Petition to Include Acid-Activated Bentonite on the National List of Allowed Substances for use in Organic Livestock Production in consideration of §205.603

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ITEM A

Petition for synthetic non-organic agricultural substance to be included on the National List of allowed substances for use in organic livestock production in consideration of §205.603.

ITEM B

Information concerning substance being petitioned:

1. **Substance name**

   Chemical name: **Acid-activated bentonite (CAS # 98561-46-7)**
   (Trade name: **POULTRY GUARD®**)

   Manufacturing Ingredients:

   As bentonite clay [$Si(Al_{1.4}Mg_{0.3}Fe_{0.}3)O_{10}(OH)_{20}$•1.8Ca]; 50-60% by wt.
   As sulfuric acid [H_2SO_4]; 40-50% by wt.

2. **Manufacturer's name and contact information**

   Trinico-Ag Inc.
   216-1 South Swing Road
   Greensboro, NC 27409
   Ph: Office (336)617-8990
   Email: johnbower@trinicoag.com

3. **Intended use of the substance**

   The primary use of the product (**Acid-activated bentonite**) is to reduce the level of ammonia[1] generated by certain urease producing bacteria commonly found in poultry litter. Additionally, it has been found to reduce populations of darkling beetles[1] and pathogens[2,3] in poultry litter, but no claims for these properties are being made in the present petition. In its finished form the acid-activated bentonite described here is composed of odorless, virtually dustless, free flowing granular particles which are spread over poultry litter by means of manually operated or tractor propelled broadcast spreaders. The rate of addition is typically about 100 lbs./1000 ft^2 of litter surface area (0.8 – 1.6 oz./ft^2). The product is added to the poultry litter only once at the beginning of each new growout cycle.

4. **Livestock**

   The product will be primarily used for poultry operations where ammonia levels are a problem from the standpoint of animal health and performance.

5. **Source and manufacturing procedure**

   The finished product of the present petition (**Acid-activated bentonite, CAS #98561-46-7)** is prepared by treating naturally occurring bentonite clay with sulfuric acid.

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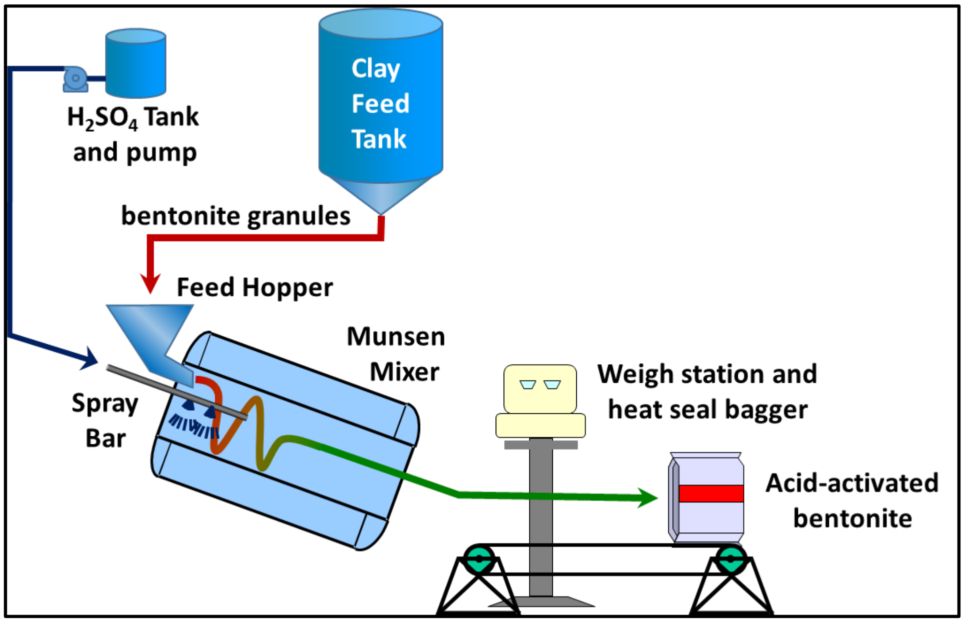
[1] See Appendix, Attachment #1: paper, G.W. McWard, D.R. Taylor, ***Acidified Clay Litter Amendment*****, J. Appl. Poultry Research, 9, 518-529 (2001)**

[2] J.B. Payne, E.C. Kroger, and S.E. Watkins; ***Evaluation of Litter Treatments on Salmonella Recovery from Poultry Litter*****, J. Appl. Poultry Research; 11, 239-243 (2002).**

[3] J.L Vicente, S.E. Higgins, B.M. Hargis and G. Tellez, ***Effect of Poultry Guard Litter Amendment on Horizontal Transmission of Salmonella enteritidis in Broiler Chicks*****, Internat'l J. Poul. Sci., 6, 314-317 (2007).**

The product is manufactured by [**spraying 46 weight percent concentrated sulfuric acid (CAS # 7664-93-9) onto a preweighed bed of bentonite clay granules (CAS #1302-78-9) as they are tumbled in a Munsen mixer (Diagram 1, below). After a short period of mixing, the acid-activated granules are transferred to a bagging line where 50 lb. aliquots are loaded into high melt-temperature plastic bags and heat sealed.]** Bagged product (40 bags) is then loaded onto 40 x 48 inch pallets and stretch wrapped for storage and/or shipment. Petitioner notes that small amounts of crystalline quartz (CAS # 14808-60-7) occur naturally in the bentonite clay used to make the finished product.

Diagram 1. Schematic for Production of Acid-Activated Bentonite.



6. Previous reviews by state or private certification programs

There have been no previous reviews by state or private certification programs regarding this substance. However, this and similar products have been extensively evaluated and recommended for use in reducing ambient ammonia levels in poultry growing operations by various university extension services (see Table 1, below): Univ. Alabama[4,5,6], Univ. Arkansas[7, 8, 9], Auburn Univ.[10], Mississippi State Univ.[11]…

[4] J. Blake, J. Hess, ***Poultry Guard® as a Litter Amendment***, Alabama Cooperative Extension Service, **Publ. ANR-1209** (May, 2001).

[5] J.Blake, J. Hess, ***Aluminum Sulfate as a Litter Amendment***, Alabama Cooperative Extension Service, **Publ. ANR-1202** (April, 2001).

[6] J.Blake, J. Hess, ***Sodium Bisulfate (PLT) as a Litter Amendment***, Alabama Cooperative Extension Service, **Publ. ANR-1208** (May, 2001).

[7] S. Watkins, ***Litter Conditioning for a Healthy Flock***, Univ. Arkansas Cooperative Extension Service, **Avian Advice, 3(2)** (Summer 2001).

[8] S. Watkins, M. Wilson, & J. Comelson, ***Litter Amendments as a Tool for Optimizing Poultry House Clean Out***, Univ. Arkansas Cooperative Extension Service, **Avian Advice, 5(2)** (Summer 2003).

[9] J. Payne, S. Watkins, ***Evaluation of Litter Treaments on Salmonella Recovery in Poultry Litter***, Univ. Arkansas Cooperative Extension Service, **Avian Advice, 7(2)** (Summer 2005).

[10] B. Lott, J. Donald, ***Ammonia Can Cause Serious Losses Even When You Can't Smell It***, Poultry Engineering, Economics & Management Newsletter, Issue 19, Sept. 2002, Auburn Univ. & U.S. Poultry & Egg Assoc.

[11] B. Lott, ***Will ammonia really hurt broiler performance?***, Mississippi State Univ. Extension Service, **Chicken Talk, Info. Sheet 1639** (Feb. 2003)

Univ. Delaware[12], North Carolina State Univ.[13], and Univ. Georgia[14].

Table 1. University Extension Reviews: Acidifying Litter Amendments

Title	Reference No.	Summary
Poultry Guard® as a Litter Amendment	(#4)	Discusses beneficial effects of using acid-activated bentonite, aluminum sulfate, or sodium bisulfate for controlling ammonia levels in poultry raising operations (better bird performance, reduced ammonia-related stress, reduced energy usage, prolonged litter reuse).
Aluminum Sulfate as a Litter Amendment	(#5)	
Sodium Bisulfate (PLT) as a Litter Amendment	(#6)	
Litter Conditioning for a Healthy Flock	(#7)	Discusses acidifying litter amendments and how to use them.
Litter Amendments as a Tool for Optimizing Poultry House Clean Out	(#8)	Presents data showing reduced bacterial, mold and yeast counts on floors of turkey brood houses after treatment with acidifying litter amendments.
Evaluation of Litter Treatments on Salmonella Recovery in Poultry Litter	(#9)	Presents data showing reduction of salmonella levels in poultry litter after treatment with acidifying litter amendments.
Ammonia Can Cause Serious Losses Even When You Can't Smell It	(#10)	Discusses inverse relationship between ammonia levels and bird weights & recommends use of solid litter amendments to reduce ammonia levels
Will ammonia really hurt broiler performance?	(#11)	Discusses relationship between optimum ventilation procedures when using litter amendments for ammonia control.
Litter Amendments: Their Role & Use	(#12)	Discusses best methods for litter management (ventilation, drinkers, and litter amendments).
Poultry Litter Amendments	(#13)	Discusses manufacturers' recommendations on use of acidifying litter amendments as well as additional benefits and some differences between the three types.
Litter Quality and Broiler Performance	(#14)	Discusses the benefits of using acidic litter treatments including: better bird performance, enhanced composition of the litter for fertilizer value, limiting emission of ammonia and odors from poultry houses, & reduced foodborne pathogens

7. Information regarding any regulatory authority registrations

This product has not been registered by EPA, FDA, or any State regulatory agencies. Petitioner is aware of NOP's prior rulings regarding the use of sulfuric acid for the pH adjustment of manure. However, petitioner solicits consideration under **§ 205.603** in the case of this product for the following reasons: (1) acid-activated bentonite is a solid substance which is much easier and safer to use in its intended application than liquid sulfuric acid; compared to concentrated liquid sulfuric acid, this product does not generate noxious fumes or pose a significant health hazard (skin burns) to applicators that would require the wearing of specialized protective equipment; (2) while a drop in pH of poultry litter does occur during initial application of the product, this is not the intended function for the product; the intended function is to reduce ambient ammonia levels in poultry

[12] B. Malone, ***Litter Amendments: Their Role & Use***, Univ. Delaware Cooperative Extension, Fact Sheet (Nov. 2005).
[13] S. Shah, P. Westerman, & J. Parsons, ***Poultry Litter Amendments***, North Carolina Cooperative Extension Service, **Publ. AGW-657** (2/10/2006).
[14] C. Ritz, B. Fairchild, & M. Lacy, ***Litter Quality and Broiler Performance***, Univ. Georgia Extension, **Bull. 1267** (April 2014).

houses for a period of time during which the growing young chicks are most susceptible to the deleterious effects of ammonia on their health and well-being; (3) the use of the product is entirely consistent with NOP objectives per **§ 205.238 Livestock health care practice standards** to reduce stress and promote well-being of animals. As noted in ref. #1: the use of this product results in statistically significant improvements in performance and adjusted feed conversion; better carcass grades; fewer breast blister; fewer foot pad lesions; and better air sac quality (fewer plaques). Finally, (4) use of the product is consistent with EPA goals and regulations aimed at controlling green-house gases (incl. ammonia) because the product sequesters ammonia which then doesn't enter the biosphere as a green-house gas.

8. Chemical Abstract Service number

The finished product as prepared according to Diagram 1 (above) is **Acid-activated bentonite (CAS # 98561-46-7)**. A product label is attached (see **attachment#1: Product Label**).

9. Physical properties and mode of action

Acid-activated bentonite (CAS #98561-46-7) of the present petition is composed of odorless, virtually dustless, free flowing granular particles. Typically, the product has a density of 50-55 lbs/ft^3, pH (5% slurry in DI water) 0.43, granular mesh size (16/48 mesh). Once formed, the probability of environmental contamination is quite low. If spilled, it can simply be swept up and reused. If reuse is not possible, acid-activated bentonite should be disposed of in a secured sanitary landfill[15] or neutralized with lime or sodium bicarbonate.

All three types of sulfate-based poultry litter amendments including acid-activated bentonite (acidified clay) of the present petition initially decrease litter pH during which time the product is removing gaseous ammonia from the ambient air and generating sulfate salts. The acidity initially associated with these materials degrades such that litter pH returns to near neutral values after about 14 days followed by more gradual increase to pH 8.5 typical of normal (untreated) poultry litter. The EPA has concluded that sulfate ions pose no threat to the terrestrial environment[16] or to the aquatic environment so long as the resultant pH of the aquatic environment is in the pH range 6.5-9.0[17].

When used according to manufacturer's directions, there is no known potential for detrimental chemical interactions of acid-activated bentonite with any other materials used in organic poultry farming. Although sulfuric acid is used in the preparation of acid-activated bentonite and sulfuric acid can negatively affect aquatic life at low pH, the subject material of the present petition is applied to poultry litter at very low levels (0.8 – 1.6 oz./ft^2). Over the course of time (i.e. normal time for growout of various poultry raised on poultry litter), any sulfuric acid originally associated with the acid-activated bentonite is degraded to non-acidic sulfate species as indicated by an increase in litter pH over time (Figure 1, below). Since the final litter pH ends up at or near pH 8.5 it would not be expected to cause any negative pH effects on the aquatic environment even in the highly unlikely situation where treated poultry litter ended up being dumped directly into waterways.

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[15] U.S. Dept. of Labor, OSHA, Occupational Health Guideline for Sulfuric Acid, Sept. 1978.
[16] **EPA R.E.D. Facts, *Mineral Acids*, EPA-738-F-93-125, p. 16** (Dec. 1993).
[17] **Ibid. p. 18**

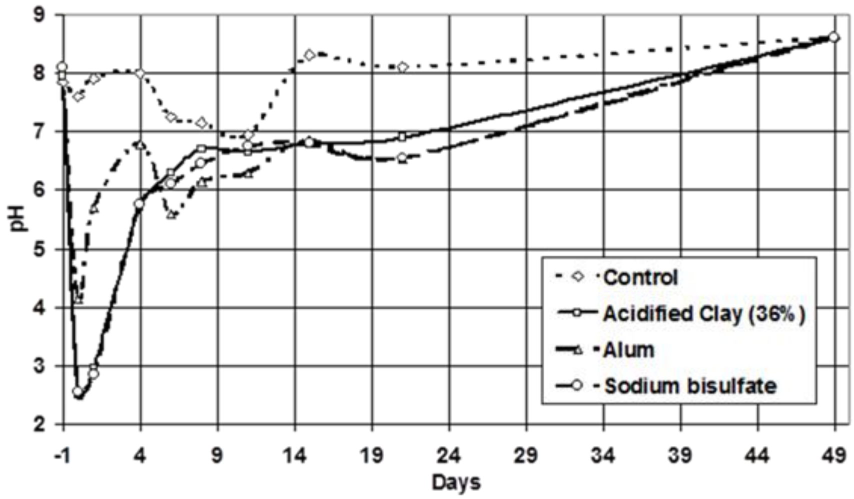

Figure 1. Litter pH over time for acid-activated bentonite (acidified clay). [Taken from attachment #1; paper: G.W. Ward, D.R. Taylor, Acidified Clay Litter Amendment, J. Appl. Poultry Research, 9, 518-529 (2001).]

While the exact nature of the sulfate species being formed in poultry litter is uncertain, the most likely candidate would be ammonium sulfate formed by the reaction between ammonia gas in the environment and sulfate species in the acid-activated bentonite. It would enter the agroecosystem as a component of the poultry litter being applied to crop fields as fertilizer[18]. Because of its low level in the poultry litter (petitioner estimates[19] upper limit of 5% by wt.), it would not be expected to exert a any effect on soil organisms or soil conditions beyond those normally encountered when applying poultry litter to fields other than slightly increasing the nitrogen content of the litter.

Poultry litter that is not burned for fuel is generally applied to crop fields by growers. Growers must utilize a Nutrient Management Program to make sure the phosphorous and nitrogen values do not exceed certain upper limits based on the type of crop they are growing and the current values for the soils in which they are planting.

10. Safety Information/MSDS

An **MSDS** for the petitioned product is attached (**attachment #3**). Petitioner is not aware of any substance report by the National Institute of Environmental Health Studies regarding acid-activated bentonite (CAS # 98561-46-7).

Because acid-activated bentonite is a solid, granular substance that is virtually odorless and dustless, it is relatively safe and easy to use. Granules which might come into contact with the skin or clothing can simply be brushed off. However, it is an acidic substance and care should be taken to ensure granules of the product do not come into contact with the eyes (wear protective glasses or goggles) or remain in contact with the skin (or clothing) for extended periods of time. Eye contact may cause severe irritation or burns. Skin contact may cause irritation or burns. Direct ingestion may cause severe irritation or burns to the mouth, throat and gastrointestinal tract. The product contains some naturally occurring crystalline quartz, a very small fraction of which is in the respirable range. However, testing during the normal use of the product indicates that exposure to respirable crystalline silica during the normal use of the product is well below the OSHA Permissible Exposure Limit (PEL) and ACGIH Threshold Limit Value (TLV) for exposure to crystalline silica.

[18] EPA, Background Report, ***Ammonium Sulfate***, AP-42 Section 6.18 (1996, Modified 10/25/2005),
[19] See Appendix, attachment #4 for math calculations.

11. Research Information

As discussed above (Section 2.3), the primary use for acid-activated bentonite is to reduce ammonia levels in poultry houses containing poultry litter. Figure 2, below shows that ammonia levels are significantly reduced by treating the litter with acid-activated bentonite (here referred to as acidified clay).

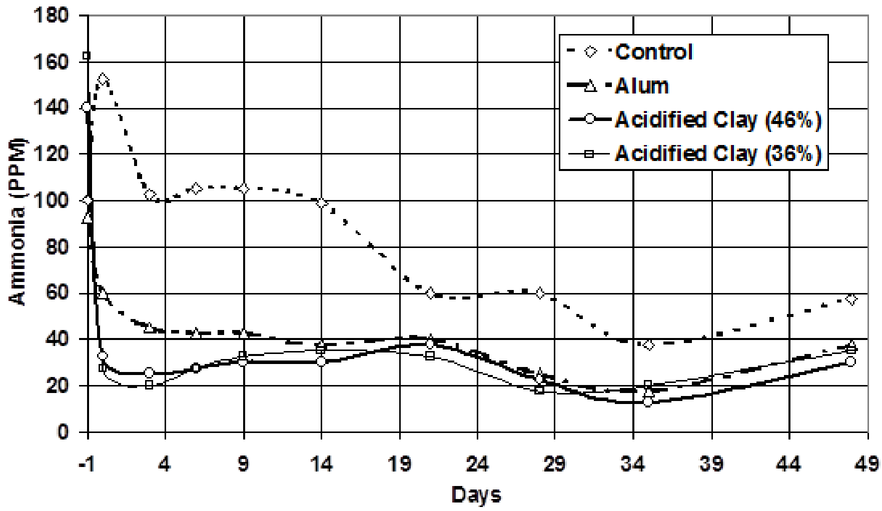

Figure 2: Ammonia levels over time for acid-activated bentonite (acidified clay) at two acid levels, and alum versus untreated control. [Taken from attachment #1; paper: G.W. Ward, D.R. Taylor, Acidified Clay Litter Amendment, **J. Appl. Poultry Research**, **9**, 518-529 (2001).]

Ammonia in poultry operations is well-known to adversely affect bird health and performance[20,21] and economics[22, 10]. The use of acidified litter amendments like acid-activated bentonite to control ammonia production in poultry operations has a number of positive attributes[23] including:

(1) The product is most active during that period of time during which the growing young chicks are most susceptible to the deleterious effects of ammonia on their health and well-being.

(2) The use of the product is entirely consistent with NOP objectives per **§ 205.238 Livestock health care practice standards** to reduce stress and promote well-being of animals. As noted in reference #1 the use of this product results in statistically significant improvements in performance and adjusted feed conversion; better carcass grades; fewer breast blister; fewer foot pad lesions; and better air sac quality (fewer plaques).

(3) The use of the product is consistent with EPA goals and regulations aimed at controlling green-house gases including ammonia because the product sequesters ammonia; accordingly, it does not enter the biosphere as a green-house gas.

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[20] A. Beker, S. Vanhooser, J. Swartzlander, R. Teeter, ***Atmospheric Ammonia Concentration Effects on Broiler Growth and Performance***, **J. Appl. Poultry Res.**, **13, 5-9 (2004)**

[21] T. Rollins, H. Barnes, **http://www.worldpoultry.net/Breeders/Health/2010/10/Harmful-effects-of-ammonia-on-birds-WP008071W/ (Oct. 25, 2010).**

[22] W. Dozier, ***Influence of Ammonia on Broiler Performance***, **The Poultry Informed Professional, 66, 1-2 (Oct. 2002)**

[23] B. Malone, ***Managing Built Up Litter***, Proceedings 2004 Virginia Poultry Health & Management Seminar, April 13, 2004

12. Petition Justification Statement: See below.

12.1. Reasons for inclusion on the National List, §205.603.

Poultry raised on their own litter generate ammonia which is detrimental to their health and performance when levels exceed 25 ppm in their environment. The acid-activated bentonite of the present petition has been shown to reduce ammonia levels thereby insuring a number of concomitant health and well-being benefits (see Section 2.11, above**).**

Although not claimed for the purposes of this petition, the product has also been shown to reduce darkling beetle adult and larvae populations in the litter. Darkling beetles are known vectors for diseases in poultry[24,25] including transmission of *Salmonella*[26]. Besides their role in disease transmission, darkling beetles can also damage structural components in poultry houses[27] thereby increasing producer costs when repairs need to be made. Finally, the product of the present petition has been shown to reduce levels of various *Salmonella* species present in poultry litter[2,3].

Petitioner could not find any current ruling regulating ammonia emissions from poultry houses, but petitioner is aware that the US EPA is currently obtaining information on ammonia emissions from concentrated feeding operations (cattle feed lots and poultry houses) with an eye toward eventual regulation. Because of their ability to reduce ammonia emissions from poultry houses acidifying litter amendments such as acid-activated bentonite can play a positive role in helping to mitigate this problem[28]. Furthermore, as already mentioned, since acid-activated bentonite can reduce *Salmonella* levels in the litter as well as horizontal transmission between chickens, it could be expected to help in reducing instances of *Salmonella* cases from humans eating *Salmonella*-tainted poultry. Finally, as previously discussed, the sulfate species in acid-activated bentonite will be transformed primarily into semi-synthetic ammonium sulfate over the course of time it is in contact with poultry litter. Accordingly, the N content of the litter will have been increased somewhat, and more importantly, transformed into a less volatile form of nitrogen. This means the nitrogen (fertilizer) value of the litter will have been improved while at the same time insuring the sequestered ammonia is in non-volatile form that can be used for plant nutrition rather than being released to the environment as a greenhouse gas (ammonia).

Use of acid-activated bentonite is, therefore, consistent with at least two NOP provisions under **§ 205.238 Livestock health care practice standards**: namely, (1) establishment of appropriate … practices to minimize the occurrence and spread of diseases and parasites; and (2) provision of conditions … [to reduce] stress appropriate to the species (in this case reduction of ammonia levels which are stressful to poultry).

[24] ***Darkling beetle a huge industry disease threat,*** B. Harmon, Feedstuffs, January 9, 2011.

[25] ***Transmission of Enteric Pathogens of Turkeys by Darkling Beetle Larva*, **R.C. Axtell, et al., J. Appl. Poultry Res. 3:61-65 (1994).

[26] ***Transmission of Salmonella to broilers by contaminated larval and adult lesser mealworms*,** N. C. Hinkle, et al., Poultry Science 88:44–48 (2009)

[27] ***Lesser Mealworm, Litter Beetle, Alphitobius diaperinus (Panzer) (Insecta: Coleoptera: Tenebrionidae)*** J. C. Dunford & P.E. Kaufman, EENY-367, Univ. FL, Institute of Food & Agricultural Sciences, revised June 2006.

[28] ***Ammonia Emissions from Agricultural Operations: Livestock***, S. Bittman & R. Mikkelsen , http://www.cacca.org/files/file_gallery/230-ccaarticlebettercropsnh3emissionfinalpdf-2010-09-24-15-270446.pdf

12.2. Alternatives in terms of practices or other available materials:

Two other solid acidifying litter amendments are currently offered for reducing ammonia levels in poultry houses: alum, $Al_2(SO_4)_3 \bullet 15\ H_2O$ and sodium bisulfate, $NaHSO_4$. Neither of these substances is listed under **§ 205.603 Synthetic substances allowed for use in organic livestock production**.

It is well known that increased ventilation can enhance litter conditions as well as bird health and performance[14,15] by helping to keep the litter dry and by removing ammonia; poultry litter amendments are never promoted or recommended as the *sole* means of achieving these conditions but are a viable *adjunct* to achieving these goals. Furthermore, during colder winter months, the use of litter amendments is almost a necessity to help mitigate costs associated with heating ventilation air coming into the poultry houses. Finally, it must be remember that increased ventilation simply removes more ammonia to the outside air where it now becomes a problem as a greenhouse gas. Better that it should be sequestered in the litter as semi-synthetic ammonium sulfate useful for its fertilizing capability.

The NOP stipulates certain conditions for organically grown animals and poultry: (1) feeds must be free of antibiotics or other synthetic substances unless explicitly allowed under § 205.603 and (2) animals must have access to the outdoors which allows for exercise and freedom of movement. However, neither of these provisions by themselves would necessarily reduce (or eliminate) the need for using poultry litter amendments under appropriate conditions. Whether feeds are free of antibiotics or other synthetic substances or not, birds will still excrete urea in their droppings, and urease bacteria in the litter will still generate ammonia as a by-product of its metabolization. Furthermore, although organically grown poultry must have access to the outdoors, the reality is that growing conditions for organically grown and commercially grown broilers are virtually identical. The NOP does not stipulate bird density, so bird densities could be equal. And even though birds grown organically do have access to the outdoors while commercially grown broilers do not, the organically grown birds become acclimated to the indoors and spend very little time outdoors. As a result, birds of either category live out their lives under virtually identical conditions, and would therefore receive the same benefits from the use of poultry litter amendments. Petitioner does note his understanding that “free range” chickens do, in fact, experience different growing conditions that either organic or commercially raised chickens.

Petitioner could find no examples where the use of acidifying litter amendments under appropriate conditions was not recommended.

12.3. Compatibility with a system of sustainable agriculture:

Spent (exhausted) acid-activated clay of the present petition is entirely compatible with sustainable agriculture by virtue of the fact that the substance is transformed during contact with gaseous ammonia into fertilizer (i.e. poultry litter + ammonium sulfate + bentonite clay granules) that can be used to maintain soil fertility. If anything, poultry litter treated with acid-activated clay will have a somewhat higher fertilizer value because of the extra ammonium sulfate it would not otherwise possess. Also, as noted above (Section 2.11), by sequestering ammonia before it escapes to the atmosphere, additional benefit is obtained by the agricultural community which is struggling to control ammonia emissions by any means possible.

The caveat, of course, is that ammonium sulfate is considered a synthetic substance by NOP and therefore prohibited for use in organic farming. Presumably this means any poultry litter containing ammonium sulfate generated by contact with acid-activated bentonite could not be used in organic farming and would have to be restricted to use in “regular” non-organic farming. However, petitioner suggests a possible interpretation that might still allow for use of such litter in organic farming. The reasoning would be: (1) the ammonia generated by the urease bacteria in the poultry litter was organically produced (i.e. – it was not produced synthetically by the Haber process); (2) therefore, the resultant ammonium sulfate in the litter is not a fully synthetic material but rather a semi-synthetic material; (3) the semi-synthetic ammonium sulfate was not added to the litter as a separate ingredient, but rather was generated *in-situ* by reaction between the organically produced ammonia and the acid-activated bentonite in the litter. Therefore, the case could be made that litter containing ammonium sulfate generated by this particular means falls into a gray zone open to a favorable interpretation by NOP.

13. **Confidential Business Information Statement:** This petition does not contain any Confidential Business Information (CBI)

APPENDIX

Attachment #1

G.W. Ward, D.R. Taylor, Acidified Clay Litter Amendment, **J. Appl. Poultry Research**, **9**, 518-529 (2001).

Acidified Clay Litter Amendment

G.W. McWard [1]
Global Poultry Consulting, Inc., Buford, GA 30519
Phone: (770) 932-2054
FAX: (770) 932-4945
D.R. Taylor
Oil-Dri Corporation of America, Vernon Hills, IL 60061
Phone: (847) 634-3090
FAX: (847) 634-4595

Primary Audience: Poultry Producers, Flock Supervisors, Production Managers, Researchers

Summary

Two broiler floor pen experiments lasting 50 and 48 days, respectively, were conducted to compare the performance of a new, granular, acidified clay poultry litter amendment (Poultry Guard^{TM}) and two other commercially available products. In Experiment 1, ammonia levels and litter pH were compared for the acidified clay, sodium bisulfate, and alum versus an untreated control. All three litter treatments reduced ammonia levels for up to 30 days relative to the untreated control. Broiler performance benefits and reduced beetle counts were obtained for all three litter amendments. In Experiment 2, the effect of increasing acid loading of the acidified clay (from 36% to 46% acid) were compared to alum and untreated control. As before, the litter amendments provided benefits (ammonia, litter pH reduction, and broiler performance gains) when compared to the untreated control. The 46% acid loaded clay gave a statistically significant improvement in adjusted feed conversion and breast blister scores but was matched by the 36% acid-loaded clay and alum treatments in most other areas.

Key Words: Acidified clay litter amendment, ammonia control, poultry litter

2000 J. Appl. Poultry Res. 9:518-529

Description of Problem

During the crucial first weeks in the lives of newly hatched chicks, it is important that their physical environment be maintained in such a way that maximum survival rates are achieved. However, the pH, temperature and moisture of litter create an environment that supports rampant bacterial growth. Aside from pathogens which can cause various life-threatening diseases, certain classes of benign bacteria may also be a problem because some bacteria can employ various enzymes to decompose uric acid. Ammonia, a by-product of this process, is subsequently released to the environment.

Without adequate ventilation [1], it is quite possible for ammonia levels to climb as high as 40-70 PPM in the ambient air. At these levels, it ceases to be a mere nuisance and can actually reduce growth performance [2] by impairing the normal respiratory function of the young chicks [3]. Generally speaking, if ammonia levels are kept below about 25 PPM, the adverse effects on the growing chicks are minimized [4]. Although ammonia levels can be controlled by evacuating air from the broodhouses and replacing it with

[1] To whom correspondence should be addressed

clean, fresh outside air, this process can be a problem during the colder winter months. Depending on the age of the chicks, temperatures in the broodhouse must be maintained at 70-85 °F; if too much fresh (cold) air is required to keep ammonia levels down, energy costs (to heat the fresh air) can become prohibitive.

One comprehensive solution to the ammonia problem would be to reduce bacterial activity [5] in the poultry litter. Recently, a number of granular poultry litter amendments have been developed which significantly reduce ammonia levels in broiler houses. Whether they act by reducing bacterial activity in the litter or by chemically binding the ammonia is not known with certainty. Obviously, the economics for using ammonia control products can be extended if they provide additional benefits beyond simply reducing energy costs during colder winter months. More recently, it is becoming apparent that these amendments offer other benefits; they are increasingly being used to alleviate the deleterious effects of high ammonia levels on broiler performance [4, 6, 7, 8]. Such benefits might include improvements in weight gain, feed conversion, carcass quality and health or even reduction in darkling beetle populations. Sodium bisulfate [9], ($NaHSO_4$), alum [10] or aluminum sulfate ($Al_2(SO_4)_3 \bullet x\ H_2O$), and, as discussed in more detail below, acidified clay are examples of currently available ammonia control products.

While specific chemistries vary, the aforementioned amendments share a similar characteristic; they all generate low pH values through the release of sulfuric acid when placed in water. This ability is important because most bacteria cannot grow well under acidic conditions. While diluted liquid phosphoric acid solution has been added directly adding to poultry litter just before new chicks were placed, it did require special equipment in the form of pressure spray systems and trained applicators. For this reason, it was often applied by firms specializing in this type of work. In contrast, granular materials can be conveniently applied by hand or properly calibrated mechanically propelled push cart, tractor-mounted spinner, or drop-type fertilizer spreaders. Application rates for these products depend somewhat on house condition but generally they fall in the range 50 to 100 lbs./ 1000 ft². Taking into account manufacturers' recommend application rates and pricing ($220 - $460 /ton), treatment costs are calculated to fall in the range of $11 - $16 /1000 ft².

Materials and Methods

Testing for these experiments was carried out at the Colorado Quality Research, Inc. facility (Wellington, Colorado) under the direction of Dr. Carey L. Quarles. The environmentally controlled facility was divided into four identical chambers of 10 pens each. Pens had concrete floors and measured 42 square feet. The litter used in these experiments had been used for at least four prior flocks; the average layout time between flocks was five to seven days. All litter was blended together prior to placement in the chambers. The litter was not top-dressed prior to use. In order to prevent bird migration, each pen was checked to assure no openings greater than 1 inch existed for approximately 18 inches in height between pens. Each chamber was separated by floor-to-ceiling partitions and had separate air exhaust/heating systems. The ventilation and environmental conditions (except for ammonia concentrations) were identical for each of the four chambers. All treatments were assigned 10 pens each with 60 birds/pen (600 birds/treatment; ~ 0.7 square feet per bird). Treatments were assigned to chambers within the house using a randomized block design. This design has a limitation regarding replicate allocations. Individually environmentally controlled chambers per replicate would have allowed for a more accurate determination of differences between litter amendments; however, this environment was not possible with this facility.

The analyses of materials used in these studies are appears in Table 1.

Experiment 1: This study was conducted to determine whether or not broilers grown on litter treated with the acidified clay product exhibited improved performance (body weights and feed efficiency) and carcass quality when compared to broilers grown on untreated litter or litter treated with two different litter treatment products (alum and sodium bisulfate). This experiment also attempted to determine the difference in ammonia and litter pH levels at various intervals prior to and after application of the litter treatments. Finally, Experiment 1 sought to find

TABLE 1. Analysis of study materials

PROPERTY	ACIDIFIED CLAY (36%)	ACIDIFIED CLAY (46%)	ALUM	SODIUM BISULFATE
Chemical type	Acidified clay		Acidic salts	
pH (5% slurry)	0.33 - 1.1	0.43	2.78	1.47
Acid Strength[A]	33.3%	40.2%	37.1%	40.2%
Bulk Density[B]	50.4 - 50.7	55.3	60.7	84.8
Application Rate[C] (pounds/1000 ft²)	112	93	100	93

[A] Titration of a 1% solution to phenolphthalein endpoint with 0.1 N NaOH.
[B] ASTM Method E727-91.
[C] For both experiments, litter treatments were applied at a rate calculated to deliver 37 pounds H_2SO_4 / 1000 ft² of floor space.

out whether darkling beetle counts were affected by these treatments.

Acidified clay (Poultry Guard™ [11]) was prepared by adding 36% by weight concentrated sulfuric acid to Oil-Dri LVM attapulgite/montmorillonite clay granules. New, unopened 50-lb. bags of alum [12]; and sodium bisulfate [13] were obtained from the field and used as received. All litter treatments were applied by broadcast spreader to the top of the litter one day prior to the receipt of chicks at the test facility. The alum product was lightly raked into the litter at the time of application; the sodium bisulfate product was lightly sprayed with water after application.

Experiment 2: This study was conducted to determine if broilers grown on litter treated with acidified clay product which had been impregnated with different levels of concentrated sulfuric acid (36% and 46% by weight, respectively) exhibited improved performance (body weights and feed efficiency) and improved carcass quality as compared to broilers grown on untreated litter or litter treated with alum. This experiment also attempted to measure the difference in ammonia and litter pH levels at various intervals prior to and after application of the litter treatments. The acidified clay products were prepared in the laboratory by adding 36% or 46% by weight concentrated sulfuric acid to Oil-Dri LVM attapulgite/montmorillonite clay granules [11]. A new, unopened 50-lb. bag of alum [12] was obtained from the field and used as received. The acidified clay products (36% and 46%) were applied immediately prior to chick placement; alum was applied five days prior to chick placement and lightly raked into the litter immediately after application.

Production Practices and Measurements

For Experiment 1, straight run day-old chicks (Cobb x Hubbard) were obtained from a commercial hatchery. For Experiment 2, straight run day-old chicks (Cobb x Cobb) were obtained from the CQR Research Hatchery. Birds were vaccinated for Newcastle and Infectious Bronchitis (NCB) at approximately 14 days of age via the drinking water. No other vaccinations or treatments, except the coccidiostat, were administered during the study.

Water was provided *ad libitum* throughout the study via one hanging, ~14-inch diameter automatic bell drinker per pen. For approximately the first 4 d of age, a floor-placed gallon drinker was also placed in each pen. Drinkers were checked twice daily and cleaned as needed to assure a clean water supply to birds at all times. Feed was provided *ad libitum* throughout the study via two hanging, ~17-inch diameter tube feeders per pen. A chick feeder tray was also placed in each pen for approximately the first 4 d. All feed added and removed from pens was weighed and recorded. The starter diets were fed from days ~0-18; the grower diets were fed from days ~18 - 42; and the finisher diets were fed from days ~42 - 49. All diet changes were conducted at the same time for all groups.

Lighting followed a commercial program. Ventilation and heat were provided, and adjusted as necessary, to maintain bird comfort, keeping in mind that high ammonia levels were desirable. These housing conditions simulated field conditions. For both experiments, litter treatments were applied at a rate calculated to deliver 37 lbs. of sulfuric acid / 1000 square feet of floor space (see Table 1).

Ammonia Measurements: For both experiments, ammonia levels in the air at bird level were measured in two pens of each chamber (8 measurements / interval) at 10 different intervals. Ammonia levels were measured using a Matheson Toxic Gas Detector, Model 8014KA and Precision Gas Detector Tubes #105SC (5-260 ppm). Ammonia levels measured after heating the chambers but with minimal ventilation prior to chick placement ranged between 75 and 120 ppm.

Litter pH Measurements: Samples were collected using a special scoop and template that only allowed litter to a depth of 1.9 cm ± 0.635 cm to be collected. Approximately 200 g samples were obtained and the pH determined (Fisher Accumet pH meter; slurry of ~ 30 grams of litter and equal volume of DI water) on the same day the sample was collected. The samples were collected from two pens for each treatment, and the same pens were sampled at each collection interval. At each collection interval, 3 samples/ pen were obtained: one each at diagonally opposite corners of the pen, and one at the center of the pen. These samples were then combined and homogenized before taking a pH reading. Ten collection intervals were employed for Experiment 1 and eight collection intervals were employed for Experiment 2.

Darkling Beetle Counts: Darkling beetle counts were conducted only in Experiment 1. On days 12 and 34 sampling was conducted over a 24-hour period to determine the relative number of adult and larval stage beetles present in each pen. One beetle trapping chamber (rigid polyvinylchloride (pvc) pipe 15.2 cm long x 1.9 cm in diameter containing rolled corrugated paper) was placed in the litter under the feeder in each pen and then removed ~24 hours later. The contents from each chamber were then placed in individual bags in a freezer until the counts were conducted. The number of adult and larval stage beetles were counted and recorded for each pen.

Clinical Observations: At 7 d following vaccination for NewCastle disease and bronchitis and again at the conclusion of the studies, 6 birds per pen were removed and necropsied to score the thoracic air sacs for lesions. The scoring system used to score air sac lesions was as follows:

0 = Clear 1 = Cloudy 2 = Plaque formation
3 = Severe plaque formation

Mortality Data: All mortalities/removals from Day 0 to study end were recorded and necropsied to determine probable cause of death. Birds unable to get to feed and water were culled to relieve suffering at the discretion of the investtigator. Bird weight, day of removal, reason for culling, and necropsy findings (including sex) of all dead and culled birds was recorded on pen mortality and necropsy records. If a decomposed bird or skeleton was discovered, the weight was not recorded and a necropsy was not conducted.

Body Weights and Feed Conversion: Birds were weighed by pen and sex on day 50 (Experiment 1) or day 48 (Experiment 2). Pens were selected and weighed in successive order by chamber. Performance data was summarized by average weight of males and females. The average feed conversion was calculated using the total feed consumption in a pen divided by the total weight of surviving birds. Adjusted feed conversion was calculated using the total feed consumption in a pen divided by the total weight of surviving birds and weight of birds that died or were removed from that pen.

Processing Data: After the final weights were obtained, all birds in 4 pens for each treatment were processed (661 birds for Experiment 1; 535 birds for Experiment 2). Processing was conducted following the day final weights were obtained. Processing data included the following:

- Foot pad scores:
  0 = normal (no burn, scab or lesion)
  1 = Pad Burn (dermis only)
  2 = Pad Scab (healing) on one or both feet
  3 = Pad Lesion (open sore) on one or both feet

- Carcass grade (individual) — USDA Grades A, B, C or condemned

- Breast blister scores: (individual) —
  0 = none present
  1 = small (< ¼ inch)
  2 = large (> ¼ inch)

*Statistical Analysis***:** All data were analyzed using the one-way ANOVA procedure [14]. Percentage data were subjected to arsine transformation [15] prior to analysis. Differences among treatment means were determined with Duncan's multiple range test and considered significantly different when $p < 0.05$.

Results and Discussion

*Ammonia Reduction***:** In Experiment 1 (Figure 1) ammonia levels for the pens treated with the acidified clay, alum and sodium bisulfate litter amendments were clearly reduced relative to the (untreated) control pens. As shown, all three litter amendments provide control for about 30 days after which time they begin to lose efficacy.

In Experiment 2 (Figure 2), similar reductions in ammonia levels were achieved when using 36% and 46% acid dosage clay and alum. Although it appears the 36% acid dosage product performed as well as the 46% acid dosage product, it must be kept in mind that litter amendments were applied on an acid equivalency basis (i.e.- they were all applied at a rate theoretically calculated to deliver 37 pounds of H_2SO_4 / 1000 ft² floor space). Accordingly, less of the 46% acid dosage product was applied than the 36% dosage product (93 lbs. versus 112 lbs., respectively) because the concentration of acid in the former was about 20% more than in the latter.

*Litter pH***:** Figures 3 and 4 show the effect of the litter amendment products on litter pH. Since all are relatively acidic in the presence of water, they cause a large drop in litter surface pH when initially applied. In Experiment 1 (Figure 3), initial litter values near pH 8 were reduced to the pH 2-4 range after application of the litter amendments. Because the acidified clay and the sodium bisulfate are more acidic (Table 1), they apparently cause larger initial pH drops than the alum. Litter pH values rebound to near-neutral levels within 5 to 7 d after application. However, litter pH for the treated litters is lower (i.e. – more acidic) than the untreated control for at least the first 21 d of the study.

In Experiment 2 (Figure 4), the litter had a higher initial value (pH 9.3) to begin with. As before, both the 36% and the 46% acidified clay formulations cause large initial drops in the litter pH that then rebound after 4 or 5 d. In this experiment, the reduction of litter pH by the alum was not as great as in the preceding experiment. This finding may result from the alum applied

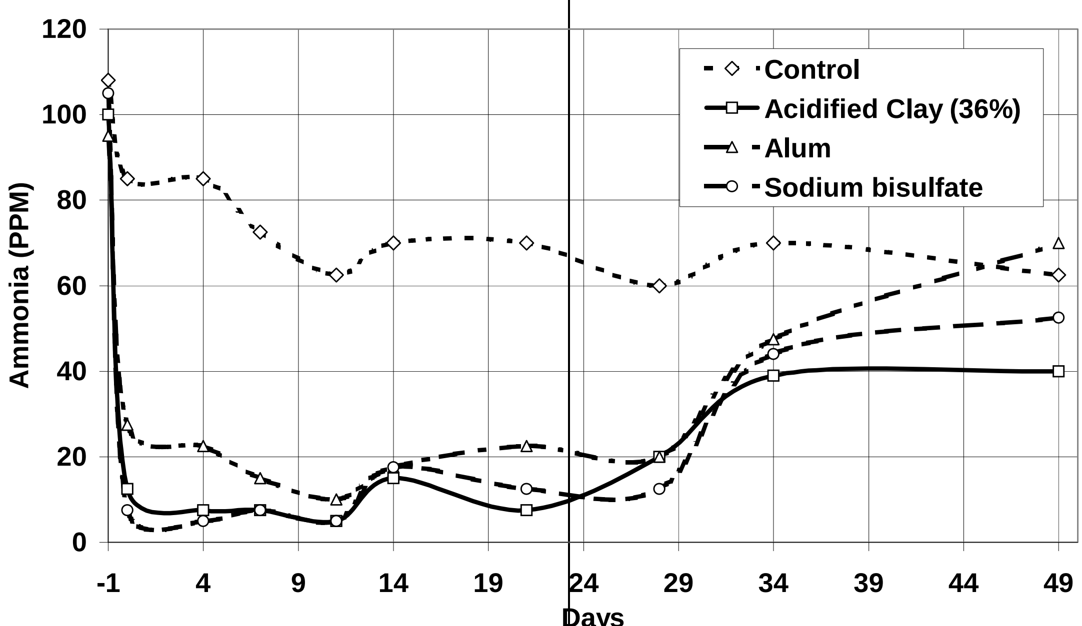

FIGURE 1. Experiment 1: Ammonia levels over time for acidified clay, alum and sodium bisulfate versus untreated control.

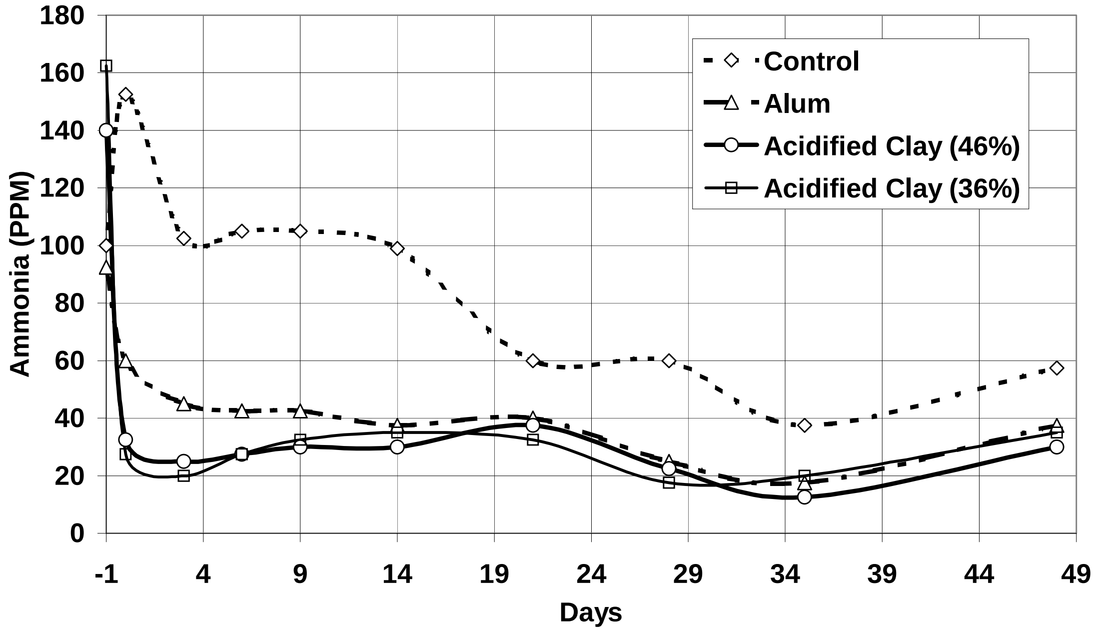

FIGURE 2. Experiment 2: Ammonia levels over time for acidified clay at two acid levels, and alum versus untreated control.

being 5 days prior to bird placement in this experiment, but only 1 day prior to bird placement in the previous. It can be speculated that the initial drop in litter pH may have already rebounded by the time we began taking pH measurements on the day prior to bird placement.

Since all three products supply acidic protons to the environment (in the form of H^+ or

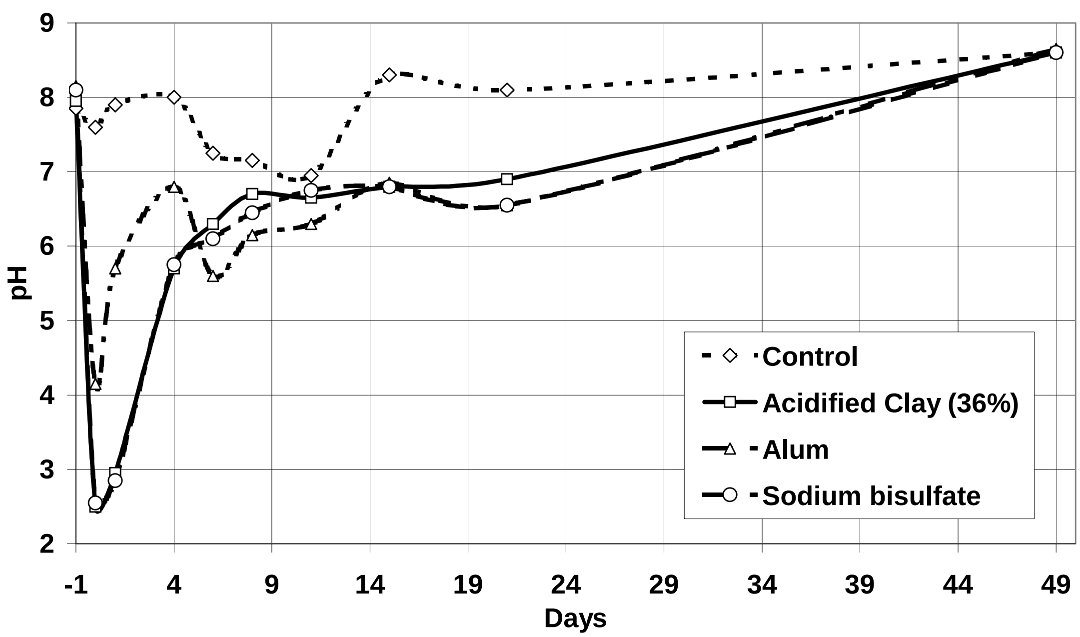

FIGURE 3. Experiment 1: Litter pH over time for acidified clay, alum and sodium bisulfate versus untreated control.

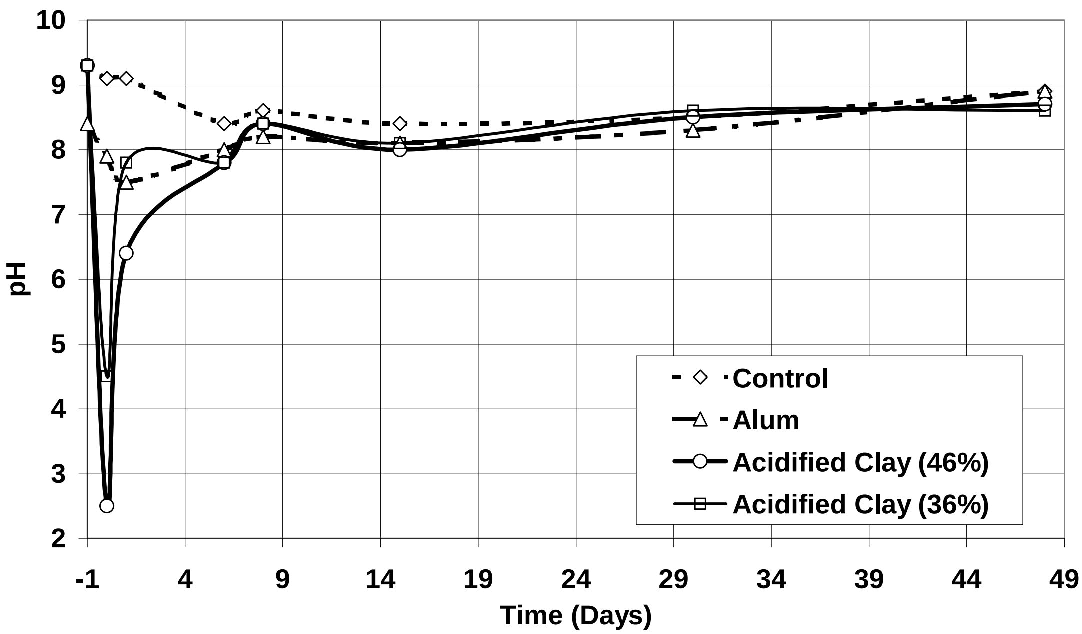

FIGURE 4. Experiment 2: Litter pH over time for acidified clay at two acid levels, and alum versus untreated control.

H_3O^+), reaction with gaseous ammonia (NH_3) to form nonvolatile ammonium cations (NH_4^+) is undoubtedly one mechanism by which ammonia is removed from the environment. However, as is clear from an examination of the graphs in Figures 1 vs 3, and Figures 2 vs 4, ammonia reduction does not directly correspond with reduction in litter pH. The effect of these litter amendments on ammonia levels lasts well beyond the time when litter pH values have nearly returned to pre-application levels.

This finding suggests that pH reduction, while it undoubtedly plays a key role in controlling ammonia levels in broiler houses via ammonia neutralization, may not be the sole mechanism operative in this process. These agents do share another characteristic: all release sulfate ions to the environment. It can be speculated that this sulfate load may, in some way, interfere with the metabolic processes of the bacteria in the litter thereby reducing their ability to generate ammonia.

Broiler Performance and Carcass Quality: Data from Experiment 1 comparing the performance and carcass quality of broilers raised over (untreated) control litter versus broilers raised over litter treated with the various litter amendments are contained in Tables 2 through 6. Performance (average weight, Table 2, column 2) was significantly improved for birds raised over both the acidified clay and sodium bisulfate products relative to the control group; however, birds raised over the alum were not significantly affected. Feed conversion for broilers raised over the litter amendments adjusted only for mortalities were not significantly different from the control group. However, a significant improvement in feed conversion was obtained for birds raised over the acidified clay when adjusted to take into account both mortalities and the heavier weights of these birds (Table 2, column 4).

Carcass quality parameters (carcass grade, breast blister scores and foot pad scores; Tables 3, 4, 5, respectively) were significantly improved for birds raised over the acidified clay amendment. Carcass grade was also significantly improved by both alum and sodium bisulfate, however, the other carcass quality parameters were unaffected. Both acidified clay and sodium bisulfate significantly improved air sac scores (Table 6) but alum did not.

In Experiment 2, weight gain (Table 7, column 2) and feed conversions for broilers raised over the acidified clay products and alum were significantly better than the control group,

TABLE 2. Experiment 1: Effect of litter treatments on performance[A].

TREAT No.[B]	AVERAGE WEIGHT (kg)	ADJUSTED FEED CONVERSION	
	Males & females	Mortality[C]	Wt. & mortality[D]
1	2.606 ^{b} ± 0.128	1.819 ^{a} ± 0.018	1.819 ^{a} ± 0.018
2	2.7437 ^{a} ± 0.060	1.836 ^{a} ± 0.034	1.776 ^{b} ± 0.034
3	2.665 ^{ab} ± 0.077	1.846 ^{a} ± 0.035	1.820 ^{a} ± 0.035
4	2.704 ^{a} ± 0.067	1.839 ^{a} ± 0.024	1.796 ^{ab} ± 0.024

[A] Means in a column that do not have common superscripts are significantly different (P<.05). Means with the same letter are not significantly different.
[B] Treatment 1 = control; 2 = acidified clay; 3 = alum; 4 = sodium bisulfate.
[C] Feed conversion adjusted for mortality calculated using the total feed consumption / pen divided by total weight of surviving birds as well as birds that died or were removed from that pen.
[D] For treatments using litter amendments, feed conversions were adjusted to a constant body weight (equal to that of the control birds). In order to adjust the feed conversion to a constant body weight, we used a factor of 5 points body weight = 1 point of feed conversion.

TABLE 3. Experiment 1: Effect of litter treatments on carcass quality[A]

TREAT No.[C]	CARCASS GRADE[B] (fraction in category)			
	A	B	C	Condemnations
1	0.598 ^{c} ± 0.082	0.331 ^{a} ± 0.106	0.018 ^{a} ± 0.022	0.053 ^{a} ± 0.067
2	0.867 ^{a} ± 0.022	0.102 ^{b} ± 0.029	0.000 ^{a} ± 0.000	0.031^{a} ± 0.032
3	0.749 ^{b} ± 0.103	0.198 ^{b} ± 0.084	0.006 ^{a} ± 0.012	0.047 ^{a} ± 0.033
4	0.808 ^{ab} ± 0.053	0.139 ^{b} ± 0.065	0.00 ^{a} ± 0.00	0.05 ^{a} ± 0.03

[A] Means in a column that do not have common superscripts are significantly different (P<.05).
[B] USDA grades.
[C] Treatment 1 = control; 2 = acidified clay; 3 = alum; 4 = sodium bisulfate.

TABLE 4. Experiment 1. Effect of litter treatments on breast blister scores[A]

TREAT No.[B]	BREAST BLISTER SCORES[C] (fraction in category)		
	0	1	2
1	0.750 ^{b} + 0.063	0.148 ^{a} + 0.056	0.049 ^{a} + 0.019
2	0.921 ^{a} + 0.037	0.049 ^{b} + 0.028	0.00 ^{b} + 0.000
3	0.831 ^{ab} + 0.106	0.106 ^{ab} + 0.088	0.017 ^{b} + 0.022
4	0.833 ^{ab} + 0.024	0.095 ^{ab} + 0.042	0.019 ^{b} + 0.023

[A] Means in a column that do not have common superscripts are significantly different (P<.05).
[B] Treatment 1 = control; 2 = acidified clay; 3 = alum; 4 = sodium bisulfate.
[C] Scoring: 0 = none present, 1 = small (<¼ inch), 2 = large (>¼ inch).

TABLE 5. Experiment 1: Effect of litter treatments on foot pad quality[A]

TREAT No.[B]	FOOT PAD SCORE[C] (fraction in category)			
	0	1	2	3
1	0.319 ^{b} ± 0.299	0.315 ^{a} ± 0.117	0.300 ^{a} ± 0.191	0.066 ^{a} ± 0.071
2	0.713 ^{a} ± 0.127	0.169 ^{a} ± 0.119	0.105 ^{a} ± 0.067	0.013 ^{a} ± 0.025
3	0.511 ^{ab} ± 0.153	0.209 ^{a} ± 0.131	0.244 ^{a} ± 0.108	0.035 ^{a} ± 0.014
4	0.403 ^{b} ± 0.119	0.194 ^{a} ± 0.062	0.311 ^{a} ± 0.119	0.092 ^{a}± 0.062

[A] Means in a column that do not have common superscripts are significantly different (P<.05).
[B] Treatment 1 = control; 2 = acidified clay; 3 = alum; 4 = sodium bisulfate.
[C] Scoring: 0 = normal (no burn, scab, or lesion), 1 = pad burn (dermis only), 2 = pad scab (healing) on one or both feet, 3 = pad lesion (open sore) on one or both feet.

TABLE 6. Experiment 1: Effect of litter treatments on air sac scores[A]

TREAT No.[B]	AIR SAC SCORES[C] (Day 49)
1	0.983 [a] ± 0.748
2	0.500 [c] ± 0.569
3	0.862 [ab] ± 0.634
4	0.684 [bc] ± 0.659

[A] Means in a column that do not have common superscripts are significantly different ($P < 0.05$).
[B] Treatment 1 = control; 2 = acidified clay; 3 = alum; 4 = sodium bisulfate.
[C] Average scores of 60 birds per treatment. Scoring: 0 = Clear, 1 = Cloudy, 2 = Plaque formation, 3 = Severe plaque formation.

whether adjusted only for mortalities (column 3) or when both mortalities and heavier bird weights were taken into account (column 4). Moreover, birds raised over the 46% acidified clay product (treatment 3) exhibited significantly better feed conversions than those raised over the alum (treatment 2) or 36% acidified clay product (treatment 4). The significantly better performance by alum in this experiment contrasts with the results of the previous experiment. This may be related to the fact that in this experiment, the alum was applied a full five days prior to chick placement (according to manufacturer's recommendations) whereas in the previous experiment, it was applied only one day prior to chick placement.

While some carcass quality parameters (i.e. carcass grade, Table 8) were significantly improved for broilers grown over the litter amendments, others (breast blister scores, Table 9; foot pad scores, Table 10) were not or were, at best, only marginally improved. In seeking to explain the lack of breast blister and foot pad differentiation between the control group and the groups grown over litter amendments in the present experiment, it is speculated that litter moisture may have played a role. Although litter moistures [16] were not measured in the first experiment, they were in this experiment. As shown (Table 11), litter moistures quickly dropped below the 20% level where they remained for most of the study. Such dry conditions may have enhanced the relatively good results observed for breast blister and foot pad scores in the control group relative to treated groups in the present experiment. It is known [17] that low litter surface water activity correlates positively with reduced bacterial activity and better performance. Since the litter for all treatments came from a blend, the reason for the lower value obtained for 0-day, Treatment 2 sample cannot be explained.

Air sac scores measured at Days 22 and 48 (Table 12) were significantly improved for birds grown over litter amendments. Moreover, birds raised over the 46% acidified clay product were significantly improved relative to the other two litter treatments. In contrast with the previous experiment, birds raised over alum in this experiment did show improvement relative to the control. This finding may again be a result of having applied the alum five days prior to bird placement in Experiment 2 versus only one day prior to bird placement in Experiment 1. Control birds, on average, had air sacs that were obviously cloudy or showed plaque formation. Birds raised over the litter amendments had air sacs which

TABLE 7. Experiment 2: Effect of litter treatments on performance[A].

TREAT No.[B]	AVERAGE WEIGHT (kg) Males & Females	ADJUSTED FEED CONVERSION Mortality[C]	ADJUSTED FEED CONVERSION Wt. & Mortality[D]
1	2.4342 [b] ± 0.1055	1.8428 [a] ± 0.0315	1.8428 [a] ± 0.0315
2	2.6492 [a] ± 0.0419	1.7837 [b] ± 0.0164	1.6889 [b] ± 0.0164
3	2.6663 [a] ± 0.0304	1.7521 [c] ± 0.0183	1.6499 [c] ± 0.0183
4	2.6444 [a] ± 0.0575	1.7740 [bc] ± 0.0298	1.6814 [b] ± 0.0297

[A] Means in a column that do not have common superscripts are significantly different ($P<.05$).
[B] Treatment 1 = control; 2 = alum; 3 = acidified clay (46% acid); 4 = acidified clay (36% acid).
[C] Feed conversion adjusted for mortality calculated using the total feed consumption / pen divided by total weight of surviving birds as well as birds that died or were removed from that pen.
[D] For treatments using litter amendments, feed conversions were adjusted to a constant body weight (equal to that of the control birds). In order to adjust the feed conversion to a constant body weight, we used a factor of 5 points body weight = 1 point of feed conversion.

TABLE 8. Experiment 2: Effect of litter treatments on carcass quality[A]

TREAT No.[B]	CARCASS GRADE[C] (fraction in category)			
	A	B	C	Condemnations
1	0.7139 [b] ± 0.0416	0.1984 [a] ± 0.0289	0.0000 ± 0.0000	0.0877 [a] ± 0.0181
2	0.8706 [a] ± 0.0668	0.0966 [b] ± 0.0519	0.0000 ± 0.0000	0.0328 [b] ± 0.0255
3	0.8738 [a] ± 0.0833	0.0973 [b] ± 0.0782	0.0000 ± 0.0000	0.0289 [b] ± 0.0234
4	0.8768 [a] ± 0.0719	0.1057 [b] ± 0.0682	0.0000 ± 0.0000	0.0175 [b] ± 0.0203

[A] Means in a column that do not have common superscripts are significantly different (P<.05).
[B] Treatment 1 = control; 2 = alum; 3 = acidified clay (46% acid); 4 = acidified clay (36% acid).
[C] USDA grades

TABLE 9. Experiment 2. Effect of litter treatments on breast blister scores[A]

TREAT No.[B]	BREAST BLISTER SCORES[C] (fraction in category)		
	0	1	2
1	0.9019 [b] ± 0.0592	0.0784 [a] ± 0.0574	0.0197 [a] ± 0.0252
2	0.9565 [ab] ± 0.0512	0.0362 [ab] ± 0.0372	0.0074 [a] ± 0.0147
3	0.9493 [ab] ± 0.0630	0.0364 [ab] ± 0.0358	0.0143 [a] ± 0.0286
4	0.9926 [a] ± 0.0147	0.0000 [b] ± 0.0000	0.0074 [a] ± 0.0147

[A] Means in a column that do not have common superscripts are significantly different (P<.05).
[B] Treatment 1 = control; 2 = alum; 3 = acidified clay (46% acid); 4 = acidified clay (36% acid).
[C] Scoring: 0 = none present, 1 = small (<¼ inch), 2 = large (>¼ inch).

TABLE 10. Experiment 2: Effect of litter treatments on foot pad quality[A]

TREAT No.[B]	FOOT PAD SCORE[C] (fraction in category)			
	0	1	2	3
1	0.6425 [a] ± 0.2927	0.2833 [a] ± 0.1871	0.0741 [a] ± 0.1270	0.0000 ± 0.0000
2	0.8125 [a] ± 0.0915	0.1580 [a] ± 0.1075	0.0294 [a] ± 0.0416	0.0000 ± 0.0000
3	0.8475 [a] ± 0.1624	0.1311 [a] ± 0.1491	0.0214 [a] ± 0.0429	0.0000 ± 0.0000
4	0.8712 [a] ± 0.1674	0.0722 [a] ± 0.0991	0.0566 [a] ± 0.0685	0.0000 ± 0.0000

[A] Means in a column that do not have common superscripts are significantly different (P<.05).
[B] Treatment 1 = control; 2 = alum; 3 = acidified clay (46% acid); 4 = acidified clay (36% acid).
[C] Scoring: 0 = normal (no burn, scab, or lesion), 1 = pad burn (dermis only), 2 = pad scab (healing) on one or both feet, 3 = pad lesion (open sore) on one or both feet.

TABLE 11. Experiment 2: Effect of litter treatment on litter moisture.

TREAT No.[A]	MOISTURE CONTENT (%) VERSUS TIME (days)							
	0	1	2	6	9	16	27	43
1	44.3	32.7	26.7	14.6	20.0	16.6	19.8	30.0
2	33.4	33.9	20.1	11.5	15.4	16.2	20.6	22.9
3	42.9	40.4	20.1	11.3	16.7	16.7	22.9	24.4
4	44.0	34.8	30.2	13.5	29.2	16.8	26.4	25.8

[A] Treatment 1 = control; 2 = alum; 3 = acidified clay (46% acid); 4 = acidified clay (36% acid).

TABLE 12. Experiment 2: Effect of litter treatments on air sac scores[A]

TREAT No.[B]	AIR SAC SCORES[C]	
	(Day 22)	(Day 48)
1	1.6667 [a] ± 0.6681	1.8833 [a] ± 0.6132
2	0.5500 [b] ± 0.6490	0.7833 [b] ± 0.8456
3	0.2833 [c] ± 0.4903	0.4833 [c] ± 0.7700
4	0.3729 [bc] ± 0.4877	0.6500 [bc] ± 0.7324

[A] Means in a column that do not have common superscripts are significantly different (P<.05).
[B] Treatment 1 = control; 2 = alum; 3 = acidified clay (46% acid); 4 = acidified clay (36% acid).
[C] Average scores of 60 birds per treatment. Scoring: 0 = Clear, 1 = Cloudy, 2 = Plaque formation, 3 = Severe plaque formation.

were clear or only cloudy. These data provide compelling evidence that the birds raised over the litter amendments were subjected to a much less severe atmospheric challenge than the birds raised over the untreated control.

*Darkling Beetle Counts***:** Darkling beetles were captured on Days 12 and 34 of Experiment 1. Counts were relatively low on Day 12, so statistical analysis of these data was not conducted. However, as shown by the data in Table 13, counts of both adult and larval darkling beetles captured on Day 34 are significantly reduced in pens receiving litter amendment treatments (with exception of adult beetle count for alum, treatment 3). Similar results have been previously reported for sodium bisulfate [18]. While the exact mechanism operative in eliciting this effect is not known with certainty, it seems plausible the low pH associated with these litter amendments either causes the beetles and larvae to migrate away from the upper layers of the litter, or in some other way interrupts the beetle's life cycle thereby leading to fewer insects being captured.

TABLE 13. Experiment 1: Effect of litter treatments on darkling beetle counts[A]

TREAT No.[B]	AVERAGE COUNT (Day 34)	
	Adults	Larvae
1	37.40 [a] + 36.58	48.70 [a] + 29.19
2	12.50 [b] + 14.44	17.10 [b] + 19.85
3	21.20 [ab] + 19.16	23.10 [b] + 17.84
4	12.30 [b] + 14.17	18.70 [b] + 18.70

[A] Means in a column that do not have common superscripts are significantly different (P<.05).
[B] Treatment 1 = control; 2 = acidified clay; 3 = alum; 4 = sodium bisulfate.

Conclusions and Applications

1. Acidic, granular litter amendments such as sodium bisulfate, alum and Poultry Guard™ (46% acid loading), a new acidified clay product, provide control over ammonia generation for up to four weeks in broiler houses under the conditions of these studies when applied at a rate between 93 and 100 lbs/1000 ft².
2. Air sac scores for birds raised over litter amendments (with one exception) are significantly improved relative to the untreated control. This provides support for the supposition that the atmospheric challenge (ammonia) is significantly reduced in the case of birds raised over the litter amendments.
3. Broilers grown on litter treated with these litter amendment products exhibit significantly better feed conversions as well as enhanced weight gain and improved carcass quality in comparison to birds raised over untreated litter.
4. There is reasonably convincing evidence that some of these litter agents (i.e. - acidified clay and alum) can positively enhance foot pad quality for birds raised over litter containing these amendments. This is an extended benefit that can provide additional economic incentive for using these agents. High quality (lesion free) chicken feet are a delicacy in China (where most U.S. production is exported) and they command higher prices; in contrast, low quality feet are simply recycled into animal feed at significantly lower prices.
5. Darkling beetle counts (adults and larvae) are significantly reduced in litters treated with these litter amendments relative to untreated litters. This may provide yet another economic benefit for using these litter agents. It has been estimated that darkling beetles cause economic

losses to the poultry industry of $15 million per year due to the combined effects of their competition for food [19], disease transmission [20] [21], and (chicken house) insulation destruction [19]. In addition, control of darkling beetles is considered an important component of on-farm HACCP programs.

References and Notes

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11. Poultry Guard™ (acidified clay), Oil-Dri Corporation of America, 410 North Michigan Ave., Chicago, IL, 60611. Acidified clay granules (Experiment 1, 16 x 48 mesh particles; Experiment 2, 20 x 60 mesh particles) of attapulgite/montmorillonite clay from Ochlocknee, GA were heated to 850 °F; ("LVM" – Low Volatile Material) prior to impregnation with concentrated (96%) sulfuric acid.

12. Al+ Clear™ Poultry Grade Alum, (Experiment 1) Lot No. B8047560; (Experiment 2) Lot No. ES81208-OPG; General Chemical, Inc., 90 East Halsey Rd., Parsippany, NJ, 07054.

13. PLT®, Jones-Hamilton Co., 30354 Tracy Rd., Walbridge, OH, 43466.

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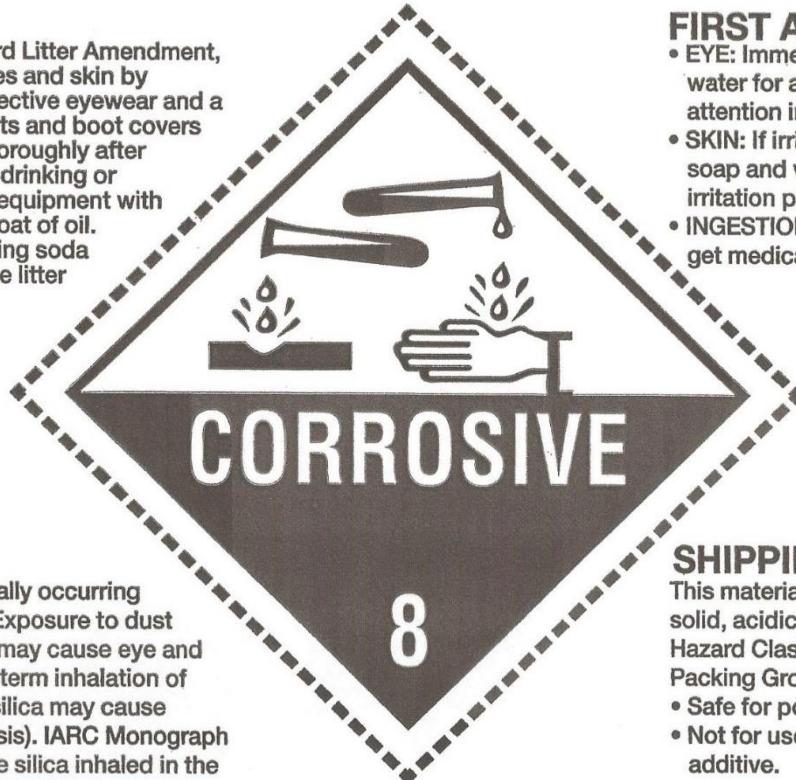
Attachment #2
Poultry Guard product label

HANDLING

When handling Poultry Guard Litter Amendment, prevent contact with the eyes and skin by wearing rubber gloves, protective eyewear and a dust mask. Long sleeve shirts and boot covers are recommended. Wash thoroughly after handling and before eating, drinking or smoking. Thoroughly clean equipment with water and spray with light coat of oil. Household ammonia or baking soda can be used to neutralize the litter amendment.

FIRST AID

- EYE: Immediately flush eyes with cool running water for at least 15 minutes. Get medical attention immediately if irritation persists.
- SKIN: If irritation exists, wash thoroughly with soap and water. Get medical attention if irritation persists.
- INGESTION: Rinse mouth out with water and get medical attention immediately.



WARNING

This product contains naturally occurring crystalline silica as quartz. Exposure to dust containing crystalline silica may cause eye and respiratory irritations. Long-term inhalation of dust containing crystalline silica may cause chronic lung damage (silicosis). IARC Monograph 68 concludes that crystalline silica inhaled in the form of quartz from occupational sources is carcinogenic to humans (Group 1). However, carcinogenicity was not detected in all industrial circumstances studied; and, under conditions of normal use, exposure to respirable crystalline silica from this product is below the OSHA Permissible Exposure Limit and the ACGIH Threshold Limit Value. To reduce exposure, use adequate ventilation and NIOSH or MSHA approved respirator.

SHIPPING

This material is regulated as: Corrosive solid, acidic, inorganic, n.o.s. (sulfuric acid), Hazard Class 8, I.D. Number UN3260, Packing Group II, IMDG # Page 8150-1

- Safe for poultry house use.
- Not for use as a direct food or drug additive.

In case of emergency,
please call 336-617-8990

Made in the U.S.A.

Manufactured By:

TRINICO AG, INC.
Greensboro, NC
(336) 617-8990

50 lbs.
(22.7 kg)

Attachment #3

Material Safety Data Sheet

POULTRY GUARD® litter amendment (#4021010)

MATERIAL SAFETY DATA SHEET

NAME OF PRODUCT POULTRY GUARD®

FILE NO.: 4021010
MSDS DATE: 9/1/2014

TRINICO AG, INC.

For Hazardous Materials [*or* Dangerous Goods]
IncidentSpill, Leak, Fire, Exposure, or Accident Call CHEMTREC
Day or night Within USA and Canada: 1-800-424-9300 CCN694186
outside USA and Canada: +1 703-527-3887 (collect calls accepted)

SECTION 1: PRODUCT AND COMPANY IDENTIFICATION

PRODUCT NAME: **POULTRY GUARD® litter amendment**

MANUFACTURER: **Trinico Ag, Inc.**
ADDRESS: **PO Box 41319**
Greensoro, NC 27404-1319

EMERGENCY PHONE: **800-424-9300**
OTHER CALLS: **336-288-0755**
FAX PHONE: **336- 288-0800**

CHEMICAL NAME: **Bentonite, acid-activated**
CHEMICAL FAMILY: **Acid-activated bentonite**
CHEMICAL FORMULA: **N/A**

PRODUCT USE: **Poultry litter amendment / ammonia control**
PREPARED BY:

SECTION 2: COMPOSITION/INFORMATION ON INGREDIENTS

INGREDIENT:	CAS NO.	% WT
Bentonite, acid-activated	98561-46-7	92-94
Quartz (crystalline silica)	14808-60-7	6-8
Quartz (respirable)	14808-60-7	0.00064 mg/m3

OSHA PEL-TWA: Bentonite	5 mg/m^3 (respirable fraction)
OSHA PEL-TWA: Quartz (crystalline silica)	10 mg/m^3/% SiO_2 + 2 TWA
OSHA PEL-TWA: Quartz (respirable)	10 mg/m^3/% SiO_2 + 2 TWA
ACGIH TLV-TWA: Bentonite	3 mg/m^3(respirable fraction) 10 mg/m^3 TWA (inhalable dust)
ACGIH TLV-TWA: Quartz (crystalline silica)	0.025 mg/m^3 TWA
ACGIH TLV TWA: Quartz (respirable)	0.025 mg/m^3 TWA

SECTION 3: HAZARDS IDENTIFICATION

EMERGENCY OVERVIEW: This product is a non-combustible mineral. Eye contact may cause severe irritation or burns. Skin contact may cause irritation or burns. This product contains some naturally occurring non-respirable crystalline silica as quartz, a very small fraction of which is in the respirable range. Oil-Dri has conducted extensive sampling during the normal use of this product. This exposure data indicate that exposure to respirable crystalline silica during the normal use of this product is well below the OSHA Permissible Exposure Limit (PEL) and ACGIH Threshold Limit Value (TLV). In addition, the company is not aware of any scientific or medical data available indicating that exposure to dust from this product under conditions of normal use will cause silicosis or cancer. Therefore adverse long-term effects would not be expected from normal use of this product. Prolonged overexposure to respirable crystalline silica may cause lung disease (silicosis). IARC, in Monograph 68, has concluded that crystalline silica inhaled in the form of quartz from occupational sources is carcinogenic to humans (Group 1); however, carcinogenicity was not detected in all industrial circumstances studied. Carcinogenicity may be dependent upon inherent characteristics of the crystalline silica or on external factors affecting its biological activity or distribution of its polymorphs.

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POTENTIAL HEALTH EFFECTS:

EYES: Direct contact with the eyes may cause severe irritation with redness, tearing and blurred vision. Possible permanent damage and blindness may result.

SKIN: Contact, especially with moist skin, may cause severe irritation and possibly burns.

INGESTION: May cause severe irritation or burns to the mouth, throat and gastrointestinal tract,

INHALATION: Inhalation of dust may cause irritation to the eyes, nose, throat and respiratory tract.

ACUTE HEALTH HAZARDS:

CHRONIC HEALTH HAZARDS: Prolonged or repeated skin contact may cause dermatitis. Inhalation of excessive concentrations of any dust, including this material, may lead to lung injury. This product contains some naturally occurring non-respirable crystalline silica as quartz, a very small fraction of which is in the respirable range. Oil-Dri has conducted extensive sampling during the normal use of this product. This exposure data indicate that exposure to respirable crystalline silica during the normal use of this product is well below the OSHA Permissible Exposure Limit (PEL) and ACGIH Threshold Limit Value (TLV). In addition, the company is not aware of any scientific or medical data available indicating that exposure to dust from this product under conditions of normal use will cause silicosis or cancer. Therefore adverse long-term effects would not be expected from normal use of this product. Excessive inhalation of respirable crystalline silica may cause silicosis, a progressive, disabling and fatal disease of the lung. Symptoms may include cough, shortness of breath, wheezing and reduced pulmonary function. The National Toxicology Program (NTP) classifies crystalline silica as a known carcinogen. The International Agency for Research on Cancer (IARC), in Monograph 68 has concluded that crystalline silica inhaled in the form of quartz or cristobalite, from occupational sources is carcinogenic to humans (Group 1). However, in making the overall evaluation, the Working Group noted that carcinogenicity was not detected in all industrial circumstances studied. Carcinogenicity may be dependent on inherent characteristics of the crystalline silica or on external factors affecting its biological activity or distribution of its polymorphs.

MEDICAL CONDITIONS GENERALLY AGGRAVATED BY EXPOSURE: Individuals with respiratory disorders such as asthma and bronchitis may be at increased risk for respiratory irritation from dust exposure.

SECTION 4: FIRST AID MEASURES

EYES: Immediately flush eyes with cool running water for at least 15 minutes, lifting upper and lower lids. Get immediate medical attention.

SKIN: Wash thoroughly with soap and water. Remove contaminated clothing and launder before re-use. Get medical attention if irritation persists.

INGESTION: Rinse mouth out with water and get immediate medical attention.

INHALATION: Remove to fresh air. If breathing has stopped, give artificial respiration. If breathing is difficult, give oxygen. Get immediate medical attention.

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SECTION 5: FIRE-FIGHTING MEASURES

FLASH POINT: Not Applicable

EXTINGUISHING MEDIA: Use any dry chemical or CO2 extinguisher that is appropriate for the surrounding small fire. Avoid applying water directly to this material. Do not get water into containers. If large fire occurs use flooding quantities of water. Aqueous runoff could be acidic and corrosive.

SPECIAL FIRE FIGHTING PROCEDURES: Firefighters should always wear self-contained breathing apparatus and full protective clothing for fires involving chemicals or in confined spaces.

UNUSUAL FIRE AND EXPLOSION HAZARDS: None

HAZARDOUS DECOMPOSITION PRODUCTS: Extremely high temperatures may generate sulfur dioxide.

SECTION 6: ACCIDENTAL RELEASE MEASURES

ACCIDENTAL RELEASE MEASURES: Wear appropriate protective clothing including gloves and eye protection. Sweep up, taking care not to generate airborne dust and collect for re-use or disposal.

SECTION 7: HANDLING AND STORAGE

HANDLING AND STORAGE: Prevent contact with the eyes, skin and clothing. Do not breathe dust. Wear protective clothing (refer to Section 8). Use only in well ventilated areas. Keep container closed when not in use. Wash thoroughly after handling and before eating, drinking or smoking. Use good housekeeping in storage and use areas to prevent dust accumulation.

Store in closed containers not susceptible to acid attack in a dry area away from flammable substances and incompatible materials. Close containers when not in use.

SECTION 8: EXPOSURE CONTROLS/PERSONAL PROTECTION

ENGINEERING CONTROLS: For operations where the exposure limit may be exceeded, local exhaust ventilation is recommended.

RESPIRATORY PROTECTION: For operations where the exposure limit may be exceeded, a NIOSH/MSHA approved high efficiency particulate respirator is recommended.

EYE PROTECTION: Chemical safety goggles and/or face shield recommended.

SKIN PROTECTION: Impervious gloves such as butyl rubber, neoprene or natural rubber are required. Use gauntlet type glove where needed to prevent skin contact.

OTHER PROTECTIVE CLOTHING OR EQUIPMENT: Appropriate protective clothing such as apron, coveralls or acid-proof suit with boots recommended where needed to prevent skin contact. Launder contaminated clothing before re-use. Discard contaminated shoes and other leather items which cannot be decontaminated. An eye wash and safety shower must be available in the work area.

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SECTION 9: PHYSICAL AND CHEMICAL PROPERTIES

APPEARANCE: Dark granules

ODOR: Slight "pickling acid" odor

PHYSICAL STATE: Solid

pH AS SUPPLIED: 0.6

SPECIFIC GRAVITY (H2O = 1): 0.9-1.0 @ 25 °C

BOILING POINT: Not applicable
MELTING POINT: Not applicable
FREEZING POINT: Not applicable
VAPOR PRESSURE Not Applicable
VAPOR DENSITY: Not Applicable
EVAPORATION RATE: Not Applicable

SOLUBILITY IN WATER: Partially soluble
PERCENT SOLIDS BY WEIGHT: 50-60%

PERCENT VOLATILE: Not applicable
VOLATILE ORGANIC COMPOUNDS (VOC): None
MOLECULAR WEIGHT: Not applicable

SECTION 10: STABILITY AND REACTIVITY

	STABLE	UNSTABLE
STABILITY:	✓	

INCOMPATIBILITY (MATERIAL TO AVOID): Do not expose to strong bases, carbides, chlorates, fulminates, nitrates, pirates, powdered metals or combustible materials.

HAZARDOUS DECOMPOSITION OR BY-PRODUCTS: Extreme heat may generate sulfur dioxide. Reacts with many metals releasing flammable hydrogen gas.

HAZARDOUS POLYMERIZATION: Will not occur.

SECTION 11: TOXICOLOGICAL INFORMATION

TOXICOLOGICAL INFORMATION: Sulfuric Acid: Oral Rat LD50 2140 mg/kg; Inhalation Rat LC50 320 mg/m3/2 hr

SECTION 12: ECOLOGICAL INFORMATION

ECOLOGICAL INFORMATION: No data avaliable

SECTION 13: DISPOSAL CONSIDERATIONS

WASTE DISPOSAL METHOD: Dispose in accordance with local, state and federal environmental regulations. If permitted, unused material may be suitable for disposal in sanitary landfill. Used material may be subject to regulation, depending on the nature of the material absorbed. Check with appropriate regulatory authority for used material containing hazardous waste.

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SECTION 14: TRANSPORT INFORMATION

PROPER SHIPPING NAME:
Corrosive Solid, Acidic, Inorganic, n.o.s. (Contains Sulfuric Acid)

UN NUMBER: UN3260

HAZARD CLASS/PACKING GROUP: 8, PG II
LABELS REQUIRED: Corrosive

SECTION 15: REGULATORY INFORMATION

CERCLA/SUPERFUND Spills of this product over the RQ (reportable quantity) must be reported to the National Response Center. The RQ for the product, based on the RQ for Sulfuric Acid of 1000 lbs, is 2,000 lbs. Many states have more stringent release reporting requirements. Report spills required under federal, state and local regulations.

SARA HAZARD CATEGORY (311/312): Acute Health and Chronic Health

SARA 313: This product contains Sulfuric Acid (CAS No. 7664-93-9). Sulfuric acid may be reportable when manufactured, processed or used as an aerosol.

TSCA: All of the components of this product are listed on the EPA TSCA Inventory or exempt from notification requirements.

EINECS: All of the components of this product are listed on the EINECS Inventory or exempt from notification requirements

EEC R&&S Phrases: Corrosive R34, S24/25, S26, S36/37/39, S45*Issued on January 31, 2008* 5 of 5 **JAPAN MITI:** All of the components of this product are existing chemical substances as defined in the Chemical Substances Control Law.

AICS: All of the components of this product are listed on the AICS Inventory or exempt from notification requirements

CANADIAN DSL: All of the components of this product are listed on the Canadian Domestic Substance List or exempt from notification requirements.

CA PROPOSITION 65: This product contains respirable crystalline silica which is known to the State of California to cause cancer.

SECTION 16: OTHER INFORMATION

NFPA RATING: Health=3 Fire=0 Reactivity=1

HMIS RATING: Health=3* Fire=0 Reactivity=1

The information in this data sheet is believed to be accurate. However, each purchaser should make its own test to determine the suitability of the product for its purposes. TRINICO AG, INC. MAKES NO WARRANTY, EXPRESSED OR IMPLIED, WITH RESPECT TO THE PRODUCT and assumes no responsibility for any risk or liability arising from the use of the information or the product. Statements about the product should not be construed as recommendations to use the product in infringement of any patent.

Attachment #4
Calculation of amount of acid activated bentonite in poultry litter applied to fields

There are several variables in determining properties of poultry litter in a chicken house:

Depth of litter: Generally averages from 2 to 6 inches, but may be as much as a foot.

Flocks per house per year:
Lighter birds (4-41/2 lbs) can have as many as 71/2 flocks per year in a house. Heavy birds (8-9 lbs.) would be around 5 flocks per year. This also is affected by the layout time between each flock. That could vary from less than a week to a month or more.

Application rates of litter amendments: The suggested rate of application for Poultry Guard is 100 lbs per 1000 square feet but, growers routinely go down to 50 lbs per 1000 square feet.

Commercial growers are required to have nutrient management plans.
These plans require an analyses of the soil, a description of vegetation produced on the land and an analysis of the litter applied. If nitrogen and phosphorus content of the litter exceed the amount that is taken up by the vegetation that is grown on the land, the rate of application must be adjusted to meet that balance.

The following are calculations based on certain assumptions that would be a normal representation of litter in a commercial chicken house.

Assumptions:

Litter: 35lbs. per cubic foot	Depth of liter: 4 inches
Application rate of Poultry Guard:	100 lbs. per 1000 square feet
Frequency of application:	5.5 times per year
1000 square feet of litter 4 inches deep	= 333 cubic feet
333 cubic feet at 35lbs. per cubic foot	= 11667 lbs of litter
100 lbs Poultry Guard in 11667 lbs. of litter	= 1% by weight.

If this was applied 5.5 times in a year that would be 550 lbs. or about 5% by weight. But remember that the litter is being increased by 1 inch of new litter generated by the chickens in each new flock.