

Physiology, Endocrinology, and Reproduction: Physiology II

109 Effects of in ovo injection of metabolic stimulants on subsequent broiler chick tissue nutrient profiles. M. M. Keralapurath*, E. D. Peebles, R. W. Keirs, D. A. Braasch, L. W. Bennett, S. K. Whitmarsh, and B. M. McGruder, *Mississippi State University, Mississippi State.*

Previous investigations have confirmed the potential effectiveness of various metabolic stimulants and physiological salt solutions in supporting broiler embryogenesis. The current study investigated the effects of the same solutions on chick posthatch performance and tissue nutrient profiles. In the first of 2 trials, 200 μ L each of a (i) 0.18 mM carbohydrate electrolyte nutrient (CEN) solution, (ii) 5 mM tripotassium citrate solution, and (iii) 1 mM creatine solution were tested in conjunction with 5.5 mM potassium chloride. In the second trial, 1.0 mM theophylline was tested as an additive to all the above treatment solutions. In both trials, a sham-injected 117 mM sodium chloride and a noninjected group were used as treatment controls. All the treatment solutions were in ovo-injected on Day 18 of incubation using an AviTech Intellilab™ injector. Chicks were weighed and tissue samples collected on Days 3 and 10 posthatch. Liver nutrient contents (glucose, glycogen, protein, and fat) and blood plasma profiles (refractive index, protein, triglycerides, glucose, and calcium) were also analyzed. The metabolic stimulant solutions used for in ovo injection did not produce any detrimental effects on the general liveability and overall performance of the broiler chicks. However, across all treatments, liver protein and fat concentrations increased and decreased, respectively, between Days 3 and 10 posthatch. Liver glucose concentration was increased by tripotassium citrate and liver moisture content was decreased by CEN. Nevertheless, CEN and sodium chloride increased plasma refractive index and, except for the potassium chloride/creatine solution, liver glycogen content was reduced by all injected solutions.

Key Words: broiler chick, in ovo injection, metabolic stimulants

110 Effect of photoperiod on upper gastrointestinal tract microbial ecology in broiler chickens. S. Dalal*¹, U. Fernando¹, K. Schwean-Lardner¹, B. Laarveld¹, H. L. Classen¹, A. V. Kessel¹, and B. Fancher², ¹*University of Saskatchewan, Saskatoon, SK, Canada,* ²*Aviagen, Huntsville, AL.*

Day-old chickens (n = 6,294) were placed in 1 of 9 rooms (12 pens per room) such that housing density would not exceed 30 kg/m², to investigate the effect of photoperiod on upper gastrointestinal tract microbial ecology. Birds were assigned to 1 of 3 lighting programs, 13 h light:11 h dark (13L), 18 h light:6 h dark (18L), and 23 h light:1 h dark (23L). At day 33, two birds from each pen were randomly killed to determine relative crop weight, pH of contents and microbial colonization. A difference ($P = 0.002$) was observed in the relative crop weight (gm/gm of body weight) for birds in the 13L (0.019) treatment compared with 23L (0.0072). Crop pH was not affected by treatment. Crop microbiota were profiled in contents from 10 chickens in each of the 13L and 23L treatments using terminal restriction fragment length polymorphism (T-RFLP) based on MspI and HaeIII digests of 16s rDNA amplicons. Phylogenetic analysis based on the DICE coefficient of similarity using

HaeIII digests indicated that individuals from the same lighting treatment showed more similarity than individuals from the different treatments. In MspI digested 16s rDNA amplicons, abundance of the 177- and 565-bp terminal restriction fragments (TRFs) corresponding to *Lactobacillus amylovorus* and *Enterococcus/Streptococcus* spp. group, respectively, were increased in crops from 13L chickens. Chickens with increased dark exposure (13L) also had a unique TRF at 574 bp representing the *Lactobacillus casei/bifermentum/salivaris* group. Analysis of HaeIII TRFs corresponded well with MspI TRFs. Increased dark exposure increased the relative size of the crop in the broiler chicken and altered the microbial profile potentially affecting functioning of the crop as a first line of protection against enteric colonization by (zoo)-pathogens.

Key Words: photoperiod, broiler, microbial ecology

111 T-2 toxin further decreases testicular function in Japanese quail selected for exaggerated adrenocortical stress response. J. M. Grizzle*¹ and D. G. Satterlee², ¹*University of Tennessee, Knoxville,* ²*Louisiana State University Agricultural Center, Baton Rouge.*

Trichothecene mycotoxin, T-2 toxin, negatively impacts growth and reproduction in avians. Selection of Japanese quail for exaggerated (high stress, HS) rather than reduced (low stress, LS) plasma corticosterone (B) response to brief restraint delays puberty and reduces cloacal gland size and foam production in adults. An experiment was conducted to determine the effects of dietary T-2 toxin on testicular function and body weight in quail of the HS and LS lines. Male quail (64 HS, 64 LS; 77 d of age) were assigned to 1 of 3 diet treatments containing 0, 8, or 12 ppm T-2 toxin. Data were collected on body weight (BW), cloacal gland area (CAREA), cloacal gland volume (CVOL), and cloacal gland foam (FOAM) production before and after a 28-day feeding period. All birds were euthanized at the end of the trial and their left testis was removed, weighed, and a portion fixed for morphological assessment. With the exception of BW, all pre-experiment measurements were less in HS birds than LS birds ($P \leq 0.05$); HS birds produced less foam (1.9 vs. 2.5 arbitrary units; AU), and had a smaller CAREA (177 vs. 191 mm²) and CVOL (1,614 vs. 1,784 mm³) than LS birds. Following T-2 toxin exposure, by 105 d of age, the stress lines continued to segregate with HS less than LS ($P \leq 0.05$) as follows: CVOL = 1,491 vs. 1,797 mm³, CAREA = 169 vs. 192 mm², and FOAM production = 1.6 vs. 2.7 AU, respectively. Within the LS line, T-2 toxin did not affect BW, CAREA, CVOL, or FOAM ($P \leq 0.05$). However, in HS birds, feeding 12-ppm T-2 toxin significantly reduced ($P \leq 0.05$) CAREA, CVOL, and FOAM as compared to the 0-ppm controls. CAREA was 204, 188, and 181 mm², CVOL was 1,951, 1,728, and 1,637 mm³, and FOAM was 3.2, 2.9, and 1.8 AU for 0, 8, and 12 ppm T-2 toxin-fed birds, respectively. Results indicate T-2 toxin exacerbates negative effects of selection for high B response to stress on male reproductive performance as measured by CAREA, CVOL, and FOAM production. Reduced fertility and reproductive performance may be expected in commercial birds exposed to high stress environments and mycotoxin contaminated feeds.

Key Words: T-2 toxin, corticosterone, Japanese quail

112 Anatomical and physiological changes in hens molted using nonfeed withdrawal. A. L. McDonald*¹, Y. Vizzier-Thaxton², S. Branton³, C. G. Scanes⁴, J. P. Thaxton², S. W. Anderson², and A. Warlick², ¹Cal Maine Foods, Jackson, MS, ²Mississippi State University, Mississippi State, ³USDA South Central Poultry Research Laboratory, Starkville, MS, ⁴University of Wisconsin, Milwaukee.

Forced molting of laying hens has been a controversial practice for years in the United States and abroad. The United Egg Producers industry association does not allow its members who participate in its animal husbandry program to molt using feed withdrawal. Much research exists regarding forced molting, but research regarding nonfeed withdrawal molting is scarce. Therefore, this study is designed to look at the physiological changes laying hens undergo each day during a commercially used nonfeed withdrawal molt. Five (5) birds were separated into 38 groups of 5 birds per pen/treatment. Hens were placed on molt feed in a staggered pattern so that all stages of molt can be compared. At day 38, three (3) birds from each treatment were evaluated for corticosterone levels, cholesterol, blood gases (pH, pCO₂, pO₂), electrolytes (cNA, cK, cCa₂, cCL, ctHb, ctCO₂), hematocrit, body weight, oviduct length and weight, follicle weight, follicle number, atrophic follicle number, ovary weight, liver weight, small intestine length and weight, gallbladder weight, spleen weight, heart weight, gizzard weight, feather data, and blood glucose. Feed data was also collected as was egg production data. Eggs were evaluated for USDA grade, weight, and specific gravity. There were differences depending upon day of treatment as well as differences from controls in the various morphological and chemical parameters measured. Egg quality did not differ nor did feather loss. The purpose of this work was to describe differences in the morphological and chemical progression of molt using industry type nonfeed withdrawal molt which had not been previously reported.

Key Words: molting, nonfeed withdrawal, physiology

113 Cytokine and chemokine gene expression in corticosterone-stressed chickens: A new indicator of stress status? S. Shini*¹, A. Shini¹, and P. Kaiser², ¹University of Queensland, Gatton, QLD, Australia, ²Institute for Animal Health, Compton, Berkshire, United Kingdom.

The expression of cytokines and chemokines in chickens during stress are poorly described and their role in the stress response has not yet been investigated. In this study we examined whether oral administration of corticosterone causes differences in cytokine and chemokine expression and whether these differences are detectable by real-time quantitative reverse transcription PCR. Moreover, we investigated if changes in cytokine and chemokine expression were associated with elevated plasma corticosterone concentrations and heterophil to lymphocyte (H/L) ratios. At 7 wk of age, 120 Hyline laying pullets were randomly divided in 3 groups and exposed for 1 wk to the following treatments in drinking water: corticosterone dissolved in ethanol, ethanol, or untreated water. Whole blood was collected at 1, 3, and 24 h, and 7 days posttreatment and used to measure total and differential leukocyte counts and to isolate heterophils and lymphocytes for RNA extraction. The chickens were then euthanized and the spleen was removed for RNA extraction. In this study we analyzed the expression of IL-1 β , IL-2, IL-4, IL-6, IL-10, IL-12 α , IL-12 β , IL-13, IL-13, IL-18, IFN- γ , TGF- β 4, CCLi1, CCLi2, CCLi3, CCLi4, CXCLi1, CXCLi2, CXCR1, and CXCR4 genes in heterophils, lymphocytes, and splenocytes. Results showed that administration of corticosterone significantly increased plasma corticosterone concentrations and H/L ratios at 1, 3, and 24 h and 10

days posttreatment. Exposure to corticosterone increased expression of chemokines CCLi2, CCLi3, CCLi4, CXCR1, and CXCR4 and cytokines IL-1 β , IL-6, IL-12 α , and IL-18 in heterophils 24 h posttreatment or at the peak of corticosterone and the H/L ratio levels. The expression of cytokine genes in lymphocytes revealed changes mainly in IL-1 β , IL-6, IL-13, and IL-18, 1 h following corticosterone treatments. Cytokine (IL-1 β , IL-6, IL-10, and IL-18) and chemokine mRNA (CXCR1 and CXCR4) expression in splenocytes were up-regulated 24 h post corticosterone treatment. The identification of a cytokine and chemokine profile in stressed chickens could contribute toward an improvement of the assessment of stress.

Key Words: cytokine, chemokine, stress response

114 Solitary and interactive effects of corticotropin-releasing hormone and vasotocin in control of corticosterone in chickens. A. Jurkevich*¹, W. J. Kuenzel¹, F. N. Madison^{1,3}, M. V. Mikhailova², and L. E. Cornett², ¹University of Arkansas, Fayetteville, ²University of Arkansas for Medical Sciences, Little Rock, ³Hope College, Holland, MI.

Arginine vasotocin (AVT) and corticotropin-releasing hormone (CRH) are 2 regulators of glucocorticoid levels and stress responses in birds. Previously we have found that CRH administered intracerebroventricularly (icv) appears to be a more potent stimulator of the hypothalamic-pituitary-adrenal (HPA) axis compared to equimolar amounts of AVT as determined by increased plasma corticosterone (CORT). The aim of this study was to examine the stimulatory effects of AVT and CRH on the HPA axis at the pituitary level to reveal possible molecular interactions between AVT and CRH regulatory pathways. Peripheral (0.5 nmol/kg, i.v.) administration of AVT to 5-week-old male broiler chicks resulted in significant increases of plasma CORT while CRH was less efficient. The combined iv administration of both peptides in subthreshold doses (0.25–0.3 nmol/kg) caused a significantly greater increase of CORT than would be expected by a simple additive effect. Such a nonlinear effect was not observed when both CRH and AVT were concurrently given icv suggesting that corticotropes and not brain mechanisms are involved. Our in vivo data with ligand-induced internalization of vasotocin VT2 or CRH receptors in corticotropes supported this assumption. The underlying mechanism was investigated by expressing CRH-R1 and VT2 receptors in a mammalian cell line. Using FRET analysis, both receptors were found forming heterodimers in the presence of CRH and AVT. Heterodimerization was accompanied by a significant augmentation of cAMP production compared to administration of CRH alone. It is concluded that the relative efficacy of CRH and AVT in stimulating the HPA axis is site-specific and their interaction at the pituitary level may critically contribute to the endocrine output of the HPA system. Supported in part by NSF grant no. IBN 01111006 and NRI grant no. 2005-35203-15850 from USDA, CSREES.

Key Words: stress, pituitary, receptor oligomerization

115 Cloning and characterization of chicken prostaglandin E receptor subtype 3 (EP3) and F receptor (FP). A. H-Y. Kwok*, Y. Wang, C. Y. Wang, and F. C. Leung, University of Hong Kong, Hong Kong, HK-SAR, China.

Belonging to the eicosanoids, prostaglandin E₂ (PGE₂) and F_{2 α} (PGF_{2 α}) are important chemical mediators regulating many vital physiological

processes, from modulation of neurotransmitter release, sodium and water re-absorption to maintenance of pregnancy. Prostaglandin E receptor subtype 3 (EP3) is believed to mainly mediate a decline in intracellular cyclic AMP (cAMP) level via coupling to Gi protein, while prostaglandin F receptor (FP) is believed to affect an increase in intracellular calcium ion level via Gq protein. Though both receptors were identified in mammals and FP in zebrafish, they have yet to be cloned in any avian species. In the present study, using reverse transcription-polymerase chain reaction (RT-PCR), the full-length cDNAs for chicken EP3 and both FP receptor isoforms were cloned from adult chicken ovary. Chicken EP3 is 370 amino acids in length and shows high amino acid identities to that of mammals (human, 57%; mouse, 66%; rat, 68%; dog, 63%; cow, 57%; rabbit, 67%). The full-length cDNAs of FP gene encodes a long and short isoforms of 364 and 258 amino acids, respectively. The long isoform shares a high degree of amino acid identities to that of mammals, including human (80%), mouse (77%), rat (78%), dog (81%), and cow (75%), and a low sequence identity to zebrafish (66%). Both long and short FP isoforms are shown to be expressed in all tissues examined. The tissue distribution profile is underway for EP3. The cloning and characterization of EP3 and FP receptors would help us to establish a molecular basis to elucidate the physiological roles of prostaglandin E₂ and F_{2α} in target tissues including their actions in chicken ovary.

Key Words: chicken, PGE₂, prostaglandin receptors, cloning PGF_{2α}

116 Sexual maturation of blind Smoky Joe chickens is not controlled by photoperiod. J. Perttula* and G. Bedecarrats, *University of Guelph, Guelph, ON, Canada.*

In birds, light can be perceived by retinal and extra retinal photoreceptors, and sexual maturation is generally induced by an increase in photoperiod. Using a genetically blind line of chickens (Smoky Joe), we previously showed that the lack of retina advances egg-laying in hens and sexual development in roosters. However, whether the change in photoperiod was instrumental in inducing maturation in blind birds was not evaluated. Thus, the objective of this study was to determine if the lack of photostimulation interferes with the premature development of blind chickens. 70 hens (43 blind; 37 sighted) and 50 roosters (25 blinds; 25 sighted) were raised from hatch and maintained under 8 h light. For females, egg production was recorded daily and age at first egg was determined. For males, 5 blind and 5 sighted roosters were sacrificed at 14, 17, 19, 21, and 23 weeks of age, and body weight (BW) as well as comb length were measured. In addition, blood and various tissues were collected. Relative testicular weight was then calculated and plasma testosterone concentrations were measured by ELISA. Histology was also performed on testis at 14, 17, and 19 weeks of age. Blind hens laid their first egg 2 weeks before sighted ones and their production remained higher throughout the experiment. No differences in BW were observed between blind and sighted roosters or between collection dates. Comb length significantly increased between 14 and 17 weeks in blind birds while no difference could be observed for the sighted birds. Testes were significantly larger in blind than sighted roosters at 17 and 19 weeks (sighted: $1.7 \pm 0.33\%$ of BW and blind: 4.12 ± 0.73 ; $P < 0.05$). Interestingly, at 23 weeks sighted males had significantly larger testes. In blind roosters, plasma levels of testosterone increased significantly ($P < 0.001$) between 17 and 19 weeks, while there was no differences in sighted ones. Similarly, histology showed advanced spermatzoa development in testes from blind animals.

In conclusion, our study shows that the premature sexual maturation

previously observed in blind chickens occurs independently of photostimulation.

Key Words: photostimulation, reproduction, sexual maturation

117 Identification and characterization of a novel receptor for chicken GHRH and the potential receptor(s) for chicken GHRH-like peptide (GHRH-L) and peptide histidine isoleucine (PHI). Y. Wang, J. Li, C. Y. Wang, and F. C. Leung*, *University of Hong Kong, Hong Kong, HK-SAR, China.*

GHRH and its structurally related peptides, GHRH-like peptide (GHRH-L, also named PRP), PHI, VIP, and PACAP, are demonstrated to play important roles in the pituitary and extra-pituitary tissues of vertebrates; however, the molecular basis of their actions in various tissues of birds remains largely unknown. In this study, 6 highly homologous receptors for GHRH-L (GHRH-LR), PACAP (PAC1), GHRH (GHRHR1 and GHRHR2), VIP and PHI (VPAC1 and VPAC2), respectively, were cloned from chicken brain (or pituitary) and their functionalities were examined in CHO cells using the pGL3-CRE luciferase reporter system. The results showed that: 1) all receptors are coupled to the cAMP-PKA pathway; 2) 2 GHRH receptors, named GHRHR1 and GHRHR2, respectively, are co-existed in the chicken genome and both of them could be activated by chicken GHRH potently and specifically; 3) Interestingly, 2 VIP receptors, namely VPAC1 and VPAC2, could be potently activated not only by chicken VIP and ovine PACAP38, but also by turkey PHI, suggesting that 2 VIP receptors may also function as the potential receptors specific for PHI in birds; and 4) chicken GHRH-L can activate its receptor, GHRH-LR; however, it also activates the 2 chicken GHRH receptors. Using RT-PCR assay, we further examined the expressions of these 6 receptors in adult chicken tissues. Interestingly, the expressions of GHRH-LR, GHRHR1 and GHRHR2 are mainly restricted to the pituitary and/or different brain regions, while PAC1, VPAC1, and VPAC2 are expressed virtually in all tissues examined. Together, our study not only identified a novel receptor for GHRH and the potential receptor(s) for GHRH-L and PHI in chickens, but also establishes a molecular basis to elucidate the physiological roles of these structurally and functionally related peptides in different tissues, including their actions in the pituitary of chicken.

Key Words: GHRH, GHRH-related peptides, GHRH-L, PHI, VIP

118 Use of dietary thyroxin as an alternate molting procedure in turkey breeder hens. V. A. L. Gulde*¹, R. Renema², and G. Y. Bédécarrats¹, ¹*University of Guelph, Guelph, ON, Canada,* ²*University of Alberta, Edmonton, AB, Canada.*

In the poultry industry, forced molting is achieved by reducing photoperiod and withdrawing feed and water for several days. Although this practice is discouraged due to serious health and welfare concerns, it remains the most effective method and alternative strategies need to be established. During initiation of a natural molt, plasma levels of thyroxin (T4) increase significantly. Furthermore, dietary T4 was shown to induce molt in caged chickens. This study was performed to elucidate whether supplemental T4 can induce molt in turkey breeder hens without feed withdrawal/avoidance. Spent hens were randomly divided into 4 groups composed of 5 floor pens (replicates) each (5 hens per pen). While a control group was maintained under a 14 h photope-

riod and fed a breeders' diet throughout, hens from the 3 other groups were supplemented with 40 ppm T4 for 10 days. Of the 3 treatment groups, 1 was maintained under a 14-h light schedule and fed breeders' diet while the 2 others were subjected to a drop in photoperiod to 6 h during or after supplementation, and were then fed a maintenance diet. Egg production, feed intake, body weight, molt and plasma levels of T4 were measured. All treated hens ceased laying within 20 days; however, several individuals spontaneously returned to lay when left on 14 h light, suggesting incomplete involution of the reproductive tract. T4 supplementation significantly reduced feed consumption ($P < 0.001$) and induced significant rapid body weight loss ($P < 0.001$). However, all hens returned to their initial body weight by experiment's end. Most treated hens initiated molt within 8 days of supplementation and all completed molt by day 37. Plasma T4 levels in treated hens increased significantly by day 3 of supplementation ($P < 0.05$) and remained significantly higher than in controls until day 9 ($P < 0.01$). Levels returned to initial values by day 35. In conclusion, dietary supplementation with 40 ppm T4 was successful in inducing molt in spent turkey breeder hens. However, a drop in photoperiod seems necessary to completely reset the reproductive system.

Key Words: molting, turkey, thyroxin

119 Brain AMP-activated protein kinase and food intake in chickens. D. M. Denbow* and P. B. Siegel, *Virginia Tech, Blacksburg.*

AMP-activated protein kinase (AMPK) is an enzyme which monitors energy levels within cells, and possibly at the whole-body level. We investigated whether genetic selection for growth altered the brain's response to changes in AMPK activity. Using chickens from lines selected from a common founder population for either low (LWS) or high (HWS) 8-week body weight, we investigated whether intracerebroventricularly (ICV) injecting compounds known to affect AMPK had a differential effect on food intake. In mammals, Compound C is reported to inhibit whereas 5-amino-4-imidazole carboxamide riboside (AICA) is reported to stimulate AMPK. ICV injection of AICA decreased food intake in 15-week-old HWS and 9-week-old LWS male chickens. In contrast, the ICV injection of Compound C increased food intake in males from both lines. In addition, the ICV injection of 5-aminoimidazole-4-carboxamide riboside (AICAR), which stimulates AMPK, also decreased food intake in LWS males. In rats, stimulating AMPK increased food intake while inhibiting its activity decreased food intake. These results with chickens are in contrast to those observed in rats. The reason for these disparate results remains to be elucidated. (This project was supported by National Research Initiative Competitive Grant no. 2007-35206-17899 from the USDA Cooperative State Research, Education, and Extension Service).

Key Words: food intake, AMPK, chickens

Processing, Products, and Food Safety

120 Sensory descriptive profiles of air and water chilled broiler breast fillets. H. Zhuang*, E. Savage, D. Smith, and M. Berrang, *ARS-USDA, Athens, GA.*

Air chilled chicken products are gaining popularity in the USA. It has been claimed that air chilling (AC) results in improved tenderness and flavor of broiler meat compared with water chilling (WC). However, there is a lack of published sensory study results to support the claims. The objective of this study was to evaluate the effect of carcass chilling methods on sensory texture and flavor descriptive profiles of broiler breast fillets deboned at 4 h postmortem. In each of 4 replications, 27 eviscerated broiler carcasses (6 weeks of age) were collected from a commercial processing line prior to chilling. After transport to the laboratory, one-third of the carcasses were hot-boned (no chill), 1/3 chilled by water immersion (0.3°C, 50 min) and 1/3 chilled by AC method (0.7°C, 150 min). The WC and AC fillets were removed from the bone at 4 h postmortem. Fillets were cooked to an endpoint temperature of 78–80°C. The sensory properties were measured by 7–9 trained panelists using 0–15 point universal intensity scales (21 attributes). Our study shows that the average intensity scores of the 9 flavor attributes analyzed ranged from 0.9 to 4.0 and there were no significant differences between the 3 treatments. The average intensity scores of the 12 texture attributes ranged from 1.5 to 7.5 and there were no significant differences between the AC and WC fillets. The average intensity scores of the texture attributes, springiness, hardness, cohesiveness of mass, bolus size, rate of breakdown, and chewiness were significantly higher in the hot-boned samples than those of either of the chilled samples, which were not different from each other. These results demonstrate that the AC method did not affect the sensory flavor and texture quality of chicken breast meat deboned 4 h postmortem compared to the WC method.

Key Words: chicken breast, air chilling, sensory

121 Effect of multiple washings in salicylic acid on the bacterial flora of the skin of processed broiler chickens. A. Hinton Jr.* and J. A. Cason, *Russell Research Center, Athens, GA.*

Experiments were conducted to determine changes in the bacterial flora of the skin of processed broilers after each of 5 consecutive washings in solutions of the keratolytic agent, salicylic acid. Skin samples from commercially processed broiler carcasses were divided into 3 groups and washed in distilled water, 10% salicylic acid, or 20% salicylic acid by agitating skin in these solutions in a Stomacher laboratory blender. After each wash, skin was transferred to fresh solutions and washing was repeated to provide samples washed 1 to 5 times in each solution. Washed skin was then stomached in Butterfield's Phosphate Buffer to recover viable bacteria remaining on the skin. Bacterial flora of the rinsates was enumerated on Plate Count Agar, *Staphylococcus* Agar, Levine Eosin Methylene Blue Agar, Lactic Acid Bacteria Agar, and *C. perfringens* Agar. Results indicated that after each of 5 consecutive washes in water, there was no significant difference in the number of bacteria recovered from skin on any of the agar media. Significantly fewer bacteria were recovered on Lactic Acid Bacteria Agar from skin after 5 washes in 10% salicylic acid than after 1 wash, but there was no significant decrease in the number of bacteria recovered on any other media after skin was washed in this solution. However, washing skin 4 or 5 times in 20% salicylic acid significantly reduced the number of bacteria recovered on Plate Count Agar and *Staphylococcus* Agar. Furthermore, no bacteria were recovered on Eosin Methylene Blue or Lactic Acid Bacteria Agars from rinsates of skin washed 4 or 5 times in 20% salicylic acid or on *C. perfringens* Agar from skin washed 3 or more times in the 20% solution. Findings indicate that successive washing of skin in salicylic acid may significantly reduce the number of bacteria recovered from the poultry skin, although bacterial popula-