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INFLUENCE OF FEEDING VARIOUS KINDS, LEVELS AND COMBINATIONS OF ANTIBIOTICS ON GROWTH AND FEED EFFICIENCY OF BROILER-STRAIN CHICKS

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INTRODUCTION

The present use of antibiotics in animal nutrition as growth promotants has been a unique phenomenon in the history of medicine. Antibiotics primarily being medicinal drugs of great potency and therapeutic value have been very effective in promoting growth by their mysterious mode of action, yet to be fully understood.

Since antibiotics which differ in their chemical and physical properties stimulate growth of chicks and poults, it is not likely their effect on metabolism can be a direct one. The only known property which these substances have in common is their antibacterial potency. So the growth promoting action of antibiotics is widely attributed to their favorable influence on the intestinal microflora as evidenced by the growth response in 'old' and contaminated environment compared with 'new' and clean one. However, the extent and nature of these changes are inconsistent for drawing definite conclusions. It does appear that the nutritional effects of antibiotics are most likely as a result of increased bacterial synthesis of essential nutrients, decreased bacterial competition with the host for the essential nutrients, and improved utilization of nutrients possibly due to reduced irritation and destruction of intestinal wall. But the exact elucidation of mode of action is still a debatable issue.

It is unlikely that under practical conditions of commercial farms, the premises could be maintained 'clean' or subclinical infections reduced to such an extent as found under experimental conditions in a laboratory. This is the reason that low level feeding of antibiotics or other growth promotants, generally 10 grams per ton of feed, are commonly used as feed additives in commercial feed formulas. Therefore antibiotics have wide acceptance as supplements to poultry feeds.

In a short span of a decade voluminous work has been done to determine the potency and mode of action of various growth promotants as feed additives. However, these results are inconsistent and so variable as many factors, namely, kinds of antibiotics (newer ones are being added continuously), environment, genetic variability, and type of diet, etc. have a profound influence on the degree of response obtained. Many recent reports have indicated a loss of response to antibiotics which were in use over a period of years and had given a consistent growth stimulation. It has been suggested that there may be a built-up resistance of antibiotic-fast microorganisms or a 'cleaning up' effect of environment. While it is rather premature to draw positive conclusions from such reports, nevertheless the possibility of decreasing response cannot be completely discounted. This emphasizes the need to study further, the new antibiotics appearing on the market, their optimum levels for supplementation and possibly their combinations in order to ascertain and further investigate their growth promoting value in poultry rations.

Though there are variations in sensitivity of microorganisms to different antibiotics with their specific spectra, their combinations and synergistic value, especially the newer products offer further scope for investigation. Also there are many conflicting reports in the literature about the combining ability of various antibiotics and their potency in growth stimulation of chicks.

Therefore in the present investigation a series of three experiments from September 1963 to May 1964 were conducted on wire in batteries employing meat-strain chicks to test the kind, levels and combination of antibiotics on growth and the efficiency of feed conversion. The following were studied: (1) effect of antibiotics on growth of broiler-strain chicks, (2) comparison of single sources and combination of antibiotics, (3) response to different levels among single sources and among combinations, (4) and the efficiency of diet utilization, that is, pounds of feed required per pound of gain in body weight.

REVIEW OF LITERATURE

The investigations during the period of 1940-48 culminating in the establishment of antibiotic growth stimulation and their supplemental value as feed additives really grew out of the earlier investigations of what was termed the "Animal Protein Factor" (APF).

During the course of these investigations, using fermentation residues for APF and its relation with vitamin B_{12}, Stockstad et al. (1949) found evidence of an auxiliary growth factor which was later identified as an antibiotic. Earlier Moore et al. (1946) demonstrated that streptomycin stimulated chick growth. The significance of his finding was overlooked due to the fact that a purified diet was used and was primarily concerned with the synthesis of vitamins by the microorganisms in the intestinal tract.

Following this discovery of antibiotic growth stimulation a number of workers have reported the results of different antibiotic feed supplementation in poultry rations, McGinnis et al. (1950, 1951) Groschke and Evans (1950), Davis and Briggs (1951) and others. The period of 1950-54 was marked by an intensive research for antibiotic growth stimulation in poultry, which has been reviewed by Braude et al. (1953), Branion et al. (1953), Jukes and Williams (1953), Stockstad (1953, 1954) and Erdheim (1955). Therefore, no attempt has been made in this presentation to review extensively the literature prior to 1954, and only pertinent references to the present work have been cited.

A number of workers have investigated the antibiotic response and its effect on intestinal microorganisms in explaining the mode of action. Sieburth et al. (1954) indicated that growth stimulation is due to greater utilization of nutrients by the host or decreased production of toxic substances by the microorganisms as all cultivable microflora were decreased in small intestines of chicks fed aureomycin or penicillin. Hauser et al. (1954, 1956) attributed antibiotic growth stimulation to increase in intestinal coliforms and reduction in lacto-bacilli. Slinger et al. (1954) and Rhodes et al. (1954) supported the idea of a positive correlation between coliform count and antibiotic growth stimulation.

Elam et al. (1954) observed a decrease in clostridia count when the diet was supplemented with penicillin. Eisenstark and Sanford (1953) and Eisenstark and Dragsdorf (1953) reported no consistent changes in the different types of intestinal microflora but they indicated an individual micro-cell enlargement possibly responsible for growth stimulation. Warden and Schaible (1962) fed cellular contents of lysed and lypolysed E. coli and antibiotics suggested an enzymetic action of antibiotics in growth stimulation.

The various studies of growth stimulation to antibiotics have also indicated a sparring action on essential nutrients. Monson et al. (1954) using limiting amounts of folic acid reported a growth stimulation to antibiotics. West and Hill (1955) observed a sparring effect of antibiotics on protein which was reduced from a level of 22 per cent to 18 per cent for optimum growth and feed efficiency.

White-Stevens and Zeibel (1954), in their studies with chlortetracycline and protein level of the diet, reported a significant curvilinear response with an optimum protein level of 20-22 per cent and 50-100 ppm of chlortetracycline supplementation, while Thayer and Meller (1955) indicated an improved nitrogen utilization in their nitrogen balance studies with aureomycin and penicillin.

Hill and Kelly (1955) observed a significant growth response to high levels of antibiotics only when the diet contained 5 per cent fish meal. Neither antibiotic nor fish meal alone were equally effective. The feed intake of chicks was equalized. Eyssen and DeSomer (1963) found a close correlation between the activity of antibiotics and intestinal fat absorption, while Nelson _et al._ (1963b) attributed growth stimulation to increased efficiency of absorption of calorgenic nutrients by decreased intestinal weights of the birds fed antibiotics.

The effect of environment on growth stimulation to antibiotics and a loss of response of late to these supplements have been reported by many workers. Waibel _et al._ (1954) reported the results of their experiments from 1950 to 1953. Penicillin and aureomycin which had given good growth response were ineffective during 1952-53. Morrison _et al._ (1954) also observed a loss of growth response in 'new' environment compared with 'old'. Libby and Schaible (1955) conducted experiments in the same laboratory from 1950-1954 and observed a loss of response to antibiotics each year. The decrease ranged from 19 per cent to 3.3 per cent during this period while controls improved as much as 19.1 per cent.

Matterson et al. (1959) conducted experiments in the same facilities over nine years using bacitracin, penicillin, and aureomycin at different levels in high energy rations, and observed a loss of response in comparison with that obtained in 1951. Heth and Bird (1962) reviewed the growth response to penicillin, chlortetracycline, and oxytetracycline in their laboratory from 1950-61. Contrary to the above reported findings they found no long term changes in growth stimulation, though variations among trials were observed in each period for both penicillin and tetracyclines.

A number of antibiotics in either crude or crystalline form have been studied and new ones as they are appearing on the market have been further investigated for their growth stimulating properties. As early as (1950) McGinnis et al. obtained significant growth stimulation to either crude or crystalline aureomycin and streptomycin. Runnels et al. (1951) reported that penicillin and streptomycin in practical broiler diets significantly stimulated growth of chicks.

Reynolds et al. (1951) working with penicillin and terramycin observed a significant growth stimulation at 2 gram per ton of terramycin and penicillin but there was no statistical difference between antibiotic fed groups. Lillie and Bird (1953), in their evaluation of crystalline antibiotics, found chloromycetin ineffective while its mycelia produced slight growth stimulation. Tomalidine also gave no response and fumagillin was toxic at levels used. Tyrothricin, gramicidin and neomycin showed evidence of growth stimulation but their effect was less consistent and of lower magnitude than the antibiotics in use. Boone and Morgan (1955) observed growth stimulation of broilers to low levels of aureomycin, penicillin, terramycin, and bacitracin.

McGinnis et al. (1958) reported the results of three experiments with turkey poults fed the commonly used antibiotics, penicillin, streptomycin, and terramycin as well as the newer ones, oleandomycin and erythromycin. The former failed to improve growth while the latter gave significant growth responses in all the three experiments. They emphasized the need for proper evaluation of the effectiveness of the antibiotics.

Biely and March (1959) compared the antibiotics chlortetracycline, penicillin, oxytetracycline, and oleandomycin in two different environments, and at two dietary levels. Oleandomycin proved more effective than all the other antibiotics irrespective of diet or environment.

Mameesh et al. (1959) obtained consistent growth response only when oxytetracycline was added to a diet containing raw hen feces. While procaine penicillin gave a growth response in one of the four experiments, various forms of sulphur compounds were inactive in promoting growth.

Wiese and Peterson (1959) compared several antibiotics in a series of four experiments. Both penicillin G and penicillin V gave a small growth response over other antibiotics. They suggested a micro-resistance to penicillin due to its use over the years. Anderson (1960) presented further evidence of the ineffectiveness of penicillin and chlortetracycline in growth stimulation while bacitracin, erythromycin, and oleandomycin produced significant increases in growth.

Trail and Casley (1960), while reporting a significant growth response to aureomycin in early stages of growth, emphasized the need for a careful experimental design and statistical analysis to prevent confounding the effects of other factors with the ones being tested. Warden and Schaible (1961) added aureomycin, terramycin, and bacitracin to a diet contaminated with fresh, dried (100°F), and autoclaved hen feces. Fresh feces depressed growth while dried and autoclaved feces gave a growth response to aureomycin and terramycin but bacitracin was only partially effective.

Yates and Schaible (1961) further tested the value of virginiamycin, terramycin, and bacitracin in batteries and floor pen reared chicks. Virginiamycin gave increased response in batteries as levels increased from 4-100 grams per ton while higher levels of zinc bacitracin and terramycin were not as beneficial as lower levels.

Heth and Bird (1961) compared both pharmaceutical and feed grades of spiramycin with chlortetracycline, oxytetracycline, and erythromycin at levels of 5-20 ppm. in two trials, one in battery and the other in floor pens. The growth response to antibiotics varied from experiment to experiment. Both grades of spiramycin compared favorably with other antibiotics. Eyssen (1962) observed a significant growth response to virginiamycin when supplemented to semi-synthetic as well as practical-type of diet.

Potter *et al.* (1962) obtained a significant increase in weight over basal when the diet was supplemented with procaine penicillin, zinc bacitracin, and erythromycin at four weeks of age but nonsignificant differences at eight weeks. This was also true for the differences in feed efficiency. Erythromycin and spiramycin proved more effective than penicillin and zinc bacitracin.

Nelson *et al.* (1962) reported a significant growth stimulation with addition of erythromycin to the diet while penicillin, tylosin, and SKF1-7980 stimulated growth in only one experiment. Feed efficiency and metabolizable energy of the diet were increased by antibiotics which stimulated growth.

Combs and Bossard (1963) reported the results of four experiments, two in batteries and two in floor pens. Virginiamycin (at 8.8 ppm) alone gave significant growth response in all the experiments when compared with chlortetracycline, oxytetracycline, oleandomycin, spiramycin, zinc bacitracin, procaine penicillin, tylosin, spontin, and erythromycin. Chlortetracycline and erythromycin each significantly stimulated growth only in one different experiment. The growth response to virginiamycin was not due to the level employed as higher levels of other antibiotics still gave less response. Nelson et al. (1963a) summarized the results of 29 experiments over a three year period. They observed a decreasing response to penicillin, bacitracin, tylosin, and erythromycin supplemented routinely. Tylosin stimulated growth markedly when first used, but the response diminished with subsequent use.

There are variable reports about the effectiveness of combination of antibiotics in their growth stimulatory value in poultry rations. McGinnis et al. (1951) reported a significant growth stimulation of chicks fed practical-type diets supplemented with aureomycin, streptomycin, terramycin, and penicillin. Penicillin proved to be most effective and streptomycin the least. A combination of penicillin, terramycin, and streptomycin gave no greater growth than penicillin alone. Davis and Briggs (1951) observed a significant growth stimulation to procaine penicillin G., aureomycin, bacitracin, terramycin, and streptomycin in most cases, but not in all. A mixture of aureomycin and streptomycin, penicillin combined with bacitracin gave no greater, but possibly less growth stimulation, than either antibiotic added alone. There was an improvement in feed efficiency to antibiotic supplementation.

Matterson _et al._ (1952) investigated the growth stimulatory value of all possible combinations in pairs of aureomycin, penicillin, terramycin, and bacitracin at three different levels. No combination of antibiotics gave a growth response significantly greater than obtained by the better of the two antibiotics when supplemented alone at a level equal to its concentration in the particular combination.

Sanford (1952) obtained superior growth with chicks fed a combination of two antibiotics and vitamin B_{12} feeding supplements than a combination of four of these supplements. Lewis and Sanford (1953) observed that combination of antibiotic B_{12} supplement Aurofac$^{(R)}$ and bacitracin appeared to be more consistent in growth promoting properties than other supplements when added to a cottonseed all-vegetable protein diet.

Wisman _et al._ (1954) reported the results of their experiment adding terramycin, penicillin, and streptomycin to a plant protein type of diet, as single source, combination and interchanging one with the other. Terramycin and penicillin produced comparable growth to 10 weeks, when fed either singly or as a mixture or when one replaced the other at three weeks of age. Streptomycin was less effective at low levels and penicillin seemingly compensated for its ineffectiveness but terramycin failed to do so. March _et al._ (1954) observed a variable growth stimulation to surface active agents supplemented singly as well as in combination with antibiotics in comparison to antibiotics alone in the diet.

Stephenson and Sullivan (1955) did not obtain any significant benefits by adding high levels of a single source or combination of antibiotics to a basal diet that already contained four grams per ton of penicillin. West (1956) in a series of nine trials compared the effects of combining 3-nitro-4-hydroxyphenyl arsonic acid with low and high levels of antibiotics. More definite and consistent improvement in growth and feed efficiency were observed at low levels than at higher levels; however, the disappearance of antibiotic response was observed during the series of trials.

Heywang (1957) observed an average increase in chick weights and feed efficiency during hot weather to a combination of one gram of procaine penicillin and three and three-fourths of either chlortetracycline or oxytetracycline. Equal results were obtained only when either of the antibiotics were fed at 50 to 100 ppm in the diet.

Gard et al. (1958) obtained improved growth response and feed efficiency to antibiotic 13184 (alone and combination) ilotycin and procaine penicillin over controls in the first experiment. However, in subsequent experiments, only antibiotic 13184 (alone and combination) gave significant growth response. Pope and Schaible (1958) reported growth stimulation to a combination of furazolidone, penicillin, and 3-nitro-4-hydroxyphenyl arsonic acid.

Monson et al. (1959) indicated a loss of response to low levels of procaine penicillin, bacitracin, oleandomycin, and attrinin. A combination of zinc bacitracin and penicillin gave only a small response in one test. These differences are of doubtful significance because there was a variation in duplicate pens.

Sherman et al. (1959) observed a significant growth response to oleandomycin supplementation of diet of chicks raised in 'old' environment at three tests in batteries and two on floor pens. While penicillin gave only a slight response, a combination of oleandomycin and penicillin under the conditions did not give a better response than obtained with oleandomycin alone.

Menge and Lillie (1960) reported variable results of the combination of antibiotics in a series of six experiments. The combination of penicillin, streptomycin, and nystatin gave a significant response over basal in the first three experiments, but failed to promote rapid growth during the fourth to sixth experiments. There was no indication of any improvement in efficiency of diet utilization to antibiotics.

Stutz (1961) obtained a significant growth response with a combination of zinc bacitracin and erythromycin; whereas, there were no significant differences in growth response when these supplements were used as single sources in his experiment. Siddiqui (1963) also observed a significant growth stimulation of meat-strain chicks to a diet supplemented with a combination of furazolidone, zinc bacitracin, and erythromycin than when these agents were used singly.

MATERIALS AND METHODS

A series of three experiments were conducted at the Kansas State University poultry farm in the poultry nutrition laboratory. A total of 936 Cobb Strain-cross, White Rock straight-run broiler chicks were used in these experiments.

Experiment 1 was initiated September 23, 1963 and ran for eight weeks. The second experiment was started January 30, 1964 and ran for eight weeks. The third and final experiment was initiated March 23, 1964 and ran for eight weeks. All the three experiments were conducted on wire in batteries.

The chicks were randomized into 24 lots of 15 chicks each, vaccinated intranasally for Newcastle and infectious bronchitis. They were wing banded, individually weighed and then randomly assigned to the different tiers in the electrically heated battery brooder. The chicks were reared till four weeks of age in these starting batteries, and then at this time were transferred to the growing batteries till eight weeks of age. Feed and water were provided ad libitum.

The 1963-64 Kansas State University chick broiler starter ration was used as the basal diet till five weeks of age and then the K.S.U. 1963-64 broiler finisher was fed from 5-8 weeks of age. The composition of the two rations is given in Tables 1 and 2 (Appendix)*. The antibiotic supplements were blended with the K.S.U. broiler basal at various levels and combinations as shown in Table 3. Antibiotics erythromycin, zinc bacitracin, and tylosin were used as supplements. The different supplements were blended homogenously by the help of appropriate mixers and stored in cans for daily feeding. Each diet was fed to two replicate lots of chicks. The quantity of feed offered and that weighed back at the time of each biweekly weighing was noted for calculating the total amount of feed consumed. The feed efficiency or the pounds of feed required for each pound of gain in body weight was

*All tables appear in the Appendix.

determined for each lot at the end of each 8-week experimental period as reported in Table 11.

Individual body weights were recorded for each biweekly period. At the end of each experiment, 8-week weights were adjusted for sex. All female weights were adjusted to male weight by noting the difference between the average weight of the females and the average weight of the males for each lot. These differences for all lots were added together and an overall average was calculated. This amount was then added to each female weight for the final adjustment. The final 8-week adjusted weights for all experiments are listed in Table 5. The weight gains for each lot in all the three experiments were adjusted for sex by adding the average male and average female weights and dividing by two. These are presented in Table 4.

Analysis of variance of the data from the three experiments was run on individual adjusted weights in each experiment separately as well as pooled together for three experiments. Also all possible pooling of data from either of the two experiments were analyzed by the analysis of variance to test the pattern of growth response to the diets fed. The diets were ranked from highest to lowest based on L.S.D. method according to the procedure described by Snedecor (1956) and are shown in Tables 6, 7, 8, and 9. The orthagonal comparisons of different levels, single and combination of antibiotics, were determined to test the overall effects of the various supplements used in these experiments and are presented in Table 10. The analysis of variance of feed conversion for the pooled data of all the three experiments is reported in Table 12.

RESULTS

An analysis of variance was run on the adjusted 8-week weights of the first experiment which indicated a significant difference at .05 level of probability between diets. A further analysis by L.S.D. method showed that under the conditions of the experiment there was no significant difference between no supplement versus supplement. However, it was observed that the higher level of 20 grams per ton of single source of antibiotics, erythromycin, and zinc bacitracin, (Diet 5 and 6) performed significantly poorer than Diets 9, 11, 8 and 7.

The analysis of variance of the data for the second experiment revealed no significant differences between diets; however, the nonsupplemented Diet 1 (negative control) gave on an average the least increase in weight when compared with supplemented diets. The data, from both experiments one and two, were pooled and an analysis of variance run on the 8-week adjusted weights. This indicated a significant interaction between experiments and diets at 0.05 level of probability while there was no significant difference between diets. This indicated a variation in the performance of the same diet in both experiments. Also a highly significant difference between experiments was observed, (Table 7).

The 8-week adjusted weights in the third experiment were also tested by the analysis of variance which revealed a significant difference between diets at .05 level of probability. The differences in the diets were further tested by L.S.D. analysis which showed Diets 9, 2 and 8 were significantly better than all other diets. Only Diets 10 and 5 were significantly superior to Diet 4, otherwise, there was no significant difference between Diets 10, 5, 6, 3, 12, 11, 7, and 1.

At the conclusion of all the experiments, an analysis of variance was run on the data pooled from the three experiments. There was no significant difference between diets, while a highly significant difference between experiments and also a significant interaction between experiments and diets was observed as shown in Table 6. However, the average weights for all the three experiments of each 12 diets showed that chicks fed antibiotic supplemented diets weighed more than the controls. This increased weight per cent of basal (negative control) ranged from 0.6 to 3.9 per cent. Diet 9 (zinc bacitracin and tylosin) gave the highest per cent increase and Diet 6 (zinc bacitracin) the least, (Table 5).

The single source of antibiotics at the level of 10 grams per ton on a combined average basis gave an increase of 1.57 per cent over the basal (negative control), while the 20 grams per ton level of these supplements increased the weights by only 0.9 per cent. The combination of antibiotics at the 20 grams level (10 g./ton of each) showed a combined average growth response of 1.57 per cent which was equal to the single sources at 10 grams per ton level. However, the combination of these antibiotics at 10 grams per ton (5 g./ton each) gave the greatest growth increase of 3.1 per cent over the basal (negative control) under the conditions of the experiment.

Further analyses of variance were run on the data pooled from experiments one and three and from experiments two and three to study all possible trends in growth stimulation to the supplements.

The results of the analysis of the data of experiments, one and three revealed a significant difference between diets at .05 level of probability while the interaction between experiments and diets was statistically nonsignificant. A highly significant difference between experiments was observed as a result of general growth differences from experiment to experiment. The L.S.D. analysis which was applied to the data to further test the differences between diets showed that Diet 9 (zinc bacitracin and tylosin) significantly increased growth over Diets 2, 10, 7, 3, 1, 12, 4, 6, and 5. There was no significant difference between Diet 9 and Diets 8 and 11 (Table 8). An orthagonal comparison of various combinations and levels of antibiotics presented in Table 10(a) indicated a significant difference at .05 level of probability between single sources versus combinations.

The pooled data from experiments, two and three also revealed a significant difference between diets at .05 level of probability, while the interaction between experiments and diets was nonsignificant. Further analysis by the L.S.D. method to locate the differences between diets, indicated nonsignificant differences between Diets 8, 2, 9, 5, 10, 6, 12, 7, 3 and 4. Diets 8, 2, 9 and 5 were significantly different from Diets 11 and 1 which is presented in Table 9. The orthagonal comparisons (Table 10(b)) of combinations and levels of the antibiotics indicated a significant difference at .01 level of probability between no supplement versus supplement; however, there was no statistically significant difference between single sources and combination of antibiotics.

Feed conversion varied between supplements and in some cases between the lots receiving the same diet. This is shown in Table 11. The poor feed conversion above three pounds reported for Experiment 1 was due to feed wastage in handling. The analysis of variance of the pooled data of all three experiments revealed no significant differences in feed conversion among diets (Table 12). The replications within experiments and the interaction between experiments and diets was also statistically nonsignificant.

DISCUSSION

The results from the series of three experiments clearly demonstrate the effective value of combination of antibiotics in stimulating growth. The two antibiotics zinc bacitracin and tylosin increased the weight of chicks more than any other supplement as compared with the basal diet. The combination of erythromycin and tylosin ranked next best for improved weight gains. These results are in agreement with Sanford (1952), Lewis and Sanford (1953), Stutz (1961) and Siddiqui (1963). A number of workers have obtained contrary results as reported by McGinnis et al. (1951), Matterson et al. (1952), Stephenson and Sullivan (1955) and Sherman et al. (1959).

However, the results obtained under the conditions of this study indicate that in some cases variations in growth response was apparent for the same antibiotic when compared between the experiments. Menge and Lillie (1960) reported similar results from a series of six experiments in which a combination of penicillin, streptomycin, and nystatin gave a significant response only in their first three experiments. West (1956), working with a combination of arsonic acid and antibiotics in broiler diets, observed a disappearance and reappearance of antibiotic response. Similarly Heth and Bird (1961) reported a variable response to antibiotics, spiramycin, chlortetracycline, and erythromycin. Stutz (1961) indicated a variation in growth response for the same antibiotic when compared between two experiments, while Heth and Bird (1962) reviewing the antibiotic growth stimulation, indicated variations among the trials.

The lower levels of combinations of zinc bacitracin and tylosin and erythromycin and tylosin were more effective in stimulating growth than the higher level combinations of these antibiotics used in the present experiments. These results are in agreement with West (1956) who reported a more definite improvement in growth at lower level of antibiotic and arsonic acid combinations than with the higher ones. Stutz (1961) and Siddiqui (1963) also obtained significant growth stimulation to lower levels of antibiotic combinations. In contrast Matterson et al. (1952) reported no beneficial results of combining antibiotics at three different levels.

Even the two higher levels of single antibiotics zinc bacitracin and erythromycin proved to be less effective than their lower levels used under the conditions of the experiment. Similar results are reported by Boone and Morgan (1955), Yates and Schiable (1961), Stutz (1961) and Combs and Bossard (1963).

Many reports have appeared in the literature indicating a variability and an apparent loss of response to different antibiotics in common use, Davis and Briggs (1951), Lillie and Bird (1953), McGinnis et al. (1959), Biely and March (1959) and Combs and Bossard (1963). In agreement with these findings, the growth response to zinc bacitracin, tylosin and erythromycin varied in each experiment during the present investigations.

It is possible that zinc bacitracin, due to its use in this laboratory for a number of years, stimulated growth to a lesser magnitude and gave the least growth response at the higher level used in these experiments. Nelson et al. (1962) reported a similar decrease in response to zinc bacitracin in their laboratory. Stutz (1961) obtained the least growth response with single source of zinc bacitracin in this laboratory, while Potter et al. (1962) contrary to these findings, indicated less growth stimulation to new antibiotics compared with penicillin and zinc bacitracin.

Tylosin, a comparatively new antibiotic not routinely used in this laboratory, gave a better response in the first and second experiment, in that order, while in the third it failed to stimulate growth. Nelson et al. (1962, 1963a) reported similar results for tylosin supplementation which markedly stimulated growth when first used but the response diminished in later trials.

Erythromycin at the lower level gave more or less a consistent response in each experiment in comparison with the other two antibiotics used during the three experiments. Potter (1962) reported more effective results obtained with erythromycin and spiramycin than procaine penicillin and zinc bacitracin, while Nelson et al. (1963a) observed a diminishing response to the routine use of erythromycin.

Though tylosin when supplemented as a single source gave less consistent growth response it proved more effective in combination. Both zinc bacitracin and tylosin and erythromycin and tylosin gave the best growth response under the conditions of the experiment. This indicates that certain supplements may be of value only in combination rather than when fed singly. Wisman et al. (1954) reported that streptomycin produced less growth to 10 weeks of age when fed singly, but a mixture of penicillin and streptomycin seemingly compensated for its ineffectiveness. Stutz (1961) indicated a beneficial value of zinc bacitracin combinations with his experiments.

The significant differences between experiments observed during the present trials indicate a need for environmental control to separate the possible effect of weather changes notably temperature on growth rate which may be confounded with the response to antibiotics. Trail and Casley (1960), using advanced statistical analysis, emphasized the need for a careful experimental design and analysis to separate the factors to be tested from the other factors confounding their effects. Monson _et al_. (1959) also observed variations in duplicate pens in their experiment.

There was no significant difference in feed efficiency when the feed conversion data of all the three experiments were pooled. However, there are indications of improved feed conversion whenever growth stimulation occurred with antibiotic supplementation. These results are in agreement with the findings of Wisman _et al_. (1954) who observed no statistical differences in feed efficiencies in their experiment. Pope _et al_. (1958), Trail and Casley (1960), Menge and Lillie (1960), Potter _et al_. (1962), Stutz (1961) and Siddiqui (1963) also reported statistically nonsignificant differences in feed conversion to antibiotic supplementation. Contrary results are reported by Davis and Briggs (1951), Gard (1958) and Nelson _et al_. (1962).

SUMMARY AND CONCLUSION

A series of three different experiments were conducted to investigate the performance of broiler-strain chicks fed diets supplemented with different kinds, levels and combination of antibiotics. A total of 936 Cobb Strain-cross White Rock straight-run broiler chicks were used in these experiments. The chicks were reared in starter batteries to four weeks of age and then shifted to the grower batteries and kept there till eight weeks of age. Body weights and feed consumption data were recorded at every biweekly interval during the 8-week experiment.

The 1963-64 Kansas State University chick broiler starter (all mash) ration was fed from 0-5 weeks and the Kansas State University broiler finisher from 5-8 weeks. Antibiotics, erythromycin, zinc bacitracin, and tylosin were used as single sources at levels of 10 grams per ton and 20 grams per ton, while their combinations, erythromycin and tylosin, zinc bacitracin and erythromycin, and zinc bacitracin and tylosin at 5 grams per ton each (10 g./ton of combination) and 10 grams per ton each (20 g./ton of combination) were added to the K.S.U. broiler basal.

The statistical analyses were run on the 8-week adjusted weights and feed conversion data and the results tabulated.

The following conclusions were drawn from this study:

1. A combination of low level of zinc bacitracin and tylosin proved superior to any other supplement used in the experiment.

2. The low level combination of erythromycin and tylosin gave the next best rate of growth.

3. A variability in growth response to antibiotics between experiments was observed.

4. Of all the single sources of antibiotics, erythromycin gave more or less a consistent growth response when compared with zinc bacitracin and tylosin.

5. The lower levels of both single sources and combinations of erythromycin, zinc bacitracin, and tylosin indicated better growth response than the higher levels used in this experiment.

6. On an average, the chicks in the supplemented groups weighed more than those in the negative control group.

7. No significant differences in feed conversion were observed between the various diets.

8. Mortality and abnormalities were found to be at a minimum.

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APPENDIX

Table 1. Composition of the 1963-64 K.S.U. broiler chick starter ration used as the basal diet in all experiments. (0-5 weeks of age)

Ingredients	Quantity used per 100 lb. (lb.)
Corn, yellow, ground	30.00
Sorghum grain, ground	31.00
Alfalfa meal, dehydrated, 17% protein	2.00
Soybean oil meal, solvent extracted, 44% protein	29.00
Fish meal, 60% protein	4.00
Soluferm - 500(R)	1.50
Calcium carbonate*	1.00
Dicalcium phosphate*	1.00
Salt (sodium cholide)*	0.50
Total	100.00
Added per 100 lb. of ration	
Trace mineral mix[1]*	23 grams
Vitamin A (10,000 USP units/gram)+	10 "
Vitamin D_3 (15,000 ICU units/gram)+	5 "
B-complex vitamin mix[2]+	46 "
Methionine (Feeding grade)+	23 "
Vitamin B_{12} (12 mg/lb)+	10 "
Choline chloride - 25% mix+	40 "
Amprol(R) (coccidiostat)+	23 "

(R)Registered trademark

*Mineral premix

+Vitamin and additives premix

[1]Trace mineral premix supplying by %: Mn 10; Fe 10; Ca, max. 14, min. 12; Cu 1; Zn 5; I_2 0.3; Co 0.1.

[2]B-complex vitamin mix supplying in mg/lb.: riboflavin 2,000; pantothenic acid 2,680; niacin 6,000; choline chloride 20,000.

Table 2. Composition of the 1963-64 K.S.U. broiler chick finisher ration used as the basal diet in all experiments. (5-8 weeks of age)

Ingredients	Quantity used per 100 lb. (lb.)
Corn, yellow, ground	34.50
Sorghum grain, ground	35.00
Alfalfa meal dehydrated, 17% protein	1.00
Soybean oil meal, solvent extracted, 44% protein	27.00
Calcium carbonate*	1.00
Dicalcium phosphate*	1.00
Salt (sodium chloride)*	0.50
Total	100.00
Added per 100 lb. of ration	
Trace mineral mix[1]*	23 grams
Vitamin A (10,000 USP units/gram)+	10 "
Vitamin D_3 (15,000 ICU units/gram)+	5 "
B-complex vitamin mix[2]+	46 "
Methionine (Feeding grade)+	23 "
Vitamin B_{12} (12 mg/lb)+	10 "
Choline chloride - 25% mix+	40 "
Amprol(R) (coccidiostat)+	23 "

(R)Registered trademark

*Mineral mix

+Vitamin and additives premix

1 Trace mineral premix supplying by %: Mn 10; Fe 10; Ca, max. 14, min. 12; Cu 1; Zn 5; I_2 0.3; Co 0.1.

2 B-complex vitamin mix supplying in mg/lb.: riboflavin 2,000; pantothenic acid 2,680; niacin 6,000; choline chloride 20,000.

Table 3. The levels and kinds of supplements used in all three experiments.

Diet	Lots		Supplements	Level (gm./ton)
1	1 & 2	K.S.U. Broiler basal +	0 suppl.	0
2	3 & 4	K.S.U. Broiler basal +	Erythromycin[1]	10
3	5 & 6	K.S.U. Broiler basal +	Zinc bacitracin[2]	10
4	7 & 8	K.S.U. Broiler basal +	Tylosin[3]	10
5	9 & 10	K.S.U. Broiler basal +	Erythromycin	20
6	11 & 12	K.S.U. Broiler basal +	Zinc bacitracin	20
7	13 & 14	K.S.U. Broiler basal +	Erythromycin	5
			Zinc bacitracin	5
8	15 & 16	K.S.U. Broiler basal +	Erythromycin	5
			Tylosin	5
9	17 & 18	K.S.U. Broiler basal +	Zinc bacitracin	5
			Tylosin	5
10	19 & 20	K.S.U. Broiler basal +	Erythromycin	10
			Zinc bacitracin	10
11	21 & 22	K.S.U. Broiler basal +	Erythromycin	10
			Tylosin	10
12	23 & 24	K.S.U. Broiler basal +	Zinc bacitracin	10
			Tylosin	10

[1] Gallimycin-10(R) a product of Abbott Laboratories, North Chicago, Illinois, supplying 10 grams of drug per pound of supplement.

[2] Baciferm-10(R) a product of Commercial Solvent Corporation, Terre Haute, Indiana, supplying 10 grams of drug per pound of supplement.

[3] Tylan-10(R) a product of Elanco Products Co., a division of Eli Lilly and Co., Indianapolis, Indiana, supplying 10 grams of drug per pound of supplement.

Table 4. Average 8-week weight gains[1] for all lots in three experiments (adjusted for sex[2]).

Diets	Lot No.	Experiment 1	Experiment 2	Experiment 3
1	1	1380	1333	1329
	2	1393	1232	1207
2	3	1332	1351	1327
	4	1342	1358	1317
3	5	1382	1309	1282
	6	1329	1355	1255
4	7	1379	1401	1197
	8	1366	1338	1247
5	9	1370	1368	1354
	10	1227	1356	1232
6	11	1319	1364	1340
	12	1310	1329	1196
7	13	1356	1341	1292
	14	1493	1340	1237
8	15	1352	1339	1381
	16	1432	1401	1290
9	17	1429	1251	1339
	18	1381	1326	1316
10	19	1405	1329	1309
	20	1308	1365	1289
11	21	1356	1313	1244
	22	1473	1296	1275
12	23	1293	1343	1302
	24	1367	1343	1261

[1]Final 8-week weight minus the initial 0-week weight.

[2]Average male weight plus average female weight divided by two.

Table 5. Average 8-week weights for all diets in three experiments (adjusted for sex).[1]

Diets	Experiment 1*	Experiment 2	Experiment 3*	Av. for 3 Experiments	Wt. % of Basal[2]
1	1549	1459	1419	1476	100.0
2	1518	1526	1487	1510	102.3
3	1545	1498	1434	1492	101.1
4	1555	1535	1395	1495	101.3
5	1490	1538	1454	1494	101.2
6	1503	1508	1443	1485	100.6
7	1572	1515	1420	1502	101.8
8	1574	1542	1472	1529	103.6
9	1604	1497	1497	1533	103.9
10	1544	1526	1458	1510	102.3
11	1589	1467	1432	1496	101.4
12	1528	1512	1433	1491	101.0
L.S.D.	66	--	50	--	--

[1] All female weights adjusted to male weight by pooling together the differences of average male weights minus average female weights for all lots and the total average difference was added to each female weight for final adjustment.

*Diets significant at $P < .05$

[2] $$\text{Weight \% of basal} = \frac{\text{Av. weight of treated group}}{\text{Av. weight of basal group}} \times 100$$

Table 6. Analysis of variance of 8-week weights with the data pooled from three experiments (adjusted for sex).[1]

Source of Variation	Degrees of Freedom	Sum of Squares	Mean Square
Experiment	2	1677886	838943**
Diets	11	251125	22830 n.s.
Diet x Experiment	22	547445	24884*
Within	900	12626726	14030
Total	935	15103182	

Table 7. Analysis of variance of 8-week weights with the data pooled from Experiment 1 and 2 (adjusted for sex).[1]

Source of Variation	Degrees of Freedom	Sum of Squares	Mean Square
Experiment	1	206483	206483**
Diets	11	172470	15679 n.s.
Diets x Experiment	11	377671	34334*
Within	600	9487916	15813
Total	623	10244540	

[1]All female weights adjusted to male weight by pooling together the differences of average male weights minus average female weights for all lots and the total average difference was added to each female weight for final adjustment.

**Significant $P < .01$

*Significant $P < .05$

n.s. Nonsignificant

Table 8(a). Analysis of variance of 8-week weights with the data pooled from Experiments 1 and 3 (adjusted for sex).[1]

Source of Variation	Degrees of Freedom	Sum of Squares	Mean Square
Experiment	1	1632524	1632524**
Diets	11	307332	27939*
Diets x Experiment	11	273115	24829 n.s.
Error	600	8271435	13786
Total	623	10484406	

[1]All female weights adjusted to male weight by pooling together the differences of average male weights minus average female weights for all lots and the total average difference was added to each female weight for final adjustment.

**Significant $P < .01$

*Significant $P < .05$

n.s. Nonsignificant

(b) Ranked diets based on L.S.D. method.[1] Showing diets ranked from high to low from pooled 8-week weights (adjusted for sex).

Diets											
9	8	11	2	10	7	3	1	12	4	6	5
1550	1523	1511	1502	1501	1496	1490	1484	1480	1475	1473	1472
———	———	———									
	———	———	———	———	———	———	———	———			
		———	———	———	———	———	———	———	———	———	———

[1]Any two diets not underscored by the same line are significantly different and any two diets underscored by the same line are not significantly different.

L.S.D. = 45 grams.

Table 9(a). Analysis of variance of 8-week weights with the data pooled from Experiments 2 and 3 (adjusted for sex).

Source of Variation	Degrees of Freedom	Sum of Squares	Mean Square
Experiment	1	654947	654947**
Diet	11	274197	24927*
Diet x Experiment	11	177252	16114 n.s.
Within	600	7510698	12518
Total	623	8617094	

**Significant $P < .01$

*Significant $P < .05$

n.s. Nonsignificant

(b) Ranked diets based on L.S.D. method[1] showing diets ranked from high to low for pooled 8-week weights (adjusted for sex).

Diets											
8	2	9	5	10	6	12	7	3	4	11	1
1507	1506	1497	1496	1492	1475	1472	1468	1466	1465	1449	1439

[1]Any two diets not underscored by the same line are significantly different and any two diets underscored by the same line are not significantly different.

L.S.D. = 43 grams.

Table 10(a). Comparisons of levels and kinds of supplements from the data pooled from Experiments 1 and 3 (adjusted for sex).

Diets	1	2	3	4	5	6	7	8	9	10	11	12	
Sum of wts. (gm)	25192	26124	25471	24703	24550	24594	25783	27180	28623	26059	26555	24981	Sum of squares
Basal vs supplement	+11	-1	-1	-1	-1	-1	-1	-1	-1	-1	-1	-1	8218 n.s.
Single vs combinations	0	+2	+2	+2	0	0	-1	-1	-1	-1	-1	-1	61628*
5 g/ton vs 10 g/ton among combinations	0	0	0	0	0	0	-1	-1	-1	+1	+1	+1	51052 n.s.
Others													186434 n.s.
Total													307332

(b) Comparisons of levels and kinds of supplements from the data pooled from Experiments 2 and 3. (adjusted for sex).

Diets	1	2	3	4	5	6	7	8	9	10	11	12	
Sum of wts. (gm)	22839	26315	24229	24188	25791	24725	24311	26350	25834	25596	23367	24569	Sum of squares
Basal vs supplement	+11	-1	-1	-1	-1	-1	-1	-1	-1	-1	-1	-1	84224**
Single vs combinations	0	+2	+2	+2	0	0	-1	-1	-1	-1	-1	-1	339 n.s.
5 g/ton vs 10 g/ton levels among combination	0	0	0	0	0	0	-1	-1	-1	+1	+1	+1	28139 n.s.
Others													158495 n.s.
Total													274197

**Significant $P < .01$ *Significant $P < .05$ n.s. Nonsignificant

Table 11. Feed conversion for all lots in three experiments at the end of the 8-week experimental period.

Diet	Lot Nos.	Experiment 1	Experiment 2	Experiment 3
			lb. feed per lb. gain	
1	1	3.12	2.42	2.53
	2	3.35	2.47	2.53
2	3	3.47	2.40	2.36
	4	3.09	2.32	2.42
3	5	3.42	2.44	2.46
	6	3.34	2.22	2.43
4	7	3.49	2.26	2.40
	8	2.94	2.35	2.61
5	9	3.51	2.31	2.47
	10	3.31	2.13	2.60
6	11	3.08	2.45	2.36
	12	3.16	2.31	2.58
7	13	3.17	2.23	2.52
	14	3.07	2.40	2.47
8	15	3.25	2.38	2.55
	16	3.19	2.44	2.42
9	17	3.21	2.36	2.58
	18	3.03	2.37	2.38
10	19	3.01	2.29	2.32
	20	3.55	2.43	2.33
11	21	3.45	2.46	2.45
	22	3.45	2.42	2.50
12	23	3.09	2.28	2.31
	24	2.87	2.39	2.33

Table 12. Analysis of variance of feed conversion with pooled data of three experiments.

Source of Variation	Degrees of Freedom	Sum of Squares	Mean Square	F-ratio
Replications (Within Experiment)	3	0.04	0.013	< 1
Diets	11	0.24	0.022	1.22 n.s.
Diets x Experiments	22	0.37	0.017	< 1
Within	33	0.61	0.018	

n.s. Nonsignificant

INFLUENCE OF FEEDING VARIOUS KINDS, LEVELS AND COMBINATIONS OF ANTIBIOTICS ON GROWTH AND FEED EFFICIENCY OF BROILER-STRAIN CHICKS

by

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M. Sc. Agri., University of Osmania, India, 1962

AN ABSTRACT OF A MASTER'S THESIS

submitted in partial fulfillment of the

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MASTER OF SCIENCE

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KANSAS STATE UNIVERSITY
Manhattan, Kansas

1965

A series of three different experiments were conducted to study the performance of broiler-strain chicks fed various kinds, levels and combination of the antibiotics, erythromycin, zinc bacitracin, and tylosin. A total of 936 Cobb Strain-cross White Rock straight-run chicks were used in these experiments. The chicks were reared from 0-5 weeks in starter batteries and then transferred to grower batteries and kept there from 5-8 weeks, the end of each experimental period.

The 1963-64 Kansas State University chick broiler starter (all mash) ration was fed from 0-5 weeks and the broiler finisher from 5-8 weeks. Antibiotics, erythromycin, zinc bacitracin, and tylosin were added to the broiler basal as single sources at two levels, 10 grams per ton and 20 grams per ton, respectively. The combination of these antibiotics at 5 grams per ton each (10 g./ton of combination) and 10 grams each (20 g./ton of combination) were used. Eleven supplemented diets and a basal (negative control) were fed to two replicate lots in each of the experiments.

Body weights and feed consumption records were maintained for each biweekly period during the entire 8-week experimental periods. The 8-week adjusted weights were tested by the analysis of variance for each experiment separately as well as pooled for all three and all possible combinations of any two experiments to study the trend of growth stimulation of the supplements. Similarly feed conversion data pooled from all the three experiments were analyzed by analysis of variance.

The following conclusions were drawn from this study:

A low level combination of zinc bacitracin and tylosin proved superior in growth stimulation to any other supplement used in these experiments, while the combination of erythromycin and tylosin was a close second; however, a variability in growth stimulation to antibiotics between experiments was observed.

Among the single sources of antibiotics, erythromycin gave the best consistent growth response in comparison with zinc bacitracin and tylosin between experiments.

The lower levels of both single sources and combination of erythromycin, zinc bacitracin, and tylosin stimulated growth more than their higher levels used in the experiment.

On an average the chicks in supplemented lots weighed more than those in the basal (negative control) lots.

No significant differences in feed conversion were observed among the various diets.