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CONTROL OF TOXIC SUBSTANCES

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Numerous toxic substances can be present in feed. However, perhaps the most commonly occurring toxic substances are, molds, mycotoxins, fat rancidity, chemical residues and pathogenic microorganisms.

Control of molds and mycotoxins

Mycotoxins are poisonous chemical substances produced by molds as they grow on feed or feed ingredients. Mycotoxins are generally very durable chemically and will not be destroyed during the feed manufacturing process. While molds are generally reduced in numbers as a result of the feed manufacturing process, molds can grow whenever conditions are favourable. Controlling mold growth and mycotoxin production can generally be accomplished by keeping moisture low, keeping feed fresh, keeping equipment clean and using mold inhibitors.

Moisture is the single most important factor in determining if and how rapidly molds will grow in feeds. Moisture in feeds comes from three sources: feed ingredients, feed milling processes and the environment in which the feed is held or stored. To control the moisture content of feeds successfully, moisture from all three sources must be managed.

Since corn and other grains are a primary source of the moisture and molds found in feed, the first important step in managing moisture in feed is to control it in the grains from which the feed is prepared. Since all feed ingredients contain moisture, they should be monitored and their moisture content recorded. It is commonly believed that the amount of moisture in grain is too small to permit mold growth except in rare and unusual circumstances. However, moisture is not evenly distributed in grain kernels. A batch of grain containing an average of 15.5% moisture may, for example, contain some kernels with 10% moisture and others with 20% moisture. The moisture content of individual grain kernels is directly related to the amount of mold growth that occurs; that is, kernels with higher moisture contents are more susceptible to mold growth. In addition to moisture, the amount of mold growth is about five times greater for broken kernels than for intact kernels.

The grinding process creates friction, which causes heat to build. If unchecked, temperature increases larger than 10.1°F (5.51°C) will cause significant migration of grain moisture encouraging mold growth. This is particularly true in cold weather when temperature differences can cause moisture to condense on the inside walls of bins. Air-assisted hammer mill systems reduce heat build-up in the product and, in turn, reduce moisture problems.

Generally, the pelleting process adds heat and 3-5% moisture to feeds in the form of steam. If the pelleting process is done correctly, this excess moisture is removed from the feed before shipment. However, if this excess moisture is not removed when the pellets are cooled, mold growth will be encouraged. Since feeds containing moisture are warmer than normal, storing hot or warm pellets in a cool bin will cause moisture to condense on the inside of the bin.

Although pelleting of feed has been shown to reduce mold counts by a factor of 100-10,000, many mold spores remain in the feed after it has been pelleted. After pelleting, the remaining spores can grow if conditions are right. Thus, the pelleting process delays, but does not prevent, the onset of mold growth and plays only a minor role in efforts to control molds. In addition, pelleted feeds may be more easily attacked by molds than non-pelleted feeds.

Time is required for both mold growth and mycotoxin production to occur. Therefore, it is important to have feeds delivered often so that they will be fresh when used. Feeds should generally be consumed within 10 days of delivery.

During and after the feed manufacturing process, feeds may come in contact with old feed that has lodged or caked in various locations within the feed manufacturing, storage and delivery systems. This old feed is often very moldy and may "seed" the fresher feed it contacts, increasing the chances of mold growth and mycotoxin formation. To prevent this problem, caked, moldy feed should be removed from all feed manufacturing and handling equipment.

The use of chemical mold inhibitors is only one of several tools useful in the complex process of controlling the growth of molds, and they should not be relied upon exclusively. The main types of mold inhibitors are

(1) individual or combinations of organic acids (for example, propionic, sorbic, benzoic and acetic acids),

(2) salts of organic acids (for example, calcium propionate and potassium sorbate) and

(3) copper sulphate.

Solid or liquid forms work equally well if the inhibitor is evenly dispersed through the feed. Generally, the acid form of a mold inhibitor is more active than its corresponding salt.

The particle size of the carriers for mold-inhibiting chemicals should be small so as many particles of feed as possible are contacted. In general, the smaller the inhibitor particles, the greater the effectiveness. Some

propionic acid inhibitors rely on the liberation of the chemical in the form of a gas or vapour from fairly large particle carriers. Presumably, the inhibitor then penetrates the air spaces between particles of feed to achieve even dispersion.

Certain feed ingredients may also affect mold inhibitor performance. Protein or mineral supplements (for example, soybean meal, fish meal, poultry byproduct meal and lime stone) tend to reduce the effectiveness of propionic acid. These materials can neutralize free acids and convert them to their corresponding salts, which are less active as inhibitors. Dietary fat tends to enhance the activity of organic acids, probably by increasing their penetration into feed particles. Certain unknown factors in corn also alter the effectiveness of organic acid inhibitors.

When mold inhibitors are used at the concentrations typically recommended, they, in essence, produce a period of freedom from mold activity. If a longer mold-free period is desired, a higher concentration of inhibitor should be used. The concentration of the inhibitor begins to decrease almost immediately after it is applied as a result of chemical binding, mold activity or both. When the concentration of the inhibitor is reduced until it is incapable of inhibiting mold growth, the mold begins to use the inhibitor as a food source and grows. In addition, feeds heavily contaminated with molds will require additional amounts of inhibitor to achieve the desired level of protection.

The heat that the feed undergoes during pelleting enhances the effectiveness of organic acids. Generally, the higher the pelleting temperature, the more effective is the inhibitor. Once mold activity commences in pellets, however, it proceeds at a faster rate than in non-pelleted feed because the pelleting process that makes feed more readily digestible by animals also makes it more easily digested by molds.

The effectiveness of copper as a mold inhibitor is difficult to document. Although copper sulphate in the diet has been shown to improve bodyweight and feed conversion in broilers, excessive levels of copper may be toxic to young animals and will accumulate in the environment. In addition, recent research has indicated that feeding copper sulphate to poultry causes the formation of mouth lesions similar to those formed by some mycotoxins. Similar mouth lesions might be formed in other animal species as well.

The possible use of inorganic binders (mineral clays) to bind mycotoxins and, prevent them from being absorbed by the animal's gut has recently received much research attention. These clay products (including zeolites, bentonite, bleaching clays from refining of canola oil and hydrated sodium calcium aluminosilicates) have been shown to change the response of rats to zearalenone and T- 2 toxin. However, it should be clearly understood that the binding of some mycotoxins may be weak or non-existent and clay products differ in their ability to bind mycotoxins. Nonetheless, many clay products are generally recognized as safe (GRAS) and are used as anti-caking or free-flow additives for feeds.

Since mycotoxins are not evenly distributed in grain or mixed feeds, it is difficult to take sample of a feed or grain that will give a meaningful result in mycotoxin analyses. Grab samples generally give very low estimates of mycotoxin content. In fact, nearly 90% of the error associated with mycotoxin assays can be attributed to how the original sample was collected. This is because only 1-3% of the kernels in a contaminated lot contain mycotoxin, and these contaminated kernels are usually not evenly distributed within the grain lot.

For whole grains, a properly taken composite sample of at least 10 lb. is required for a reasonably accurate mycotoxin analysis. Trucks can usually be sampled with a grain probe, but bins must often be sampled as grain is being withdrawn.

Screening of corn for possible aflatoxin contamination using a "black light" was a popular technique 15-20 years ago. In spite of the widespread use of black lighting to screen for aflatoxin and other mycotoxins, research has shown that the technique detects materials that are not mycotoxins, and is, therefore, inappropriate. The black light test should never be used for any kind of mycotoxin screening.

The minicolumn is a small column that contains silica gel and adsorbents to which sample extracts are applied for detection of aflatoxin. Minicolumns were also very popular for aflatoxin screening until antibody-based test kits became widely available during the last few years. If properly used, the minicolumn test is capable of giving good results for aflatoxin under certain conditions. However, like the black light, it has often been mishandled and misused. The minicolumn is no longer recommended.

Analytical techniques for the detection of mycotoxins continue to improve. Several commercial laboratories now test for a variety of mycotoxins. Although analytical costs can be a constraint, these costs may be insignificant compared to the economic consequences of production and health losses associated with mycotoxin contamination. Commercial antibody test kits for screening or quantification are currently available for aflatoxin, zearalenone, deoxynivalenol, T-2 toxin, ochratoxin A and fumonisins. These antibody methods, while still being improved, are good if properly used.

Control of fat rancidity

Why is control of rancidity important?

Fats or high-fat ingredients are added to feeds to provide energy, provide essential fatty acids, solubilise fat soluble vitamins, improve palatability, minimize dust and provide lubrication. However, along with these benefits, fats can introduce problem associated with oxidative rancidity.

Oxidative rancidity of feed fats can reduce metabolisable energy, destroy fat-soluble vitamins and

reduce palatability. In severe cases, rancidity can cause severe symptoms such as muscular dystrophy or necrotic tissue in various organs.

Oxidative rancidity is a chemical reaction in which oxygen attacks a weak point (a double bond) in the fat, forming a peroxide. These peroxides can attack other fat molecules that lead to a chain reaction as well as to the destruction of fat-soluble vitamins and other nutrients. When most of the fat molecules have formed peroxides, the chemical reaction progresses to the formation of other compounds such as ketones, aldehydes and short-chain organic acids. The oxidative rancidity process is catalysed or helped along by metal ions (particularly copper) and gains momentum as it progresses. Thus it is essential to arrest the process early and /or not allow it to begin.

The oxidative rancidity process is slowed drastically by antioxidants in fats. Antioxidants protect fat by providing a chemical compound with which oxygen or peroxides may react. The reaction of oxygen with antioxidants protects the fat but uses up one “unit” of antioxidant protection for the fat.

How rancidity is measured?

The extent to which fats are rancid is generally measured by the initial peroxide value (IPV) test. Although many laboratories perform the IPV test, results of this test can be confusing and difficult to interpret. The IPV test is reported in “milliequivalents”. A milliequivalent (meq) is a unit of measurement that allows the measurement of the degree of oxidation. While there are numerous opinions as to which IPV unit represents rancidity in fats, all seem to agree that fat testing 20 meq is rancid.

The IPV test measures the degree of rancidity at the time the test is performed. If fats are unprotected by antioxidants, rancidity values can change rapidly. As the process of oxidative rancidity continues in fats, IPV increases rapidly, plateaus and then declines. Once oxidative rancidity begins to decline, the chemical processes have progressed to another "level" of chemical deterioration.

The degree to which fats are protected from rancidity is generally measured by the active oxygen method or AOM test. This test involves performing an IPV test followed by bubbling air through the fat sample and running a second IPV test. In some applications, the AOM test involves bubbling air through the fat sample and measuring the length of time required for the sample's IPV to reach 20 meq (rancidity). However, the more common method of conducting the AOM test is to bubble air through the sample for 20 hours and then report the sample's IPV.

Control of rancidity in fats.

The prevention of oxidative rancidity involves the following steps:

1. Establish specifications with fat or ingredient suppliers, which would include specifications regarding the addition of antioxidants to fats and high-fat ingredients.
2. Collect fat samples at delivery and have the samples analysed by a reputable lab.
3. Avoid fat suppliers who have supplied off-quality fats in the past.
4. Store liquid fats in clean tanks. Tanks that have not been regularly cleaned may contain a variety of materials that promote rancidity.
5. Do not expose liquid fats to pipes or fittings containing copper (such as brass).

Control of chemical residues

National survey have indicated consumers perceive the hazard, associated with chemical residues in food to be more serious than that of most other food-related issues (Gibbons etal., 1996). Since chemical residues in foods often occur via feed, it is important to address the issue of chemical residues in feed.

Chemical residues occurring in food are generally of two broad types: antibiotics (or antimicrobials) and industrial chemicals, which would include pesticides. Since Food & Drug Administration regulations are aimed primarily at the documentation of control of antibiotics within the feed mill environment, the issue needs no further discussion except to say follow the regulations.

FDA routinely tests for 354 pesticides in foods and feeds. In 1998, 1.7% of the feed or feed-ingredient samples tested by FDA contained illegal chemical residues. Chemical residues were most often found in animal by-products, grains and plant by-products (FDA, 1998). In view of the fact that the finding of a significant chemical residue in food can cause extensive damage to the reputation of a food company as well as a feed company, it is important to address the issue of chemical residues in feeds. Furthermore, experience from previous decades has shown that those who ignore chemical residue issues may be forced to destroy literally millions of pounds of meat, milk or eggs.

As most pesticides in use today have very short half-lives and do not accumulate in animal tissues, it would appear that most of the risk of chemical residues linked to feed would be associated with older chemicals that may still be present within the agricultural environment. Therefore, residue avoidance programs for feed should address the possibility of contamination with older chemicals that have the potential for bioaccumulation within animals consuming those feeds.

Although chlorinated hydrocarbon pesticides, such as DDT, dieldrin and chlordane, were banned from widespread use more than several

decades ago, some of these materials apparently still exist and are occasionally used illegally. In addition, industrial chemicals, such as polychlorinated bi- phenyls (PCBs), are present in antiquated equipment; thus, chemical residue avoidance testing in feeds should include tests for both chlorinated hydrocarbons and PCBs.

Chemical residue avoidance programs in feed involve two steps:

1. regular testing and
2. sample retention.

Regular testing of feed ingredients will ensure that chemical residue problems are detected quickly while retention of feed ingredient samples provides companies with a means to track contamination to its source.

Regular testing of feed ingredients for chemical residues should involve testing all major feed ingredients, including liquid fat. Since testing of each individual feed ingredient load for chemical residues can be an expensive proposition, individual ingredient samples may be composited, provided testing procedures are sufficiently sensitive to detect chemical residues.

How long should feed ingredient samples be retained? As a general rule of thumb, it would appear that feed ingredient samples should be retained until products from the animals that consumed the feed have been in consumer channels for two weeks, this procedure will provide feed companies with invaluable information and documentation in the event of a chemical residue incident.

Control of pathogenic microorganisms

Sampling for microorganisms in the feed mill.

Sampling is an often over-looked area when gathering information about pathogens in the feed mill environment. Certainly, the collection of adequate samples that represent the batch being sampled is important. However, a more basic question must be addressed. Are we certain that the contamination detected in the feed came from the sample or from the hands of person collecting the sample?

At one feed mill facility, mill personnel were instructed to collect samples while researchers collected samples from many of the same locations. The data from this study are shown in Table 1. A total of 43.75% of the samples collected by the mill personnel were positive for salmonella, while only 7.32% of samples collected by the researchers were positive. These data suggest that proper sample collection is a must if one wants to have a true assessment of contamination.

1. Sampling for Salmonella at feed manufacturing facilities

	-Mill personnel collected-			-Researcher collected-		
Sample Type	Number Run	Number positive	% positive	Number Run	Number positive	% positive
Mash	14	4	28.57	5	1	20.00
Cooler	14	2	14.28	6	0	00.00
Load out	14	5	35.71	19	0	00.00
Meat meal	14	12	85.71	2	2	100.00
Fish meal	8	4	50.00	2	0	00.00
Corn/wheat	8	1	12.50	1	0	00.00
Liquid fat	8	7	87.50	6	0	00.00
All samples	80	35	43.75	41	3	07.32

While a variety of methods exist for dealing with the issue of cross contamination, perhaps one of the simplest is one developed by Jim Andrews of Holly Farms (now Tyson). Paper cups are purchased in a plastic bag. Mill employees are instructed not to touch samples and to keep cups tightly closed within the plastic bag when not in use. Samples are collected only in new paper cups. Cups are used only once and then discarded. Samples are placed in sterile plastic bags following collection for transport to the laboratory. Although simple, this method is quite effective at preventing cross contamination.

Steps toward control of microorganisms in the feed mill.

Control of microbial pathogens in feeds and feed mills involves procedures to

(1) exclude pathogens from the feed,

(2) prevent multiplication of the organism in the feed and

(3) kill pathogens within the feed and prevent recontamination.

It should be clearly understood that feed milling processes are incapable of killing certain pathogens (i.e., spore formers). Thus, these pathogens MUST be excluded for control, Furthermore, even when feed mill processes destroy pathogens, higher numbers of these pathogens in feeds require even harsher treatments. Harsher treatments cause nutritional damage to the feed as well as costing more. Thus, in reality, each of these control procedures is interdependent and must be pursued simultaneously.

Basic steps in excluding the pathogens from feed.

Pathogens may enter feed through any number of routes; however, the primary routes for entry to the feed are through ingredients, vermin within the mill or cross contamination in the mill. Ingredient quality is important as far as nutritional composition is concerned and is also important in terms of microbiological quality. Ingredients are major source of pathogen entry into the feed.

1. **Obtain clean ingredients:** The nutrients that allow animals to grow are also the nutrients that allow pathogens to survive and, in certain situations, to multiply. Animal proteins are often considered high-risk products as far as pathogen incidence is concerned. However, contamination of oil seed meals, such as soybean, cottonseed, rapeseed, palm kernel and canola, has also been observed (Williams, 1995). In addition, contamination has also been reported in grain and grain by-products, such as wheat midds (MAFF, 1993). However, any ingredient can be contaminated. Therefore, it is important that all feed ingredients be obtained from reputable suppliers that implement pathogen control measures.
2. **Verify ingredient quality:** It is important to thoroughly check grains and other ingredients for signs of infestation, such as bird or rat faeces, that can carry pathogens, before accepting any ingredient. Bagged and bulk ingredients should be

visually inspected on arrival for signs of moisture penetration, insects or rodent attack. Incoming trucks and rail cars should also be checked for cleanliness. Affected bags or bulk ingredients should be refused if standards are not met.

3. **Maintain a clean receiving area:** Feed spillage around the receiving area should be cleaned up immediately to ensure that there is no feed material to attract birds and rodents. Once ingredients have been accepted, care should be taken to prevent contamination in the receiving area. This area should be free of pests, have a hard-surfaced floor and be well drained and covered.

Catwalks in the receiving area collect dust and can be a source of contamination. Air currents and machine vibrations can cause accumulated dust and dirt to drop from the catwalks directly into feed ingredients. Regular cleanups of catwalks, false ceilings, overhead beams and girders will reduce the potential for pathogen contamination from these sources.

4. **Control Dust:** No dust or caked material should be present at any point within the mill, as these materials can provide a media for pathogen survival or growth. A dust collection system is important, as it will keep the amount of dust down. Venting to the outside, separate from intake, will remove potentially contaminated dust. The installation of filters capable of ensuring that dust laden air is not being drawn in through the ventilation system will minimize recirculation of potentially contaminated dust.

Filters should be installed on intakes through which air is being drawn to cool pellets. These filters will prevent air that is contaminated with pathogens from contaminating pellets. A schedule should be established, according to manufacturer's instructions, to replace filters frequently and routinely. Circulating air from the finished product area to the raw ingredient area will also minimize air-born contamination.

5. **Clean up feed spills:** Feed spills anywhere in the feed mill can provide a medium for pathogen spread and are a source of recontamination, as they can be tracked to other areas. This material should be thrown out, or if clean, quickly recovered. While this material can be reprocessed (pelleted), caution should be used since wet materials can encourage pathogen multiplication. Always discard wet material.

6. **Proper feed storage:** Separate bulk bin storage should be designated for either mash or pelleted feeds to prevent cross contamination of pellets by residues of feeds that have not been heat-treated. Finished products should always be packaged in new or properly sanitized bags since pathogens can survive for months on bag material.

In addition, it must be recognized that pathogens survive for long periods in the feed mill environment. Thus ingredients should be stored in structures, containers or bins that will keep out moisture. Specific bins should be designated exclusively for storage of high-risk ingredients, such as meat and bone meal. If possible, dedicated delivery lines for these high-risk ingredients are preferable.

Prevent multiplication of pathogens in feed.

The lack of moisture is the primary reason pathogens do not rapidly multiply in feeds. Thus the primary task in preventing pathogen multiplication is moisture control. Obivious sources of moisture, such as roof leaks, uninsulated pipes or areas where wind can blow rain in, must be eliminated. It should also be recognised that water should not be used to clean feed manufacturing facilities, unless there is no other alternative.

Contamination of conveying equipment may be associated with areas of high humidity or moisture. In feed mills utilising a heat-treatment process (e.g. pelleting or extrusion), the environmental conditions (i.e. temperature and humidity) of the cooler are ideal for the establishment of a microcosm of bacteria and mold. Dust and feed particles adhere to the internal surfaces of the cooler and become media for pathogen growth. As feed passing through the coolers comes into contact with these particles, the feed becomes contaminated, and this contamination spreads throughout the downstream conveying system. This route of contamination of feed/feed ingredients occurs not only in feed mills, but also in rendering plants, vegetable oilseed plants and blending operations. Implementation of a dry-cleaning process or disinfection of equipment often aids in addressing the problem.

Kill pathogens in feed and prevent recontamination.

Temperatures of 250°F (122°C) for 15 minutes are required to kill certain spore-forming pathogens. Clearly, it is virtually impossible to meet these time and temperature requirements during the feed manufacturing process. Therefore, spore-forming pathogens such as clostridium and bacillus must be dealt with in some alternative fashion.

There are only two practical methods to reliably kill pathogens in feed:

1. pelleting or extrusion, and
2. chemical treatment of the feed.

As previously mentioned, as pathogen numbers increase, longer times, elevated temperatures or higher chemical levels will be required to eliminate the organism from feed.

The pelleting process is effective at reducing the isolation rates of certain pathogens, but pelleting does not eliminate pathogens from feed and feeds can be recontaminated after the pelleting process. Although pelleting is effective at killing most salmonella in feeds, the pelleting process is highly dependent on the formula.

Certain formulas are capable of receiving tremendous quantities of heat, while others can receive little or none. Extrusion or expansion overcomes some of the difficulties of

pelleting and it involves much higher temperatures than pelleting. Thus, extrusion or expansion should be more effective than pelleting at killing pathogens. Nevertheless, regardless of the temperature, feeds exposed to heat treatment (either pelleting or extrusion) must be cooled to remove excess heat and moisture. As Shrimpton (1989) has shown, the cooling process can recontaminate feeds with pathogens, reducing the benefit of the heat-treatment process. The pelleting or extrusion process has no residual activity, so feeds can be easily recontaminated at any time they are exposed to pathogens.

Chemical preservatives have also been utilized to kill pathogens in feed and feed ingredients. Garland (1994) listed the products available for inhibition of pathogens in animal feeds. Most of these products listed contained propionic or formic acid or salts of these acids. The propionic acid products were recommended at about 3 Kg per ton, while the lone formic acid product mentioned was recommended at 6.8 Kg per ton. Although Garland was probably simply cataloguing manufacturers recommendations for the products listed and manufacturers can and do test products for efficacy, the method used for evaluating chemical preservatives can greatly influence the outcome of efficacy tests.

Westerfield et al (1970) demonstrated that propionic acid had no significant inhibitory effect on pathogens in unsterilised poultry feed, but was completely inhibitory when the same feed was sterilised. While no one in the commercial poultry industry uses sterilised feed, Rouse et al (1988) utilised sterile feed to test a commercial propionic acid product. Interestingly, even in these unrealistic test conditions, they showed that a level of 5 kg per ton was effective at eliminating pathogen levels of 10^2 , but not 10^6 bacterial cells/g of dry feed.

Vanderwal (1979) tested the ability of a number of fatty acids to kill enteric organisms in unsterilised feed with moisture of less than 16%. These data are similar to those of McCubbine (1989), who demonstrated that when chickens are fed feeds containing pathogens under controlled conditions, a dose response relationship exists with formic acid and the incidence of the pathogen isolation in caecal contents.

While encouraging, such data does not account for every eventuality . It is not uncommon for feed moisture to increase following manufacture. This moisture may be from roof or storage tank leaks or may result from condensation. Smyser and Snoyenbos (1979) demonstrated that when meat and bone meal become moist, none of the 11 (eleven) organic acids tested prevented the multiplication of pathogens. They found that only formaldehyde 0.1% prevented the multiplication of pathogens.

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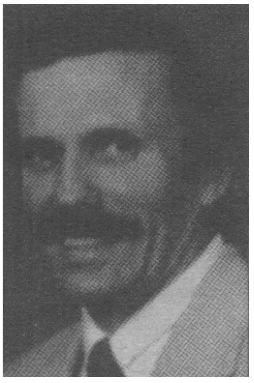

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