p = 0.0202$	12.63
	Study country	9.71	8	$p = 0.286$	4.24
	Duration of supplementation	2.41	4	$p = 0.66$	0.00
	Supplementation level	10.81	2	$p = 0.0045$	18.92
	Broiler breed used	10.8	6	$p = 0.094$	10.42
BWG	Study continent	7.76	2	$p = 0.021$	12.63
	Study country	9.71	8	$p = 0.286$	4.24
	Duration of supplementation	2.41	4	$p = 0.66$	0.00
	Supplementation level	10.8	2	$p = 0.0045$	18.92
	Broiler breed used	10.8	6	$p = 0.0944$	10.42

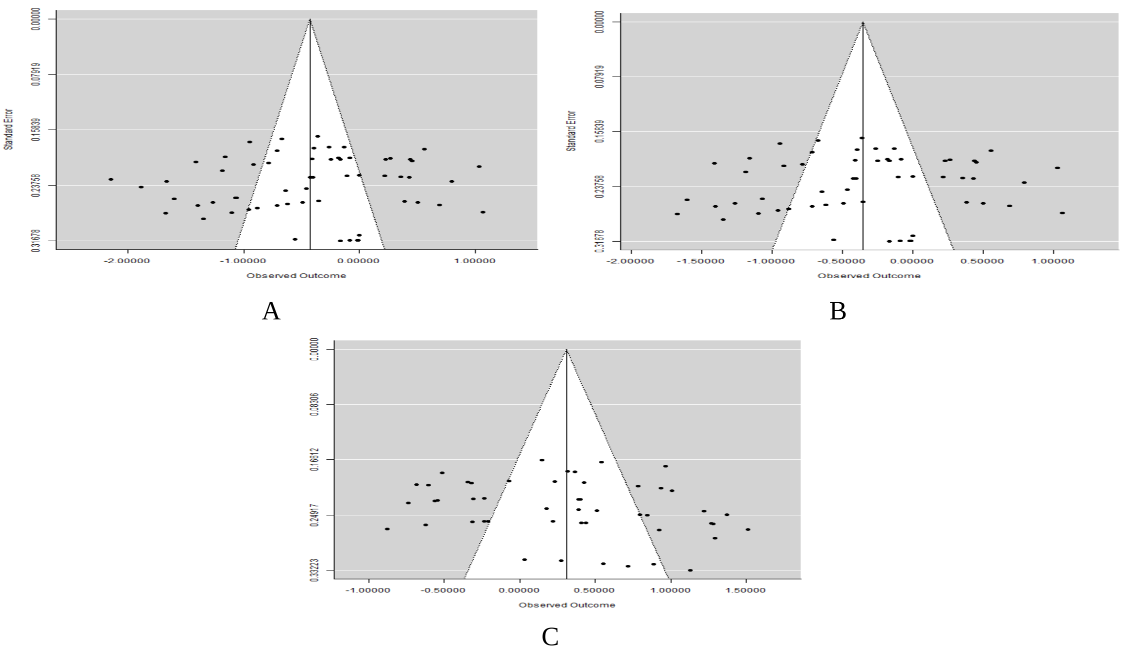

Figure 8. *Funnel plots of the effect of yeast on FC (A), FCR (B) and BWG (C) in broiler chickens*

Analysis of moderators

The current meta-analysis disclosed an important association between yeast supplementation in broilers and some elements of our selected explanatory variables. This revealed that the moderators chosen were responsible for the inconsistency of results reported in broiler chickens fed probiotic yeast supplemented diets. There is

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significant relationship between probiotic (yeast) supplementation and FC, FCR and BWG in broiler chickens for studies performed in Asian and Europe. However, studies undertaken in Europe found a bigger relationship (FC, $d = 0.993$; FCR, $d = 0.993$; BWG, $d = 0.838$) than those conducted in Asia. The effect of yeast on FC, FCR and BWG in broilers for studies conducted in Africa had no significant association. The observed disparity between the production indices of broiler chickens from studies undertaken in Africa and those performed in Europe and Asia can be partially attributed to differences in the environmental conditions of the three study continents. The environmental conditions hampering chicken performance and health include temperature, relative humidity, light and housing system (Elijah and Adedapo, 2006). Environmental temperatures have a negative impact on the survival and success of the broiler production. Broiler chickens are susceptible to environmental change because there is a narrow range of thermal conditions under which they can maintain a stable body temperature. However, no data was reported on the weather conditions of the study countries and continents as at the time the investigated was conducted. The failure of the authors of the study included in the meta-analysis to report the weather condition of the study region as at the time the study was carried out was the reason we could not identify the specific variables that might have been accountable for the differences in the productivity of broilers raised in Africa compared to those raised on the other two continents. The current meta-analysis also found that there was significant association effect of yeast on FC and FCR in broilers raised in Egypt, India, Pakistan, Iraq and turkey, whereas for BWG it was evident on studies undertaken in India, Iraq and Turkey. Yeast effect on FC and FCR was found in studies which lasted for 21, 35 and 42 days. However, there was no beneficial effect of yeast on studies that lasted 28 and 56 days. Enhancement in FC and FCR for studies that lasted for 21 days translates to an increase in body weight gain in broiler chickens. This indicate that study continent, study country, duration of supplementation are the limiting factors in several of the studies included in the meta-analysis. The level of yeast supplementation is a determinant in many studies and has led to differences in the outcomes reported by the various authors (Frizzo et al., 2011; Ezema and Ugwu, 2014). In this study, the beneficial activities of yeast on FC, FCR and BWG were shown in studies that fed diet supplemented with yeast at the rate of < 10 g/kg feed. The Ross and Cobb broiler type had enhanced FCR and BWG with less feed intake, whereas Arbor acre had enhanced FCR, which did not translate to increased body weight gain in the current meta-analysis.

Source of heterogeneity and publication bias

This meta-analysis included research conducted in several parts of the world. In this context, the generalization and validity of the conclusions reached are strong. Potential biases, however, such as publication bias and heterogeneity were evident in the present meta-analysis and this was anticipated as most of the studies were undertaken in 10 study countries drawn from 3 continents. Substantial heterogeneity was recorded, sensitivity and stratified analysis fail to fix the issue. Results of meta-regression analysis showed that explanatory variables (study continent, study country, duration of yeast supplementation, yeast supplementation level and breed of broiler used) accounted for most of the variations among the studies included in the meta-analysis. For FC, FCR and BWG, the Rosenberg Nfs is 3138, 964 and 1913 respectively, which is 9, 4 and 6 times higher the threshold of 320 (5 × 62 + 10), 255 (5 × 49 + 10) and 305 (5 × 59 + 10) required to consider the mean effect size robust. Thus, publication bias was not an issue

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in the current meta-analysis since a relatively large number of unpublished studies would be required to alter significant impacts of yeast supplementation on broiler chicken productivity.

Conclusion

This meta-analysis provided baseline information on guidelines to standardize the experimental designs of future trials on the effect of probiotic-yeast supplementation on broiler chicken productivity. Results found that adding yeast to the broiler chicken diet at a level less than 10 g/kg feed improved the productivity of broiler chickens. There was also a significant association effect between probiotic-yeast and explanatory variables. These results will encourage feed producers, poultry nutritionists, and farmers to make informed choices about the use of yeast in broiler chicken diets as an alternative feed additive instead of antibiotics. More research is, however, needed to ascertain the effect of yeast supplementation on quality characteristics of broiler chicken meat using meta-analytical approach.

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