VALUE-ADDED TRAITS OF ALFALFA

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ABSTRACT

Value-added traits of alfalfa are needed to provide farmers new high value profitable products. Processing alfalfa to obtain value added products includes three different fractionation methods: 1) wet fractionation; separation into juice fraction and a fiber fraction, 2) dry fractionation; separation into leaves and stems, and 3) fractionation by passage of the whole herbage through the digestive systems of ruminant animals, leaving a high fiber residue. Phytase from transgenic alfalfa has been tested in poultry and swine rations. Chicks supplemented with phytase from transgenic alfalfa juice or leaf meal had growth equal to chicks fed phosphorus supplemented rations. The manure from these chicks supplemented with alfalfa phytase contained less than half the phosphorus levels of manure from chicks fed inorganic phosphorus supplements. The economic value of phytase alfalfa product could generate $750 to $1500/A income from alfalfa grown in the Midwest. Alfalfa hay can be fractionated to yield stems and leaf meal. Alfalfa leaf meal has been shown to be acceptable supplement to replace a portion of alfalfa hay and soybean meal in diets of lactating dairy cattle, replace protein supplement in beef cow diets, finishing steer diets and diets of growing turkeys. Current energy costs in this country limit the use of alfalfa stems to generate electricity from gasification. The fiber portion of alfalfa can produce lactic acid and ethanol. The fiber from alfalfa manure has yielded press board and water filters capable of removing heavy metals from contaminated water.

Key Words: alfalfa, fractionation, transgenic, phytase, and enzymes

INTRODUCTION

Approximately 84 million tons of alfalfa was harvested from U.S. farms in 1999, having an estimated value of $6.2 billion, ranking behind only corn and soybeans. Alfalfa is a major field crop, which supports dairy, beef and sheep production in the U.S. Alfalfa hay acreage and production has remained constant in recent years. Alfalfa hay production is moving west with the rapid growth of the dairy industry in the West. Dairy farm numbers are declining most rapidly in dairy regions of the Midwest. Beef cow-calf production remains relatively constant on U.S. farms.

New alfalfa products of high value are needed to expand the acreage of alfalfa within the United States. Research efforts are underway to develop alfalfa with value-added traits and to develop new processing technologies.

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VALUE-ADDED PROCESSING OF ALFALFA

Three methods of forage fractionation exist:

- wet fractionation; separation into juice fraction and a fiber fraction,
- dry fractionation; separation into leaves and stems,
- fractionation by passage of the whole herbage through the digestive systems of ruminant animals, leaving a high fiber residue.

Two important conditions must be met for alfalfa fractionation to be feasible and sustainable are:

- the total value of the resulting products must be greater than the original forage plus the cost of processing;
- all fractions must have an economic value to avoid creating a waste stream.

The wet-fractionation process has two advantages for agriculture:

- forage crops can be harvested almost independent of weather, since moisture is removed by mechanical expression rather than by field drying,
- a versatile protein concentrate is obtained which can be fed to non-ruminants, including humans, as well as being used to supplement rations of ruminants such as high-producing dairy cows.

Wet fractionation of forage crops allows biomass to be produced at very competitive prices due to the high values of the co products. The fractionation process consists of expressing juice from fresh herbage. The resulting fibrous fraction is high in cell wall constituents (cellulose, hemicellulose, and lignin). It is suitable for combustion, gasification, or enzymatic hydrolysis and fermentation to ethanol or organic acids (e.g., lactic).

The juice fraction contains 25 to 30% of the dry matter in the original herbage, depending on the severity of processing. It is high in protein and solubles and is almost fiber-free. It can be used to produce both food-grade and feed-grade protein concentrates as well as other high-value products (xanthophylls for pigmenting poultry products; enzymes such as phytase, cellulases, lignin peroxidase and α-amylase and biodegradable plastics, all from transgenic alfalfas).

HIGH PHYTASE TRANGENIC ALFAFLA

Buildup of phosphorus in the environment and the resulting degradation of water resources are of mounting concern. Much of this buildup is traceable to human activities. Important among these is livestock production. Monogastric animals, such as poultry and swine, which can solubilize only a small fraction of the phosphorus in their grain-based rations while excreting the remainder, have come under increased scrutiny. Supplementation of inorganic phosphorus into rations to meet animal nutritional requirements exacerbates the problem.

Much of the phosphorus in grain is in the form of insoluble phytates. Researchers have shown that supplementing poultry and swine rations with the enzyme phytase can lead to solubilization of the phosphorus, thus eliminating the need for phosphorus supplementation and concurrently reducing the level of phosphorus in animal excrement to approximately one-half of that normally experienced.

The enzyme phytase derived from *Aspergillus niger* has to date, generally been produced in fermentation vats using genetically engineered microorganisms. It has been estimated that the cost of phytase supplementation with this material would be about three times the cost of conventional supplementation with dicalcium phosphate.

As an approach to reducing the cost of phytase production, a multi-disciplinary USDA-ARS-UW team at Madison, Wisconsin has produced transgenic alfalfa with the capability of expressing phytase (Austin-Phillips and Bingham, 1997). This phytase can be recovered with juice extracted from herbage. Leaf meal, which typically has protein and fiber contents greater than 25 % and less than 20 %, respectively, has also been used successfully.

Koegel et al. (1999) reported feeding growing chicks with alfalfa-produced phytase, which at appropriate levels can totally replace the inorganic P supplementation, Fig. 1. Replacing inorganic P with phytase resulted in a reduction of P concentration in poultry feces to less than one-half. They further reported that alfalfa phytase in the form of fresh juice, dried juice, or leaf meal was all effective with swine, Fig. 2. and poultry (data not shown).

The quantity of phytase, which can be produced in transgenic alfalfa, is on the order of 200 X 10^6 units/acre/year, equivalent to an amount able to treat 500 tons of poultry ration. At current cost of inorganic P supplementation, the value of phytase would be \$750 - \$1500 per acre-year. The value of xanthophylls and protein content of alfalfa as well as the environmental benefits would be in addition to this.

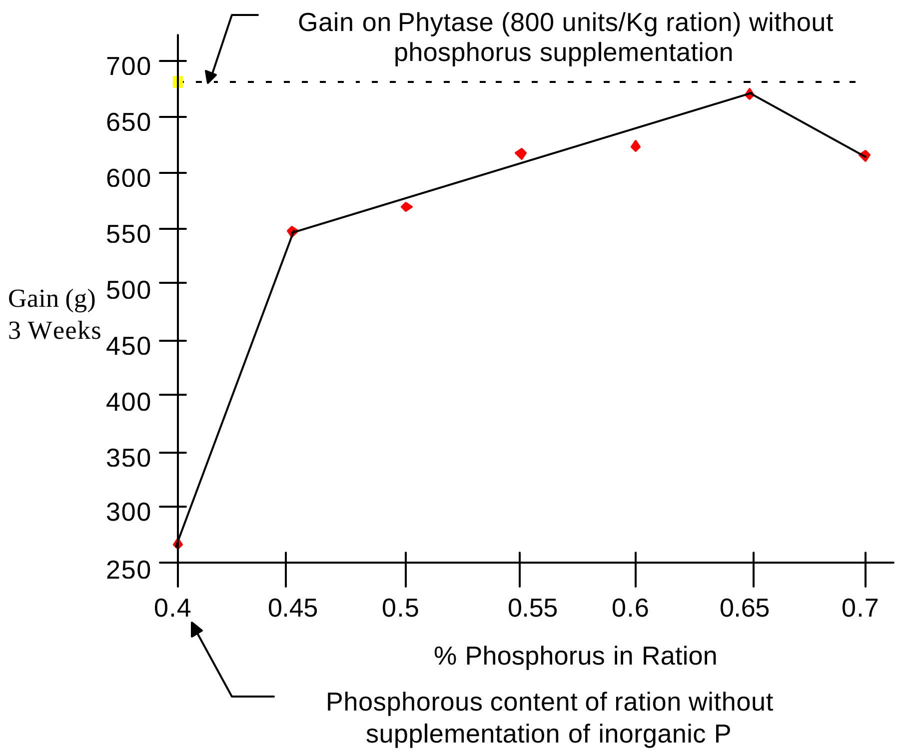

Fig. 1. Three-week gain of chicks vs. phosphorus content of ration compared to phytase with no phosphorus supplementation. SOURCE: Koegel et al., 1999.

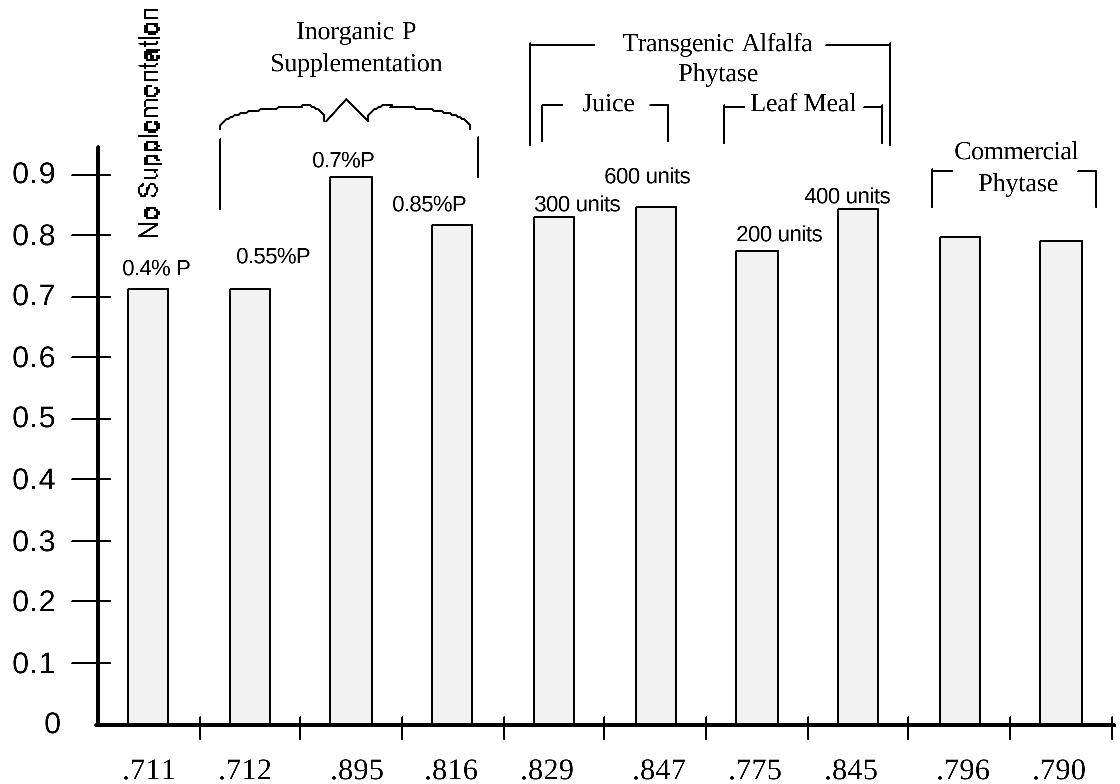

Fig. 2. Daily gain (kg) of weanling pigs fed at various levels of inorganic phosphorus supplementation or with phytase supplementation from various sources. Trial duration: 18-21 days. SOURCE: Koegel et al., 1999.

DRY FRACTIONATION OF ALFALFA HAY

Dry fractionation of alfalfa hay into leaf meal and stems used as solid fuel was pioneered by the Minnesota Valley Alfalfa Producers (MNVAP), Granite Falls, Minnesota and Northern States Power (NSP), Minneapolis, Minnesota, Martin and Oelke, 1996. The research effort to produce 75 MW of electrical energy from alfalfa stems via gasification was initiated in 1993 by Department of Energy, University of Minnesota, MNVAP and several power generation partners. The project was cut short of construction of a new power generation plant in May 1999 due to negation of the original power purchase agreement between MNVAP and NSP. NSP was under legislative mandate to generate power from a closed-loop farm grown biomass system by 2002. However, under pressure from businesses not willing to pay the alfalfa-electricity based energy prices, alfalfa processors claiming unfair research product support for MNVAP and unusual delays in approval of the power purchase agreement by the Public Utilities Commission the project was terminated (Sheaffer et al. 2000). However, animal feeding trials to investigate the use of alfalfa leaf meal (ALM) for dairy cattle, beef, and turkey feed supplement have been completed (DiCostanzo et al., 1999).

Nutrient composition of ALM obtained by manual or mechanical separation is listed in Table 1. Initial studies using ALM as a protein source or as replacement for hay were based on lower quality ALM. This was likely due to the initial mechanical adjustments that the plant had to undergo to get stems separated from leaves. Studies conducted after 1997 used ALM of quality similar to material obtained by manual separation. At this point, the plant had adopted additional measures to separate stems from leaves. This material is now available for commercial use; its composition is listed in Table 1.

Using the fiber content of ALM to predict TDN value results in estimates of TDN that range between 67 and 72%, depending on the formula used. Thus, ALM compares well to the energy content of high quality hay or small grain silages.

Table 1. Nutrient composition of alfalfa leaf meal obtained through manual or mechanical separation and its applications.

	Separation			
	Manual	Mechanical		
Application:	Laboratory 1994	Experimental 1996-1997	Experimental 1998-1999	Commercial 1998-1999
Dry matter, %	92.5	93.45	92.29	89.67
	----------% DM----------			
NDF	36.0	36.48	43.63	34.45
ADF	21.5	21.90	26.60	25.09
CP	25.2	21.89	25.78	28.23
Ash	--	11.42	12.42	14.75
Calcium	--	2.03	2.15	2.88
Phosphorus	--	0.30	0.29	0.34
Potassium	--	2.24	1.92	2.24
Sodium	--	0.09	.08	0.04
Magnesium	--	0.38	0.37	0.43
	----------ppm DM----------			
Manganese	--	66.57	64.36	87.00
Iron	--	491.10	722.60	625.00
Zinc	--	33.00	28.65	39.00
Copper	--	13.20	16.04	15.75

SOURCE: DiCostanzo et al. 1999.

Lactating cow. A feeding trial with lactating cows is included as an example of the animal feeding trials conducted by DiCostanzo et al. 1999. A 3 x 3 Latin square study (11 to 14 d adaptation, and 5 to 7 d sample collection) was conducted to determine the value of ALM pellets as a substitute for high quality alfalfa hay (Jorgensen, 1998). The forage portion of the control diet (containing no ALM) was equally derived from alfalfa hay and corn silage dry matter (DM). Alfalfa leaf meal pellets replaced 28% or 56% of the alfalfa hay, and 7% or 15% of the soybean meal (SBM) in the low- or high-ALM diets, respectively. Dietary energy (1.69 Mcal NE_L/kg DM) and protein (18%) concentration was balanced to be similar across all diets. Macro- and microelements were balanced so that diets provided similar concentrations of these elements. Consumption of ALM averaged 2.31 or 4.23 kg of ALM pellets/d for cows fed the low- or high-ALM diet, respectively. Cows fed the high-ALM diet had lower ($P < .05$) dry matter intake (DMI) than those fed the low-ALM diet; DMI of cows fed no ALM was intermediate (Table 2). As a result of lower DMI by cows fed the high-ALM diet, intakes of protein, NDF and ADF were lower in cows fed the high-ALM diet than in those fed the low-ALM diet. Milk yield, milk composition, or yield of fat, protein or lactose was not affected ($P > .05$) by substituting ALM for alfalfa hay (Table 2). Inclusion of ALM in the diet had no effect on DM digestibility (data not shown).

Table 2. Effects of substituting alfalfa leaf meal for alfalfa hay on intake and milk yield of lactating dairy cows.

Item	ALM[a] substituting alfalfa hay at, %			
	0	28	56	SE
Diet composition, % DM				
Corn silage	25.8	26.0	26.0	
Alfalfa hay, chopped	25.9	18.5	11.2	
ALM pellets	--	7.9	15.8	
Concentrates[b]	48.3	47.6	47.0	
DMI, kg/d	28.0[cd]	29.3[c]	26.7[d]	.60
Milk, kg/d	38.9	39.8	39.6	.67
Fat, %	3.69	3.47	3.64	.08
Protein, %	3.10	3.03	3.07	.03
Lactose, %	4.80	4.70	4.70	.06

[a] ALM = alfalfa leaf meal
[b] Cracked corn, distillers dried grains, soybean meal, wheat midds, molasses, tallow, urea, and minerals and vitamins to balance diets for nutrient requirements.
[c,d] Means with different superscripts differ ($P < .05$).
SOURCE: Jorgensen, 1998.

Minnesota researchers (DiCostanzo et al., 1999) concluded that ALM is a suitable substitute for hay and soybean meal in diets of lactating dairy cows, although some question remains as to the performance and body weight response to ALM supplementation during a whole lactation. As a component of starter diets, ALM has the potential to enhance intake and gain when constituting 12% of the starter DM. At greater inclusion proportions, ALM may reduce intake in young ruminants. In dairy calves (aged 4 to 40 d), this reduction in intake may or may not be accompanied by a reduction in weight gain. The latter was the case when suckling calves were offered creep feed for 80 d before weaning. In receiving (growing beef animals after shipment) diets, DMI enhancement may not be accompanied by a gain response; thus, feed DM required/kg gain may increase. This effect does not appear to carry over into the finishing period. In fact, finishing steers fed 9% of their diet DM as ALM had faster gains at greater intakes. The most "adequate" inclusion level for ALM appears to be between 7 and 12% of the diet DM in beef cattle. Effects of ALM on incidence of liver abscess in feedlot diets are somewhat inconclusive and warrant further study. Similarly, efforts to enhance the value of ALM through heating to render a bypass protein source must focus on reducing exposure time or temperature.

While solid fuel yields the highest net energy and has the lowest processing cost, its use is generally limited to electric power generation. An alternative to solid fuel is the saccharification and fermentation of the ligno-cellulosic, fiber fraction to ethanol (Koegel et al., 1999). While conversion of fiber to ethanol results in less total energy and is a more complicated process, its versatility and potential use as a transportation fuel make it an interesting alternative.

LIMITATIONS TO BIOTECH TRAIT APPLICATIONS:

Introduction of new traits into alfalfa by genetic modification has tremendous potential to improve alfalfa production and utilization by providing new products of high value, by improving the utilization of current alfalfa products (i.e., alfalfa haylage), and by improving the environment through reduction of N and P in animal manure. With exception of improving protein utilization of alfalfa silage by incorporation of the enzyme PPO, most new products require large investments in facilities to process alfalfa for either wet or dry fractionation. Until the Public Utilities Commission in Minnesota reversed its approval of MNVAP's contract with NSP of Minnesota during the fall of 1999, it appeared that alfalfa would not only be processed for value-added products (stems for electricity and leaf meal supplement for livestock), but it also would have been an example of agriculture and energy industries cooperating to provide farmers a new product that would be sustainable both economically and environmentally. Alfalfa research needs both public and private support to engage in new capital expenditures to process alfalfa into new non-feed products.

In addition, genetic modification of alfalfa will be expensive and political. Legal battles over property rights and the negative perception of the consuming public will also influence application of Biotech to alfalfa. This symposium can be the beginning of an industry-wide (public and private research and education; alfalfa, forage and grassland organizations) effort to ensure that research and development of new products is made available to the struggling rural communities of the nation.

SUMMARY AND CONCLUSIONS

Genetically modified alfalfa can be processed to provide new products of higher value than traditional alfalfa products for monogastric and ruminant animals. Processing green alfalfa to produce products from separated fiber and products from processed juice removes the traditional hindrance rain damage to product harvesting. Corn and soybean cash crop farmers will benefit from any type of alfalfa fractionation discussed. Genetic modification is expensive research; therefore, the industry must insure those value-added traits with the high potential profit for farmers are exploited. We must cooperate as an industry united to provide the public with safe, nutritious food under an environmental resource abatement goal.

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