CELL WALL CARBOHYDRATES
Livestock

Executive Summary

Cell wall carbohydrates desired for organic feed are derived from the yeast species *Saccharomyces cerevisiae*. Commonly known as baker's yeast, it has been used for many years in the food industry particularly in breads and alcoholic beverages such as wine and beer. Mannan Oligosaccharide makes up approximately 50% of the yeast's carbohydrate content found in the cell wall.

Farmers are petitioning the use of cell wall carbohydrates as a feed additive, blended with botanicals and biologics into powder, capsules, and boluses for use primarily in bovines, and suitable for other livestock as well. Their petition states that it will be used as an adsorbant toxin bunder with typical dosage levels as part of a blended product consisting of about 1/8-1/2 an ounce per day in cattle feed and mixed into the daily ration.

Cell wall carbohydrates (CWC) are used as a dietary toxin binder, primarily focusing on the mycotoxins commonly found in fungus that grows in areas of moisture. Farmers also wish to use cell wall carbohydrates, derived from non-irradiated and non-GMO yeast (*Saccharomyces cerivisiae)*, as a toxin binder for pathogenic toxins as well. Cell wall carbohydrates have proven to be a highly effective toxin binder which does not tie up critical dietary immune supporting nutrients as is the case with other toxin binders.

From the farmers' petition, toxemia is a significant problem in livestock and is a predisposing factor for many secondary infections that are currently being addressed with antibiotics, i.e. high somatic cell count, mastitis, dysentery, poor component milk, calf scours, and off-feed. It is estimated that over 25% of the world's grain supply can produce toxemia in livestock, making toxemia a growing problem and it is important for organic producers to have an effective natural tool to address this issue of relying on antibiotics to treat secondary problems.

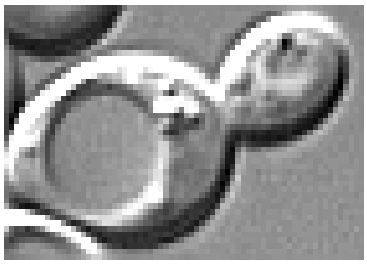
With regards to safety, the FDA has already considered *Saccharomyces cerevisiae* as safe for use in food production. Baker's yeast has particular properties that cause bread to 'rise' and beer to ferment. The discussion concerning whether or not the cell wall carbohydrate derivative wished for use in feed is synthetic or not has yet to be determined, but from a general perspective, yeast and its derivatives should be safe for the environment, the animals, and the human consumers as well.

Summary of TAP Reviewer's Analyses[1]

Synthetic/ Nonsynthetic	***Allow without restrictions?***	***Allow only with restrictions? (See Reviewers' comments for restrictions)***
Synthetic (2) Unable to determine (1)	No (3)	Yes (2) No (1)

[1] *This Technical Advisory Panel (TAP) review is based on the information available as of the date of this review. This review addresses the requirements of the Organic Foods Production Act to the best of the investigator's ability, and has been reviewed by experts on the TAP. The substance is evaluated against the criteria found in section 2119(M) of the OFPA [7 USC 6517(m)]. The information and advice presented to the NOSB is based on the technical evaluation against that criteria, and does not incorporate commercial availability, socio-economic impact, or other factors that the NOSB and the USDA may want to consider in making decisions.*

Identification

 [2]

Chemical names: Cell wall carbohydrates from *Saccharomyces cerevisiae* yeast.

Other Names: CWC, EGM, MOS, commonly known as baker's yeast.

Characterization

Composition:

Saccharomyces cerevisiae is a yeast species which has been domesticated for at least 3000 years. It stands out from most other species by its high resistance of ethanol. A small group of closely related species is used in the production of beer, wine, bread, and yeast extract (microbiological ingredient or condiment). *Saccharomyces cerevisiae* also is an important model system in genetics. Most enzymes of intermediary metabolism were first discovered and characterized in "yeast". This species was also the first eukaryote whose entire genome was sequenced.[3]

Properties:

Yeasts are microscopic fungi -- single-cell organisms of the plant kingdom which are generally about 5-10 microns in size.[4]

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9. Physical and chemical properties
9.1 Appearance: Powder
9.2 Colour: Toasty
9.3 Odour: Typical yeast odour
9.4 Changes in appearance: -
9.5 Freezing point: N/A
9.6 Boiling point/boiling range: N/A
9.7 Flash point: -
9.8 Autoflammability: -
9.9 Explosive properties: -
9.10 Specific gravity: 0.6 – 0.7
9.11 Solubility/miscibility in water: N/A

[2] http://dir.nichd.nih.gov/Interest_Groups/yeast/index.html
[3] *Eumycota: Phylum Dikaryomycota* http://instruct.uwo.ca/biology/318b/318lab02.htm#Lab%206
[4] Directly referenced from http://www.diamondv.com/articles/booklet/booklet.html

9.12 pH (concentrate) (10 g/L) at 20°C: 5.0 – 5.5
9.13 Further instructions: -

[5]

How Made:

Yeasts such as *Saccharomyces cerevisiae* are naturally existing single-celled fungi that multiply by **budding**, or in some cases by division (**fission**), although some yeasts such as *Candida albicans* may grow as simple irregular filaments (**mycelium**). They may also reproduce sexually, forming asci which contain up to eight haploid **ascospores**. *Saccharomyces cerevisiae* has thick-walled, oval cells, around 10 µm long by 5 µm wide.[6]

Specific Uses:

Yeast usage has found applications in many areas. One particular area of interest is in cattle grazing fescue pastures. Much of the eastern and southern United States has endophyte-infected fescue as the main source of forage protein and energy. While new lines of endophyte-free fescue exist, it is unlikely that there will be wide-spread replanting of fescue areas. There is a renewed interest in year-round or extended grazing to reduce the feed cost of cow-calf production programs. Yeast products may assist in digestion of forages. The yeast *Saccharomyces cerevisiae* is produced by fermenting selected liquid and cereal grain raw ingredients with bakers yeast. Yeast delivery by means of a mineral supplement to improve animal performance would be less labor intensive than replanting a vast acreage of pastures.

Yeast cultures have been shown to positively affect animal performance and mineral consumption. Studies in Florida and California resulted in improved feed intake, production, and reduced rectal temperatures during summer heat stress in dairy. Other research trials have shown that yeast cultures have also increased rumen bacteria numbers and improved the digestion of feedstuffs in both beef and dairy animals. Both mineral consumption and absorption have been positively affected by the addition of yeast culture to free-choice mineral mixes. Finally one 1986 study showed improved weight gains in yeast culture fed cattle grazing fescue pasture.

As mentioned previously, much of the eastern and southern United States has endophyte-infected fescue as the main source of forage protein and energy. Yeast products, such as Saccharomyces cerevisiae, may assist in digestion of forages. In some studies pregnant heifers appeared to gain slightly more weight if they had access to a free-choice mineral supplement containing yeast when compared to a control mineral. In this particular study, however, the control heifers lost less weight during the interval of calving and peak lactation although this may have some relation to milk production differences. Cow-calf pairs consuming yeast-mineral mixes resulted in increased weaning weights. This is obviously a benefit.

In a study by Boyles and Co-workers at Ohio State University gestating heifers appeared to gain slightly more weight if they had access to a free-choice mineral supplement containing yeast than a control mineral (Table 1). There also appeared to be slightly more body-weight gain for the yeast- supplemented heifers compared to controls during early-spring grass growth.

[5] *Material safety data sheet (91/155/EEC)* http://www.begerow.de/pmm/english/sdb-pdf/hefen.pdf
[6] *Saccharomyces cerevisiae* http://www-micro.msb.le.ac.uk/video/Scerevisiae.html

Table 1. Analyses of Animal Body Weight Gain During Different Periods of the Year and Stages of Production.

	Control	Yeast	SE'
July-February Gestation Period, lb/mo	35.8	40.7	1.59
Feb-April Calving-Peak Lactation	-63.3	-76.1	4.70
April-June Spring Grass Growth	21.8	30.9	4.00

'Standard error.

The period of calving, peak lactation, and rebreeding is a very critical time in the production stage of beef cattle. In the same study, body condition was critically evaluated during the months of April, May, and June. Body condition based upon pounds to inches in height was found to be similar between control and yeast-supplemented cattle (Table 2). All heifers had hip height measurements of approximately 50 inches in April. Milk production in May was 15.4 and 15.6 lb per day for control and yeast-supplemented heifers, respectively (Table 2). Milk production for heifers consuming the yeast-mineral mix appeared to be greater. Weaning weights and weight per day of age appeared to be improved by availability of a yeast-mineral mix.

Table 2. Height, Weight-to-Height, and Milk Production of Spring-Calving Heifers Supplemented Yeast While Grazing Fescue.

	Control	Yeast
Height, inches	49.8	49.6
April, Weight/Height (lb/in)	18.4	18.6
May, Weight/Height (lb/in)	18.8	19.5
June		
Milk[1], lb/day	15.4	15.6
Milk[2], lb/day	9.6	12.9
Weaning Weight[3], lb	382.1	408.9
Weight/Day of Age[3], lb	1.9	2.0

[1]Based on weigh-suckle-weigh measurements, 5/28/98.
[2]Based on weigh-suckle-weigh measurements, 8/26/98.
[3]Adjusted for birth weight.

Finally, this study showed that yeast inclusion increased total mineral supplemental intake. Total supplemental mineral intake was 0.23 and 0.40 lb per day for the control and yeast-mineral, respectively. The yeast-mineral intake was 4.8 ounces per day, and the total yeast consumption per day was 1.2 ounces per day. The difference in total supplement intake between treatments was 0.19 ± 0.072 lb per day.

From a different perspective, use of yeast has been shown to have a positive influence on intake in newly received stocker and feedlot cattle. Yeast appears to be useful in reducing stress effects in these cattle and has been shown to be of benefit in getting fresh cattle started on feed somewhat faster.

As you can see, evidence exists that use of yeast cultures does have a positive influence on cattle performance.

Practical Considerations for DFM

In general, most would agree that DFM based on bacteria must be "live." In light of this, they must survive processing, storage and the gut environment. In contrast, the need to provide a high number of "live" yeast *(Saccharomyces cerevisiae)* has been the subject of many debates. As previously mentioned, some products guarantee live yeast cells (e.g., 1 ´ 109 cfu per g) and are fed at low inclusion rates (only 10-20 grams per day) but other products suggest that live organisms are not required for beneficial effects. The metabolites present in the culture extracts have been suggested to be the "active" ingredients. One study reported that heating (such as in the pelleting process), but not irradiation, decreased the ability of an *Aspergillus oryzae* extract to stimulate rumen bacterial growth and activity. Another trial reported that the stimulatory effect of yeast on numbers of rumen cellulolytic bacteria was diminished when yeasts were heated. The debate on the need for live yeasts will continue unless more definitive studies addressing this issue are conducted.

Direct-fed microbial products are available in a variety of forms including powders, pastes, boluses, and capsules. In some applications, DFM may be mixed with feed or administered in the drinking water. However, use of DFM in the latter manner must be managed closely since interactions with chlorine, water temperature, minerals, flow rate, and antibiotics can affect the viability of many organisms. Non-hydroscopic whey is often used as a carrier for bacterial DFM and is a good medium to initiate growth. Bacterial DFM pastes are formulated with vegetable oil and inert gelling ingredients. Some fungal products are formulated with grain by-products as carriers. Some DFM are designed for one-time dosing while other products are designed for feeding on a daily basis. However, there is little information comparing the efficacy of administering a DFM in a single massive dose compared to continuous daily dosing. The need for a bacterial DFM to actually attach and colonize gut surfaces in order to have a beneficial effect is also questionable. However, in certain applications, the argument could be made that a DFM organism need only produce its active component (without colonization) to be beneficial. Additionally, dose levels of bacterial DFM have varied. Studies can be found where *L. acidophilus* have been fed at levels ranging from 106 to 1010 colony forming units (cfu) per animal per day. A 1980 study suggested that feeding more than 107 cfu per head per day may cause lower nutrient absorption due to overpopulation of the gut.

Tolerance of DFM microorganisms to heat is important since many feeds are pelleted. In general, most yeast, *Lactobacillus*, *Bifidobacterium*, and *Streptococcus* are destroyed by heat during pelleting. In contrast, bacilli form stable endospores when conditions for growth are unfavorable and are very resistant to heat, pH, moisture and disinfectants. Thus, bacilli are currently used in many applications that require pelleting. Over-blending can sometimes compensate for microbial loss during pelleting, but this is not an acceptable routine practice. Future improvements in strain development may allow use of heat-sensitive organisms in pelleted feeds. Bacterial products may or may not be compatible with use of traditional antibiotics and thus care should be taken when formulations contain both types of additives. For example, some species of bacilli are sensitive to virginiamycin, and lactobacilli are sensitive to chlortetracycline and penicillin. Information on DFM and antibiotic compatibility should be available from the manufacturer.

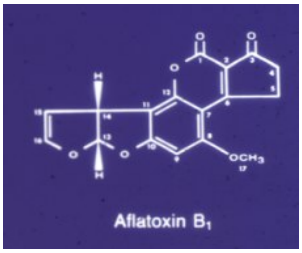
Viability of DFM products has improved over the past several years but it is highly advisable to adhere to storage recommendations. For example, products should be kept away from moisture, excess heat, and light. Future research on new DFM products will need to address viability if oxygen sensitive microorganisms are to be developed for commercial purposes.

Conclusions

The use of microbial products has been shown to have merit. Obviously research is still required to better grasp application and how cattle respond to microbial product feeding in different situations.[7]

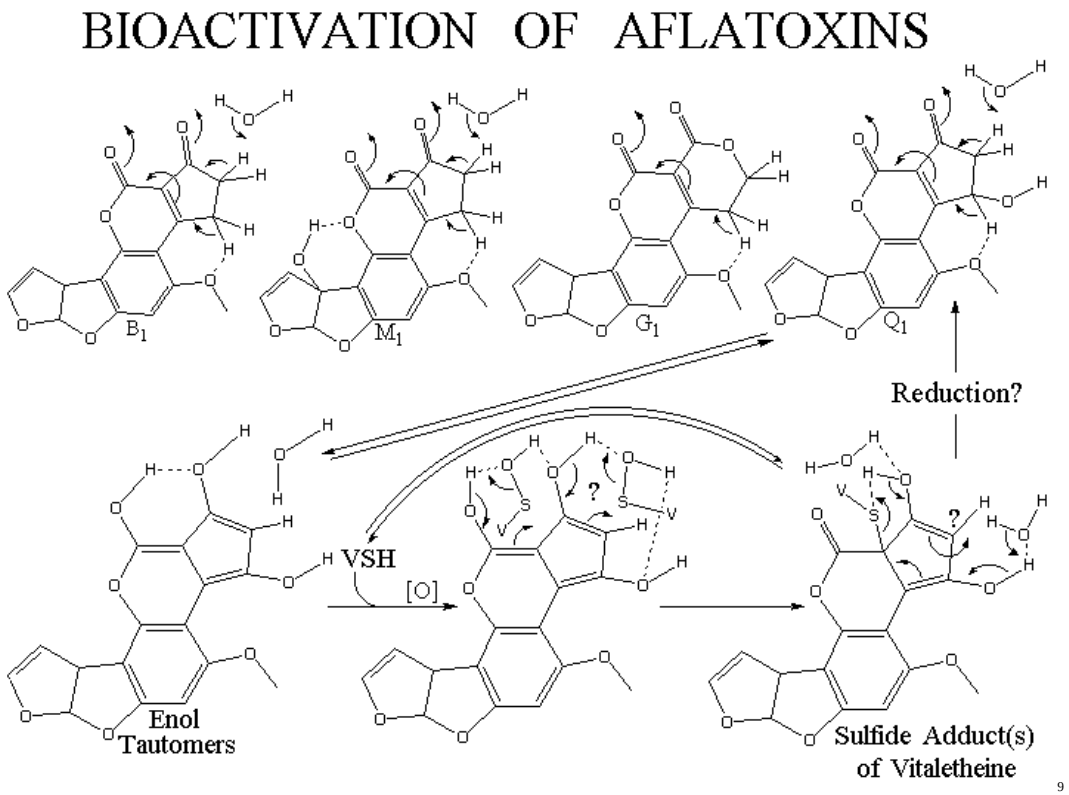
Action:

Aflatoxins are naturally occurring toxins that are metabolic byproducts of fungi, *Aspergillus flavus*, and *Aspergillus parasiticus*, which grow on many food crops under favorable conditions.
Mycotoxin literally means poison from a fungi and are named on the basis of the fungus that produces them, thus "Aflatoxin" uses the "A" for Aspergillus and "fla" for the species "flavus" along with the word toxin.[8]



Probable Reactions of Carcinogenic Aflatoxins with the Vitaletheine Modulators

Aflatoxins can undergo many of the same reactions with sulfenic acids characteristic of reactions with dimedone or vitamin C. Adjacent keto- groups make any hydrogen on the intervening carbon acidic, favoring tautomerization of this structure to an enol. Like the enol tautomer of dimedone, these probably react with sulfenic acids to alkylate the sulfur, thereby generating a sulfide-linked adduct.



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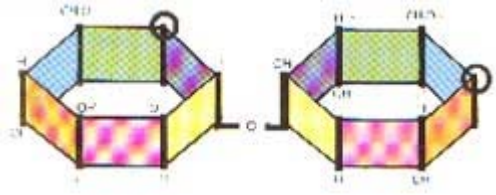
[7] *YEAST USAGE CAN POSITIVELY AFFECT FEEDING AND SUPPLEMENTATION* http://www.cattletoday.com/archive/2002/February/CT188.shtml
[8] *Food Safety Research: A Focus On Aflatoxin Contamination* http://www.nal.usda.gov/fsrio/research/fsheets/fsheet01.htm
[9] *Aflatoxins and Carcinogenesis Through Alkylation of Vitaletheine Modulators?* http://www.highfiber.com/~galenvtp/vtlafltx.htm

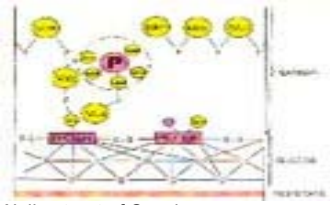
Aflatoxin, the fungal **carcinogen** first identified in 1960, is now recognized as the prototypical laboratory **carcinogen**. It causes mutations in the p53 tumor-suppressor gene as well as mutations which are involved in the majority of human cancers. Aflatoxin has been shown to contaminate tobacco products. Tobacco-related cancers, including those associated with ETS, often show the same p53 mutations associated with aflatoxin exposure. The US Food and Drug Administration (FDA) does not regulate aflatoxin contamination of tobacco. Aflatoxin was first identified in 1960 as one of the most potent carcinogens known, and has been recognized as a teratogen, mutagen, **carcinogen**, immunosuppressant, and potent inhibitor of protein synthesis. Aflatoxin is likely accelerating the spread of AIDS through commodity and tobacco contamination. The FDA began regulating aflatoxin on agricultural commodities, such as peanuts, corn, and grains, in 1966. Federal and state laws prohibit interstate shipment of contaminated aflatoxin commodities exceeding 20 parts per billion (ppb).[10]

MANNAN-OLIGOSACCHARIDE

This unique oligosaccharide is derived from Paecilomyces sp. and *Saccharomyces cerevisiae*. It can effectively bind and absorb various pathogens; thus, blocking the colonization of pathogens in the Gastrointestinal (GI) Tract and reduce their infection.



Mannan-oligosaccharide



Cell Wall extract of Saccharomyces. cerevisiae

Live Saccharomyces cerevisiae yeast culture (minimum 1.0 x 10^{11} CFU / Kg). Live yeast cultures are rich in enzymes, fatty acids, and Vitamin B complexes and unknown growth factors, which stimulate the activity and proliferation of cellulytic bacteria. Yeast cultures can also absorb mycotoxins and improve the digestion and absorption of minerals including Phosphorus, Magnesium, Calcium, Copper, Potassium, Zinc, and Manganese. Yeast by itself is also a probiotic to expel pathogens.[11]

M.O.S.500: a naturally derived extract from the cell wall of Saccharomyces cerevisiae, is a food grade ingredient and fermentation additive. The mannan oligosaccharide content is approximately 50% of the carbohydrate fraction. MOS is a Mannanoligosaccharide derived from the cell wall of the yeast Saccharomyces cerevisiae. Mannan is a sugar recognized by certain bacteria, including many strains of E. coli and salmonella. In the oligosaccharide form however, the mannan is not available for the pathogen to grow. When MOS is added to calf diets, lectins of these pathogens are tricked into attaching to the mannan sugar instead of the carbohydrates attached to the intestinal villi. These lectins are then flushed out without being able to metabolize the sugar, (see diagram) resulting in a "cleansing" effect of the intestinal wall and preventing permanent damage to the villi (finger-like protrusions on the intestinal wall containing sights for nutrient absorption). This allows improved animal performance.[12]

One mode of action for mannan-based oligosaccharides involves interference with colonization of intestinal pathogens. Cell surface carbohydrates are primarily responsible for cell recognition. At the simplest level is the role of carbohydrates in blood types which are differentiated by cell coat sugars. Bacteria have lectins (proteins or glycoproteins) on the cell surface that recognize specific sugars and allow the cell to attach to that sugar. These sugars can be found on the epithelial cell surface. Binding of Salmonella, Escherichia coli and Vibrio cholera has been shown to be mediated by a mannose-specific lectin-like substance on the

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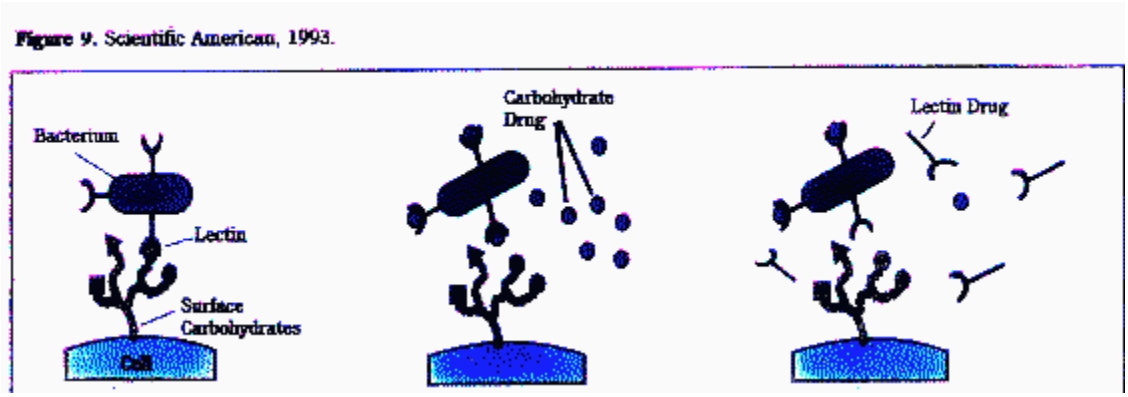
[10] *MONITORING AND REMEDIATION OF AFLATOXIN AND MYCOTOXIN LEVELS ON TOBACCO AS A HARM REDUCTION STRATEGY* http://216.239.51.100/search?q=cache:T_7GClSLYegC:www3.who.int/whosis/fctc/Submissions/F3940387.doc+mycotoxin+as+a+carcinogen&hl=en&ie=UTF-8

[11] *CENMOS: Naturally Binds Pathogens and Mycotoxins in the Feed.* http://www.cenzone-europe-turkey.com/htm/en/e_cenmos.htm

[12] Directly referenced from http://savacaf.com/library/t00144.html

bacterial cell surface.[13] Mannan-oligosaccharides are thought to block the attachment of pathogenic bacteria to the animal's intestine and colonization that may result in disease, while acting as a nutrient to other beneficial bacteria. It is also thought to stimulate the animal's immune system, thereby further reducing the risk of disease.[14]

Using just 2 grams per feeding in the milk replacer or 2-4 pounds per ton in the calf starter, cost is only $0.01 per feeding or about $0.50 per bag of milk replacer. MOS is an excellent and inexpensive way to naturally improve your calf program.



In addition to binding pathogens, Bio-Mos trials with calves have shown a statistically significant reduction in respiratory problems. This is appears to be because Bio-Mos modulates the immune system to increase macrophage and immunoglobin activity.[15]

Among the toxigenic species of the genus *Fusarium*, *F.proliferatum* is known to produce the largest spectrum of mycotoxins, including beauvericin, fumonisin, fusaproliferin and moniliformin. These secondary metabolites are produced within the cytoplasm and must be exported to the exterior to exert their toxic effects. ABC transporters are likely candidates for the secretion of these compounds. In S*accharomyces cerevisiae*, analyses of the entire genome sequence has revealed the presence of 29 ABC-transporter genes, that have been grouped into 6 classes based on the topology of the deduced proteins. Degenerate primers were designed that are based on the sequences from the genes in the two largest classes, and these primers were used to amplify PCR fragments from several toxigenic *Fusarium* spp. Sequence analyses revealed that these fragments show homology to different genes. Subsequently, a BAC library of *F. proliferatum* (constructed in pBeloBAC 11 and consisting of 10 genome equivalents with 50kb-100kb inserts was screened by PCR. Several positive clones were identified and sequence analysis revealed complete genomic copies of two ABC transporters, *Fp*ABC1 and *Fp*ABC2, with strong homologies to the so-called PDR- and MDR-like ABC transporters, respectively. Northern blot analysis was performed to study the expression of ABC1and ABC2 under different conditions including those inducive for mycotoxin production as well as under conditions of fungicide stress. Several ABC-like gene homologs derived from published EST sequences were also included in these experiments.[16]

The complete yeast genome sequence revealed the presence of seven ORFs whose predicted protein products show significant amino-acid sequence homology to the aryl alcohol dehydrogenase (AADH) of the lignin-degrading filamentous fungus, *Phanerochaete chrysosporium.* This paralagous gene set consists of 6 telomere-associated genes, each of which is on a different chromosome, that comprise a gene set. The seventh gene is at an internal site and is not relted to the 6-mameber family at the nucleotide sequence level. Enzyme assays demonstrated that *S.cerevisiae* has an aryl alcohol dehydrogenase activity and that, as

[13] Directly referenced from http://www.nutriteck.com/bulk/mosyeast.html
[14] Directly referenced from http://www.biomatrixinternational.com/prodsheet/ProdSheet%20MOS%20104.pdf
[15] Directly referenced from http://www.vigortone.com/probiotics.htm
[16] *ABC transporters in Fusarium proliferatum.* http://www.hgmp.mrc.ac.uk/research/fgsc/fungalgenetics2001/biochemabs.htm

in *P.chrysosporium*, this activity is elevated in stationary phase cultures. All seven genes have been deleted separately and the septuple mutant constructed. Biochemical analysis demonstrated that this septuple *AAD* gene deletant was completely unaffected in its aryl alcohol dehydrogenase activity. The *AAD* ORFs also display similarity with the *Aspergillus* norsolinic acid dehydrogenase, an enzyme in the aflatoxin biosynthetic pathway. For this reason, the possible involvement of these genes in sterol biosynthesis was investigated in collaboration with Steve Kelly (Node **N8**, lipid metabolism), but without positive result. The Norwich laboratory is currently investigated the possible involvement of these genes in polyketide biosynthesis and a more direct phenotype is being sort by investigating the sensitivity of the deletants to various inhibitors that are structurally related to aryl alcohols. Again, no positive resultsd were obtained. As a result of further *in silicio* analysis, we examined the response of the *AAD* genes to oxidative stress. The genes on chromosomes VI and X are expressed in response to glutathione antagonists. This response has been shown to be Yap1p-dependent. Since the ORF on chromosome VI is interrupted by a stop codon, the apparent seven-fold redundancy of the *AAD* gene-set reduces to one, in respect of the response to oxidative stress.[17]

Combinations:

Comparative feeding and group metabolic trials were conducted on sexed cockerels of ROSS hybrid to study the effect of biologicals containing mannan- oligosaccharides (b_1) and *Enterococcus faecium* M-74 (b_2) and their combinations (b_3) in starters BR1 (a single level of proteins) and in feed mixtures BR2 for broiler production with two levels of proteins (a_0 - 20.85%, a_1 - 18.22%), as exerted on growth, feed consumption and basic nutrient digestibility. The live weight of chickens receiving feed mixtures BR2 with lower protein level (a_1) was lower by 1.28% on day 35, and by 2.53% on day 42, than in group (a_0) with higher protein level. The differences were statistically insignificant. The average live weight of chickens at 21 days of age was higher by 2.3% - 2.2% in experimental groups b_1, b_2, b_3 in comparison with control (b_0). This difference was also statistically insignificant. The group of chickens **receiving the combination of mannan-oligosaccharides and *Enterococcus faecium*** M-74 showed the live weight higher by 4.44% at the age of 42 days than control (b_0) at ($P < 0.1$). The live weight of chickens was significantly ($P < 0.1$) higher when the bacteria *Enterococcus faecium* M-74 were used in diets BR2. This positive effect of biologicals on chicken weight was determined in diets BR2 with higher and lower protein levels. The statistically significantly ($P < 0.1$) lowest feed consumption per 1 kg of weight gain (expressed in kg) was recorded in the group of chickens (b_3) that received feeds with the combination of mannan-oligosaccharides + *Enterococcus faecium* M-74. The difference against control (b_0) was (- 4.87%) at 35 days of age and (- 4.34%) at 42 days of age. A significant difference ($P < 0.1$) was also calculated for total feed consumption per 1 kg of weight gain for feeding periods 1st to 35th day and 1st to 42 day of chicken age. Biologicals based on mannan-oligosaccharides and *Enterococcus faecium* M-74 had positive effects on the consumption of BR2 feeds at higher and lower protein levels. The effect of protein levels in BR2 diets on N retention and fiber digestibility coefficient was statistically significant. N retention was higher by 5.61% in groups of chickens receiving BR2 diets with lower protein level (a_1) at ($P < 0.05$). Fiber digestibility of this group was higher by 19.14% at ($P < 0.1$). Statistically significantly higher ($P < 0.05$) N retention (by 5.93%) was determined in the group of chickens receiving feeds with combinations of biologicals containing mannan-oligosaccharides + *Enterococcus faecium* M-74 (b_3) in comparison with control (b_0). Groups (b_2) and (b_3) had statistically significantly higher ($P < 0.1$) coefficients of fiber digestibility against control (b_0): by 13.14% and 14%, respectively. The lower percentage content of proteins in BR2 diets was reflected in lower N output in droppings. N output in groups of chickens receiving feeds with lower protein level (a_1) was lower by 10.03% (in g) against control (a_0). Lower average values of N output in droppings (in g) per 1 kg of weight gain were determined in groups of chickens receiving BR2 diets with the combination of biologicals based on mannan-oligosaccharides + *Enterococcus faecium* M-74 (b_3).[18]

Over 30 trials have looked at the ability of MOS to stimulate faster growth rates in calves and have shown positive results varying from 5 to 35% better growth rates. Many of these trials have been carried out on

[17] ***The aryl alcohol dehydrogenase (AAD) gene set*** http://mips.gsf.de/proj/eurofan/eurofan_1/n9/
[18] Directly referenced from http://www.vuvz.cz/old/English/Knihy/oligosachar.htm

university farms where the challenge is obviously lower and responses are typically lower. However, as the summary of 14 trials with 900 calves below shows, MOS has proven effective even in these cases.[19]

	No. Calves	Days	Control	MOS	Improvement
University of Tenn	48	28	25.24	25.63	10.3%
Institute Animal Nutrition, Poland	24	30 d	32.67	44.24	35.4%
North American Biosciences C.	29	35 d	27.95	37.27	33.3%
North American Biosciences C.	28	35 d	26.06	30.82	18.3%
R&L Veal, Ohio	67	42 d	57.32	64.92	13.2%
Nippei, Japan	17	42 d	59.52	76.94	29.3%
Milk Specialties	240	56 d	47.70	50.70	6.7%
North American Biosciences C.	36	56 d	74.07	78.70	6.3%
Federal University R.G.S.	24	56 d	45.67	53.08	16.2%
California State, Fresno	162	60 d	44.71	56.59	26.6%
University of Sao Paulo	36	60 d	41.03	50.22	22.4%
Continental Grain	96	60	58.33	62.96	7.9%
Colorado State University	53	63 d	63.38	66.53	5.0%
Measurement was based on Total Weight Gain (lbs) Mean Average Improvement					17.1%

Status

Historic Use by Organic Farmers:

Effects of Mycotoxins in Animal Feeds (NC State University)

Swine

Swine are sensitive to mycotoxins, especially nursing or nursery-age swine. In general, mycotoxins cause reductions in feed intake, growth performance, and immune function when levels are relatively low. Producers must be aware that if one toxin is identified in a sample, the chances are high that other toxins are present. Some toxins may not have been identified as of yet, but research on known mycotoxins provides insight into the expected effects in swine and potential methods to reduce those effects. Table 3 contains a summary of the maximum permissible concentrations of mycotoxins in swine feeds.

Aflatoxin B1 has been the most extensively studied. Twenty to 200 ppb will cause a decrease in feed intake and growth performance, which can be partially offset by increasing specific dietary nutrients such as lysine or methionine. In severe cases (1,000 to 5,000 ppb) of aflatoxicosis, one can expect acute effects

[19] Directly referenced from http://savacaf.com/library/t00144.html

including death. Aflatoxin M1 appears in milk of sows consuming aflatoxin-contaminated diets and may affect piglets nursing those sows.

Feed concentrations of deoxynivalenol (DON) of 300 to 500 ppb are often associated with feed refusal, decreased weight gain, and increased incidence of infectious diseases. DON levels greater than 1000 ppb, will cause feed refusal or decrease in feed intake resulting in severe weight loss. It appears that pigs will often consume a sufficient amount of contaminated feed to induce vomiting. In fact, DON is also called vomitoxin because of its association with swine vomiting.

T-2 toxin has detrimental effects on swine performance, but no effect levels have not been determined for commercial production environments. However, field observations indicated that T-2 and related compounds are associated with decreased productivity at feed concentrations of 200 ppb or less.

Zearalenone will significantly affect the reproductive performance of swine. Prepuberal gilts are the most sensitive to zearalenone. The symptoms commonly observed when feeding diets contaminated with zearalenone include a reddening and increased size of the vulva, and increased size of mammary tissue. Zearalenone will cause embryonic mortality at certain stages of gestation. Fertility problems are often associated with zearalenone concentrations of 100 to 200 ppb in sow feeds.

Poultry

Aflatoxin affects all poultry species. Although it generally takes relatively high levels to cause mortality, low levels can be detrimental if continually fed. Young poultry, especially ducks and turkeys, are very susceptible. As a general rule, growing poultry should not receive more than 20 ppb aflatoxin in the diet. However, feeding levels lower than 20 ppb may still reduce their resistance to disease, decrease their ability to withstand stress and bruising, and generally make them unthrifty.

Laying hens generally can tolerate higher levels than young birds, but levels should still be less than 50 ppb. Aflatoxin contamination can reduce the birds' ability to withstand stress by inhibiting the immune system. This malfunction can reduce egg size and possibly lower egg production. In addition, one must pay special attention to the use of contaminated corn in layer rations because eggs are promptly used as human food and aflatoxin metabolites have been found in egg yolks.

Mycotoxin levels found in most field situations tend to be low. Yet the combination of low levels of mycotoxins with the stresses associated with commercial production situations and/or exposure to disease organisms can produce effects in poultry which are subtle, indirect, and sometimes ill-defined. Since the effects of mycotoxins on poultry are dependant upon the age, physiological state, and nutritional status of the animals at the time of exposure, and since mold growth at various points within the feed production and distribution system can magnify mycotoxin problems, mycotoxicoses can be difficult to diagnose in field situations.

Mycotoxins produced by the mold genus *Fusarium* include: T-2 toxin and it's chemical relatives (trichothecenes), deoxynivalenol (DON), fumonisin, and zearalenone. Other animals tend to be more sensitive to the effects of fumonisin, deoxynivalenol, and zearalenone when compared to poultry. Nevertheless, detection of these mycotoxins within poultry rations indicates that the ration or the ingredients within the ration have been subjected to mold activity. Since numerous other mycotoxins, as well as reduced nutritive value and palatability of feeds, are generated by mold activity, the presence of fumonisin, deoxynivalenol, or zearalenone in poultry feeds is cause for concern.

T-2 toxin and trichothecenes can cause mouth and intestinal lesions as well as impair the birds' immune response, causing egg production declines, decreased feed consumption, weight loss, and altered feather patterns. While much is yet to be learned, T-2 toxin and related compounds are currently thought to be the most potent Fusarium mycotoxin for poultry.

DON alone has few effects in poultry. However, in field situations the DON level is sometimes associated with reduced feed consumption in layers and broiler breeders. This means that DON may be an indicator that T-2 or other unknown *Fusarium* mycotoxins are present.

Dairy Cattle

Aflatoxin-contaminated feed not only reduces animal performance and overall health, but it also creates risks of residues in milk. Aflatoxin is secreted into milk in the form of aflatoxin M1 with residues approximately equal to 1 to 2 percent (1.7 percent average) of the dietary level. This ratio is not influenced greatly by milk production level since higher producing cows consume more feed and have a slightly higher transmission rate. Due to risks of milk residues, dietary aflatoxin should be kept below 25 ppb. This level is conservative due to: (1) nonuniform distribution of aflatoxin in grain and feed, (2) uncertainties in sampling and analysis, and (3) the potential for having more than one source of aflatoxin in the diet. Replacement animals may tolerate 50 to 100 ppb aflatoxin.

In dairy cattle DON is associated with reduced feed intake, lower milk production, elevated milk somatic cell counts, and reduced reproductive efficiency. Milk production loss appears to occur when diets contain more than 300 ppb DON. Although controlled research has shown no cause and effect relationship between DON levels and reduced milk production, field observations have shown that reductions in milk output of 25 pounds per cow were seen when DON was 500 ppb or more. This suggests that DON may serve as a marker for feed that was exposed to a situation conducive to mold growth and mycotoxin formation. The possible presence of other mycotoxins, or factors more toxic than DON, seems likely. Dietary levels of 300 to 500 ppb DON in dairy feeds indicate mycotoxin problems and warrant attention.

Zearalenone causes estrogenic responses in dairy cattle, and large doses of this toxin are associated with abortions. Other responses of dairy animals to zearalenone may include reduced feed intake, decreased milk production, vaginitis, vaginal secretions, poor reproductive performance, and mammary gland enlargement in virgin heifers. Establishment of a tolerable level of zearalenone for dairy cattle is difficult, and is at best only a guess based on a meager amount of data and field observations. As with DON, zearalenone may serve as a marker for toxic feed. It is suggested that zearalenone not exceed 250 ppb in the total diet.

In dairy cattle T-2 toxin has been associated with feed refusal, production losses, gastroenteritis, intestinal hemorrhages, and death. T-2 has also been associated with reduced immune response in calves. Data with dairy cattle are not sufficient to establish a tolerable level of T-2 in the diet. Therefore, a practical recommendation may be to avoid T-2 in excess of 100 ppb in the total diet for growing or lactating dairy animals.

Fumonisin is another commonly isolated mycotoxin. However, fumonisin has only recently been isolated and only enough data exist to know that levels in excess of 20,000 ppb are potentially toxic to ruminants.

Beef Cattle

Aflatoxin and other mycotoxins can have considerable effects on beef cattle although the problems are usually less critical than for swine and poultry. Consumption of feeds highly contaminated with aflatoxin may reduce growth rate and increase the amount of feed required per pound of gain. Calves are generally more sensitive to feed contamination than adult cattle. In affected calves, some cases have revealed severe rectal straining and a prolapsed rectum. Lactating cows show a significant reduction in milk yield. Research has shown that high levels of aflatoxin can also cause liver damage in adult cattle. Feeding a high level of aflatoxin may also depress immune function, resulting in disease outbreaks.

Based on the feeds available, those contaminated with aflatoxin should be fed at the lowest level possible and for the shortest period of time practical. The effects of aflatoxin fed to cattle depend on the level of

aflatoxin in the ration, the length of the feeding period, and the age of the animal. If aflatoxin-contaminated feeds must be fed to beef cattle, follow these guidelines (on a dry matter basis):

1. Creep feeds and diets for gestating and lactating beef cows should contain less than 20 ppb of aflatoxin.
2. Unstressed, growing-finishing cattle in excess of 400 pounds may be fed diets containing up to 100 ppb of aflatoxin.
3. Diets for stressed feeder cattle should contain no more than 20 ppb of aflatoxin. Stressful conditions include weaning, shipping, extreme heat or cold, diseases, and parasites.
4. Animals destined for slaughter should receive aflatoxin-free diets for at least 3 weeks before slaughter.

Since cattle in the southeast are typically fed high forage diets, they are usually fed grain only as a supplement. Thus a relatively high level of aflatoxin can occur in the grain before it exceeds the tolerable dietary level. In general, cattle will eat about 2.5 percent of their body weight as dry matter. This can be used to calculate the contribution of grain to their total ration, and the tolerable level of aflatoxin in the grain. For example, growing calves weighing 600 pounds will consume about 15 pounds of total feed (600 lb multiplied by 2.5% equals 15 lb). If they are fed 3 pounds of grain plus forage-to-appetite, the grain will make up about 20 percent of their total diet (3 lb divided by 15 lb equals 20%). In this case the grain may contain up to 500 ppb of aflatoxin (100 ppb divided by 20% equals 500 ppb). Aflatoxin levels allowable in the grain, given different rates of inclusion in the beef ration. (NC State University)[20]

Knowing that aflatoxin has such an impact on the well-being of a farm in general, the use of cell wall carbohydrates derived from the yeast, *Saccharomyces cerivisiae*, in order to inhibit this deadly fungal toxin is a key component for the protection of a farm's inhabitants.

OFPA, USDA Final Rule:

Saccharomyces cerivisiae, nor any of its derivatives have been officially listed anywhere in the NOP final rule. As in section 205.600 of the NOP final rule, "any synthetic substance used as a processing aid or adjuvant will be evaluated against the following criteria: (2) the substance's manufacture, used and disposal do not have adverse effects on the environment and are done in a manner compatible with organic handling." The cell wall carbohydrates of *Saccharomyces cerivisiae* are not explicitly listed in section 205.603 as a synthetic substance allowed for use in organic livestock production, nor is it listed in section 205.604 as a prohibited substance. The classification of *Saccharomyces cerivisiae* and any of its derivatives as a synthetic substance for use in organic farming has yet to be determined.

FDA:

Note: The following law pertains to the Requirements for Specific Standardized Eggs and Egg Products. Relevant information regarding *Saccharomyces cerivisiae* has been highlighted and italicized.

TITLE 21--FOOD AND DRUGS

[20] *UT-Aflatrol* http://www.ublcorp.com/aflatrol.html

CHAPTER I--FOOD AND DRUG ADMINISTRATION, DEPARTMENT OF HEALTH AND HUMAN SERVICES (CONTINUED)

PART 160--EGGS AND EGG PRODUCTS--Table of Contents

Subpart B--Requirements for Specific Standardized Eggs and Egg Products

Sec. 160.105 Dried eggs.

(a) Dried eggs, dried whole eggs are prepared by drying liquid eggs that conform to Sec. 160.115, with such precautions that the finished food is free of viable Salmonella microorganisms. They may be powdered. Before drying, the glucose content of the liquid eggs may be reduced by one of the optional procedures set forth in paragraph (b) of this section. Either silicon dioxide complying with the provisions of
Sec. 172.480 of this chapter or sodium silicoaluminate may be added as an optional anticaking ingredient, but the amount of silicon dioxide used is not more than 1 percent and the amount of sodium silico-aluminate used is less than 2 percent by weight of the finished food. The finished food shall contain not less than 95 percent by weight total egg solids.

(b) The optional glucose-removing procedures are:

(1) Enzyme procedure. A glucose-oxidase-catalase preparation and hydrogen peroxide solution are added to the liquid eggs. The quantity used and the time of reaction are sufficient to substantially reduce the glucose content of the liquid eggs. The glucose-oxidase-catalase preparation used is one that is generally recognized as safe within the meaning of section 201(s) of the Federal Food, Drug, and Cosmetic Act. The hydrogen peroxide solution used shall comply with the specifications of the United States Pharmacopeia, except that it may exceed the concentration specified therein and it does not contain a preservative.

(2) Yeast procedure. The pH of the liquid eggs is adjusted to the range of 6.0 to 7.0, if necessary, by the addition of dilute, chemically pure hydrochloric acid, and controlled fermentation is maintained by adding food-grade baker's yeast (Saccharomyces cerevisiae). The quantity of yeast used and the time of reaction are sufficient to substantially reduce the glucose content of the liquid eggs.

(c) The name of the food for which a definition and standard of identity is prescribed by this section is ``Dried eggs'' or ``Dried whole eggs'' and if the glucose content was reduced, as provided in paragraph (b) of this section, the name shall be followed immediately by the statement ``Glucose removed for stability'' or ``Stabilized, glucose removed''.

(d)(1) When either of the optional anticaking ingredients specified in paragraph (a) of this section is used, the label shall bear the statement ``Not more than 1 percent silicon dioxide added as an anticaking agent'' or ``Less than 2 percent sodium silicoaluminate added as an anticaking agent'', whichever is applicable.

(2) The name of any optional ingredient used, as provided in paragraph (d)(1) of this section, shall be listed on the principal display panel or panels of the label with such prominence and conspicuousness as to render such statement likely to be read and understood by the ordinary individual under customary conditions of purchase.

(e) Label declaration. Each of the ingredients used in the food shall be declared on the label as required by the applicable sections of parts 101 and 130 of this chapter.

[42 FR 14462, Mar. 15, 1977, as amended at 58 FR 2883, Jan. 6, 1993]

Note: The following law pertains to the Requirements for Specific Standardized Eggs and Egg Products. Relevant information regarding *Saccharomyces cerivisiae* has been highlighted and italicized.

TITLE 21--FOOD AND DRUGS

CHAPTER I--FOOD AND DRUG ADMINISTRATION, DEPARTMENT OF HEALTH AND HUMAN SERVICES (CONTINUED)

PART 160--EGGS AND EGG PRODUCTS--Table of Contents

Subpart B--Requirements for Specific Standardized Eggs and Egg Products

Sec. 160.145 Dried egg whites.

(a) The food dried egg whites, egg white solids, dried egg albumen, egg albumen solids is prepared by drying liquid egg whites conforming to the requirements of Sec. 160.140 (or deviating from that section only by not being Salmonella free). As a preliminary step to drying, the lysozyme and avidin contents may be reduced. If lysozyme and avidin levels are reduced, cation exchange resins regulated for use under Sec. 173.25 of this chapter shall be used. As a further preliminary step to drying, the glucose content of the liquid egg whites is reduced by adjusting the pH, where necessary, with food-grade acid and by following one of the optional procedures set forth in paragraph (b) of this section. If the food is prepared from liquid egg whites conforming in all respects to the requirements of Sec. 160.140, drying shall be done with such precautions that the finished food is free of viable Salmonella microorganisms. If the food is prepared from liquid egg whites that are not Salmonella free, the dried product shall be so treated by heat or otherwise as to render the finished food free of viable Salmonella microorganisms. Dried egg whites may be powdered.

(b) The optional glucose-removing procedures are:

(1) Enzyme procedure. A glucose-oxidase-catalase preparation and hydrogen peroxide solution are added to liquid egg whites. The quantity used and the time of reaction are sufficient to substantially reduce the glucose content. The glucose-oxidase-catalase preparation used is one that is generally recognized as safe within the meaning of section 201(s) of the Federal Food, Drug, and Cosmetic Act. The hydrogen peroxide solution used shall comply with the specifications of the United States Pharmacopeia, except that it may exceed the concentration specified therein and it does not contain a preservative.

(2) Controlled fermentation procedures

(i) Yeast procedure. Food-grade baker's yeast (Saccharomyces cerevisiae) is added to the liquid egg whites and controlled fermentation is maintained. The quantity of yeast used and the time of reaction are sufficient to substantially reduce the glucose content.

(ii) Bacterial procedure. The liquid egg whites are subjected to the action of a culture of glucose-fermenting bacteria either generally recognized as safe within the meaning of section 201(s) of the Federal Food, Drug, and Cosmetic Act or the subject of a regulation established pursuant to section 409 of the act, and the culture is used in conformity with such regulation. The quantity of the culture used is sufficient to predominate in the fermentation and the time and temperature of reaction are sufficient to substantially reduce the glucose content.

(c)(1) Dried egg whites in which the lysozyme and avidin have been reduced shall not be nutritionally inferior, as defined in Sec. 101.3(e)(4)(i) of this chapter, and shall be considered nutritionally equivalent to untreated egg whites if they meet the conditions that the biological quality of the protein contained is equal to or greater than that of untreated egg white from the same batch of liquid egg

white.

(2) Compliance with the biological quality of protein requirement of paragraph (c)(1) of this section shall be determined by the analytical method prescribed in ``Official Methods of Analysis of the Association of Official Analytical Chemists,'' 14th Ed. (1984), section 43.253-43.257, ``Protein Efficiency Ratio, Rat Bioassay, Final Action,'' which is incorporated by reference. Copies may be obtained from the Association of Official Analytical Chemists International, 481 North Frederick Ave., suite 500, Gaithersburg, MD 20877-2504, or may be
examined at the Office of the Federal Register, 800 North Capitol Street, NW., suite 700, Washington, DC.

(d) When the dried egg whites are prepared from liquid egg whites containing any optional ingredients added as whipping aids, as provided for in Sec. 160.140(a), the common names of such optional ingredients shall be listed on the principal display panel or panels of the label with such prominence and conspicuousness as to render the names likely to be read and understood by ordinary individuals under customary conditions of purchase.

(e) The name of the food for which a definition and standard of identity is prescribed in this section is alternatively ``Dried egg whites'', Egg white solids'', ``Dried egg albumen'', or ``Egg albumen solids''. If the lysozyme and avidin content is reduced as provided in paragraph (a) of this section, the name shall be immediately preceded or followed by the statement ``lysozyme and avidin reduced'' when the dried egg whites are sold as such. When the dried egg whites are used in a fabricated food, the statement ``lysozyme and avidin reduced'' may be omitted from any declaration of ingredients required under Sec. 101.4 of this chapter.

(f) Label declaration. Each of the ingredients used in the food shall be declared on the label as required by the applicable sections of parts 101 and 130 of this chapter.

[42 FR 14462, Mar. 15, 1977, as amended at 51 FR 11435, Apr. 3, 1986; 51 FR 25362, July 14, 1986; 54 FR 24895, June 12, 1989; 58 FR 2883, Jan. 6, 1993; 63 FR 14035, Mar. 24, 1998]

Note: The following law pertains to food additives permitted for direct addition to food for human consumption.

TITLE 21--FOOD AND DRUGS

CHAPTER I--FOOD AND DRUG ADMINISTRATION, DEPARTMENT OF HEALTH AND HUMAN SERVICES (CONTINUED)

PART 172--FOOD ADDITIVES PERMITTED FOR DIRECT ADDITION TO FOOD FOR HUMAN CONSUMPTION--Table of Contents

Subpart I--Multipurpose Additives

Sec. 172.896 Dried yeasts.

Dried yeast (Saccharomyces cerevisiae and Saccharomyces fragilis) and dried torula yeast (Candida utilis) may be safely used in food provided the total folic acid content of the yeast does not exceed 0.04 milligram per gram of yeast (approximately 0.008 milligram of pteroyglutamic acid per gram of yeast).

Note: The following law pertains to food additives permitted for direct addition to food ofor human consumption—in particular, baker's yeast.

TITLE 21--FOOD AND DRUGS

CHAPTER I--FOOD AND DRUG ADMINISTRATION, DEPARTMENT OF HEALTH AND HUMAN SERVICES (CONTINUED)

PART 172--FOOD ADDITIVES PERMITTED FOR DIRECT ADDITION TO FOOD FOR HUMAN CONSUMPTION--Table of Contents

Subpart I--Multipurpose Additives

Sec. 172.898 Bakers yeast glycan.

Bakers yeast glycan may be safely used in food in accordance with the following conditions:

(a) Bakers yeast glycan is the comminuted, washed, pasteurized, and dried cell walls of the yeast, Saccharomyces cerevisiae. It is composed principally of long chain carbohydrates, not less than 85 percent on a dry solids basis. The carbohydrate is composed of glycan and mannan units in approximately a 2:1 ratio.

(b) The additive meets the following specifications on a dry weight basis: Less than 0.4 part per million (ppm) arsenic, 0.13 ppm cadmium, 0.2 ppm lead, 0.05 ppm mercury, 0.09 ppm selenium, and 10 ppm zinc.

(c) The viable microbial content of the finished ingredient is:

(1) Less than 10,000 organisms/gram by aerobic plate count.

(2) Less than 10 yeasts and molds/gram.

(3) Negative for Salmonella, E. coli, coagulase positive Staphylococci, Clostridium perfringens, Clostridium botulinum, or any other recognized microbial pathogen or any harmful microbial toxin.

(d) The additive is used or intended for use in the following foods when standards of identity established under section 401 of the Act do not preclude such use:

Use	Limitations
(1) In salad dressings as an emulsifier and emulsifier salt as defined in Sec. 170.3(o)(8) of this chapter, stabilizer and thickener as defined in Sec. 170.3(o)(28) of this chapter, or texturizer as defined in Sec. 170.3(o)(32) of this chapter.	Not to exceed a concentration of 5 percent of the finished salad dressing.
(2) In frozen dessert analogs as a stabilizer and thickener as defined in Sec. 170.3(o)(28) of this chapter, or texturizer as defined in Sec. 170.3(o)(32) of this chapter.	In an amount not to exceed good manufacturing practice.
(3) In sour cream analogs as a stabilizer and thickener as defined in Sec. 170.3(o)(28) of this chapter, or texturizer as defined in Sec. 170.3(o)(32) of this chapter.	Do.
(4) In cheese spread analogs as a	Do.

stabilizer and thickener as defined in Sec. 170.3(o)(28) of this chapter, or texturizer as defined in Sec. 170.3(o)(32) of this chapter.	
(5) In cheese-flavored and sour cream-flavored snack dips as a stabilizer and thickener as defined in Sec. 170.3(o)(28) of this chapter, or texturizer as defined in Sec. 170.3(o)(32) of this chapter.	Do.

(e) The label and labeling of the ingredient shall bear adequate directions to assure that use of the ingredient complies with this regulation.

[42 FR 14491, Mar. 15, 1977, as amended at 45 FR 58836, Sept. 5, 1980]

Note: The following law pertains to direct food substances affirmed as generally recognized as safe. Relevant information regarding *Saccharomyces cerevisiae* has been highlighted and italicized.

TITLE 21--FOOD AND DRUGS

CHAPTER I--FOOD AND DRUG ADMINISTRATION, DEPARTMENT OF HEALTH AND HUMAN SERVICES (CONTINUED)

PART 184--DIRECT FOOD SUBSTANCES AFFIRMED AS GENERALLY RECOGNIZED AS SAFE--
Table of Contents

Subpart B--Listing of Specific Substances Affirmed as GRAS

Sec. 184.1983 Bakers yeast extract.

(a) Bakers yeast extract is the food ingredient resulting from concentration of the solubles of mechanically ruptured cells of a selected strain of yeast, Saccharomyces cerevisiae. It may be concentrated or dried.

(b) The ingredient meets the following specifications on a dry weight basis: Less than 0.4 part per million (ppm) arsenic, 0.13 ppm cadmium, 0.2 ppm lead, 0.05 ppm mercury, 0.09 ppm selenium, and 10 ppm zinc.

(c) The viable microbial content of the finished ingredient as a concentrate or dry material is:

(1) Less than 10,000 organisms/gram by aerobic plate count.

(2) Less than 10 yeasts and molds/gram.

(3) Negative for Salmonella, E. coli, coagulase positive Staphylococci, Clostridium perfringens, Clostridium botulinum, or any other recognized microbial pathogen or any harmful microbial toxin.

(d) The ingredient is used as a flavoring agent and adjuvant as defined in Sec. 170.3(o)(12) of this chapter at a level not to exceed 5 percent in food.

(e) This regulation is issued prior to general evaluation of use of this ingredient in order to affirm as GRAS the specific use named.

Note: The following law pertains to anti-Saccharomyces cerevisiae (S. cerevisiae) antibody (ASCA) test systems. Relevant information has been highlighted and italicized.

TITLE 21--FOOD AND DRUGS

CHAPTER I--FOOD AND DRUG ADMINISTRATION, DEPARTMENT OF HEALTH AND HUMAN SERVICES (CONTINUED)

PART 866--IMMUNOLOGY AND MICROBIOLOGY DEVICES--Table of Contents

Subpart F--Immunological Test Systems

Sec. 866.5785 Anti-Saccharomyces cerevisiae (S. cerevisiae) antibody (ASCA) test systems.

(a) Identification. The Anti-Saccharomyces cerevisiae (S. cerevisiae) antibody (ASCA) test system is an in vitro diagnostic device that consists of the reagents used to measure, by immunochemical techniques, antibodies to S. cerevisiae (baker's or brewer's yeast) in human serum or plasma. Detection of S. cerevisiae antibodies may aid in the diagnosis of Crohn's disease.
(b) Classification. Class II (special controls). The special control is FDA's ``Guidance for Industry and FDA Reviewers: Class II Special Control Guidance Document for Anti-Saccharomyces cerevisiae (S. cerevisiae) Antibody (ASCA) Premarket Notifications.''

[65 FR 70307, Nov. 22, 2000]

Note: The following law pertains to food additives. Relevant information has been highlighted and italicized.

TITLE 21--FOOD AND DRUGS

CHAPTER I--FOOD AND DRUG ADMINISTRATION, DEPARTMENT OF HEALTH AND HUMAN SERVICES (CONTINUED)

PART 170--FOOD ADDITIVES--Table of Contents

Subpart A--General Provisions

Sec. 170.**3** Definitions.

For the purposes of this subchapter, the following definitions apply:
(a) Secretary means the Secretary of Health and Human Services.
(b) Department means the Department of Health and Human Services.
(c) Commissioner means the Commissioner of Food and Drugs.
(d) As used in this part, the term act means the Federal Food, Drug, and Cosmetic Act approved June 25, 1936, 52 Stat. 1040 et seq., as amended (21 U.S.C. 301-392).
(e)(1) Food additives includes all substances not exempted by

section 201(s) of the act, the intended use of which results or may reasonably be expected to result, directly or indirectly, either in their becoming a component of food or otherwise affecting the characteristics of food. A material used in the production of containers and packages is subject to the definition if it may reasonably be expected to become a component, or to affect the characteristics, directly or indirectly, of food packed in the container. ``Affecting the characteristics of food'' does not include such physical effects, as protecting contents of packages, preserving shape, and preventing moisture loss. If there is no migration of a packaging component from the package to the food, it does not become a component of the food and thus is not a food additive. A substance that does not become a component of food, but that is used, for example, in preparing an ingredient of the food to give a different flavor, texture, or other characteristic in the food, may be a food additive.

(2) Uses of food additives not requiring a listing regulation. Substances used in food-contact articles (e.g., food-packaging and food-processing equipment) that migrate, or may be expected to migrate, into food at such negligible levels that they have been exempted from regulation as food additives under Sec. 170.39.

(f) Common use in food means a substantial history of consumption of a substance for food use by a significant number of consumers.

(g) The word substance in the definition of the term ``food additive'' includes a food or food component consisting of one or more ingredients.

(h) Scientific procedures include those human, animal, analytical, and other scientific studies, whether published or unpublished, appropriate to establish the safety of a substance.

(i) Safe or safety means that there is a reasonable certainty in the minds of competent scientists that the substance is not harmful under the intended conditions of use. It is impossible in the present state of scientific knowledge to establish with complete certainty the absolute harmlessness of the use of any substance. Safety may be determined by scientific procedures or by general recognition of safety. In determining safety, the following factors shall be considered:

(1) The probable consumption of the substance and of any substance formed in or on food because of its use.

(2) The cumulative effect of the substance in the diet, taking into account any chemically or pharmacologically related substance or substances in such diet.

(**3**) Safety factors which, in the opinion of experts qualified by scientific training and experience to evaluate the safety of food and food ingredients, are generally recognized as appropriate.

(j) The term nonperishable processed food means any processed food not subject to rapid decay or deterioration that would render it unfit for consumption. Examples are flour, sugar, cereals, packaged cookies, and crackers. Not included are hermetically sealed foods or manufactured dairy products and other processed foods requiring refrigeration.

(k) General recognition of safety shall be determined in accordance with Sec. 170.30.

(l) Prior sanction means an explicit approval granted with respect to use of a substance in food prior to September 6, 1958, by the Food and Drug Administration or the United States Department of Agriculture pursuant to the Federal Food, Drug, and Cosmetic Act, the Poultry Products Inspection Act, or the Meat Inspection Act.

(m) Food includes human food, substances migrating to food from food-contact articles, pet food, and animal feed.

(n) The following general food categories are established to group specific related foods together for the purpose of establishing tolerances or limitations for the use of direct human food ingredients. Individual food products will be included within these categories according to the detailed classifications lists contained in Exhibit 33B of the report of the National Academy of Sciences/National Research

Council report, ``A Comprehensive Survey of Industry on the Use of Food Chemicals Generally Recognized as Safe'' (September 1972), which is incorporated by reference. Copies are available from the National Technical Information Service (NTIS), 5285 Port Royal Rd., Springfield, VA 22161, or available for inspection at the Office of the Federal Register, 800 North Capitol Street, NW., suite 700, Washington, DC 20408:

(1) Baked goods and baking mixes, including all ready-to-eat and ready-to-bake products, flours, and mixes requiring preparation before serving.

(2) Beverages, alcoholic, including malt beverages, wines, distilled liquors, and cocktail mix.

(**3**) Beverages and beverage bases, nonalcoholic, including only special or spiced teas, soft drinks, coffee substitutes, and fruit and vegetable flavored gelatin drinks.

(4) Breakfast cereals, including ready-to-eat and instant and regular hot cereals.

(5) Cheeses, including curd and whey cheeses, cream, natural, grating, processed, spread, dip, and miscellaneous cheeses.

(6) Chewing gum, including all forms.

(7) Coffee and tea, including regular, decaffeinated, and instant types.

(8) Condiments and relishes, including plain seasoning sauces and spreads, olives, pickles, and relishes, but not spices or herbs.

(9) Confections and frostings, including candy and flavored frostings, marshmallows, baking chocolate, and brown, lump, rock, maple, powdered, and raw sugars.

(10) Dairy product analogs, including nondairy milk, frozen or liquid creamers, coffee whiteners, toppings, and other nondairy products.

(11) Egg products, including liquid, frozen, or dried eggs, and egg dishes made therefrom, i.e., egg roll, egg foo young, egg salad, and frozen multicourse egg meals, but not fresh eggs.

(12) Fats and oils, including margarine, dressings for salads, butter, salad oils, shortenings and cooking oils.

(13) Fish products, including all prepared main dishes, salads, appetizers, frozen multicourse meals, and spreads containing fish, shellfish, and other aquatic animals, but not fresh fish.

(14) Fresh eggs, including cooked eggs and egg dishes made only from fresh shell eggs.

(15) Fresh fish, including only fresh and frozen fish, shellfish, and other aquatic animals.

(16) Fresh fruits and fruit juices, including only raw fruits, citrus, melons, and berries, and home-prepared ``ades'' and punches made therefrom.

(17) Fresh meats, including only fresh or home-frozen beef or veal, pork, lamb or mutton and home-prepared fresh meat-containing dishes, salads, appetizers, or sandwich spreads made therefrom.

(18) Fresh poultry, including only fresh or home-frozen poultry and game birds and home-prepared fresh poultry-containing dishes, salads, appetizers, or sandwich spreads made therefrom.

(19) Fresh vegetables, tomatoes, and potatoes, including only fresh and home-prepared vegetables.

(20) Frozen dairy desserts and mixes, including ice cream, ice milks, sherbets, and other frozen dairy desserts and specialties.

(21) Fruit and water ices, including all frozen fruit and water ices.

(22) Gelatins, puddings, and fillings, including flavored gelatin desserts, puddings, custards, parfaits, pie fillings, and gelatin base salads.

(23) Grain products and pastas, including macaroni and noodle products, rice dishes, and frozen multicourse meals, without meat or

vegetables.

(24) Gravies and sauces, including all meat sauces and gravies, and tomato, milk, buttery, and specialty sauces.

(25) Hard candy and cough drops, including all hard type candies.

(26) Herbs, seeds, spices, seasonings, blends, extracts, and flavorings, including all natural and artificial spices, blends, and flavors.

(27) Jams and jellies, home-prepared, including only home-prepared jams, jellies, fruit butters, preserves, and sweet spreads.

(28) Jams and jellies, commercial, including only commercially processed jams, jellies, fruit butters, preserves, and sweet spreads.

(29) Meat products, including all meats and meat containing dishes, salads, appetizers, frozen multicourse meat meals, and sandwich ingredients prepared by commercial processing or using commercially processed meats with home preparation.

(30) Milk, whole and skim, including only whole, lowfat, and skim fluid milks.

(31) Milk products, including flavored milks and milk drinks, dry milks, toppings, snack dips, spreads, weight control milk beverages, and other milk origin products.

(32) Nuts and nut products, including whole or shelled tree nuts, peanuts, coconut, and nut and peanut spreads.

(33) Plant protein products, including the National Academy of Sciences/National Research Council ``reconstituted vegetable protein'' category, and meat, poultry, and fish substitutes, analogs, and extender products made from plant proteins.

(34) Poultry products, including all poultry and poultry-containing dishes, salads, appetizers, frozen multicourse poultry meals, and sandwich ingredients prepared by commercial processing or using commercially processed poultry with home preparation.

(35) Processed fruits and fruit juices, including all commercially processed fruits, citrus, berries, and mixtures; salads, juices and juice punches, concentrates, dilutions, ``ades'', and drink substitutes made therefrom.

(36) Processed vegetables and vegetable juices, including all commercially processed vegetables, vegetable dishes, frozen multicourse vegetable meals, and vegetable juices and blends.

(37) Snack foods, including chips, pretzels, and other novelty snacks.

(38) Soft candy, including candy bars, chocolates, fudge, mints, and other chewy or nougat candies.

(39) Soups, home-prepared, including meat, fish, poultry, vegetable, and combination home-prepared soups.

(40) Soups and soup mixes, including commercially prepared meat, fish, poultry, vegetable, and combination soups and soup mixes.

(41) Sugar, white, granulated, including only white granulated sugar.

(42) Sugar substitutes, including granulated, liquid, and tablet sugar substitutes.

(43) Sweet sauces, toppings, and syrups, including chocolate, berry, fruit, corn syrup, and maple sweet sauces and toppings.

(o) The following terms describe the physical or technical functional effects for which direct human food ingredients may be added to foods. They are adopted from the National Academy of Sciences/ National Research Council national survey of food industries, reported to the Food and Drug Administration under the contract title ``A Comprehensive Survey of Industry on the Use of Food Chemicals Generally Recognized as Safe'' (September 1972), which is incorporated by reference. Copies are available from the National Technical Information Service (NTIS), 5285 Port Royal Rd., Springfield, VA 22161, or available for inspection at the Office of the Federal Register, 800 North Capitol Street, NW., suite 700, Washington, DC 20408:

(1) ``Anticaking agents and free-flow agents'': Substances added to finely powdered or crystalline food products to prevent caking, lumping, or agglomeration.

(2) ``Antimicrobial agents'': Substances used to preserve food by preventing growth of microorganisms and subsequent spoilage, including fungistats, mold and rope inhibitors, and the effects listed by the National Academy of Sciences/National Research Council under ``preservatives.''

(**3**) ``Antioxidants'': Substances used to preserve food by retarding deterioration, rancidity, or discoloration due to oxidation.

(4) ``Colors and coloring adjuncts'': Substances used to impart, preserve, or enhance the color or shading of a food, including color stabilizers, color fixatives, color-retention agents, etc.

(5) ``Curing and pickling agents'': Substances imparting a unique flavor and/or color to a food, usually producing an increase in shelf life stability.

(6) ``Dough strengtheners'': Substances used to modify starch and gluten, thereby producing a more stable dough, including the applicable effects listed by the National Academy of Sciences/National Research Council under ``dough conditioner.''

(7) ``Drying agents'': Substances with moisture-absorbing ability, used to maintain an environment of low moisture.

(8) ``Emulsifiers and emulsifier salts'': Substances which modify surface tension in the component phase of an emulsion to establish a uniform dispersion or emulsion.

(9) ``Enzymes'': Enzymes used to improve food processing and the quality of the finished food.

(10) ``Firming agents'': Substances added to precipitate residual pectin, thus strengthening the supporting tissue and preventing its collapse during processing.

(11) ``Flavor enhancers'': Substances added to supplement, enhance, or modify the original taste and/or aroma of a food, without imparting a characteristic taste or aroma of its own.

(12) ``Flavoring agents and adjuvants'': Substances added to impart or help impart a taste or aroma in food.

(13) ``Flour treating agents'': Substances added to milled flour, at the mill, to improve its color and/or baking qualities, including bleaching and maturing agents.

(14) ``Formulation aids'': Substances used to promote or produce a desired physical state or texture in food, including carriers, binders, fillers, plasticizers, film-formers, and tableting aids, etc.

(15) ``Fumigants'': Volatile substances used for controlling insects or pests.

(16) ``Humectants'': Hygroscopic substances incorporated in food to promote retention of moisture, including moisture-retention agents and antidusting agents.

(17) ``Leavening agents'': Substances used to produce or stimulate production of carbon dioxide in baked goods to impart a light texture, including yeast, yeast foods, and calcium salts listed by the National Academy of Sciences/National Research Council under ``dough conditioners.''

(18) ``Lubricants and release agents'': Substances added to food contact surfaces to prevent ingredients and finished products from sticking to them.

(19) ``Non-nutritive sweeteners'': Substances having less than 2 percent of the caloric value of sucrose per equivalent unit of sweetening capacity.

(20) ``Nutrient supplements'': Substances which are necessary for the body's nutritional and metabolic processes.

(21) ``Nutritive sweeteners'': Substances having greater than 2 percent of the caloric value of sucrose per equivalent unit of sweetening capacity.

(22) ``Oxidizing and reducing agents'': Substances which chemically oxidize or reduce another food ingredient, thereby producing a more stable product, including the applicable effect listed by the National Academy of Sciences/National Research Council under ``dough conditioners.''
(23) ``pH control agents'': Substances added to change or maintain active acidity or basicity, including buffers, acids, alkalies, and neutralizing agents.
(24) ``Processing aids'': Substances used as manufacturing aids to enhance the appeal or utility of a food or food component, including clarifying agents, clouding agents, catalysts, flocculents, filter aids, and crystallization inhibitors, etc.
(25) ``Propellants, aerating agents, and gases'': Gases used to supply force to expel a product or used to reduce the amount of oxygen in contact with the food in packaging.
(26) ``Sequestrants'': Substances which combine with polyvalent metal ions to form a soluble metal complex, to improve the quality and stability of products.
(27) ``Solvents and vehicles'': Substances used to extract or dissolve another substance.
(28) ``Stabilizers and thickeners'': Substances used to produce viscous solutions or dispersions, to impart body, improve consistency, or stabilize emulsions, including suspending and bodying agents, setting agents, jellying agents, and bulking agents, etc.
(29) ``Surface-active agents'': Substances used to modify surface properties of liquid food components for a variety of effects, other than emulsifiers, but including solubilizing agents, dispersants, detergents, wetting agents, rehydration enhancers, whipping agents, foaming agents, and defoaming agents, etc.
(30) ``Surface-finishing agents'': Substances used to increase palatability, preserve gloss, and inhibit discoloration of foods, including glazes, polishes, waxes, and protective coatings.
(31) ``Synergists'': Substances used to act or react with another food ingredient to produce a total effect different or greater than the sum of the effects produced by the individual ingredients.
(32) ``Texturizers'': Substances which affect the appearance or feel of the food.

[42 FR 14483, Mar. 15, 1977, as amended at 47 FR 11835, Mar. 19, 1982; 53 FR 16546, May 10, 1988; 54 FR 24896, June 12, 1989; 60 FR 36595, July 17, 1995]

[21]

Regulatory: EPA/NIEHS/Other Sources

EPA:
EPA registers and regulates antimicrobial pesticides under the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA). To obtain registration, manufacturers of antimicrobial products must meet the basic standards, the foremost being: 1) that the product will not cause unreasonable adverse effects to human health or the environment, and 2) that product labeling and composition comply with the requirements of FIFRA. Moreover, manufacturers are required to submit to EPA detailed and specific information concerning the chemical composition of their product; effectiveness data to document their claims against specific microorganisms and to support the directions for use provided in labeling; labeling

[21] All above laws regarding epinephrine and its legal use were directly copied and pasted from the government archives found on the web under relevant sections that pertained to this research. No alterations were made except certain significant information within the original text was highlighted for convenience purposes as previously noted. http://www.accessdata.fda.gov/scripts

that reflects the required elements for safe and effective use; and toxicology data to document any hazards associated with use of the product.

Recently, increased concern has emerged regarding whether public health products used to kill microorganisms pathogenic to man on inanimate surfaces and objects in hospitals, schools, restaurants, and homes work as claimed on the label. The private and public sector communities, including competitor registrants, have made the Agency aware of sterilizers and hospital disinfectants which may be ineffective. EPA has responded to this situation by developing a comprehensive strategy to improve the regulation of antimicrobial pesticides

Since public health products are crucial for infection control, and because of the increased controversy regarding product effectiveness, the Agency is conducting pre-registration confirmatory and post-registration enforcement testing of certain public health products. More specifically, EPA has entered into an Interagency Agreement with the FDA, and is jointly testing all sterilants except gases (registered and those seeking registration) and registered products which make unsubstantiated claims of controlling the bacterium which causes tuberculosis (including sterilants and hospital disinfectants). These two types of public health products are the most crucial to infection control and their failure could pose grave danger to the public and the medical community.

Furthermore, EPA has greatly improved communications with the public, all levels of government, academia, user communities, industry, health professionals, trade organizations, and independent testing groups. Also, EPA has committed funds to ensure that the tests used to demonstrate the efficacy of antimicrobial products are reliable and reproducible; is in the process of developing a complaint system to handle concerns regarding ineffective products; amplified internal controls to ensure the integrity of data submitted by registrants; and is currently publishing a quarterly newsletter designed to educate the general public about the status and direction of the regulation of antimicrobial products. The Agency is actively working to ensure that all antimicrobial products sold and distributed in the marketplace are effective in protecting public health and the environment from potential health risks.[22]

OSHA:
Three HB vaccines have been commercially marketed and available. The original Heptavax B®, a plasma-derived product, was introduced in the United States in 1982. This vaccine is available only in limited amounts and reserved for patients with specific medical conditions. The other two preparations are recombinant vaccines prepared from yeast cultures (***Saccharomyces cerevisiae***) that have been genetically altered to produce the hepatitis B surface antigen. Recombivax HB® was the first vaccine created using recombinant DNA technology and has been available since 1987. In 1989 a Belgium firm received a license in the U.S. for their Engerix-B® vaccine. The major difference between the products is the number of steps used in recovery and purification of the antigen from the yeast cultures, thus resulting in different dosage amounts (see Table 1). Both vaccine preparations provide adequate immunity. [23]

I. Exempt Experiments

Some recombinant DNA work is exempt from the Guidelines (Section III-E). These experiments should be reported on Appendix A of the Internal Processing Form from Office of Research Services when applying for a grant. All such research must be conducted using Biosafety Level 1 Practices (BL-1) (see reverse side). This group includes (but is not limited to) experiments that:

1. use as host-vector systems *E. coli* K-12, *Saccharomyces cerevisiae*, *Saccharomyces uvarum*, or *Bacillus subtilis*, and their plasmids;

[22] Information was referenced from http://www.epa.gov/pesticides/citizens/antimic.htm#regulation
[23] *Hepatitis B: Vaccination Information* http://www.dentalcare.com/soap/journals/dh_news/dhn0903/dn09n05.htm

2. rDNA molecules containing less than one-half of any eukaryotic genome that are propogated and maintained in cells in tissue culture.[24]

NIOSH: In order to test the effects of any substance on human health, NIOSH runs many of its tests on *Saccharomyces cerevisiae*, one of the first organisms to have its entire genome code understood. The outcomes and results shown by the yeast are often used as precursors to rules set in order to establish the safety measures of any substance on human health.

NOSB: Cell Wall Carbohydrates material is scheduled to be petitioned in September of 2002.
Category: Livestock
Petitioned use of material: Substitute for Antiobiotics

NTP, IARC: not listed as a known carcinogen.[25]

NIEHS:
Our long term objectives are to understand the molecular mechanisms of bypass replication of UV damaged DNA templates in the yeast *Saccharomyces cerevisiae*. In yeast, bypass replication is under the control of the *RAD6- RAD18* gene complex. Genetic evidence indicates the existence of two branches -- a mutagenic branch and a non-mutagenic branch of bypass replication, and the RAD6-RAD18 protein complex controls both of these processes. Genetic and biochemical studies focus on identifying the various components of the bypass pathway, defining their biochemical activities, and reconstitution of bypass replication *in vitro*. Other studies focus on the identification of protein substrates of RAD6-RAD18 complex, and determination of how ubiquitination by this complex modulates the activities of proteins involved in bypass replication. [26]

Like many other recent advances in genetic understanding, today's work was done using common bakers yeast, called *Saccharomyces cerevisiae*, the living substance that makes bread dough rise. This yeast has been a model system for much molecular genetic research for more than three decades because its basic cellular mechanisms also exist in mammals. The Resnick laboratory, where the work was performed, pioneered the use of yeast over 25 years ago in genetic and molecular studies to understand how DNA double-strand breaks, the major source of radiation-induced genetic damage and change, are produced and repaired by cells.[27]

Status Among U.S. Certifiers

NOFA: "The following medications are allowed with a 5 day withholding:
- non-steroidal anti-flammatory (i.e. banamine)
- antihistamines (e.g. epinephrine, adrenaline)
- anesthetics"[28]

State Organic Certifiers:
Oregon does not have specific limitations on materials used for crops and livestock. If the materials comply with USDA regulations, they are deemed acceptable for use in the state of Oregon. (Contact- Ron McKay) [29]

Pennsylvania is in accordance with guidelines proposed by OMRI.
(Contact- Martha Melton- state certifier) [30]

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[24] *Safe Handling of Biological Hazards* http://ehs.unc.edu/manuals/LabManual/lsm13.htm
[25] Chemical Hygiene Plan, Appendix C: *Carcinogens*
http://www.chem.pacificu.edu/ChemHyg&Invent/CHP/CHP%20App%20C%20Carcinogens.html
[26] *EHS University of Texas* http://www.envmed.rochester.edu/wwwrlp/niehsc/utexas/prakashl.html
[27] *What Protects us from Radiation?* http://www.niehs.nih.gov/oc/news/resnat.htm
[28] VOF Organic Meat & Egg Production – NOFA Vermont http://www.nofavt.org/sht02_stds7.cfm
[29] Information was referenced from a phone interview with Ron McKay, State Certifier, June 5, 2002.

Minnesota does not have specific limitations on materials used for crops and livestock. If the materials comply with USDA regulations, they are deemed acceptable for use in the state of Minnesota. (Contact- Mary Hanks- state certifier) [31]

International

Canadian General Standards Board:
7.2 Feed

7.2.1 The diet shall be nutritionally balanced to meet the nutritional requirements in accordance to the needs of the animal and shall be of good nutritional quality.

7.2.2 All organic feed shall be produced and processed in accordance with the following specifications. Livestock, in general, shall receive 100% of foodstuffs from organic sources. However, feeds consisting of no less than 85%, calculated on a dry matter basis, from organic sources for ruminants and no less than 80% from organic sources for non-ruminants may be used at the discretion of the certification body. When an unforeseeable major occurrence (e.g the natural disaster or any other such as flood, drought, or extreme weather) limits the availability of organic feed, a certification body may permit a refinement in the minimum acceptable percentage of an animal's rations to come from a transitional product when available, or when a transitional product is unavailable, from non-organic feed beyond the 15-20% limits shall be identified as such if sold within six (6) months of consuming such feed.

7.2.3 The following products shall under no circumstances be included or added to a livestock animal's diet: feed medications, including all hormones and antibiotics used to promote growth, synthetic appetence modifiers, preservation agents (subject to par. 7.2.5), colouring agents, urea, animal by-products (slaughterhouse waste), dung, droppings or other animal waste, medicated feeds, genetically engineered and/or modified organisms (GEO/GMO) or their products, feeds that have been defattened using solvents (hexane, etc.), chemically-extracted feeds (soy, canola, or other meals) or feeds to which other chemicals or prohibited substances have been added.

7.2.4 Compounds produced from genetically engineered and/or modified organisms (GEO/GMO), their products or related gene technology, are not permitted as ingredients of livestock feed. The following groups of ingredients, if obtained by a synthetic process, must have the approval of a certification body: vitamins, trace elements, and pure amino acids. A certification body shall list conditions under which these ingredients may be authorized. These conditions are represented by the following categories:

- *a. deficiencies that are specific to an enterprise or to a feed stock*
- *b. livestock of specific type and age*
- *c. specific abnormal circumstances beyond the control of the operator, and*
- *d. biogeographical requirements.*

7.2.5 The following silage preservation products are permitted as part of the production plan: bacterial or enzymatic additives and agricultural food by-products (e.g. molasses), with the exception that genetically engineered and/or modified organisms (GEO/GMO), or their products, are not permitted.[32]

IFOAM: not specifically mentioned in approved list[33]

Japan: not specifically mentioned in approved list[34]

[30] Information was referenced from a phone interview with Martha Melton, State Certifier, June 5, 2002
[31] Information was referenced from a phone interview with Mary Hanks, State Certifier, June 12, 2002
[32] *Organic Agriculture.* Canadian General Standards Board
[33] Directly referenced from http://www.ifoam.org/standard/ibs_final02.html
[34] Directly referenced from http://www.fas.usda.gov/gainfiles/200004/25647377.pdf

European Union: not specifically mentioned in approved list[35]

News
June 1, 2001
Yeast culture receives EU approval
Alltech's **Yea-Sacc^{1026}** is the only yeast culture to gain European Union approval as a performance-enhancing yeast additive for dairy cows, fattening cattle, and calves. The Standing Committee for Animal Nutrition (SCAN) granted the approval.

The approval is in accordance with EU Council Directive 70/524/EEC (Council Regulation No 937/2001 of May 11, 2001). Alltech developed Yea-Sacc^{1026} from a naturally-occurring strain of *Saccharomyces cerevisiae* yeast. In the early 1980s, Alltech researchers observed that this particular strain of yeast had beneficial effects on rumen function and cattle performance. Because yeast has been safely used in human food for many years, its inclusion in animal feed is acceptable to consumers.[36]

Section 2119 OFPA U.S.C. 6518(m)(1-7) Criteria

1. *The potential of the substance for detrimental interactions with other materials used in organic farming systems.*

Considering that baker's yeast, *Saccharomyces cerivisiae,* is safely used in foods that are ingested by humans on a daily basis, and since the FDA has declared it as being entirely safe for use, there should be no real problem with the yeast or any of its derivatives in a general view.

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10. Stability and reactivity
10.1 Conditions and materials to avoid: None
10.2 Conditions of reactivity:: Stable
10.3 Hazardous decomposition products: None
10.4 General instructions: -
[37]

1368 *In vitro* a.atoxin binding characteristics of an esteri.ed glucomannan product. J.W. Evans* and M. Kudupoje, *Alltech BiotechnologyInc., Nicholasville, KY.*
Mycotoxin binders have been shown to reduce the deleterious effects of aflatoxin in animals. A series of experiments were conducted to examine the binding characteristics of an esterifed glucomannan (EGM) derived from the yeast cell wall of *Saccharomyces cerevisiae*. The EGM has been shown to reduce the toxic effects of aflatoxin in livestock. In the first study, an *in vitro* binding assay was used to determine the saturation point of aflatoxin binding by EGM in water or phosphate buffer. The binder (0.1%) was mixed with increasing concentrations of toxin (2,4,6,8 and 10 ppm in water/18, 20, 22, 24 and 26 ppm in buffer), centrifuged and unbound toxin concentrations were determined in the supernatant. The saturation point was defined as the minimum binder concentration at which maximum binding is reached. The saturation point for aflatoxin binding was greater in phosphate buffer than in water (8 vs. 24 ppm). In addition, aflatoxin binding responded to phosphate concentrations in a quadratic fashion ($P<0.001$). Another trial was conducted to determine binding strength in water and phosphate buffer. Aflatoxin (1 ppm) and EGM (0.1%) were mixed,incubated at 37.C for 1 hr, and centrifuged. Toxin concentrations were determined in

[35] Directly referenced from http://www.foodstandards.gov.uk/multimedia/pdfs/elist_numbers.pdf
[36] Directly referenced from http://www.alltech-bio.com/alltech%5CAlltech2.nsf/pages/News_Yeast_culture_receives_EU_approval
[37] *Material safety data sheet (91/155/EEC)* http://www.begerow.de/pmm/english/sdb-pdf/hefen.pdf

the supernatant and pellet after three methanol extractions. Binding was stronger in the phosphate buffer than in water as indicated by lower aflatoxin recovery in the buffer (568 vs. 279 ppb,P <0.001).
A third study compared the saturation points of the EGM and a clay binder (CB). Both binders were mixed in water with increasing toxin concentrations until saturation was reached. The binding capacity of EGM in water was more than four times higher than CB (5.2 vs.1.2 μg/mg binder,P <0.001). This series of studies showed that EGM is able to bind aflatoxin and not mask or destroy it. In addition, aflatoxin binding by EGM is phosphate dependent and more efficient at binding aflatoxin than binding by the CB tested.[38]

2. *The toxicity and mode of action of the substance and of its break down products or any contaminants, and their persistence and areas of concentration in the environment.*

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3. Description of hazards
Not a controlled hazardous product.

11. Toxicological information
11.1 General: Not applicable – non-toxic food ingredient

14. Transport information
None

15. Regulatory information
None

16. Other relevant information
The particulars correspond to our current state of knowledge and experience. We describe our products with regard to necessary safety requirements but not in conjunction with guaranteed properties and quality descriptions.[39]

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3. *The probability of environmental contamination during manufacture, use, misuse, or disposal of the substance.*

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12. Ecological information
12.1 General: -
13. Disposal considerations
13.1 Disposal of materials: Can be treated as garbage.
13.2 Disposal of packaging: Use licensed disposal facility. Follow local regulations[40]

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4. *The effects of the substance on human health.*

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4. First aid measures
4.1 Inhalation: n. a.
4.2 Skin exposure: Wash with water
4.3 Eye exposure: Wash with water
4.4 Ingestion: Non-toxic
4.5 General instructions: -

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[38] *ASAS/ADSA Animal Health.* http://www.adsa.org/jointabs/iaafs128.pdf
[39] *Material safety data sheet (91/155/EEC)* http://www.begerow.de/pmm/english/sdb-pdf/hefen.pdf
[40] *Material safety data sheet (91/155/EEC)* http://www.begerow.de/pmm/english/sdb-pdf/hefen.pdf

5. Fire-fighting measures (Fireproofing)
Not applicable

6. Accidental release measures
6.1 Personal precautions: Wear dust mask in poorly ventilated areas
6.2 Environmental precautions: -
6.3 Methods for cleaning up: Sweep area and rinse with water

7. Handling and storage
7.1 Handling: -
7.2 Storage:
Requirements for storage rooms and containers: Always store in original container or in a clean covered container.
Storage class: -
VbF class: -

8. Exposure control / personal protection:
8.1 Recommended control equipment: -
8.2 Control parameters: -
8.3 Recommended personal protection:
respiratory: Wear dust mask in poorly ventilated areas
hands: N/A
eyes: Normal industrial eye precautions should be followed
skin: N/A
Revised: 10 February 2000 Version No.: 1 Page 2 of 2[41]

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5. *The effects of the substance on biological and chemical interactions in the agroecosystem, including the physiological effects of the substance on soil organisms(including the salt index and solubility of the soil), crops and livestock.*

As aforementioned, *Saccharomyces cerivisiae* is a common baker's yeast and it should not have any harm or detrimental effects to anything in the environment. However, yeast is effected by Potassium Sorbate:

Potassium sorbate is widely used as a preservative in food products. The inhibition kinetics of yeast growth by potassium sorbate was determined in order to predict the inhibition mechanism and develop an antimicrobial activity model. Linear regression resulted in uncompetitive growth inhibition using different concentrations of nutrients in liquid media containing yeast extract, malt extract (YM broth) and potassium sorbate. At 30°C, constant Ks of YM broth to yeast was 0.254%, maximum growth rate (μmax) of yeast was 0.008 min-1, and inhibition constant (Ki) of potassium sorbate to yeast growth was 0.324%. This statistical method could reliably determine the growth inhibition mechanism and kinetics.

RESULTS & DISCUSSION

POTASSIUM SORBATE EFFECTIVELY INHIBITED THE GROWTH OF yeast. The pH of culture broth slowly decreased at the stationary growth stage, however, did not significantly change during the logarithmic phase The pH of culture broth is very important to the inhibitory activity of potassium sorbate. Undissociated potassium sorbate has stronger inhibitory activity than the dissociated form (Sofos and Busta, 1981; Eklund, 1983). Because the pH was not changed during logarithmic growth and the growth rate (μ) was calculated from growth data during the logarithmic phase, the values of the growth rate (μ) and the parameters (β_1, β_2, and β_{12}) of the statistical growth of microorganisms in food products model were not affected by pH of the culture broth.

A parameter test for the full model showed that the parameters 1/[S], [I], which represent β_1 and β_2, respectively, and the intercept were significant ($p<0.0001$, Table 2). However the parameter [I]/[S], which represents the interaction, β_{12}, was not significant. The variable x_{12}, which is [I]/[S], did not contribute significantly to the model when it included x_1 and x_2. Therefore, only x_1 and x_2 were selected as main variables in the growth inhibition model.

$$\frac{1}{} = \frac{K_s}{} \frac{1}{} + \frac{[I]}{} + \frac{1}{} \quad (9)$$

[41] *Material safety data sheet (91/155/EEC)* http://www.begerow.de/pmm/english/sdb-pdf/hefen.pdf

$\mu \quad \mu_{max} [S] \quad \mu_{max}K_i \quad \mu_{max}$

With this reduced model (Eq. 9) in which [I] and $[S]^{-1}$ were the main variables, after discarding [I]/[S], K_s of YM broth, μ_{max} of yeast, and K_i of potassium sorbate were 0.254%, 0.008 min^{-1}, and 0.324%, respectively. Because Eq. (9) has only x_1 and x_2 as main variables compared to the full model (Eq. 7), the available parameters for the model were β_0, β_1, and β_2. This result strongly suggested that the growth inhibition of yeast by potassium sorbate followed the uncompetitive inhibition model (Table 1). The relationship between the inverse growth rate constant (1/μ), the concentration of potassium sorbate [I], and the different concentrations of nutrient in the broth media [S] showed the parallel slope, typical of the uncompetitive inhibition model (Fig. 3). **Overall potassium sorbate, thus, inhibits the growth of yeast by an uncompetitive inhibitory mechanism.**[42]

6. The alternatives to using the substance in terms of practices or other available materials.

There are other toxin binders available such as bentonite, diatomaceous earth, and charcoal but the use of these is not desired. These other options can tie up critical dietary immune supporting nutrients while simultaneously binding to the toxins.

Prevention of Feed Contamination

Controlling mold growth and mycotoxin production is very important to the feed manufacturer and livestock producer. Control of mold growth in feeds can be accomplished by keeping moisture low, keeping feed fresh, keeping equipment clean, and using mold inhibitors. Grains and other dry feed such as hay should be stored at a moisture level 14 percent or less to discourage mold growth. Aeration of grain bins is important to reduce moisture migration and to keep the feedstuffs dry.

Moisture Control

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: (1) feed ingredients, (2) feed manufacturing processes, and (3) 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 controlled.

Moisture in Feed Ingredients

Since corn and other grains are a primary source of the moisture and molds found in feed, the first important step in controlling 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 controlled. 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 percent moisture may, for example, contain some kernels with 10 percent moisture and others with 20 percent 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 were 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. Thus the fraction of commercial grain, known as broken kernels and foreign matter, can be expected to have a higher mold and mycotoxin content than the portion composed of whole kernels.

Moisture in Feed Manufacturing Processes

Grains are commonly ground with a hammer mill to aid in mixing and handling, to improve digestibility, and to improve the pelleting process. This grinding process creates friction, which causes heat to build up.

[42] *Modeling the Growth Inhibition Kinetics of Baker's Yeast by Potassium Sorbate Using Statistical Approaches.* http://www.confex.com/ift/JFSonline6s83RAqM/pdfs/jfsv63n1p0012-0014ms557H.pdf

If unchecked, temperature increases greater than 10 degrees Fahrenheit will cause significant migration of grain moisture encouraging mold growth. This is particularly true in cold weather when temperature differences cause moisture to condense on the inside walls of bins. Air-assisted hammer-mill systems reduce heat buildup in the product and, in turn, reduce moisture problems.

The pelleting process involves mixing steam with the feed, pressing the mixture through a die, and then cooling the pellets to remove heat and moisture. Generally, heat and 3 to 5 percent moisture are added to the feed during the pelleting process in the form of steam. If the pelleting process is done correctly, this excess moisture is removed from the feed before shipment. If, however, 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 to 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 nonpelleted feeds.

Moisture and Feed Storage Environment

To control mold growth, obvious sources of moisture in the feed handling and storage equipment must be eliminated. These sources may include leaks in feed storage tanks, augers, roofs (either at the barn or at the feed mill), and compartments in feed trucks.

A fact about feed moisture often overlooked is that it changes in relation to the feed's environment. Since animals kept in confinement housing add moisture to their environment by respiration and defecation, the air in these houses can be very humid. Feed that was initially very low in moisture content will gain moisture when placed in a humid environment. The humidity in confinement housing should therefore be controlled by providing adequate ventilation.

Keeping Feed Fresh

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

It is equally important to manage the feed delivery system to ensure that feeds are uniform in freshness. Field surveys have shown that poultry farms producing birds with the poorest performance were those with the most feed in their feeder pans. On these farms, the feeds contained the greatest amount of moisture and had the highest number of molds. If the feeder system is allowed to keep the feed pans full at all times, the feed in the pans will be significantly older than that in the storage tank. The animals will tend to eat primarily the feed in the top layer, and the feed at the bottom of the pans will age, providing greater opportunities for molds to grow. The animals' performance may suffer as a result. To prevent this problem, the feeder system should be turned off weekly. The animals will then be forced to clean out all of the feed in the feeders before it becomes excessively old.

A similar principle applies to feed storage tanks. The feed next to the wall is last to exit the tank and therefore stays in the tank the longest. The feed in contact with the wall is also the only portion of the feed that changes appreciably due to temperature. These factors make feed in contact with the wall susceptible to moisture migration and mold growth. It is best to maintain two feed tanks so that one tank can be completely emptied and cleaned before it is refilled with new feed.

Equipment Cleanliness

When feed is manufactured and delivered to farms, it may come in contact with old feed that has lodged or caked in various areas of the feed 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.

Use of Mold Inhibitors

Types of Mold Inhibitors

The use of chemical mold inhibitors is a well-established practice in the feed industry. However, mold inhibitors are 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 sulfate. 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.

Dispersion

Many factors influence the effectiveness of mold inhibitors, and proper attention to these factors can enhance the benefits they provide. Mold inhibitors cannot be effective unless they are completely and thoroughly distributed throughout the feed. Ideally, this means that the entire surface of each feed particle should come in contact with the inhibitor and that the inhibitor should also penetrate feed particles so that interior molds will be inhibited.

The particle size of the carriers for mold-inhibiting chemicals should be small so that 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 vapor from fairly large particle carriers. Presumably, the inhibitor then penetrates the air spaces between particles of feed to achieve even dispersion.

Effect of Feed Ingredients

Certain feed ingredients may also affect mold inhibitor performance. Protein or mineral supplements (for example, soybean meal, fish meal, poultry by-product meal, and limestone) 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.

Time Dependence

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 that are heavily contaminated with molds will require additional amounts of inhibitor to achieve the desired level of protection.

Influence of Pelleting

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

Copper Sulfate

The practice of recommending copper sulfate as a treatment for fungal diseases in animals goes back many decades. The effectiveness of copper as a mold inhibitor is difficult to document. Although copper sulfate in the diet has been shown to improve body weight and feed conversion efficiency 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 sulfate 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.

Animal Management

If unacceptable mycotoxin levels occur, removal of the contaminated feed is preferable. While it is often not possible to completely replace the ration, particularly the forage ingredients, obviously, moldy feeds should be removed. Acidic diets may intensify the effects of mycotoxins and should be avoided in these situations. Increasing nutrients such as protein, energy (fats and carbohydrates), and vitamins in the diet may also be advisable. The addition of antioxidants to the animal assists in dealing with the effects of mycotoxins.

The possible use of inorganic binders (mineral clays) to bind mycotoxins, and prevent them from being absorbed by the animal's gut, has received a lot of research attention recently. These clay products (which include zeolites, bentonite, bleaching clays from refining of canola oil, and hydrated sodium calcium aluminosilicates [HSCAS]) have been shown to change the responses of rats to zearalenone and T-2 toxin. However, it should be clearly understood that binding of some mycotoxins may be weak or nonexistent and that clay products differ in their ability to bind mycotoxins. While one HSCAS product called NovaSil has been shown to bind aflatoxin protecting animals against aflatoxicosis, under FDA regulations these clay products cannot be sold as mycotoxin binders. Nonetheless, many clay products are GRAS (Generally Recognized As Safe) and are used as anticaking or free-flow additives for feeds[43]

7. Its compatibility with a system of sustainable agriculture.

Saccharomyces cerivisiae, commonly known as baker's yeast has been used in breads and beers for years. The FDA has declared it as a very safe substance for food purposes. When discussing a system of sustainable agriculture, there are many things that should be considered: the animal's welfare, the environmental effects of the substance, as well as the overall effects in relation to human health.

The cell wall derivatives taken from the yeast should demonstrate the same effects that the yeast itself demonstrates. The question as to whether or not the cell wall carbohydrate derivative is a synthetic substance has yet to be determined. But on the whole, it should not be a threat to the environment, to the consumers, or to the animal's themselves.

On the other hand, mycotoxins are a serious threat to both the humans and the animals they attack. Found in fungus, these mycotoxins have proven to disrupt the normal digestive functions of humans and animals. Mycotoxins reside in feed and can be passed to humans through dairy from farm animals possessing the toxin. With regards to beef cattle, mycotoxins become an economic threat, in addition to the threat to the animal itself. Growth rates of the animal can be stunted and it may actually take more feed

[43] ***Understanding and Coping with Effects of Mycotoxins in Livestock Feed and Forage.*** http://www.ces.ncsu.edu/drought/dro-29.html#mycos

per pound of gain. This is all due to the fact that mycotoxins find their home in the gastrointestinal tract of the animal and inhibit normal digestion. The poison can be hindered by the carbohydrate yeast derivative found in *Saccharomyces cerivisiae* which binds to the mycotoxin and inhibits it from harming the host.

There are methods and precautions that can be followed in order to maintain good feed and prevent the invasion of mycotoxins, but cell wall carbohydrates derived from *Saccharomyces cerivisiae* are a feed additive that can also help prevent the gastrointestinal problems associated with mycotoxin poisoning. Hence, *Saccharomyces cerivisiae*, and its particular carbohydrate derivatives taken from the cell wall should, in fact, be compatible with a system of sustainable agriculture.

TAP Reviewers' Discussion

Reviewer 1 *[Ph.D, Food Microbiology. Assistant Professor of Food Microbiology. Research interests include biochemical and genetic analysis of enzyme systems influencing microbiological activity and applications of rDNA techniques. Midwest U.S.]*

Observations/Conclusions

Identification
An animal dietary toxin binder made up of cell wall carbohydrates (CWC), isolated from the yeast *Saccharomyces cerevisiae*. These CWC are composed of approximately 50% mannan oligosaccharide and will be as an antibiotic substitute.

Characterization
It is not clear as to how this feed additive will be extracted and processed. Are we talking about the addition of *Saccharomyces cerevisiae* cells (Direct Microbial Product-DMP) or only the cell wall carbohydrates? If it is just the CWC, then there needs to be a description of what this process involves in order to determine whether the additive is compatible with organic farming standards.

Synthetic or Non-Synthetic?
The yeast itself is non-synthetic and has been used for millions of years as fermentation aids without adverse effects. *Saccharomyces cerevisiae* is considered a GRAS substance by the FDA and is used as baker's yeast and brewer's yeast. However, the compound that is being petitioned here is not the yeast, but its CWC, specifically the mannan oligosaccharide portion that has been shown to actively bind toxins and pathogenic microorganisms. How will the CWC be extracted and purified from *Saccharomyces cerevisiae*? What types of by-products will come out of the process and what kind of disposal method will be used? The process must not have adverse effects on the environment.

According to the USDA-OFPA, the term "synthetic" means a compound that is manufactured by a chemical process or a process that chemically changes a substance extracted from naturally occurring plant, animal or mineral sources ...." The CWC in question is obviously naturally a part of the yeast, hence non-synthetic with that regard. However, because I do not have a description of the actual extraction process, I do not know if the process would chemically change the CWC from its natural state in the yeast (in which case it would be considered "synthetic"). Thus, based on the materials I received in the TAP review and citations, I cannot make a judgement on whether or not the CWC is synthetic.

Effectiveness
Mannan oligosaccharides have been shown to be recognized and bound by mold and bacterial toxins. Furthermore, it is thought that in the intestinal tract of animals, the lectins of pathogenic bacteria such as *Salmonella, E. coli* and *Vibrio cholerae* would bind the mannan oligosaccharides instead of the host intestinal epithelial cells, thus preventing their colonization of the host intestinal tract. The carbohydrate also acts as a nutrient source to other beneficial bacteria in the gut and is also thought to stimulate the animals' immune system. The adsorbed toxins and pathogens are then flushed out of the animal's system naturally. The use of this feed supplement would substitute the use of sub-therapeutic levels of antibiotics

commonly used in non-organic farming, a practice that has been blamed for increasing numbers of antibiotic-resistant pathogens.

Although the practice of using sub-therapeutic levels of antibiotics in farm animals in this country has been widespread, many conventional farmers are now turning to the use of DMF supplements, such as probiotics, to promote health in animals. Yeasts, including *Saccharomyces cerevisiae*, are often used as part of a cocktail of probiotics. From all the studies that have been cited in the TAP report, it appears that the mannan oligosaccharides from *Saccharomyces cerevisiae* are very effective as dietary toxin binders as well as supplemental nutrient source in animals.

Allow with Restrictions?
The *Saccharomyces cerevisiae* strain used must be a wild-type strain and not a genetically engineered one. Since the yeast itself is a natural product, the use of the cells as a DMP should be allowed. If only the CWC extract is to be used, the product must not be irradiated and the process used to obtain the extract must be in a way that complies with organic handling standards.

Reviewer 1 Recommendations Advised to the NOSB

Unable to determine if Synthetic or Nonsynthetic.

For Livestock, the substance should be Added to the National List with restrictions.

Reviewer 2 *[Ph.D, Swine Nutrition; Minor in Biochemistry. Research interests in swine nutrition and management. Southeast U.S.]*

I am in general agreement with the TAP review and believe that it is fairly complete and provides sufficient background information and relevant research pertaining to the use of Cell Wall Carbohydrates (CWC).

I believe that Cell Wall Carbohydrates (CWC) from non-irradiated and non-GMO *Saccharomyces cerivisiae* should be considered as a synthetic substance allowed for use in organic livestock production as a dietary toxin binder. The basis for this judgment include the following:

1. *Saccharomyces cerivisiae* is a naturally-occurring yeast species with no documented evidence of harmful effects on the environment.
2. FDA already considers *Saccharomyces cerivisiae* as safe for use in food production. CWC, more commonly known as baker's yeast, is presently used in the production of breads and alcoholic beverages.
3. Research has demonstrated that CWC can have a positive influence on livestock and poultry performance. In 14 trials summarized in the TAP review, CWC (in particular mannan oligosaccharide or MOS) improved growth rate in calves by an average of 17.1%. There is also a substantial body of research not cited in the TAP review that demonstrates CWC can improve animal performance. Although the mode of action of CWC is not entirely understood, it is thought to elicits its effects through:
    a. Binding and absorbing pathogens, such as Salmonella and E-coli, in the gastrointestinal (GI) tract, thus minimizing the colonization of pathogens in the GI tract.
    b. Improving the digestion and absorption certain of minerals, such as calcium, phosphorus, magnesium, copper, zinc, potassium, and manganese.
    c. Binding mycotoxins produced by molds present in animal feedstuffs and minimizing the secondary effects of mycotoxins. In addition, unlike other toxin binders that are presently available to livestock and poultry producers, CWC do not tie up other dietary nutrients that are important in immune processes.

d. Stimulating the animal's immune system to increase macrophage and immunoglobulin activity.

In summary, I see no reason to not allow Cell Wall Carbohydrates (CWC) for use in organic livestock production as a dietary toxin binder, provided it is derived from non-irradiated and non-GMO *Saccharomyces cerivisiae*.

Reviewer 2 Recommendations Advised to the NOSB

The substance is Synthetic.

For Livestock, the substance should be Added to the National List with restrictions.

Reviewer 3 *[PhD, Animal Science; Associate Professor, Animal Science--Anaerobic and Gastrointestinal Microbiology. Research interests include ruminal anaerobic physiology and biochemistry. Southeast U.S.]*

Introduction. This TAP report describes the potential use of cell wall carbohydrates derived from non-genetically modified yeast as a substitute for antibiotics in animal feeding systems and as a binder of mycotoxins found in contaminated feedstuffs. Cell wall carbohydrates and other yeast-derived products have the potential to be useful in certain organic animal production systems. *However, the report has, in the reviewer's opinion, significant ambiguities that raise serious concerns regarding the application of cell wall carbohydrates in animal feed.*

Intent of OFPA and NOP Rules. The feeding and care of domestic livestock involves the complex interaction of animal metabolism and behavior with feed material, environment, and human intervention. In many regards, the organic production of livestock is a more challenging undertaking than producing crops organically. *Although the OFPA and NOP rules provide some guidance for producing organic animal products, there is considerable vagueness about definitions and intent. The resultant uncertainty raises critical questions with regard to animal husbandry practices and the consideration of substances for inclusion in organic production.* For instance, the OFPA states that producers "….shall not use growth promoters…..and synthetic trace elements….to stimulate growth or production of such livestock" (section 6509 (c) (3)). The NOP Final Rule further states that organic producers must "not…provide feed supplements or additives in amounts above those needed for adequate nutrition and health maintenance for the species at its specific stage of life" (205.237 (b) (2)). Although one obvious intent of these regulations is to prohibit the use of hormones and anti-microbial compounds, it is much less clear how a broad range of other potential feed components (namely, the wide array of supplements and additives) are to be considered.

Perhaps most problematic are the phrases "to stimulate growth or production" and "adequate nutrition". The National Research Council Committee on Animal Nutrition has established nutrient requirements for various domestic animals, and these requirements define nutrient inputs needed by animals at different ages and production levels (e. g., milk output, tissue deposition). In this sense, "adequate nutrition" is defined by the desired production level. But nutrients available from a particular combination (diet) of organically produced feedstuffs may not meet the nutrient demands dictated by a particular production level or physiological state, and the only way to meet that demand is by inclusion of a feed supplement(s) or additive(s). Although the supplement/additive may be natural and/or organically produced, it is clearly 'promoting' and 'stimulating' growth and production beyond a level constrained by either the quantity and/or the quality of the basic diet. *In this sense, it is not clear whether the intent of the NOP is to permit producers to design feeding strategies to maximize production or whether producers are to be constrained by the feed and environmental circumstances inherent to their specific production system.* It should be realized that maximizing production, *even if done using organic materials,* is not always a sustainable

practice in all scenarios. *Consideration of these issues is critical to determining whether the use of compounds such as cell wall carbohydrates is appropriate in organic animal production.*

Yeast-derived cell wall carbohydrates. Live yeast, non-viable yeast cultures, and sub-cellular fractions of yeast cells have been fed to ruminants and non-ruminants for over 20 years. The animal production responses to these materials in animal rations is variable, but there is a body of literature which generally supports the contention that yeast-products enhance production and provide health benefits in certain production scenarios. Arguably, the most significant responses are observed under intensive management regimes and at high levels of production. Although their use is becoming more accepted and widespread, there is still very little information on the precise mode of action for most of these products. Given the variable nature of methods to produce these materials and the often undefined nature of the materials involved, this lack of information is not surprising. Consequently, whole yeast and complex mixtures of yeast-derived materials have a multitude of effects and probably a range of modes of action. The actions of more purified components is generally better understood. For instance, the interference of binding of pathogenic bacteria caused by mannan-oligosaccharides is perhaps the best-supported mode of action thus far described.

Yeast and yeast-derived products are usually derived from large industrial scale fermentations that typically use maize as the major feedstock for growth of the microorganisms. Whole yeast are then separated from the fermentation liquid by centrifugal force and dried. At this point, a variety of processing schemes are employed to generate products ranging from intact whole yeast to sub-cellular fractions of yeast including carbohydrates from cell walls. The report, however, did not provide details on specific production and processing methods for either culture of yeast of purification of the cell wall carbohydrates.

Ambiguities and potential problems. Beyond the uncertainties about the intent of the OFPA and NOP rules (see above), there are a variety of specific concerns that careful reading of the report reveals.

Report does not focus on a particular product or use application. As mentioned previously, there are a wide variety of yeast and yeast-derived products available to animal producers. However, the report *does not explicitly state* a specific product or class of products that is being considered. There are numerous references made to other types of yeast products. Thus, although the title is 'cell wall carbohydrates', the application is written so ambiguously that, if approved, a whole *range* of products could be theoretically employed. One assumes that this would *not* be the intent of an approval. This situation is troubling and makes one wonder whether the report is purposely vague.

Most of the report discusses the use of cell wall carbohydrates as binders of mycotoxins (pp. 5-12). Yet, the explicitly petitioned use of the material is as a "substitute for antibiotics" (p. 25). Since mycotoxin binding and anti-microbial activity are completely *separate issues*, serious questions are raised about the actual intended use of the cell wall carbohydrates. Given the fact that many antibiotics are fed to animals as growth promoters, it seems that the substitute of cell wall carbohydrates would not be permitted under the NOP rules. The report itself (e. g., p. 9 and 27) makes explicit note of the "performance-enhancing" nature of yeast additives.

Lack of information on production of cell wall carbohydrates. As noted above, most industrial fermentations involving yeast are based on maize as a feedstock. Genetically modified corn is increasingly being used as a substrate for fermentations and it is entirely possible, and probable given the nature of the processing steps, that any yeast or yeast-derived products would contain residual maize substrate and, consequently, genetically modified material. In addition, even if the material from a particular fermentation tank was intended for organic, the same (or another) tank in the plant could have genetically altered material in it at some point. Again, the report makes no mention of production methods and so it is impossible to evaluate the potential contamination of materials intended for organic use.

Use as a mycotoxin binder may not be justified. Even if one assumes that cell wall carbohydrates can be produced in a manner consistent with the intent of organic agriculture rules, another question becomes whether this material is actually needed or appropriate for use in organic animal agriculture. As the report details (pp. 10-12), mycotoxin contamination is mainly a problem related to particular production systems (e. g., swine, poultry, beef feedlots) that feed significant amounts of concentrate (grain). In addition, young, non-ruminant animals are the population most susceptible to mycotoxin poisoning. Also, animals in conventional, highly intensive operations may be more susceptible not only because of higher concentrate feeding, but because they are typically subject to higher levels of stress and disease than is probably seen in organic operations.

As the report notes (pp. 30-34), management practices designed to minimize formation of mycotoxin in feedstuffs during storage or processing and withdrawal of contaminated feeds are effective methods by which to prevent toxicosis problems. Therefore, given the 'whole-system' nature of most organic cultural practices, the emphasis on appropriate technology and management, and an emphasis on forage and pastures as feedstuffs for animals, it is not clear whether the use of yeast-derived products in organic systems is justified. This seems to be especially the case for adult ruminant animals.

Tone and organization of report detract from overall quality of presentation. In many respects, the report seems to have been pieced together from a variety of literature from a particular manufacturer and in places (e .g., bottom of p. 7) is self-serving product promotion. There are numerous examples of scientific studies being cited but no literature citations are provided (pp. 3-4). Some of the material is not even relevant to the issues at hand (p. 8). In places, terms are not defined (e. g., DFM). Overall, one is left with the sense that large portions of the report were prepared in a rushed fashion. This does not serve the review purpose.

Summary. Cell wall carbohydrates from yeast appear to prevent mycotoxin poisoning in certain production systems. Other yeast-derived products can be used to enhance animal growth and could replace antibiotics that are currently used in some production systems. It appears that such supplementation is most beneficial in intensive animal operations and at high production levels which are often characteristic in intensive systems. However, the application for use of yeast products, as outlined in this report, is too broad; namely, it is not at all clear whether purified cell wall carbohydrates, partially purified yeast walls, whole yeast cultures, or some combination of these is being proposed for use. In addition, there is the question of whether the intent of the OPFA and NOP permits the use of such materials on the basis of their growth-promoting effects. There is a lack of description about how the cell wall carbohydrate material would be produced and the potential problem of contamination with genetically altered material. The report also suffers from an unfocused presentation that, at times, appears to be taken directly from promotional literature.

(Reviewer 3 Conclusions)

Given (i) the lack of information about production methods, (ii) ambiguity about intended uses of cell wall material and yeast-derived products, (iii) questions about whether use of such materials is justified under most organic production regimes, and (iv) the very vague definitions surrounding organic animal nutrition and production, the reviewer recommends that cell wall carbohydrates and other yeast-derived products not be recommended for inclusion on the National List at this time. In the reviewer's opinion, each of the four points noted above must be very carefully evaluated prior to further consideration of yeast-derived products, including cell wall carbohydrates. As a matter of emphasis, there needs to much more thoughtful consideration given to the specific intent of feeding animals supplements and additives in sustainable production systems. It is suggested that even if approval were granted after such consideration, the application of cell wall carbohydrates be restricted to particular animal species (i. e., non-ruminants).

Reviewer 3 Recommendations Advised to the NOSB

The substance is Synthetic.

For Livestock, the substance should be Excluded from the National List.

TAP Conclusion

Of the three reviewers, two advised the addition of cell wall carbohydrates to the National List with restrictions, while one advised that they be excluded at this time. Two ruled cell wall carbohydrates to be synthetic and one was unable to determine whether or not they are synthetic. Concerns included the lack of information on how exactly yeast derivatives were processed in order to derive the cell wall carbohydrates, the possible growth-promoting effects of yeast products, and ambiguity as to what the cell wall carbohydrates would be used for.