

# Sugarcane Derived Polyphenol Feed Additive Supplemented to the Wheat Diet Improved Broiler Performance

<sup>1</sup>\*N. Sekh and <sup>1</sup>C. Oshea

<sup>1</sup>The University of Nottingham (UK), Department of Bioscience- Animal Nutrition Division, United Kingdom

\*Corresponding author email: drnajimsekh@gmail.com

## ABSTRACT

*This study examined effects of incremental levels of sugarcane derived polyphenol on most commonly used cereals in the poultry feed industry; wheat and maize. Both cereals rapidly digestible starch, which is usually digested completely than protein itself whereas polyphenol slows down glucose absorption in rat without compromising feed intake. Polyphenol may enhance depositing of muscle in broilers due to synchronous availability of the plasma glucose and amino acid. In this experiment, 240 Ross 308 male broilers (5 birds/cage; n=6) were given 8 different diets, wheat and maize; without or with polyphenol inclusion level at 0.5%, 2% and 4% respectively. Feeds were given ad-libitum in 2 phases as grower (day 11-24) and finisher (day 24-38). Parameters assessed were body weight (BW), average daily feed intake (ADFI), feed conversion ratio (FCR), drip loss%, shear force and meat color. Birds were euthanized at day 38 for meat quality inspection using Pectoralis muscle. The research revealed that 2% polyphenol in wheat diets significantly reduced ADFI (day 24-38) as compared to unsupplemented wheat diets ( $p < 0.0001$ ). At the same level, polyphenol numerically improved FCR (day 24-38) in broilers fed wheat diets (FCR 1.93) and maize diets (FCR 1.63) as compared to unsupplemented wheat diets (FCR 1.98) and maize diets (FCR 1.70) respectively. However, 4% polyphenol inclusion level had adverse effect on BW (day 17 and 38) and ADG (day 10-38, day 24-38) in broilers fed wheat diets as compared to unsupplemented wheat diets. On other hand, maize diets supplemented with different level of polyphenol did not have any significant effect on BW, ADG, ADFI and FCR in broilers as compared to unsupplemented maize diets. Also, there was significantly higher ADFI (day 24-38) in broilers fed wheat diets as compared to maize diets ( $p < 0.0001$ ). Regarding meat qualities, there were not any effect of polyphenol on drip loss%, shear force and meat color. However, cereal type itself had significant effect on meat color appearance as maize fed broilers had more yellowness value of meat ( $b^*$ ;  $p < 0.0001$ ) than broilers fed wheat diets. In conclusion, polyphenol supplementation found to be useful for broilers mainly on wheat diets at 2% inclusion level.*

**Keywords:** FCR, Drip loss%, Meat color, Polyphenol, Ross 308

## **INTRODUCTION**

Wheat and maize are most commonly used cereals in the poultry feed (Selle et al., 2003). Both of them have rapidly digestible starch which is usually digested completely (Weurding. R et al., 2003, Liu et al., 2013b) than protein. It is suggested that feed efficiency can be improved by slowing down the starch digestibility and accelerating protein digestibility due to higher protein and energy utilization in broilers at the site of muscle synthesis (Giuberti et al., 2013, Liu et al., 2013b, Selle et al., 2015). Polyphenol, on other hand, has shown to slow down glucose absorption in rat without affecting the feed intake. Polyphenol can play a role for synchronous availability of plasma glucose and amino acid for depositing muscle protein efficiently in broilers as suggested by Geiger (et al., 1950), Weurding. R (et al., 2003), and Liu (et al., 2013).

Besides, there are evidences from animal and human studies that dietary polyphenols have useful gut modulatory effects. It has been observed that polyphenol improves villus height: crypt depth ratio in the duodenum in piglet (Sehm et al., 2006). This is in agreement with broiler's research by Viveros (et al., 2010) where there was an improvement of villus height in the small intestine due to polyphenol rich products. Moreover, Caspary (et al., 1992) had demonstrated that increased villus height leads to an improvement of gut function as a result of increased absorption surface, expression of brush border enzyme and nutrient transport mechanism. Additionally, polyphenol has shown to inhibit proliferation of pathogens in gastrointestinal tract of human (Duda. A et al., 2012).

At molecular level, dietary polyphenol can reduce inflammation (Blanch. C et al., 2012) through modulating nuclear factor kappa (NF- $\kappa$ B). In several studies, flavonoid-rich products have shown anti-inflammatory effect both in vivo and in vitro (Gessner et al., 2013). In addition, polyphenol can improve growth performance and beneficial fatty acids level whereas it can also reduce lipid oxidation and cholesterol value in broilers as mentioned by Starcević. K (et al., 2014). Polyphenols have shown to minimize the adverse reaction of lipid peroxidation by decreasing the level of malondialdehyde (MDA) and improving antioxidant status in blood and tissue (Labban, L et al., 2014) in human. Jung (et al., 2010) reported that gallic acid which is a polyphenol found in grape, wine and tea improved nutritional value of poultry meat by increasing arachidonic acid and docosahexanoic acid level and also, water-holding capacity of breast meat in broilers. Likewise, dry rosemary leaves and rosemary oil; rich in polyphenol found to improve sensory value of meat and significantly decreased the malondialdehyde concentration (Yesilbag et al., 2011) in chicken breast muscle. In another experiment, hesperidin and genistein supplemented on broiler ration enhanced the fatty acid profile (PUFA, n-6:n-3 PUFA ratio) of breast muscle (Kamboh and Zhu et al., 2013) in poultry.

However, Voljc (et al., 2013) suggested that addition of only 3 gm per kg of a product containing polyphenol was not sufficient to prevent the adverse effect of lipid peroxidation

in the broiler meat. Furthermore, Viveros (et al., 2011) had observed negative effect of polyphenol on body weight in 21-days old chickens whose diets were supplemented with grape seed extract at the level of 7.2 gm per kg feed as compared to birds from other level of supplementation. Besides, significant drop in FCR (1.43 vs 1.51) was also reported in chicken fed grape pomace at the rate of 60 g per kg of feed.

It was speculated that sugarcane derived polyphenol supplementation on wheat diet improves growth performance and meat quality in broilers. Wheat is rich in viscous, non-starch polysaccharide (Xylan) which has shown to affect growth performance in poultry at higher degree as compared to less viscous diet like maize (Marquardt. et al., 1994, Jia et al., 2009, Rodriguez et al., 2012). The higher transit time and lower intestinal passage rate in birds due to viscous digesta ascribed to wheat diets may provide polyphenol advantage to act as antioxidant efficiently, and may even improve function of intestinal villi and enzymatic action, thus enhancing the feed efficiency. The objective of this research was to find out optimum inclusion level of sugarcane derived polyphenol supplement to improve broiler's performance and meat quality.

## **MATERIALS AND METHODS**

This study was carried out from June 2019 to September 2019. The experiment was conducted at the University of Sydney, Australia on behalf of Poultry Research Foundation. Later, the raw data was analyzed at the Department of Bioscience at the University of Nottingham, UK. Meanwhile, all birds were kept in battery cage system throughout research period at the Poultry Research Foundation. All procedures used in this research were approved by the Animal Ethics Committee of The University of Sydney and were in accordance with Australian Code for the care and use of animals for scientific purpose.

### **Experimental design and dietary treatments**

The research was conducted as a completely randomized design; comprising two different cereals (wheat and maize) as a source of energy along with four inclusion level of polyphenol (0%, 0.5%, 2% and 4%) making a total 8 treatment diets as shown in Table 1. Thus, it was 2×4 factorial array of dietary treatments offered to 240 Ross 308 male broilers (5 birds/cage; n=6). Chicks had free access to water and feed. Chicks were provided common wheat-soybean based starter diet until day 10. Afterwards, broilers were offered experimental grower diets from day 11 to 24, and finisher diets from day 24 to 38 as shown in Table 2. The sugarcane derived polyphenol product had a syrup like consistency and contained 3.5 g polyphenol per kg product. All diets were formulated to satisfy nutrients requirement as per the recommendation of Ross 308 guidelines. Exogenous enzymes were not added, and energy value were adjusted with soybean oil. Digestible amino acid level was adjusted with canola meal, soybean meal and synthetic amino acids. Average body weight (BW), average daily gain (ADG), average daily feed

intake (ADFI) and feed conversion ratio (FCR) were recorded on a weekly interval basis. The FCR values were adjusted for body weight of bird that died during research period. Table 1. Experimental diets based on growth stage, cereal type and polyphenol inclusion level

Diets	Stage	Cereal Type	Polyphenol %
1	Grower / Finisher	Wheat-based diet	0%
2	Grower / Finisher	Wheat -based diet	0.5%
3	Grower / Finisher	Wheat- based diet	2%
4	Grower / Finisher	Wheat-bdased diet	4%
5	Grower / Finisher	Maize-based diet	0%
6	Grower / Finisher	Maize-based diet	0.5%
7	Grower / Finisher	Maize-based diet	2%
8	Grower / Finisher	Maize-based diet	4%

Table 2. Dietary formulation, calculated and analyzed nutrient composition of the diets based on maize or wheat for broiler chicken from 11 to 38 days' post- hatch

Ingredient	Wheat grower diet (11-24 days post hatch)	Wheat finisher diet (24-38 days post hatch)	Maize grower diet (11-24 days post hatch)	Maize finisher diet (24-38 days post hatch)
Wheat/Maize	590	689.5	558	605
Soyabean meal	272	152	281	240
Canola meal	37	45	66.5	40
Vegetable oil	36	54	32	56
Limestone	9	8.45	8	8.35
Dicalcium Phosphate	21	16	20	16
Salt	1.5	1.3	1.5	1.3
Sodium Bicarbonate	5.25	4.6	5.25	4.6
Lysine Hcl	2.75	3.25	2.45	3.25
Methionine	2.7	2.4	2.5	2
Celite	20	20	20	20
Threonine	0.8	1.5	0.8	1.5
TMV premix	2	2	2	2
TOTAL	1000	1000	1000	1000
ME (MJ/Kg)	12.44	12.99	12.44	12.99
CP%	22.2	18.5	22.2	18.5

## **Meat quality assessment**

Birds were sacrificed when they reached day 38 following the recommendation of euthanasia of experiment animals. Meat quality was determined taking one bird per cage. To measure drip loss%, approximately 25 g of samples were taken, trimmed off any noticeable fats and weighted and then calculated as weight of the fresh meat sample (g) minus weight of the sample stored at 4°C at 7 days' post-slaughter which was later expressed into percentage. Meat color was determined on day 0 and day 7 post-slaughter using CIELAB method (International Commission on Illumination, 1976) where triplicate color measurement for Lightness (L\*), redness (a\*) and yellowness (b\*) of the Pectoralis muscle were assessed using a Minolta Lab CR-10 colorimeter. Using a Warner Bratzler Shear attachment on a Stable Micro Systems TAXT2 Texture Analyzer, shear force value was also calculated.

## **Statistics**

Data were tested for normality using the univariate procedure of SAS (SAS Inst. Inc., Cary, NC) and was analysed as a completely randomized design incorporating 8 dietary treatments. The cage served as the experimental unit for growth performance data and one bird per cage for the meat quality variables. Data were analysed using the generalized linear model procedure of SAS. Preplanned contrasts for growth performance and a meat quality variables were evaluated between cereal types (unsupplemented Wheat vs Maize) and polyphenol level (0.5%, 2% or 4% versus 0% polyphenol). Finally, data are presented as least square means± standard error of the mean (sem). Differences were considered significant at  $P < 0.05$ .

## **RESULTS**

### **Growth performance**

Wheat fed broilers had higher FCR than maize fed birds ( $p < 0.001$ ) in last week (day 24-38) which was due to higher ADFI (day 24-38) in wheat fed birds. Both wheat and maize fed birds gained similar BW at day 38 ( $p = 0.018$ ) (Table 3). It has been observed that higher ADFI (day 24-38) was improved by polyphenol at 2% level of inclusion in wheat diets as compared to unsupplemented wheat diets ( $p < 0.0001$ ). However, maximum supplement level (4% polyphenol) on wheat diets had adverse effect on BW (day 17 and 38) and ADG (day 10-38, day 24-38) as compared to unsupplemented wheat diets. Contrary to this, maize diets supplemented with different level of polyphenols didn't have any significant effect on BW, ADG, ADFI and FCR in broilers as compared to unsupplemented maize diets. Overall, 2% polyphenol inclusion level numerically improved FCR at day 24-38 ( $p < 0.001$ ) in both wheat and maize fed broilers as compared to the unsupplemented wheat and unsupplemented maize diets respectively.

Table 3. Effect of polyphenol on body weight (BW), average daily gain (ADG) and feed conversion ratio (FCR) in broilers from 10- 38 days post-hatch fed wheat and maize diets

Parameter	Wheat diet 0% Pp	Wheat diet 0.5% Pp	Wheat diet 2% Pp	Wheat diet 4% Pp	Maize diet 0% Pp	Maize diet 0.5% Pp	Maize diet 2% Pp	Maize diet 4% Pp	SEM	P-value
BW d10	396	385	366	365	380	363	374	372	10	0.238
ADG d10-17	68bc	65b	64b	67bc	71ac	74a	70ac	69bc	2	0.003
ADFI d10-17	73	74	74	72	74	75	74	73	1	0.791
FCR d10-17	1.06b	1.14a	1.14a	1.06b	1.03b	1.01b	1.04b	1.06b	0.02	0.002
BW d17	875cd	838abc	817a	831ab	877cd	883d	866bcd	852abcd	15	0.019
ADG d17-24	99	97	100	94	99	98	98	97	2	0.451
ADFI d17-24	163	161	156	153	165	160	157	158	6	0.850
FCR d17-24	1.63	1.66	1.56	1.62	1.65	1.62	1.59	1.62	0.05	0.908
BW d24	1571	1518	1514	1490	1573	1570	1554	1533	23	0.103
BW d32	2461	2418	2354	2295	2507	2541	2537	2496	62	0.071
BW d38	3184bc	3074ab	3045ab	2983a	3155bc	3162bc	3239c	3166bc	50	0.018
ADG d32-38	120	109	1155	115	108	103	117	112	8	0.871
ADG d10-38	100bc	96ab	96ab	94a	99bc	100bc	102c	100bc	2	0.029
ADG d24-38	115bc	111ab	109ab	107a	113abc	114abc	120c	117bc	3	0.040
ADFI d24-38	229a	224ab	211bc	217ab	193d	194d	196d	199cd	4	<.0001
FCR d24-38	1.98a	2.01a	1.93a	2.04a	1.70b	1.70b	1.63b	1.71b	0.03	<0.001

a, b, c, d: means within columns not sharing common letters are significantly different ( $P < 0.05$ )

### Meat quality

As shown in Table 4, there were not effect of polyphenol on drip loss%, shear force, and meat color. However, dietary cereal type itself had significant effect on the meat color. At day 0 post-slaughter, the unsupplemented wheat fed chicken had more L\* value than unsupplemented maize diets ( $p=0.04$ ). In addition, at day 0 ( $p<0.0001$ ) and day 7 ( $p<0.001$ ) post-slaughter, cereal type had significant effect on the yellow color trait of the meat where maize fed broilers had higher b\* value (more yellowness color of meat) than wheat fed broilers.

Table 4. Effect of cereals and polyphenol (Pp) inclusion level on drip loss (%), shear force (N) and meat color (day 0 and day 7) in broilers post-slaughter

Parameter	Wheat diet 0% Pp	Wheat diet 0.5% Pp	Wheat diet 2% Pp	Wheat diet 4% Pp	Maize diet 0% Pp	Maize diet 0.5% Pp	Maize diet 2% Pp	Maize diet 4% Pp	SEM	P-value
Drip loss (%)	5.01	5.37	7.58	4.75	6.67	6.98	5.97	6.22	0.91	0.345
Shear force (N)	35.8	36.4	32.3	47.3	35.3	36.5	35.5	34.7	4.34	0.412
Meat color (Day 0)										
L*	54.56ab	52.4bc	56.3a	54.63ab	50.85c	50.99c	52.14bc	52.71abc	1.27	0.04
a*	2.49	2.67	1.78	3.38	2.24	2.74	2.13	1.90	0.37	0.08
b*	3.61a	3.47a	4.29a	4.55a	9.32b	8.24b	9.28b	9.39b	0.73	<0.001
Meat color (Day 7)										
L*	54.06	51.12	54.73	53.29	51.78	52.40	51.27	50.61	1.3	0.28
a*	2.19	2.18	1.56	2.51	1.71	2.23	1.85	2.08	0.34	0.57
b*	6.04a	5.83a	6.88a	6.95a	11.01b	10.87b	10.48b	10.69b	0.68	<0.0001

a, b, c: means within columns not sharing common letters are significantly different ( $P < 0.05$ ) / L\*-Lightness, a\*-redness, b\*-yellowness

## DISCUSSION

It is well documented that feed ingredients rich in soluble non-starch polysaccharide (NSP) like wheat diet results in poor performance in broilers as compared to less viscous diet like maize. Moreover, such adverse effect has been reported higher on young species (Kalmendal et al., 2011). In this research, there was no effect of cereal type (unsupplemented maize and wheat) during early growing phase of broilers from day 10 to 17 in terms of ADG and FCR. This contrasts to the findings by Marquardt (et al., 1994), Jia (et al., 2009) and Rodriguez (et al., 2012). This outcome on early growth performance might appeared due to the fact that all broilers were given same basal diets, wheat-soybean meal starter, at the beginning of trial. It was only after day 10 that maize and wheat diets were given separately to the experimental groups. Therefore, it is likely that experimental diets might have taken time to have impact on birds.

There was increased ADFI (day 24-38) in broilers fed unsupplemented wheat diets which in turn had increased FCR as compared to unsupplemented maize fed broilers. Similar observation was reported by Peng (et al., 2003) where birds fed wheat diets had higher

FCR than maize fed birds and it was reported due to higher NSP content of the wheat diets. On day 38, both wheat and maize fed broilers attended similar BW ( $p=0.018$ ). Therefore, it may be concluded that broilers on wheat diets were compensating nutrients requirement by increasing feed intake to meet its genetic potential.

Wheat diets supplemented with maximum level of polyphenol i.e. 4% had negative effect on BW (day 17, day 38) and ADG (day 10-38, day 24-38) as compared to unsupplemented wheat diets. Similar outcomes have been reported by Jansman (et al., 1989) and Nyachotti (et al., 1997) stating relatively higher dietary concentration of polyphenol may reduce performance. On other hand, wheat diets containing 2% polyphenol level had beneficial effect on broilers. At this level, there was decreased ADFI (day 24-38); without getting affected ADG (day 24-38) and final BW (day 38) as compared to unsupplemented wheat diets. At this level, polyphenol might have increased villus height leading to an improvement of gut function as a result of increased absorption surface, expression of brush border enzyme and nutrient transport mechanism as suggested by Duda. A (et al., 2012).

It is also concluded that 4% polyphenol inclusion level on wheat diets had most detrimental effect on broilers. Furthermore, it might be concluded that polyphenol gets tightly regulated when supplemented to wheat diet ascribed to the viscosity of digesta due to soluble NSP. As suggested by Jansman (et al., 1989), Ortiz (et al., 1993) and Surai. P.F (et al., 2014) excess polyphenol in wheat diets may have been poorly absorbed, quickly transformed into range of metabolites which may have formed complexes with lipoprotein due to its highly reactive hydroxyl group, ultimately reducing nutrient digestibility in presence of viscous intestinal digesta. Further research is required to understand the relationship in between polyphenol, cereal type and its effect in terms of dose.

In addition, maize diets supplemented with different levels of polyphenol did not have any effect in broilers as compared to unsupplemented maize diets. However, maize diets supplemented with 2% polyphenol inclusion level numerically improved FCR (1.63 vs 1.7) at day 24-38 as compared to unsupplemented maize diets. Broilers fed maize diets may have higher nutrients absorption in intestine, thus increasing opportunities for nutrient utilization as maize is comparatively less viscous and lower in fiber to digest (Jacob. J et al., 2015).

Overall, supplementation of polyphenol had multiple effects on broilers. Similar outcomes have been reported by Rohn (et al., 2006), Brenes (et al., 2010), and Chamorro (et. al., 2013). It has been also stated (Hodek et al., 2002) that the physiological effects of polyphenol not only depend on one but various factors like type/subtype, concentration, absorption, metabolic transformation of polyphenol. The average FCR of commercial male Ross 308 (day 24-38) according to the Ross Performance Objective Guideline (2019)



is 1.74. As sugarcane derived polyphenol, at 2% inclusion level, numerically lowered the FCR in both diets, and in maize diets it is even lower than Ross Guideline, there is need for research whether it is achievable so in farm condition.

Regarding meat qualities, there were no effect of polyphenol and cereal types on drip loss% and shear force. This contrast with finding by Ao and Choct (et al., 2004) who reported that wheat diets fed group of birds had lower drip loss% value than maize fed birds. As suggested by Mir. N (et al., 2017) broiler chicken meat quality is dependent on multiple factors, thus a very complex process. Absence of effect on drip loss% and shear force when compared in between wheat and maize fed birds in this study might have happened due to muscle pH, temperature, glycogen, stress level to which chickens were exposed were similar for all treatment groups before and after slaughter.

Lastly, cereal type itself had significant effect on color trait of the meat in this study where  $b^*$ (yellowness) value was higher in broilers fed maize diets as compared to the wheat diets. This outcome is supported by Smith (et al., 2002) who stated that wheat fed birds produce paler meat than from the maize fed birds. This result may be due to pigments (xanthophylls and carotene) present in maize diets. Color of meat is a valuable attribute as consumers think yellow meat is reasonably healthy (Sunde et al., 1992). Consumers prefer broilers meat with more yellow color (Mateo and Carandang et al., 2006) and hence this can affect broiler meat purchase decision (Garcia et al., 2013). However, limited studies have been conducted correlating the color and nutritional value of the poultry meat.

## **CONCLUSION**

Phytogenic feed additives like sugarcane derived polyphenol supplementation in the poultry feed have drawn interest of animal nutritionists; with more companies manufacturing such products globally. In this research, polyphenol supplementation has not only shown to be beneficial, but also proven to be detrimental with higher inclusion rate. On other hand, interaction of typical polyphenol with various feed ingredients to be used in feed formulation is largely unknow. Their metabolites and consequent effect on microflora with various farm mangement condition is yet to be explore as future research directive. Lastly, the cost-benefit or return on investment analysis should be major criteria for incorporating such exogenous products.

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