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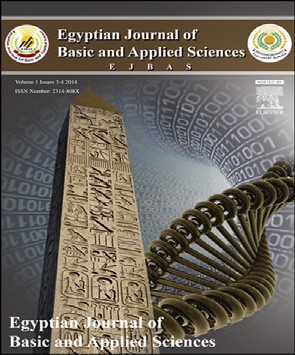
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Full Length Article

Quality evaluation of egg composition and productivity of layers in EM (Effective Microorganisms) treatments: A field report



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## a b s t r a c t

Layers divided in to three groups; Group 1 (Control group) e The layers fed with standard commercial food masses with Biobantox, BMD and formaldehyde. Group 2 (EM Treatment 1) e The layers fed with standard commercial food masses with AEM solution (5 L/ton feed). Group 3 (EM Treatment 2) e The layers fed with AEM (5 L/ton feed) treated com- mercial food masses + AEM treated (2 L/1000 L) drinking water. Increased monthly average egg production, egg weight and the decreased mortality ratio and egg wastages identified with the group 3 EM treated layers. The values of egg shell weight, thickness, yolk weight,

yolk index and Haugh index found higher in group 3 EM treated layers. The content of calcium, sodium, potassium, magnesium, iron, zinc and total protein identified maximum in the group 3 EM treated layers. The concentration of total cholesterol, saturated fat, trans-fat found reduced and the monounsaturated, polyunsaturated found higher in EM group 2 and 3 layers. The total cost identified as Rs. 110.80, 38.00 and 52.50 (in Indian Ru- pees) for group 1, group 2 and group 3 treatments respectively. It has inferred from the present findings that, the effect of EM treatments in commercial layers of the egg pro- duction showed good quality parameters and economic value.

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# Introduction

Poultry products are good sources for human population. However, poultry industry confronted due to incidence of various infectious diseases still chaos for the poultry in- dustries. Among these, salmonellosis is infectious and most important zoonotic disease. It transmits vertically and de- creases the production in the infected flocks. Mortality in susceptible birds may reach up to 90% [[1]](#_bookmark7). The use of hybrid layers over the world promoted for production of eggs and meat. The hybrid layers usually start laying about 20 weeks of age and peak egg production reached during the first produc- tion cycle. The average production rate of commercial layers usually remains close to 0.9 eggs per day [[2]](#_bookmark8). For the control of several infectious diseases, the use of antibiotics is increasing day by day. This generous use of antibiotics causes not only severe problems (Bacterial resistance, Dysbiosis) in poultry but also responsible for harmful residual effects in meat and eggs thus leading to public health hazards [[3]](#_bookmark9). In this connection, use of microorganisms is efficiency to reduce the effect of chemical hazards. Of these, several microbes such as *Lactoba- cillus plantarum*, *Lactobacillus casei* and *Streptoccus lactis* (lactic acid bacteria), *Rhodopseudomonas palustrus* and *Rhodobacter spaeroides* (photosynthetic bacteria), *Saccharomyces cerevisiae* and *Candida utilis* (yeasts), *Streptomyces albus* and *Streptomyces griseus* (actinomycetes), and *Aspergillus oryzae*, *Penicillium sp*. and *Mucor hiemalis* (fermenting fungi) [[4,5]](#_bookmark10) previously used in several poultry farm applications such as sustainable agricul- tural, industrial, odor control, waste management and envi- ronmentation. But, studies related to the applications of Effective Microorganisms- (EM) in the poultry field are too limited. Therefore, the present study tried to identify the effect of EM-microbes in egg production, mortality ratio and egg quality analyses with the commercial layers.

# Materials and methods

### *Preparation of AEM (Activated-EM) solution*

The commercially available EM stock solution [Contains, *Lac- tobacilli* sp. (>105 CFU ml—1), *Rhodopseudomonas* sp. (>105 CFU ml—1) and *Saccharomyces* sp. (>105 CFU ml—1)] bought from ECO- Pro, Auroville, Pondicherry, Tamilnadu, India. The

shelf life of the EM stock solution is one year. One part of EM stock- solution and one part of organic jaggery (1:1 V/W ratio) mixed with 50 L of chlorine free ground water. The prepared solution transferred to the food grade plastic container and

tightly closed with plastic lid and kept in shade at ambient room temperature (20e40 ◦C) for 10 days ([Fig. 1](#_bookmark3)). During the fermentation, the lid opened daily once to release pressure.

After 10 days, the AEM solution verified by pleasant sweet sour smell and the acidic pH nature (pH 3.5e4.0).

### *Treatments*

The field experiment carried out at PSP poultry farm, Mettu- patty, Namakkal District, Tamilnadu, India for three months. 25 weeks old *Gallus domesticus* layers selected and divided into three groups.

* + 1. *Group 1 (Control group)* The layers to fed with standard commercial food masses with Biobantox, BMD (Bacitracin methylene disalicylate) and formaldehyde.

### *Group 2 (EM treatment 1)*

The layers fed with standard commercial food masses with AEM solution (5 L/ton).

### *Group 3 (EM treatment 2)*

The layers fed with AEM (5 L/ton) treated commercial food masses + AEM treated (2 L/1000 L) drinking water.

### *Data collection*

After completion of 30 days AEM and chemical treatments (End of the October, 2012), data analysis such as egg produc- tion, mortality ratio and egg wastage analysis taken on the daily basis.

Egg Production = Number of eggs (in numbers)/Total number of birds × 100

Mortality = Number of birds died/Total number of birds × 100 Egg wastage = Number of broken and waste eggs per shed

Fig. 1 e Preparation of AEM solution.

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| Table 1 e Allometric analysis of AEM and chemical treatments in commercial layers. | Parameters Egg Mortality Egg Egg Egg length Egg width Egg shell Egg shell Egg albumin Egg yolk Yolk Haugh production (%)a ratio (%)a wastage (%)a weight (g) (mm) (mm) thickness (mm) weight (g) weight (g) weight (g) index unit (HU) | Group 1 82.81 3.86 38.31 60.4 ± 2.60 55.40 ± 0.60 45.79 ± 0.92 1.41 ± 0.004 6.1 ± 0.29 34.09 ± 2.27 15.02 ± 2.72 0.47 ± 0.03 91.67 ± 3.90  Group 2 82.92 2.48 37.09 61 ± 4.51 54.10 ± 0.51 46.10 ± 0.27 1.39 ± 0.120 5.8 ± 0.90 35.08 ± 2.07 15.83 ± 4.09 0.48 ± 0.92 92.86 ± 3.76  Group 3 83.20 2.59 37.545 61.6 ± 5.83 55.8 ± 0.83 46.01 ± 0.61 1.44 ± 0.004 6.2 ± 1.01 35.03 ± 3.91 15.95 ± 2.93 0.52 ± 0.93 92.91 ± 3.09 | a Values are average of three months data. |

### *Weight and allometric analysis of the egg samples*

After 30 days of treatments 10 eggs selected randomly for the results analysis of the following parameters; the weight of eggs and their parts e albumen, yolk and egg shell with membranes determined with a precision electronic balances. The long and short axes of eggs measured with a technical caliper. The yolk and albumen heights determined with a tripod micrometer with a precision of 0.01 mm. The egg shell thickness with shell membrane measured with a special AMES micrometer in the equatorial region and both poles (sharp and blunt). The arithmetic mean of the three mea- surements kept as egg shell thickness.

### *Haugh units*

The HU value was identified with the following formulas *HU* = *100\*log(h* + *7.57* — *1.7\*EW0.37)*, where *h* is the height of thick albumen at the boundary with the yolk; EW e the egg weight [[6]](#_bookmark11).

### *Yolk index*

Yolk quality was evaluated through the yolk index: *YI* = *h/D*, where *h* is the yolk height and *D* e yolk diameter [[7]](#_bookmark12).

### *Estimation of mineral content*

Mineral content of the total egg content (Yolk and albumen) sample tested after dry ashing of samples at 550 ◦С and dissolution in hydrochloric acid. Elements, such as calcium, potassium, magnesium, iron and zinc determined with the atomic absorption spectrophotometer Analist 800, *Perkin Elmer* at Omega laboratories, Namakkal.

### *Estimation of the fatty acid composition*

The lipid and fatty acid composition of the yolk determined by fat extraction with methanol: chloroform (2:1) by the method of [[8]](#_bookmark13) Bligh and Dyer at Omega laboratories, Namakkal.

### *Estimation of biochemical analysis*

The level of total carbohydrates, total protein and total cholesterol content determined standard colorimetric methods [[9]](#_bookmark14) at Omega laboratories, Namakkal at Tamilnadu, India.

### *Cost analysis*

The cost analysis of antibiotics, EM stock solution and jaggery calculated for preparing 1 ton feed masses and the results were expressed as in Indian Rupees.

### *Statistical analysis*

Statistical analysis such as ±SEM calculated with MS office 2003.

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| Table 2 e Mineral content, biochemical composition and fatty acid analysis of AEM and chemical treatments in commercial layers. | | | | |
| S. no | Test parameters (mg/100 gms) | Group 1 | Group 2 | Group 3 |
| 1. | Calcium | 44.5 ± 0.50 | 45.5 ± 0.5 | 64.5 ± 0.50 |
| 2. | Phosphorus | 124.0 ± 1.00 | 125.0 ± 5 | 122.0 ± 2.00 |
| 3. | Sodium | 15.0 ± 0.00 | 15.5 ± 0.5 | 21.0 ± 1.00 |
| 4. | Potassium | 41.0 ± 1.00 | 42 ± 2.00 | 43.0 ± 1.00 |
| 5. | Magnesium | 02.0 ± 0.00 | 2.0 ± 0.00 | 2.5 ± 0.00 |
| 6. | Iron | 1.05 ± 0.05 | 1.25 ± 0.05 | 1.50 ± 0.00 |
| 7. | Zinc | 0.75 ± 0.05 | 0.82 ± 0.025 | 0.95 ± 0.05 |
| 8. | Total protein | 5.43 ± 0.03 | 5.51 ± 0.24 | 6.44 ± 0.06 |
| 9. | Total carbohydrate | 1.27 ± 0.025 | 1.40 ± 0.235 | 1.10 ± 0.025 |
| 10. | Total cholesterol | 363.00 ± 11.50 | 355.00 ± 0.03 | 320.00 ± 20.00 |
| 11. | Calories (K.cal) | 108.00 ± 2.00 | 112.50 ± 0.03 | 127.50 ± 7.5 |
| 12. | Total fat | 9.3 ± 0.10 | 9.25 ± 0.98 | 10.3 ± 0.15 |
| 13. | Monounsaturated fat | 4.0 ± 0.0 | 4.24 ± 0.04 | 5.55 ± 0.05 |
| 14. | Polyunsaturated fat | 1.42 ± 0.02 | 3.0 ± 0.00 | 2.9 ± 0.10 |
| 15. | Saturated fat | 3.05 ± 0.05 | 2.50 ± 0.00 | 2.55 ± 0.05 |
| 16. | Trans fat | 1.54 ± 0.005 | 1.01 ± 0.01 | 0.75 ± 0.05 |

# Results

The percentage occurrence of maximum monthly average egg production (88.28%, 84.28% and 83.20%) identified with the group 3 EM treated layers in all the three consecutive months over the group 1 and group 2 treated layers. Further, the layer mortality ratio and egg wastages found decreased when compared with the group 1 and group 2 treated layers. The range of maximum egg weight (61.6 ± 5.83 g) identified with the group 3 EM treated layers. Further, the results of the egg

height and width not showed and major variations between the groups. Further, the maximum range of egg shell weight (6.2 ± 1.01 g) and shell thickness (1.44 ± 0.004 mm) identified with the group 3 EM treated layers. Additionally, the results of

the albumin weight not showed any major differences be- tween the chemical and EM group treated layers. Further the value of yolk weight (15.95 ± 2.93 g), yolk index (0.52 ± 0.93) and

Haugh index (92.91 ± 3.09) found higher in group 3 EM treated

layers when compared with the control layers ([Table 1](#_bookmark4)). The

phosphorous identified with the group 2 EM treated layers. The maximum concentration of the total protein (6.44 ± 0.06 mg/100 g of egg) identified with the group 3 EM treated layers. Further, the concentration of total cholesterol reduced (320.00 ± 20.00 mg/100 g of egg) when compared with the group 2 and control (Group 1) layers. In addition, the maximum range of energy value (127.50 ± 7.5 Kcal) identified with the group 2 EM treated layers. Further, the maximum concentration (1.40 ± 0.235 mg/100 g of egg) of total carbohy-

drate identified with the group 1 EM treated layers. The

monounsaturated (5.55 ± 0.05 mg/100 gms), polyunsaturated (3.0 ± 0.00 mg/100 gms) found higher in EM group 3 and group 2 layers than the chemical (Group 1) treated layers. Further, the saturated (2.55 ± 0.05 mg/100 gms) and trans fat

(0.75 ± 0.05 mg/100 gms) concentrations found decreased than

the chemical group treated layers (Group 1) ([Table 2](#_bookmark5)). The total cost identified as Rs.110.80, 38.00 and 52.50 (Indian Rupee) for group 1, group 2 and group 3 respectively ([Table 3](#_bookmark6)).

maximum concentration of calcium (64.5 ± 0.50 mg/100 g of

egg), sodium (21.0 ± 1.00 mg/100 g of egg), potassium (43.0 ± 1.00 mg/100 g of egg), magnesium (2.5 ± 0.00 mg/100 g of egg), iron (1.50 ± 0.00 mg/100 g of egg) and zinc (0.95 ± 0.05 mg/

100 g of egg) identified in the group 3 EM treated layers when compared with the control and group 1 treatments. Similarly, the maximum (125.0 ± 5 mg/100 g of egg) concentration of the

# Discussion

Egg is a biological structure intended by nature for reproduc- tion. It protects and provides complete diets for the devel- oping embryo and serves as the principle source of food for the first few days of the chick's life. Eggs are special cells found

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| Table 3 e Cost analysis of EM and chemical treatments in commercial layers. | | | |
| Parameters | Ingredients | Cost of ingredients/ton/L of feed production  (in Indian Rupees) | Total grand  (in Indian Rupees) |
| Group 1- layers treated with chemicals | I) Biobandox | 62.00/- | 110.80/- |
|  | II) BMD | 9.80/- |  |
|  | III) Formaldehyde | 39.00/- |  |
| Group 2- layers treated with AEM in | AEM solution (5 L of AEM/ton of feed masses) | 38.00/- | 38.00/- |
| food masses |  |  |  |
| Group 3- layers treated with AEM in | AEM solution (5 L of AEM solution/ton of feed | 52.50/- | 52.50/- |
| drinking water and food masses | masses + 2 ml of AEM/L of drinking water) |  |  |

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in female animals and nearly all animals produce them, some animals including birds lay their eggs external to their body and it is unquestionably one of the most nutritionally balanced foods for man. In poultry product egg is a major product composed of albumin (58%), egg yolk (31%) and egg shell (11%). In addition, albumen contains half their contents of the egg portion and the yolk contains the major portion of the vitamins and lipids [[10,11]](#_bookmark15). In general, the quality of the egg started with the consumer's needs and the treatment

methods which give general characteristics of the egg with

intact shell, shape size, appearance and the internal quality of the egg parts. In addition, the several antibiotics also used as potential agents to cure several diseases including the mor- tality decline in the livestock's [[12]](#_bookmark16). But, the efficiency of an-

tibiotics is good in poultry products. Unfortunately, the

consumer's views are that edible poultry tissues contami- nated with harmful concentrations of drug residues. Because of this, the present study tried to identify the effect of effective microorganisms in the egg quality, mortality and production ratio in the commercial layers. The results of the present study suggested that, the average percentage production of eggs found higher in EM treated layers than the chemical treated layers; this might be due to the excessive beneficial

EM- microbes in the gut motility region, thereby increasing the uptake and absorption of the nutrients [[13]](#_bookmark17). Similar reports are also identified with the *Bacillus subtilis* treated layers [[14]](#_bookmark18). Further, the results of the mortality ratio also found decreased when compared with the chemical treated layers this might be due to the increased survival nature of the effective mi- croorganisms by altering the intestinal flora [[15]](#_bookmark19) and the suppressing nature of the pathogenic bacteria and improving the immune potency [[16,17]](#_bookmark20). Further, the results of the external parameters such as egg weight, height, width, shell weight, yolk weight and Haugh units anonymously varied between the chemical and EM treated layers. This agrees with the earlier reports [[18]](#_bookmark21). The results of the egg shell thickness showed the increased range values than the normal chemical control layers this might be due to the increased assimilation of the calcium in the intestinal tract [[14]](#_bookmark18). This may also responsible for reducing the egg wastages in the EM treated layers. Similarly Mahdavi et al. [[19]](#_bookmark22) reported the improved egg shell thickness in hens supplemented with the probiotics. The results of the yolk index found higher in the EM treated layers than the chemical treated layers. Enhancing the yolk index related to the stability of the yellow pigments in the mem- brane of the yolk between the lipid molecules and this might be due to the increased uptake of xanthophylls from the corn [[20]](#_bookmark23). The results of the mineral intakes found higher calcium, magnesium, iron, zinc and potassium and this might be due to the acidic nature environment nature of the microbial flora in the intestinal tract this may increase the mineral ionization and thus increase the mineral absorption ratio [[14]](#_bookmark18). The re- sults of the cholesterol concentration decreased than the other biochemical constituents and this might be due to assimilating the cholesterol in the gastrointestinal tract for their own cellular metabolism thus reducing the level of cholesterol absorption [[21]](#_bookmark24). Similarly, Kalavathy et al. [[22]](#_bookmark25) showed that lactic acid bacterial strains may alter the entero hepatic cycle and reduce cholesterol through assimilating dietary cholesterol into the bacterial cells and the bile salt

hydrolyzes in the intestine. Further, the saturated and trans fat found decreased in the EM treated layers this might be due to the inhibit hydroxymethyl-glutaryl-coenzyme A, an enzyme involved in the gastrointestinal tract [[23]](#_bookmark26). The cost analysis of chemical and EM treatments showed tremendous gain in the reduction of the chemical usages.

# Conclusion

It has concluded that, the effect of EM technology in poultry applications are helpful to improve the cost value and the quality parameters of the egg compositions.

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