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INDUSTRY SUMMARY. Hen diuresis (excessive urination) syndrome has emerged over the past five years as a significant cause of mortality in the US broiler breeder industry. The condition affects hens in production and is characterized by transient muscle weakness in the vent region, transient diuresis (most commonly occurring in the early to mid-morning), and often urate deposits on the skin below the vent. Affected hens are often seen straining to lay an egg, which suggests uterine contraction, is also inhibited.

Related hen mortality, often reaching one percent or more per week, is believed to be primarily the result of male aggression, which causes trauma of the vent region. When male aggression is not present, mortality is generally lower (~0.5 percent a week) and is the result of E. coli peritonitis infections. The exact association between the cause of mortality and this syndrome is unknown, but may be the consequence of transient partial to full oviduct prolapse. Up to this point, cases have been generally sporadic, although in at least three instances the problem has been widespread within individual complexes. Evidence of the condition at low levels has been identified in many flocks with normal mortality.

Based on work done prior to this study, evidence suggests that the underlying problem is metabolic and related to egg production. This problem is difficult to study because of the difficulty of collecting urine in birds, since birds evacuate feces and urine together from the cloaca. In order to study this syndrome, a reliable method to collect urine from chickens is needed. The ultimate goal of this pilot study was to determine if surgical colostomy of hens would provide a means for collecting urine for analysis.

In this study, colostomy was successfully performed. The colon was surgically separated from the urinary tract and attached to a new surgical opening in the abdominal wall of the birds. Over the course of several days, we collected urine samples from clinically "unaffected" and "affected" birds, which had undergone the colostomy surgery. Analysis of the samples gave an indication of how long after colostomy urine is affected. Based on normalization of urine sodium levels, colostomy surgery appeared to affect urine content for around 32 hours after the surgery. Based on this finding, sample analysis began at this time point. Two urine values were statistically different between "affected" and "unaffected" groups. "Affected" birds had higher sodium and lower potassium urine values compared to "unaffected" birds. In addition, as was expected, urine output was higher in the "affected" group. Our findings support our hypothesis that this syndrome has a metabolic basis.

The information generated from this pilot study can be used in future studies to obtain paired blood and urine samples for detailed research on the characterization and cause of this condition. This information could potentially lead to a preventative strategy or treatment program for this condition and give the industry a better understanding of the metabolic needs of the modern broiler breeder.
MATERIALS AND METHODS

Animals
A total of 20 hens (10 affected and 10 unaffected) were selected from one commercial broiler breeder flock which was reporting birds affected with this syndrome. “Affected” hens were classified as those hens with transient muscle weakness in the vent region and urate deposits (pasting) on the skin below the vent. Transient diuresis was considered to be present if the bird was visibly wet on the skin below the vent. The bird may or may not have had visible urates in the region below the vent. If the bird was not wet but did have visible urates present on the skin below the vent, then it was considered affected but the transient diuresis was not present at that time. For the purpose of this study we selected birds with visible urates on the vent region and did not require the presence of transient diuresis at the time of selection for the bird to be considered affected. Ten "unaffected" birds were also chosen from the same farm. Definition of unaffected was based on the lack of all the previously mentioned clinical signs. Selected birds were transported to the Poultry Research and Diagnostic Laboratory (PRDL, Pearl, MS) in poultry type crates and/or pet carriers. Once at the PRDL, a physical exam was performed on each animal and clinical findings were noted. Weights were obtained for proper pharmaceutical dosing. Birds were then rinsed in warm water and a commercial dishwashing detergent.

Bird Housing
Birds were housed individually in isolator units at the PRDL. Birds were fed the same once daily commercial ration that they were being fed in the field and had access to water at all times. Isolator units were cleaned daily and all birds were observed every 8 hours by laboratory personnel. Birds remained in the isolator units for a total of 8 days.

Surgical Preparation and Colostomy Procedure
The colostomy procedure that was performed was adapted from a previously published protocol described in Manangi, Clark and Coon (2007). Beginning approximately one hour prior to surgery, each bird was administered a dose of 10 mg/kg enrofloxacin (Baytril 100mg/ml) q24h orally for prophylactic prevention of secondary bacterial infections. This medication was continued post-operatively at the same dose. One single dose of butorphanol (1 mg/kg IM) was also administered prior to surgery for analgesia. Each patient was then physically restrained for anesthetic induction via face mask with 5% isoflurane and 2L flow oxygen. Isoflurane gas anesthesia has been shown to have fewer side effects than injectables, so it was the preferred option for anesthetizing the chickens in this study (Greenlees 1990). Once anesthetic induction was achieved, the patient was intubated with a non-cuffed endotracheal tube (4.0 Fr) and maintained with 2.5% isoflurane and a 1.5 L flow oxygen. The surgical area was gently plucked and aseptically prepared using diluted chlorhexidine surgical scrub solution. Birds were then moved to the surgery room. Birds were placed on right lateral recumbency with the left leg placed and secured cranially in order to optimize surgical access. Sterile drapes were placed. One surgical incision was performed on the left caudal flank, paramedially, and approximately 2.5 cm cranial to the cloaca. Skin was incised using a number 15 scalpel blade. The incision was extended through the subcutaneous tissue, abdominal musculature, air sac, and
peritoneum using sharp and blunt dissection. Occasional hemorrhage was controlled by manual compression. Once the coelomic organs were identified, two sterile cotton tipped applicators were placed in the cloaca. The cloaca was lifted at the level of surgical incision allowing identification of the colon. The colon was exteriorized using Debakey atraumatic forceps. A doyen intestinal forceps were then placed at the level of distal colon and at the levels of the cranial coprodeum. Two contralateral hemoclips were placed cranial to the distal doyen intestinal forcep to allow closure of the coprodeum. To the best of our knowledge, this is the first reported use of hemoclips for colostomy in chickens. The distal colon was incised cranial to the hemoclips. Hemorrhage and appropriate closure of the distal colon was assessed prior to intracoelomic replacement of exposed organs. The remaining colon (cranial to the incision) was sutured to the body wall. All suture material used was absorbable, PDS 4-0 (Johnson and Johnson) and suture patterns were everting.

A commercially available colostomy bag ring adapter (Hollister New image ring adapter reference #15204, 57mm (2 1/4”) to 70 mm (2 ¾ “)) was placed around the cloaca. The ring was secured using a nonabsorbable suture nylon (ethicon, Johnson and Johnson) in single continuous and single interrupted suture patterns. The surgical area was coated with silver sulfadiazine cream 1%. A commercially available colostomy bag (Hollister New Image drainable pouch reference #18104, 70mm, 2 ¾ in) was then attached to the ring adapter and a tail clamp was applied to the bag. Once all procedures were complete, anesthesia was discontinued and animals were allowed to recover under supervision.

Post Operative Care
Following surgery, birds were recovered quietly in a holding cage and observed continuously. Once birds were completely recovered, meloxicam 1.5 mg/mL (1 mg/kg PO q24h) was administered for the remainder of the study for pain management. Enrofloxacin (10 mg/kg, PO q24h) was also continued post operatively to minimize risk of secondary bacterial infections. Drug administration was initially performed via syringe, but in order to minimize induced stress associated with manipulation, drugs were administered using a red rubber tube. Birds were observed every 8 hours and records were kept regarding egg production, fecal output, and overall bird condition. Incision sites were kept patent by cleaning the colonic opening using cotton tip swabs and sterile saline, as needed.

Urine Collection
Urine was collected every 8 hours post colostomy via the drainable colostomy pouches. Pouches were removed from the birds at each collection time and urine was drained and measured. Urine volume from each bird was recorded for each collection time. Urine was then submitted for processing and laboratory analysis.

Urine Processing and Analysis
Immediately following collection, whole urine was analyzed at the PRDL using commercially available urine test strips (Pro Advantage Urine Reagent Strip, NDC) to detect glucose, bilirubin, ketones, blood, pH, protein and urobilinogen. Refractometry was used to measure specific gravity. Free/soluble Na+, K+, Ca++, and Mg++ ions all co-precipitate with uric acid and thus must be re-suspended and solubilized into the homogenized urine to obtain an accurate assessment of urinary mineral content (Wideman et al. 1989a,b). To accomplish this, an aliquot of homogenized whole urine was mixed with an equal volume of .5 M LiOH (442410 Sigma-Aldrich Lithium hydroxide reagent grade, 98%) to dissolve uric acid precipitates and liberate trapped minerals and electrolytes. This solubilized urine was used for all subsequent analyses. Urine samples were stored frozen (-20°C) until analysis could be done at the Baptist Health Systems laboratory. Urine was analyzed for calcium, chloride, protein, creatinine, protein/creatinine ratio, glucose, magnesium, phosphorus, potassium, sodium, urea nitrogen, and uric acid using standard Modified Jaffe, Enzymatic, and Spectrophotometric methods. Statistical analyses were then performed on a subset group of all urine parameters obtained to determine if there were differences between "affected" and "unaffected" birds.

Blood Collection and Analysis
At the conclusion of the study and prior to euthanasia, 3mL of whole blood was collected from each bird via the brachial vein. Serum was extracted and submitted to the PRDL for blood chemistries. Chemistry parameters included: glucose; BUN; creatinine; sodium; potassium; chloride; calcium; phosphorus; total protein; albumin; aspartate aminotransferase (AST); alanine aminotransferase (ALT); alkaline phosphatase (ALP); bilirubin; cholesterol; gamma-glutamyltransferase (GGT); creatine kinase (CK); and magnesium. Statistical analyses were performed on all blood chemistry parameters to determine if there were differences between "affected" and "unaffected" birds.

Euthanasia and Necropsy Examination
At the conclusion of the study, all birds were euthanized at the PRDL by lab trained personnel via CO₂ gas. Birds immediately underwent necropsy examination by American College of Poultry Veterinarians (ACPV) board certified poultry pathologists. Special attention was focused on the surgical site, and any lesions presumed to be related.

Tissue Collection and Histopathology
Tissues (liver, heart, pancreas, small intestine, spleen and kidney) were collected from all birds at the time of necropsy examination. Tissue samples were fixed in 10% neutral buffered formalin, and processed routinely for histopathological examination. Paraffin-embedded tissues were sectioned at 4–5 µm and stained with hematoxylin and eosin. Subsequently, von Kossa stains were performed on a kidney section to help determine mineral composition of tissues.

Statistical Methods
Each outcome was assessed using PROC UNIVARIATE in SAS for Windows 9.3 (SAS Institute, Inc., Cary, NC) to determine if the data was normally distributed. If an outcome was determined to be normally distributed, a t-test was conducted using PROC TTEST in
SAS for Windows 9.3 to determine if there was a significant difference in the values for normal and affected birds. If the outcome was not normally distributed, a Wilcoxon Rank Sum test using PROC NPAR1WAY was used to make comparisons between values for normal and affected birds. An alpha level of 0.05 was used to determine statistical significance.

IACUC
Mississippi State University (MSU) complies with the provisions of the Institutional Animal Care and Use Committee as specified by the Animal and Plant Health Inspection Service, USDA in 9 CFR Part 1 (1-91). This project was assigned IACUC #13-036.

RESULTS
Physical Examination
Urates were present around the vent region of all of the "affected" birds, but varied in severity. Six out of ten birds had only minor amount of urates; two had moderate amount of urates; and two birds had heavy amount of urates. Vent tone was also considered. Four of the "affected" ten birds had minimal to no vent tone as determined by digital vent palpation. Four of the ten "affected" birds also had some degree of pecking/cannibalism induced trauma associated with the vent area. Four birds from of each treatment group had palpable hard shelled eggs just inside the vent region.

Anesthesia and Colostomy Surgeries
Colostomy was performed on all ten of the "unaffected" birds and on nine of the "affected" birds. One of the "affected" birds was found dead in the cage prior to surgery. Aspiration pneumonia/tracheal obstruction was suspected, as necropsy examination revealed feed in the trachea and an absence of other gross lesions. All remaining birds underwent anesthesia and surgery without any obvious immediate complications. The pharmaceutical regimen used appeared to provide sufficient analgesia, antiseptic, anesthesia, and antibacterial agents. Recovery from gas anesthesia using 2.5% isoflurane with a 1.5 L flow of oxygen was almost immediate.

Feces Production
Surgical stomas were successfully kept patent and passage of feces was observed for all birds. However, some produced more feces than others.

Urine Collection
Urine collection was easily performed using the ring adapter and drainable pouch. The particular sized adapter and pouch that were used were ideal for both collection of urine and also the removal of any eggs that were laid during the project duration. Pouches were rinsed after each collection, as they became soiled with urates, and were re-attached in most cases. If a pouch was determined to be excessively soiled, it was replaced with a new pouch.

Urine Normalization and Subset Determination
All urine was analyzed as described in the materials and methods. Data was examined and a sample set was selected based on "normalization" of the sodium levels post-
Sodium level was selected because it showed a clear pattern of "normalization" following colostomy. Based on these levels, urine normalization appeared approximately at the fourth collection, or 32 hours post-operatively. Sample collection was initiated at this time, followed by the five immediate subsequent sample collections. A total of six samplings, or two 24 hour periods were included to account for potentially inherent variations due to normal circadian rhythms.

Urine Statistical Analysis
Using results from the selected sample sets, data was analyzed using statistical methods described above. Based on the analysis, "affected" birds had significantly lower potassium levels ($P = 0.0409$) and significantly higher amount of sodium ($P = 0.0377$) compared to the "unaffected" birds. Numerically, but not statistically, there were many differences between the two treatment groups. One worth noting was urine volume. The mean urine volume for the "affected" birds (35.83 mL +/- 14.86) was greater than for "unaffected" birds (25.17 mL +/- 8.91), however, the difference was not significant ($p=0.0716$).

Blood Results and Statistical Analysis
Based on the analysis of our single blood collection from birds in our study, there were no significant differences between "affected" and "unaffected" birds identified using statistical methods described above.

Necropsy Results
All abnormal findings were recorded. Fifty percent (5/10) of the "unaffected" birds and 67% (6/9) of the "affected" birds were sexually inactive (based on the presence of regressed ova) by the end of the trial. It was unclear what the reproductive status was prior to surgery, so this abnormality may or may not have been attributed to the surgery. Forty percent (4/10) "unaffected" and 33% (3/9) "affected" birds had mild to moderate peritonitis. Again, it was unclear how many birds had peritonitis prior to the surgery, so this lesion may or may not be attributed to the surgery. Twenty percent (2/10) "unaffected" and 33% (3/9) "affected" birds had necrotic tissue around the rectal "stump". Necrosis of this tissue was considered normal, as necrosis and atrophy was anticipated following excision of this tissue. Twenty percent (2/10) "unaffected" and 44% (4/9) "affected" birds had an enlarged colon (megacolon). Twenty percent (2/10) of the "unaffected" birds had varying degrees of dehiscence of the surgical stoma site with corresponding fecal contamination of the abdomen. Twenty-two percent (2/9) of the "affected" birds had mild to moderate stricture of the stoma with evident fecal blockage. Eleven percent (1/9) of the "affected" birds had subcutaneous ventral edema and 11% (1/9) had incisional pooling of blood. Ten percent (1/10) "unaffected" and 11% (1/9) "affected" birds had what was described as flaccid hearts. Ten percent (1/10) "unaffected" and 22% (2/9) "affected" birds had either round or cecal worms. The author’s concluded that 47% of birds developed definitive lesions related to surgical complications (dehiscence of surgical site with subsequent fecal contamination; megacolon; ventral subcutaneous edema; incisional pooling of blood; and stricture of stoma with fecal blockage). Lesions associated with the surgical procedures were expected in the birds and it is believed that none of the lesions reported adversely affected the sample analysis.
Histopathology
Kidneys from the ten "unaffected" birds were observed microscopically with the following findings. Four birds were reported as histologically "normal." Five had minimal-mild multifocal lymphocytic interstitial nephritis. Four birds had mild to moderate lymphocytic periureteritis, and one bird had mild to moderate intratubular mineral casts.
As for the nine "affected" birds, the following kidney microscopic changes were reported. Only two birds were said to have "normal" kidneys. Seven had minimal to moderate multifocal lymphocytic interstitial nephritis, and one bird had intravascular bacteria. Three birds had mild to moderate lymphocytic periureteritis. Two birds had intratubular mineralization/mineral casts and another two had multifocal renal tubular mineralization. Mild interstitial fibrosis (n=1), mild-moderate multifocal renal tubular degeneration (n=1), or rare intratubular cellular casts (n=1) were also occasionally detected. Von Kossa stains were subsequently performed on a kidney section with intratubular mineral casts and demonstrated that the mineral deposits were calcium salts.

DISCUSSION
To date there have been no reports in the literature regarding “hen diuresis syndrome” in chickens. An extensive physiological investigation of a broiler breeder diuresis outbreak (1983-1984 eastern US) has been previously reported (Wideman et al. 1989a,b). This previous outbreak is very similar to the cases seen thus far in the southeastern US over the past four years, but the underlying cause is suspected to be different. In that outbreak, Arkansas infectious bronchitis virus (IBV) was strongly suspected to be the underlying inciting cause. Arkansas IBV is still commonly present and may be a contributing factor to this condition, but the current field picture does not suggest that it is the sole contributing factor. In addition to IBV (Condron 2005, Wideman et al. 1989a,b), electrolyte imbalances, mycotoxins (oosporein, ochratoxin, citrinin) (Glahn 1989), and physiological stress have been reported as causes for diuresis. The transient nature of the diuresis, consistent onset in spring/summer season, consistent onset at or near peak production, lack of significant kidney damage, and lengthy duration of the condition once established in a flock indicates toxins and most known infectious agents are unlikely candidates as the inciting cause. Cloacitis, also known as vent gleet, caused by a fungal infection, has been reported to cause wet, urate stained vents. Birds with this condition have been reported to have a foul odor which is not present in the condition under investigation. Although not reported in the literature, it has been observed that impaction of the lower intestine can cause diuresis and urate staining of the vent, as well. The role of dietary electrolyte balance in acid-base balance has been studied by multiple investigators. It has been reviewed primarily in relation to heat stress (Borges 2003a,b, 2007) and general stress (Olanrewaju 2007). Ketone production during starvation has been demonstrated (Hevia 1979). Lactic acidosis has been studied as it related to Fatty Liver Kidney Syndrome (Blanave 1979) in layers and Sudden Death Syndrome in broilers.
Techniques for urine collection in poultry have been described by over 50 authors. Most of these studies were concerned with nutritional studies involving the excretion of nutrients in the urine, and not necessarily a clinical urine analysis. A limiting factor
involving urine studies in chickens is the difficulty of separating urine from feces. This obstacle has been circumvented by different methods including, cannulation, catheterization, or surgical exteriorization of ureters and by using funnels designed to fit into an urodeum (Davis, 1927; Coulson and Hughes, 1930; Hester et al., 1940; Hart and Essex, 1942; Dixon and Wilkinson, 1957; Ainsworth, 1965; Buss et al., 1980 and Wideman 1982) to separate urine from feces. These procedures appear to be useful for short-term collection, but they have side effects such as polyuria, suture breakdown (Fussell, 1969), and fistulation into the cloaca, especially in ureteral exteriorization (Ainsworth, 1965; Tao et al., 1969), in which birds need to wear a harness for sample collection.

Colostomy, a procedure that involves surgically severing the colon just anterior to the rectum and exteriorizing, allows researchers to keep birds for potentially long-term experiments to collect urine and feces separately without the problem of cross-contamination between the two. Various colostomy techniques have been reported in the literature (Rothchild 1947, Dixon 1958, Colvin et al. 1966, Paulson 1969, Fussell 1969, Okumura 1976, Dingle and McNab 1985, Belay et al. 1993, and Jirjis et al. 1997, and Manangi et al. 2007). Complications and side effects are rarely reported in this literature. However, the most consistent complications are recurring blockage, scabbing or closure of the opening, short-term survivability, colon protrusion, and colon withdrawal or retraction into abdomen. Post-op polyuria or diuresis for 3 days after the surgical procedure has been reported (Hester et al. 1940, Colvin et al. 1966, Paulson 1969, and Manangi et al. 2007). Postoperative infection have been reported, but antibiotic therapy for 3-5d minimized infection (Manangi et al. 2007). In our study, enrofloxacin was administered q24 h to accomplish this goal.

In the colostomy technique described by Drs. Manangi, Clark and Coon, 2007, their sample size was 10 broiler breeder hens with a post-operative complication rate of 30%. Based on this information, we selected a sample size of 10 "affected" birds and 10 "unaffected" birds to ensure that the colostomy procedure could be accomplished and urine could be successfully collected in both groups. In addition, the urine analysis parameters that were measured can serve as a foundation for sample size calculations for future studies.

For urine collection, various methods have been used (Ariyoshi and Morimoto 1956, Paulson, 1969, Richardson et al. 1960, Fussell 1969, Belay et al. 1993, and Jirjis et al. 1997). The collection devices mentioned in these studies may only be suitable for short term studies and require frequent removal of urine from the collection fittings to reduce the discomfort of the birds. This may also not be appropriate for urine collection in egg-laying birds such as broiler breeders. Because our study involved broiler breeder hens, it was important to have a proper urine collection system for which both eggs and urine can be trapped. Manangi et al. 2007, used colostomy bags of suitable size for broiler breeder type birds. Bags were placed around the cloaca and the tail of the bag was allowed to rest on the cage floor. Stress was therefore minimized.
Urine evaluation has not been widely reported in the literature due to the difficulty in urine collection; however a review of avian urology is available (Styles 1998). The effects of low pH on uterine contraction have been studied in laying hens (Kupittayanant 2010) and in humans (Pierce 2003).

We attempted to minimize several pitfalls and limitations that were considered prior to conducting this study. One of these included the nature by which diuresis presents itself clinically in broiler breeders. This condition seems to be seasonal (spring & early summer) and it appears to be subjectively more severe in the early morning hours. Therefore, birds with the syndrome are most prevalent in the spring time. For this reason, our study was conducted during late spring/early summer (May), and birds were selected early in the day. Another limitation was the relatively small sample size which may have resulted in insufficient statistical power to detect differences in some outcomes between "unaffected" and "affected" birds. However, the primary goal of this trial was to demonstrate the feasibility of the colostomy technique and to establish relevant criteria for future research focus. Despite the limited sample size, statistically significant differences were detected for two parameters, urinary potassium and sodium levels.

As previously discussed, this pilot study primarily served to answer the major possible pitfall which is related to the colostomy procedure itself as a means of urine collection. Urine collection capabilities to further assess this condition relied upon performing a successful colostomy in the hens. For our purposes, colostomy along with placement of collection bags served as an adequate means for urine collection. In addition, contrary to reports from the literature of possible negative anesthetic responses including tetanic convulsions during recovery, anesthetic complications did not occur in our study. The post-operative complications (dehiscence of surgical site with subsequent fecal contamination; megacolon; ventral subcutaneous edema; incisional pooling of blood; and stricture of stoma with fecal blockage) related to the surgical procedure did not affect the ability of the investigators to collect the urine samples or adversely affect the quality of the urine samples in the time required for this investigation.

In the literature, transient urine findings were described postoperatively. We considered that this may have an effect on the urine samples during the initial days after surgery, as there may need to be a recovery period to allow for normalization of urine flow. Because of this, it was important to establish a time line of when urine appeared to "normalize." The Na+ levels appeared to become stable after around 32 hours after surgery, so that time could be considered a potential time to start collecting urine for future studies. It has also been well established in the literature that colostomy prevents the back flow of urine into the intestinal tract and therefore prevents water absorption that takes place in the ceca and colon of the birds. This may have an effect on urine output in which colostomized birds have more water in the urine compared to non colostomized birds. In order to reduce the impact of this bias, the interpretation of the urine output of "affected" birds was compared to colostomized "unaffected" birds, as well.

CONCLUSION
Our study accomplished the primary goal of determining whether or not colostomy was an adequate method for urine collection in broiler breeders with "hen diuresis syndrome." Despite the small bird numbers used in our study, statistically significant differences were determined between the two groups. At this point, it is unclear why "affected" birds have higher urine Na+ and lower urine K+ levels compared to "unaffected" birds from the same flock. Potassium deficiency can affect muscle contraction, therefore the transient loss of muscle tone may be related. However, the cause of hypokalemia is unclear. Can Na+ or K+ levels be altered to minimize this syndrome in the field? How much of a role do water have on these values? There are many questions that remain and future studies are needed to answer the many questions surrounding this unusual syndrome. In addition, for future studies, paired urine and blood chemistries are preferred, as we feel that only then will researchers be able to fully interpret the effects of the syndrome in today's modern broiler breeder type bird.

References


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LIST OF PUBLICATIONS AND PRESENTATIONS.
This work was presented in two presentations at the 2014 AAAP meeting in Denver, CO on Tuesday, July 29, 2014. Part one was titled "Investigation of Potential Inciting Causes of Hen Diuresis Syndrome" and was presented by Dr. Bradley Turner. Part two was titled "Utilizing Urinalysis Post-Colostomy for Investigating Potential Inciting Causes of Hen Diuresis Syndrome" and was presented by Dr. Kelli Jones. This work has also been submitted (October 2014) for publication consideration to the journal Avian Diseases. Re-prints will be forwarded if this work is accepted for publication.