Effect of effective microorganisms on broiler chicken performance and ammonia production in poultry house

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SUMMARY

A study was conducted between January and March 2001 to assess the effects of Effective Microorganisms (EM) as feed additive in broiler chicken production on growth performance. The experiment involved 210 day-old broiler chicks which were randomly allocated to 14 pens of 15 birds each. There were seven treatments which were replicated twice. The main treatment, EM, was provided to experimental birds in two levels, 10 mlEM/l and 20 mlEM/l in drinking water or by spraying on litter material, or both in water and litter. Mortalities and causes were recorded. Furthermore, ammonia concentration in the poultry house was measured from the litter samples. There were significant ($P<0.05$) differences between treatments for growth performance and carcass yield. The slaughter weights for EM treated groups were 1372±9.1 g and 1658.6±9.2 g for 10 mlEM/l and 20 mlEM/l respectively compared to 1207±9.1 g for the control. The average daily weight gains were 31.9±0.2 g/d and 38.7±0.2 g/d for 10 mlEM/l and 20 mlEM/l, respectively, compared to 27.9±0.2 g/d for the control. The dressing percentages were 77.9, 79.1 and 73.9 for 10 mlEM/l, 20 mlEM/l and control groups respectively. Mortality was not significantly ($P>0.05$) affected by EM treatments while, ammonia levels in the litter treated with EM were generally lower than that of the untreated control group. It is concluded that supplementation of EM in drinking water and spraying in litter has growth promoting effect to broilers and the optimum performance was achieved at the rate of 20 mlEM/l provided in drinking water and sprayed on the litter.

Key words: Effective microorganisms, probiotics, broiler, carcass

INTRODUCTION

The practice of feeding livestock with sub-therapeutic levels of antibiotics (i.e. used for animal growth promotion rather than specifically to treat diseases) has been practised for over 50 years (EMRO, 2010). Antibiotics affect micro flora by altering the metabolism of microorganisms, and suppressing microbial growth in the gut. Usage of antibiotics has negative effects on animal's health and production such as residues in tissues, development of resistance in microorganisms, allergies and genotoxicity (Markovic *et al.,* 2009). Moreover, in recent years, consumer demand for natural and organic foods has risen steadily, not only in developed countries but also in developing ones. As the demand for organic and naturally produced livestock by consumers increases, antibiotic use in animal feeds is expected to decrease and will be limited to therapeutic treatment of diseases, or will be eliminated outright in evolving animal production systems (Flint and Garner, 2009).

Effective microorganisms (EM) are a combination of useful regenerative micro-organisms that exist freely in nature and are not manipulated in any way. Effective microorganisms mainly include five families of micro-organisms i.e. Lactic acid bacteria, Yeasts, Actinomycetes, Photosynthetic bacteria and Fungi (EMRO, 2010). According to World Health Organization probiotics are "Live microorganisms which when administered in adequate amounts confer a beneficial health effects to the host" (Zonis, 2008). Available literature suggests that the use of microbial preparations such as EM, have some beneficial effects in poultry production such as improvements in growth rate, feed efficiency, prevention of intestinal infections, and improved nitrogen utilization (Safalaoh, 2006; EMRO, 2010). Furthermore, when EM is used in rearing sheds, not only help to suppress disease causing organisms, but also rapidly eliminate and control ammonia produced from droppings and as a result the air quality is improved significantly. Use of probiotics including effective microorganisms is limited in developing world, Africa in particular. The objectives of this study were to evaluate the

performance of broiler chicken supplemented with EM and assess the effectiveness of EM in suppressing foul smell in poultry houses.

MATERIALS AND METHODS

A total of 210 day-old broiler chicks (Hybro chicks) were used in this experiment. Birds were vaccinated against Newcastle and Gumboro diseases in the first week of their life. All hygienic procedures were observed and no antibiotic was used for all experimental birds for the whole experimental period of six weeks. However, in the first week coccidiostats were used due to the outbreak of coccidiosis.

Chicks were housed in a closed well ventilated house, with a deep litter floor of rice hulls. The birds were randomly allocated to 14 pens each with 15 chicks. Commercial broiler starter mash was fed as basal diet and was supplied from 1st to 21st day and growers mash was fed from 22nd to 42nd day. The starter mash and growers mash contained 21% CP and 19% CP, respectively, on dry matter basis. Feeds and water were given *ad libitum* for the whole trial period.

Effective Microorganisms (EM)

SAiON EM-1 original stock solution, developed in Austria, was used as the main treatment. It contains Photosynthetic bacteria (*Rhodopseudomonas palustris)*, Lactic acid bacteria (*Lactobacillus pluntarum* and *Lactobacillus casei)*, Yeast (*Saccharomyces cerevisae)* with pH 3.5. EM-1 is an inactive solution, so to make it active, water and molasses were added.

One litre of EM-1 was mixed with one litre of molasses and 18 litres of clean dechlorinated water. The resulting solution is hereby referred to as EM-2 and was put in a 20 litre bucket. The bucket was covered properly using a lid with small aperture to facilitate gas release due to fermentation process. The solution was kept in a cool place for 10 days before being used. After 10 days the solution was observed visually and it appeared yellowish-brown in colour with a characteristic smell upon smelling and sour test. The pH of the resulting solution was 3.54. Those properties indicated that the solution was ready to be used. The prepared solution was valid for 30 days.

Treatment application

Two levels of EM-2 i.e. 10 mlEM/l of water and 20 mlEM/l of water were allocated randomly to experimental birds by mixing it in drinking water and/or spraying on litter. There were seven treatments and each was replicated twice (Table 1). Treatment one (T_1) was the control diet whereby no EM was included either in the litter or drinking water. For T_2 and T_5 EM was sprayed to the litter only, while for T_3 and T6 it was mixed in water only and for T_4 and T_7 it was sprayed to both water and litter. Inclusion of EM in litter was done only once, but was added daily in drinking water.

Table 1. Treatment combinations

EM-2 Concentration	
10 ml/l	**20 ml/l**
T_2- Litter only	T_5- Litter only
T_3 Water only	T_6- Water only
T_4 Water and Litter	T_7- Water and Litter

Data collection and analysis

Growth performance

Body weight of all birds was measured in grams on the first day just after arrival, as initial weight and subsequent body weights were taken on weekly basis for six consecutive weeks. Overall body weight gain was determined as the difference between final body weight and initial body weight. Daily weight gain was determined as the difference between final weight and initial weight divided by the number of experimental days (42 days).

Carcass evaluation

At the end of the experiment a total of 70 live chickens were randomly selected (5 birds per pen from each replicate) and slaughtered for carcass evaluation. Carcass yield was determined after evisceration and removal of feet and the resulting weights were expressed as the proportion of live weight at slaughter i.e. the dressing percent.

Diseases and mortality rates

The experimental birds were closely monitored for health status, especially feeding behaviour, respiratory behaviour and fecal material consistency. The effect of EM treatment on mortality was assessed based on the percentage of live and dead birds. A Chi square test was applied to test the significance of treatment effects on mortalities.

Testing for ammonia concentration in litter

Four **s**amples of litter materials from each replicate were collected randomly from the pen at two weeks intervals. The samples were mixed thoroughly and subjected to laboratory for ammonia determination on the same day. The litter material was dissolved and filtered and the fluid was used to determine the ammonia-N concentration using Kjeldahl technique without digestion. It was assumed that ammonia (NH_3) in litter material was free. Five ml fluid + Sodium hydroxide (NaOH) was distilled into boric acid using Kjeltec apparatus and blank distillation was included. The blank comprised of 5 ml of distilled water + NaOH. The litter fluid and blank distillate were titrated against 0.1M HCl to estimate N content and NH_3 concentration was calculated using the equation below adapted from Doto (2002).

$$NH_3(mg/l) = \frac{N \times (V_{\textbf{Litter fluid}} - V_{\textbf{Blank}}) \times \text{Molecular weight of } NH_3 \times 100)}{\text{Volume of litter fluid}}$$

Where: N = Normality of HCl, acid used during titration.

V = Volume of HCl acid used in litter fluid or blank titration.

Statistical analysis

Growth performance, carcass characteristics and ammonia concentration data were analyzed using the General Linear Model procedure of SAS (2002). There were no significant differences between treatments on initial body weights. Hence, the effect of treatment combinations on final body weights, average daily weight gain, carcass yields and ammonia concentration were analyzed using the following model:

$$Y_{ijkl} = \mu + C_i + M_j + (CM)_{ij} + (P_{ij})_k + \epsilon_{ijkl}$$

Where: Y_{ijkl} = Observation i^{th} concentration, j^{th} medium and k^{th} replication,
μ = General mean
C_i = Effect of i^{th} concentration (1 = 10 mlEM/l, 2 = 20 mlEM/l)

M_j = Effect of j^{th} method of application (medium) (1 = litter, 2 = water, 3= water and litter)

$(CM)_{ij}$ = Interaction between j^{th} medium and i^{th} concentration

$(P_{ij})_k$ = Effect of k^{th} replication within i^{th} concentration and j^{th} medium ϵ_{ijkl} = Random effect peculiar to each bird

RESULTS

Growth performance

Least square means for final body weight and weight gain are shown in Table 2. The results show that there were significant differences (P<0.05) between treatments and birds on 20 mlEM/l had generally higher values compared to those on 10 mlEM/l. Spraying of EM on litter and simultaneously adding it in water did not significantly influence growth rate when EM was applied at the rate of 10 mlEM/l as compared to adding it in water only or adding it in litter only. Conversely, media combinations in the 20 mlEM/l treatment significantly improved both final body weight and growth rates. In general birds which received EM in water + litter (T_7) at the rate of 20 mlEM/l were much heavier (1658.6 ± 9.2 g) at 42 days and had higher average daily gain (38.7 ± 0.2g) than birds in the control (T_1) group and other treatments.

Carcass evaluation

The results show that there were significant differences (P<0.05) between treatments on carcass weight (Table 3). The interaction between medium and EM levels was also significant. Generally, birds receiving 20 mlEM/l in water or in litter had higher carcass yield and higher dressing percent than those in which 10 mlEM/l were added except for T_4, whilst the control group had the lowest carcass values. The results show that the highest values for carcass weights (1121.1 ± 11.3 g) and dressing percent (79.1%) were obtained from birds receiving EM in water + litter (T_7) at the rate of 20 mlEM/l.

Table 2. Effects of EM application in the litter and water on growth performance of broiler chickens

EM Levels	Treatment	Medium	Parameter	
			Final body Weight (g) (Mean±SE)	**Weight gain (g/day) (Mean±SE)**
0 mlEM/l	T_1	Control	207 ± 9.1^{f}	27.9 ± 0.2^{f}
10 mlEM/l	T_2	Litter	1260.8 ± 9.2^{e}	29.2 ± 0.2^{e}
	T_3	Water	1339.8 ± 9.1^{d}	31.1 ± 0.2^{d}
	T_4	Litter + water	1372.6 ± 9.1^{d}	31.9 ± 0.2^{d}
20 mlEM/l	T_5	Litter	1406.8 ± 9.1^{c}	32.7 ± 0.2^{c}
	T_6	Water	1495.5 ± 9.5^{b}	34.8 ± 0.2^{b}
	T_7	Litter + water	1658.6 ± 9.2^{a}	38.7 ± 0.2^{a}

^{abcdef} Least square means with different superscript letters in the same column are significantly different (P<0.05).

Mortality rate

The results of mortality rates for the whole experimental period are presented in Table 4. The results show that a total of 27 birds (12.8%) died. High mortality rates occurred in the first week where 19 chicks died. Higher mortality rate (20%) was observed in the control treatment compared to 8.8% for 10 mlEM/l and 14.4% for 20 mlME/l. However, the differences were not statistically different (P>0.05). Likewise, there were no significant differences among the different modes of applications (medium) although slight higher mortality was experienced in treatment where EM was applied in water.

A total of 13 dead bodies were submitted to the Department of Veterinary Pathology, Sokoine University of Agriculture for pathological examination. The investigation results suggested that the cause of death were mainly huddling due to low temperatures. One bird in the control group was disposed due to lameness in the fourth week of experimental period.

Ammonia concentration in litter

There were significant differences (P<0.05) between treatments on ammonia concentration in the litter. Pens in which birds were receiving 20 mlEM/l in water + litter (T_7) had low ammonia levels of 28 mg/l, 26 mg/l and 22 mg/l for the second, fourth and sixth weeks respectively,

compared to the control group (T_1) which had 46.1 mg/l, 50.1 mg/l and 70.1 mg/l in the same periods (Table 5). However, in the second week, pens with T_6 and T_7 had lower ammonia levels compared to the control and other treatments. In the control treatment, ammonia concentration increased by 8% in the fourth week and 34% by end of sixth week. The general trends are shown in Figure 1.

Table 3. Effects of EM application on litter and water on carcass weight and dressing percent of broiler chickens

EM Levels	Treatment	Medium	Parameter	
			Carcass weight (g) (LSM ± SE)	**Dressing percent (%± SE)**
0 mlEM/l	T_1	Control	1036.8 ± 11.6^{e}	73.9 ± 1.4^{e}
10 mlEM/l	T_2	Litter	1060.9 ± 11.1^{d}	75.7 ± 1.4^{cd}
	T_3	Water	1067.4 ± 11.1^{bc}	76.2 ± 1.4^{c}
	T_4	Litter + water	1093.9 ± 11.1^{c}	77.9 ± 1.4^{b}
20 mlEM/l	T_5	Litter	1090.1 ± 11.1b^{c}	77.2 ± 1.4^{b}
	T_6	Water	1076.9 ± 11.2^{b}	77.5 ± 1.4^{b}
	T_7	Litter + water	1121.1 ± 11.3^{a}	79.1 ± 1.4^{a}

^{abcde} Least square means with different superscript letters in the same column are significantly different ($P<0.05$).

Table 4. Mortality rate of birds reared under different EM levels and medium

Factor	Levels	Initial number	Died	Mortality rate (%)	$\chi 2$	P-value
EM concentration	Control	30	6	20	2.833	0.242
	10 mlEM/l	90	8	8.8		
	20 mlEM/l	90	13	14.4		
Medium	Control	30	6	20	3.676	0.298
	Water	60	10	16.6		
	Litter	60	5	8.3		
	Litter + water	60	6	10		

Table 5. Effects of EM treatment on ammonia (NH3) concentration in litter materials at different period

EM level	Treatment	Medium	NH_3 mg/l		
			2nd	4th	6th
0 mlEM/l	T_1	Control	46.1[a]	50.1[a]	70.1[a]
10 mlEM/l	T_2	Litter	40.1[b]	38.1[b]	34[b]
	T_3	Water	38.1[bc]	32[bc]	30[bc]
	T_4	Litter + water	34[bc]	30[bc]	28[bc]
20 mlEM/l	T_5	Litter	36.1[bc]	30[bc]	26[cd]
	T_6	Water	30[d]	28[c]	24[cd]
	T_7	Litter + water	28[d]	26[c]	22[d]
Standard error			1.6	2.4	1.8

[abcde] Least square means with different superscript letters in the same column are significantly different (P<0.05).

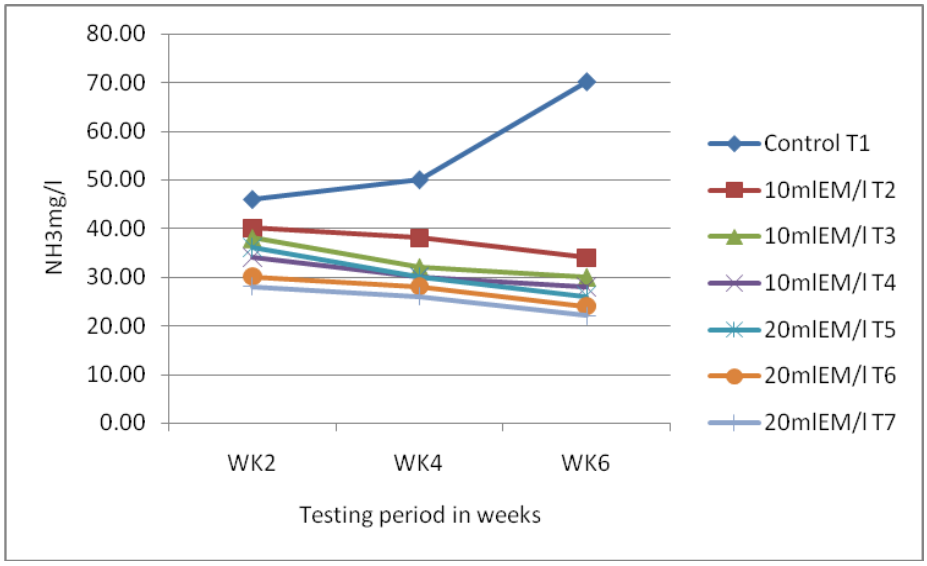

Figure 5. Trend of ammonia concentration in litter materials during the experimental period

DISCUSSION

The results show clearly that adding EM in drinking water and spraying on the litter materials concurrently, enhanced the growth of broiler chickens during the entire experimental period compared to non-EM control group. Effective microorganism supplemented birds were heavier than the control group and had heavier carcasses. Similar observations was reported by Willis and Reid (2008) who found that live body weight gain and carcass yields are significantly higher in broilers supplemented with probiotics. Related results have been reported by Bozkurt *et al.* (2009) and Ashayerizadeh *et al.* (2009).

The enhanced growth can be explained by the probiotic effects particularly that of *Saccharomyces cerevisiae, Lactobacilus acidophilus* and a mixture of *Lactobacilli* which were components of the EM preparation used in this study. These microorganisms have been found to be naturally rich source of protein, minerals and vitamin B-complex (Shareef and Al-Dabbagh, 2009), hence encourage growth of the beneficial microflora in the gastrointestinal tract, which in turn, improves protein efficiency ratios and/or nitrogen utilization (Weijiong and Yongzhen, 2001; Safalao, 2006). To the contrary, Flint and Garner (2009) and Safalao and Smith (2001) reported that addition of EM at the rate of 15 g/kg of feed elicited no beneficial effects on body weight gain while an inclusion rate of 30 g/kg of EM with or without antibiotic resulted in improved body weight gain suggesting that the effects of EM are dose dependent.

With regard to mortality rates, there were no significant differences between treatments on

mortality rates. However, the general trends were lower mortalities in EM treated groups. The observed trend is consistent with other previous studies on the potential benefits of probiotics in stimulating poultry immune system (Dharne, 2008; Anjum *et al.*, 2011; Dunkley, 2008; Patterson and Burkholder, 2003). Higher mortalities were recorded during the early stage of growth (week 1) and huddling resulting from unexpected intermittent power cuts, especially during the night was established to be the main cause of death.

It is apparent that there were progressive reduction of ammonia levels in litter materials, during the experimental period for EM probiotic supplemented birds. The lowest level was observed in T_7 (20 mlEM/l in litter + water treatment). Conversely, there was progressive increase of ammonia concentration in litter materials in the control group. The observed trend agrees with those of Bhola (2011) and Li *et al.*(1998) who observed concentration of toxic ammonia (NH_3) inside a poultry house keeping EM treated birds to be 26.5 ppm compared to 87.6 ppm for non-EM control groups. The level of ammonia concentration observed in (T_7) i.e. 22 mg/l is within the safety level recommended by SCD Probiotics (2009) and Teraganix (2011). According to these authors the average poultry house should have about 20-29 ppm (i.e. 20 - 29 mg/l) of ammonia. The decreasing patterns of ammonia concentration observed in litter material treated with EM indicate the ability of probiotics to suppress malodours in poultry houses. This is due to the efficiency of probiotic to transform NH_3 into less toxic substances and may thus be considered as functional anti oxidant.

It is concluded that the study demonstrated that supplementing EM in drinking water and spraying in litter materials has growth promoting effects. According to this study the highest growth performance is achieved when EM is added at the rate of 20 mlEM/l in drinking water and sprayed in the litter. Furthermore, this study concludes that ammonia levels in birds receiving EM either in water or in litter or both litter and water is generally lower than that of untreated group and EM treatment levels do not significantly affect birds mortality.

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REFERENCES

Anjum DA, Tahir H, Farzana R, Ghulam G, Tariq J. Influence of Effective Microorganisms on Health and Immune System of Broilers under Experimental Conditions. EM Research organization (EMRO). Retrieved from http://www.emrojapan.com on 04 June 2011, 2011.

Ashayerizadeh O, Dastar B, Shams SM, Ashayerizadeh A, Mamooee M. Influence of Antibiotic, Prebiotic and Probiotic Supplimentation to Diets on Carcass Characteristics, Hematological Indices and Internal Organs Size of Young Broiler Chickens. *J Anim and Vet Advances* 8 (9): 1772–1776, 2009.

Bhola KD. Effective Microorganisms (EM) for Animal Production. Retrieved from http://www.pdfcari.com /Effective. Microorganism (EM) for Animal Production. html on 01 March 2011, 2011.

Bozkurt M, Kucukyilmaz K, Cath AU, Cinar M. The effect of single or combined dietary supplementation of prebiotics, organic acid and probiotics on performance and slaughter characteristics of broilers. *SA J Anim Sci* 39 (3): 197 – 205, 2009.

Dharne VH. Maintaining Gut Integrity. The poultry site Article 978. Avitech, Animal healthpvt. LTD. Retrieved from http://www.thepoultrysite.com/ article/978/ Maintaining Gut Integrity on 26 March 2011, 2008.

Doto SP. The effect of probiotics on feed intake, digestibility and rumen environment in non lactating cows. MSc dissertation, Sokoine University of Agriculture, 2002.

Dunkley C. The Use of Probiotics and Prebiotics in Poultry Feeds. Poultry site, article 1108, University of Georgia. Retrieved from http://www.thepoultrysite.com/article/1108 on 26 March 2011, 2008.

EMRO (EM Research Organization). Working with EM Technology for Sustainable Environment. Retrieved from http://www.emsustains.co.uk on 5 August 2010.

Flint JF, Garner MR. Feeding beneficial bacteria: A natural solution for increasing efficiency and decreasing pathogens in animal agriculture, Poultry Science Association. MicroBios Inc., Ithaca, NY 14850. *J Appl Poult Res* 18: 367-378, 2009.

Li WJ, Ni YZ, Umemura H. Effective Microorganisms for Sustainable Animal Production in china. Retrieved from http://www.infrc.or.jp on 30 June 2010, 1998.

Markovic R, Sefer D, Krstic M, Petrujkic B. Effects of different growth promoters on broiler performance and gut morphology. *Arch Med Vet* 41: 163-169, 2009.

Patterson JA, Burkholder KM. Application of Prebiotics and Probiotics in Poultry production. *Poult Sci* 82: 627 – 631, 2003.

Safalaoh ACL. Body weight gain, dressing percentage, abdominal fat and serum cholesterol of broilers

supplemented with a microbial preparation. *Afr J Food Agric and Devel* 6: (1), 2006.

Safalaoh ACL, Smith GA. Effective Microorganisms (EM) as an alternative to antibiotics in broiler diets: Effects on broiler performance, feed utilization and serum cholesterol. In Proceedings of the 6th International Conference on Kyusei Nature Farming, South Africa, 2001.

SAS (Statistical Analysis System) Version 9.2 Copyright © 1989-1996. SAS Institute Inc., Cary, NC, USA, 2002.

SCD (Sustainable Community Development). Probiotics. The odour problem in poultry production. Retrieved from http://www.scdprobiotics.com on 30 June 2010, 2009.

Shareef AM and Al-Dabbagh ASA. Effect of probiotic (*Saccharomyces cerevisiae*) on broiler chicks. *Iraqi J Vet Sc* 23: 23-29, 2009.

Teraganix. Poultry Odor Control with Effective Microorganisms. Retrieved from http://www.teraganix.com/EM on 01 March 2011, 2011.

Weijiong L, Yongzhen N. Use of Effective Microorganisms to Suppress Malodours of Poultry Manure. *J Crop Prod* 3: (1) 215 – 221, 2001.

Willis WL, Reid L. Investigating the Effects of Dietary Probiotics Feeding Regimens on Broiler Chicken Production and Campilobacter jejuni presence. *Poult Sci* 87: 606 – 611, 2008.

Zonis S. Probiotics, Prebiotics and Synbiotics. Retrieved from www.thenibble.com/ reviews/nutri/probiotic on 29 June 2010, 2008.