

Understanding the Benefits of Humic Substances in Animal Nutrition

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Introduction

Humic substances have been used in Indian medicine (“Ayurveda”) for approximately 3,000 years due to their immuno-modulatory, antioxidant, diuretic, antihypertensive, and hypoglycemic effects (as reviewed by Winkler and Ghosh, 2018). They are found in sediments and mined around the world. The objective of this paper is to provide an overview of humic substances, discuss their chemical properties, and explore the role of their use in animal nutrition.

Peat, a source of humic substances, is considered a natural material that has long been used in agronomy as well as human and animal medicine. Reed-Sedge Peat (RSP) used in the US in livestock and poultry feed is defined by AAFCO (73.241-249) as a natural substance that is partially a mixture of decomposed plant material from reeds, sedges, grasses, and some hypnum mosses that are found in wetlands and containing one third to two thirds peat fibers. Furthermore, AAFCO states that RSP should not contain more than 15% moisture and is free from all harmful micro-organisms. It is approved for use in animal feeds, not to exceed 5% of the total daily ration, as a carrier for liquid products and premixes or as a nutritional diluent for lowered energy diets.

What the definition does not indicate is that some RSP being fed commercially has been decomposing for millions of years around the world and, depending on the mine and location, the material can differ. For example, peat and freshwater RSP are in the same family. Peat is partially decayed vegetation forming near wetlands, bogs, and swamps while freshwater RSP is generally much older than other types of peat and is considered to consist of richer, decomposing forests and wetlands. The humus level is higher and the humic acid level is likely higher in RSP deposits (Table 1). It is important to understand the differences, as excess salt and toxic minerals can also be present in saltwater sources.

Table 1. Comparison of Reed-Sedge Peat vs. Freshwater Reed-Sedge Peat

	Reed-Sedge Peat	Freshwater Reed-Sedge Peat
Appearance	Brown	Rich Black
Constitution	30-60% Plant Fibers; Lower Humus	Fully Decomposed; High Humus
Carbon	Low-Medium Carbon	Medium-High Carbon
Richness	Low Density	High Density
Material Composite	Reeds, Sedges, Bacteria	Ancient Forests, both flora and fauna

Naturally Hidden Benefits – Humic Substances

Reed-sedge peat is scientifically classified as a humic substance or decomposed organic matter, which are the most common forms of organic carbon found in the natural environment. Throughout this decomposition, the remaining matter contains humic acid, fulvic acid, ulmic acid, and humin carbon structures (Table 2).

Table 2. Summary of Humic Organic Substances (adapted from Trckova et al., 2005).

Humic Substance	Description of Humic Substances
Humic Acids	Medium to high molecular weight, aromatic polyfunctional compound with cation and anion exchange sites. Degree of decomposition can make it up to 50% or more of the peat product. Dark brown in color with solubility in alkaline conditions.
Fulvic Acids	Lower molecular weight, but similar in structure to humic acid. Yellow in color but soluble across all ranges of pH.
Ulmic Acids	Dark in color and soluble in alcohol, but not well investigated.
Humates	Salts of humic acid that contain Ca, NA, Al, and Fe in the exchange site versus H.
Humins	The fraction of humic substances that is not soluble in water at any pH value and in alkali. Black in color.

Humic substances are highly-abundant organic compounds formed in soils. The key function of humic acid is to transfer the nutrients from the soil to the living organism. Additionally, humic acid acts as a vasodilator, increasing cell wall permeability. The role of fulvic acid is to capture the minerals and place an electric charge on them for ready uptake by the organism. Known reactive groups of fulvic acid are carboxyls, hydroxyls, carbonyls, phenols, and quinones, which are fundamental in fulvic acid's ability to chelate and act as an antioxidant.

Humic Substances for Animal Nutrition

Traditionally peat-type products have been used as bedding sources in livestock but are also noted to have preventive and therapeutic values regarding gastrointestinal issues, such as diarrhea, dermatitis and other skin disorders, and inflammatory conditions such as arthritis. Humic acids stabilize the intestinal microflora, ensuring an improved nutrient utilization and feed efficiency. This leads to an increase in the live weight of animals without increasing the amount of feed (as reviewed by Písařková et al.). An earlier review by Trckova et al. (2005) summarized the growth and performance benefits in multiple species:

- Improvement in broiler body weight gain by 5 to 7% and reduction in mortality by 3 to 5%
- Supplementation to feed rations of layers resulted in increased egg production, enhanced feed conversion efficiency and reduced mortality rate
- Improvements in body weight gain and feed conversion ratio (FCR) in diets void of antibiotics in nursery pigs
- Beneficial effects against different viral infections in multiple species

Additional findings suggest humic substances are beneficial as a feed additive for cattle. A commercially available humic substance in Australia fed to feedlot cattle demonstrated a 12.8% improvement in average daily gain (ADG) and a 14.7% reduction in dry matter (DM) feed intake, resulting in improved FCR and a 15.2% reduction in the number of days on feed (Cusack, 2008).

In laying hens, humic substances increased hen body weight (BW) gain by 44.7% during 22-40 weeks of lay, while also lowering cholesterol content in the yolk by 13.2% (Yalcin et al., 2006). Further work in turkey breeders also demonstrated improvements in hen weight, but also feed conversion, egg production and quality that resulted in improved fertility and hatchability when supplemented 200 mg/kg or greater of humic acid was fed (Ibrahim, 2016). When fulvic acid is fed at 0.6%, broilers had an improved final BW, gain, and FCR compared to control-fed birds, resulting most likely from the improvements in endogenous enzymes (amylase, lipase, and protease) and

other antioxidant and immune parameters (Mao, 2019). Additionally, feeding humic substances linearly improved overall BW gain, FCR, carcass weight, and meat quality traits in broilers when fed from 0.5-1.5% humic substances for 42 days (Ozturk et al., 2011). Further research within the USA (Edmonds et al., 2014) reported that feeding a humic substance in combination with butyric acid not only improved growth and feed efficiency, but also reduced mortality when fed alone or in combination with butyric acid.

In growing-finishing pigs, fulvic acid quadratically increased ADG, gain to feed (G:F), final BW, hot carcass weight (HCW), and malondialdehyde (MDA) from 30 kg to slaughter (Bai et al., 2013). Furthermore, through broken line regression, a range of 0.14-0.65% of humic acid is needed to optimize both growth performance and meat quality. Further work also demonstrated that backfat thickness was decreased in finishing pigs by feeding 0.2-0.6% fulvic acid (Chang et al., 2014), most likely correlated to the changes in different endocrine parameters (growth hormone, insulin, leptin, thyroid hormone, thyroxine, and LDL cholesterol) measured. Growth responses could be potentially also linked to improvements in gross energy (GE) and phosphorus digestibility, as Kunavue and Lien (2012) demonstrated when fulvic acid was fed alone or in combination with a probiotic to growing pigs (20 kg). Their trial also demonstrated an improvement in ADG compared to a NC (negative control) diet without an antibiotic and similar to an antibiotic PC (positive control).

Immune Modulation

It has been well documented that humic substances are involved in several different pathways of the immune system. They have been used in natural medicine for centuries as they not only exhibit anti-inflammatory and anti-viral properties but also have the ability to reduce bacterial growth. Used in vitro or in vivo, the E. coli LPS (lipopolysaccharides) challenge model is one of the classical models to understand the role of different substances on the inflammatory response. In-vitro work with human stem cells indicated that humic acid could potentially inhibit LPS-induced inflammatory pathways (Gau et al., 2000). Humic acid fed in combination with butyric acid (0.25, 0.22%, respectively) had a beneficial response in an E. coli LPS-induced challenge. Pigs that were challenged and fed the combination had a 62% reduction in cortisol levels than if fed alone or the control fed pigs that were challenged (Weber et al., 2014). Furthermore, in 4-wk old broilers, birds fed humic acid had a decreased number of blood heterophils which resulted in a decreased heterophil: lymphocyte ratio, which is indicative of a lower level of stress. However, this response did not continue when birds were sampled at 5 weeks of age (Rath et al., 2006).

It is important to note that the researchers fed higher levels than typically fed in today's commercial programs, attempting to induce tibial dyschondroplasia, which did not occur, but reduced growth rate was the result. Furthermore, fulvic acid can have both pro-inflammatory and anti-inflammatory properties. As reviewed by Winkler and Ghosh (2018), pro-inflammatory properties include the ability to increase antibody titers, reduce bacterial wound size, and on a cellular level, increase reactive oxygen species (ROS) and nitric oxide (NO) production and complement fixation.

Oxidative Stress

Oxidative stress occurs when there is an imbalance of highly ROS compared to antioxidants. For instance, when ROS is high, glutathione (GSH) and superoxide dismutase (SOD) cannot counteract the problem, and thus cellular damage occurs, which is harmful to the animal. It is well established that humic substances are widely utilized both in agronomy and environmental pollutant treatments for their oxidative and chelation properties, but the mechanism is not well known. However, Aeschbacher et al. (2012) determined that humic substances contain phenolic electron-donating moieties that cover a wide range of oxidation potentials. Furthermore, their work demonstrated that terrestrial-aquatic humic substances, such as RSP, have greater antioxidative properties than terrestrial or microbially derived humic substances. When growing broilers were fed fulvic acid, their plasma levels of SOD, glutathione peroxidase (GPx) and MDA increased. These elevated levels indicate a more robust oxidative state for broilers, which also in return resulted in improved immune function, whereas serum IgG, A, and M were elevated when fulvic acid was fed at 0.6% (Mao, 2019).

Mycotoxin Mitigation

Even though the FDA does not recognize any substances as mycotoxin binders, different additives have the natural ability to chelate, or bind molecules, including mycotoxins and their substrates. However, mycotoxin mitigation has moved beyond binding, to include mycotoxin modifiers, in which a substance would modify the chemical structure of mycotoxins in attempts to reduce their toxicity.

Humic substances are one of these well-documented natural substances that have a strong affinity to bind various substances, such as heavy metals, herbicides, minerals, mutagens, monoaromatic and polycyclic compounds, and even *Bacillus subtilis* bacteria. Recent work indicates that humic acids also could diminish the adverse effects of aflatoxin in livestock. For example, humic acids provide protection against mycotoxins by increasing their ability to form a protective film on the mucous epithelia of the gastro-intestinal tract against infections and toxins (Kuhnert et al., 1991). The macro-colloidal structure of humic acid ensures good shielding on the mucous membrane of the stomach and the gut, the peripheral capillaries, and damaged mucous cells. As a result of this process, the resorption of toxic metabolites is reduced or fully prevented, especially after infections, in cases of residues of harmful substances in animal feed. Through a comprehensive analysis of 27 mycotoxin binders commercially available, De Mil et al. (2015) demonstrated that binders containing mixed-layered smectites and humic acids had the greatest affinity to reduce free zearalenone concentrations in-vitro.

In-vivo research in broilers have shown that feeding as little as 0.1% humic acid to broilers fed diets contaminated with 100 ppb aflatoxin can perform similarly to the positive control-fed birds (Arafat et al., 2017). Furthermore, humic acid also protected the liver weight and linearly decreased aflatoxin B1 residues as supplemented levels of humic acid increased. Interestingly they also observed that aflatoxin contamination reduced antibody titers to the given vaccine, but was restored with humic acid supplementation, most likely as a result of also maintaining the immune system as demonstrated in similar blood differential counts to the positive control-fed birds. Similar findings were reported by Ghahri et al. (2010), who found broilers fed aflatoxin-contaminated feed had improved BW gain and FCR, while protecting the liver and bursa against toxicity damage, by feeding as low as 0.2% humic acid. These results indicate that humic acid substances go beyond their natural chelation characteristics to add value to livestock nutritional strategies.

Environmental Impact Opportunities

Environmental regulations and restrictions are ever increasing, and animal agriculture is being heavily targeted for both methane and ammonia emissions. Ruminants, especially cattle, are the most widely spread livestock around the world. There have been several studies conducted that have demonstrated that feeding humic substances can reduce ammonia production in cattle without negatively impacting their performance. For instance, Terry et al. (2018) reported an 8.5% linear ($P = 0.04$) reduction in ammonia production when levels were increased from 0-3.0 g/d, even with a product that contained only 50.7 and 4.4% humic and fulvic acids, respectively compared to previous work ranging from 61.8-89.8% organic acids. Furthermore, when pigs were fed humic substances, ammonia emissions were reduced up to 18%, dependent on dose (Ji et al., 2006).

Summary

There are a variety of research publications indicating the advantages of utilizing humic substances, either topically or as a feed additive, due to their natural abilities to reduce inflammatory pathways, chelation properties, bind mycotoxins, etc. Results in animal feeding trials have been reported with inconsistent responses in growth performance and efficiency, but this appears to be due primarily to the source or quality of the humic substance or improper formulation. Additionally, the benefits of feeding humic substances appear to be synergistic or enhanced when fed in addition to other acids or feed additives, such as butyric acid. Ultimately, the use of humic substances or RSP gives a potential economic advantage to livestock and poultry producers when a high-quality managed source is used and dosing is determined according to supporting research.

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