# TABLE XII

Effect of Different	Levels of	Linoleate	on th	e Growth	of Male
and Female Chicks	(17)				
			_		

Dietary	% Calories	6-w	6-week	
linoleate	as	body v	body weights <sup>1</sup>	
%	linoleate	Males	Females	
0.0	0.0	362 <sup>a</sup>	363 <sup>a</sup>	
0.15	0.5	392 <sup>bc</sup>	359 <sup>a</sup>	
0.3	0.9	423 <sup>cde</sup>	378 <sup>bcd</sup>	
0.6	1.8	453 <sup>ef</sup>	409 <sup>de</sup>	
1.2	3.6	479 <sup>fg</sup>	439 <sup>de</sup>	
2.4	7.3	509 <sup>g</sup>	439 <sup>de</sup>	

<sup>1</sup>Means having the same superscript do not differ significantly  $(P \leq .01)$ .

Since increased dietary energy is usually accomplished by substituting corn (rich in linoleic acid) for other cereal grains or fibrous byproducts (usually low in linoleic acid), or through the increased addition of supplemental fats (which may range from low-linoleic acid sources such as tallow to high-linoleic acid sources such as the vegetable oils), it becomes apparent that at least a portion of the response to increased dietary energy levels may be the result of increased levels of linoleic acid per se. Further studies are needed to determine the extent of this response in broilers to market age.

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#### TABLE XIII

Effect of Level of Dietary Linoleate on Growth of Male Broiler Chicks (18)

Dietary	Added lino	Weight	
source of linoleate	% of diet	% of calories	gain (g)
None	0	0	496 <sup>a</sup>
Safflower oil	0.7	2.1	540 <sup>0</sup>
	1.5	4.2	465
	2.9	8.1	586 <sup>cd</sup>
	4.4	11.7	591 d
	5.8	15.2	$601^{d}_{1}$
*	11.7	27.1	594 <sup>d</sup>
Corn oil	1.7	4.8	587 <sup>cd</sup>
1	3.4	9.2	584 <sup>ca</sup>
	5.2	13.2	$610^{d}$
•	6.9	16.9	604 <sup>d</sup>

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## ABSTRACT

Major research efforts by university and industry workers throughout the world have been directed toward improving the utilization of soybean protein by the calf. This paper reviews the literature and summarizes the questionable characteristics and methods of improving utilization of soya protein sources for young calves. Current application of soya protein in calf milk replacers is discussed.

# INTRODUCTION

Interest in utilizing vegetable protein in milk substitutes for calves has increased as a result of the potential economy in calf raising and the increasing importance of milk protein for humans. The vast majority of investigations have utilized soybean protein due to its potential nutritional value and its abundance.

Numerous reports have indicated reduced performance of calves when fed milk replacers containing soya flour (1-4). However, chemical modification has resulted in good performance (5-7). Diarrhea has been reportedly increased in calves when soya flour was added to milk replacers (1,3,8), but some workers have not indicated this is a problem (2,4,6,7). Several reports have demonstrated reduced performance of calves when fed milk replacers containing soya protein concentrates (4, 9-13); however, other reports have noted good results (3) particularly at lower protein replacement levels (10,11). Some reports have found that diarrhea increased when soya protein concentrate was added to milk replacers (8,12), yet others found no problem, or found even improved fecal consistency (3,4,9,10,13,14).

Utilization of soya protein by calves presumably varies due to factors inherent within various soya protein sources. This paper reviews the literature and attempts to summarize the questionable characteristics and methods of improving utilization of soya protein sources for the young calf. Current application of soya protein in calf milk replacers is discussed.

# **QUESTIONABLE CHARACTERISTICS**

## Digestibility of Nutrients and Passage of Digesta

Reduced digestibility of total nitrogen, fat and ash of milk replacers containing a soya protein concentrate and a soya meal has been reported (4,9) (Table I). Protein digestibility for the soy protein concentrate and soya meal was estimated to be approximately 75% and 50%, respectively. Carbohydrate digestibility of soya protein concentrate containing replacers was less than (9) or equivalent to (4) that of milk protein concentrate replacers. Carbohydrate digestibility of a soya meal containing replacer was somewhat less than an all milk protein replacer (4). Milk replacer containing a soya protein concentrate yielded lower dry matter, nitrogen and gross energy digestibility than that from a milk protein replacer (15). The calculated digestibility of soya protein was 79% in that experiment. It has been suggested that soya protein sources may reduce digestibility by causing intestinal absorption capacity not to improve in early life (8) as determined by xylose absorption studies (Fig. 1).

As calves age, improved digestibility of nutrients is apparent, with extreme variation among calves (16) (Fig. 2). Utilization of soya flour containing milk replacers was not satisfactory until the calves were approximately 25 days of age (17). Marked increases in nitrogen, fat and ash digestibility were found as calves increased from 1 to 2 weeks of age (4). Digestibility of nitrogen and fat of a soya protein concentrate containing formula increased from the third to the fifth week on experiment, but no change was noted for calves fed a soy meal (4) (Table I). Calves started on experiment at an average age of 30 days yielded equivalent performances when fed milk or soya protein

## TABLE I

Apparent Digestibility of Components of Milk R	(4) teplacers
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Week of experiment	Component	Protein source Milk Soy prot. conc. Soy meal		
		(%)	(%)	(%)
3	Nitrogen	92	80	66
5		95	85	67
3	Fat	96	75	84
5		96	86	84
3	Ash	94	78	70
5		92	77	70
3	NFE	98	98	94
5		99	96	95
3 5	Soy protein		72 79	51 52

replacers (4).

When concentrates and hay were made available to calves in addition to milk replacers, the differences in growth rates of calves fed milk or soya protein replacers diminished (4).

Inhibition of abomasal emptying, an increased rate of passage and flow of digestion through the small intestine, abnormal water and salt exchange, and decreased nitrogen absorption have been reported when various soya protein sources were fed to calves (18,19). The flow rate from the abomasum of dry matter and protein from a soya protein concentrate was slower than that from milk or fish protein sources (14), but was faster with a soya flour than with milk protein (20).

Decreased nutrient digestibility and/or absorbability and abnormal digestion flow, resulting from the use of soya protein in calf milk replacers, have been cited as contributors to poor calf growth and diarrhea.

#### **Trypsin Inhibitor**

Trypsin inhibitor activity of soy protein sources has been negatively correlated with calf growth (21,22). Researchers have suggested that this is brought about by reduction of the level and concentration of pancreatic trypsin and chymotrypsin secretion in the calf (3,23,24) (Fig. 3). Some evidence indicates that normal conditions in the abomasum (HCl and pepsin secretion) may reduce trypsin inhibitor activity over time (25,26) (Fig. 4). However, alkaline conditions such as those normally found in the small intestine



FIG. 1. Mean peak xylose concentrations in plasma at 2 to 2.5 hr after an oral dose of 5 g xylose/kg body weight, in five calves fed milk, soy flour, soy protein concentrate, or Promocaf from 1 through 5 wk of age (modified from 8).



FIG. 2. Effect of age on mean and individual digestibility of protein (16).



FIG. 3. Total trypsin and chymotrypsin activities in intestinal digesta collected from calf 1 with different levels of soybean trypsin inhibitor extract added to whole milk. Collection times 0, 2 and 5 are 30 min intervals just before, and 2 and 5 hours after the a.m. feeding (25).

may increase trypsin inhibitor activity of soya through the "release" of bound inhibitor (16) (Fig. 5).

The exact role of trypsin inhibitor in calf nutrition remains unanswered. The growth rate of calves fed for 3 or 5-6 weeks was not related to the total trypsin (r=-.20) and chymotrypsin (r= -.19) content of the pancrease (3) which indicates that these measurements may be misleading. Some data suggests there is no effect of trypsin inhibitor in the calf (27).

The literature would indicate that trypsin inhibitor may play only a partial role in the response of calves fed soya protein sources. The effect of processing soya protein sources on trypsin inhibitor activity is discussed in a following section.

## Antigenicity

Digestive disturbances noted when soya products were fed to calves indicated there may be gastrointestinal allergic response (18,19). Changes in intestinal morphology became apparent after 7 days of feeding soya protein containing milk replacers (28).

Serum antibody response was recorded within 3 weeks of initial feeding of soya protein milk replacers, and previously sensitized calves responded to reintroduction of soya protein in the replacer with marked increases in antibody level (28) (Fig. 6). The presence of an antibody to soya protein is common in the neonatal calf; this antibody originated from the colostrum (29). The production of antibodies in calves fed heated soybean flour coincided with an increased rate of digesta flow (30), which was specifically associated with feeding soya products containing the antigenic proteins, glycinin and Beta-conglycinin (31).

Differences in calf performance and health cited in the literature, from various soya protein sources, could be a result of the antigenic properties of the soya protein sources. The effects of processing soya protein sources on antigenicity is discussed in this paper in a following section.



FIG. 4. Comparative decrease in trypsin-inhibiting activity of RSBM and SBM<sub>4,2</sub> treated with HCl (pH 1.6) and with pepsin; RSBM incubated with HCl (pH 1.6) (...o...); RSBM incubated with HCl (pH 1.6) containing 0.2% pepsin (...o...); SBM<sub>4,2</sub> incubated with HCl (pH 1.6) (...o...); and SBM<sub>4,2</sub> incubated with HCl (pH 1.6) containing 0.2% pepsin (...o...) (27).

#### **Carbohydrate Content**

The nutritional value of carbohydrate from soya protein sources is questionable, since the young calf is capable of extensive utilization of lactose and glucose with little or no value from starch (32-34). Soya flour in milk replacer lowered amylase concentration (23,33). No increase in concentration occurred in the first 21 days of life (33), further suggesting the limited availability of carbohydrate for calves in soy protein sources.

Digestibility of the carbohydrate fraction for milk replacers containing various soya protein sources indicates that considerable carbohydrate disappeared (4,9,27), presumably due to intestinal fermentation. Carbohydrate digestibility of soya protein concentrate containing milk replacer was equal to that of a milk protein replacer, but carbohydrate digestibility in a soya meal replacer was some-



FIG. 5. Effect of pH during sample preparation of the anti-tryptic activity of soy flour (16).



FIG. 6. (a) Mean  $\log_{10}$  titres of groups of sixteen calves fed either a high-fat baby calf food containing no soya protein ( $\blacksquare$ ), or 'Gold Top' baby calf food (BOCMS) containing either 10% Sorbosoy ( $\bullet$ ) or 25% Hisoy ( $\blacktriangle$ ) (British Arkady Ltd). (b) Log<sub>2</sub> haemagglutination titres of two calves fed a milk replacer containing 40% HESM on two occasions (diet S) separated by a period of no-soya protein diet (diet C). Hatched area indicates a period of severe scouring occuring in both animals (29).

what less (4) (Table I).

The starch level should not exceed 10% in milk replacers fed to calves under 3 weeks of age (35). Since soya flour contains 30-40% carbohydrate, the replacement of milk protein in a quantity up to 50% in calf milk replacers should not be detrimental to calf performance. The level of carbohydrate in soya protein concentrates (15-20%) could be advantageous at high soya protein concentrate replacement levels.

### **Oil Content**

Crude and refined soybean oils in milk substitutes have severely retarded growth and dramatically increased diarrhea in young calves (36-38) (Fig. 7). Hydrogenation of the soybean oil improved calf performance and health to be equivalent to those fed butterfat containing controls. Indications



FIG. 7. Effect of type of oil in the reconstituted milk diet of calves on the changes in body weights (37).

of deterioration during storage were cited (38); however, no direct cause of the deleterious effects was found.

Further investigation using unsaturated vegetable oil (corn oil) produced detrimental performance and health in calves (39). Reduced digestibility (40), and reduced vitamin concentration in plasma (41) of calves was especially noted when stored vegetable oil was used. The rate of abomasum emptying was not influenced by unsaturated vegetable oil (corn oil), nor were there indications of an adverse effect on gastric secretion of HCl or proteolysis (42). In this same study, fecal dry matter was markedly lower for calves fed milk replacer containing unsaturated vegetable oil.

The mechanism by which highly unsaturated fats cause poor growth and diarrhea in young calves has not been elucidated. Since soya flour contains approximately 3% fat, the replacement of milk protein in a quantity up to 50% in calf milk replacers should not be detrimental to calf performance. The level of fat in soya protein concentrates is negligible and therefore presents no concern.

#### Hemagglutinin Activity

Raw soya protein contains hemagglutinin, which is toxic to rats (43). Hemagglutinin is described as playing as important a role as trypsin inhibitor in the inhibition of growth rate (44). The major cause of inhibition of growth associated with hemagglutinin was attributed to reduced feed intake. Hemagglutinin's sensitivity to heat, acid, alkaline and pepsin make it easy to inactivate this toxin (26, 25); therefore, when processed soy protein sources are utilized, this growth-inhibiting factor should be of little concern.

## METHODS OF IMPROVEMENT

## **Amino Acid Supplementation**

Methionine is the limiting amino acid for rats, chicks and pigs when soy protein is offered, and is therefore assumed



FIG. 8. Mean cumulative changes in body weight of calves fed diets A, B, C, negative control, and commercial replacer (modified from 5).



FIG. 9. Body weight gains of the calves fed Milkvit, and heated (HSD), partly heated (PHSD), and raw soybean milk replacers (RSD) (modified from 9).

to be of equal importance in the preruminant calf. Actual requirements for the calf have only recently been established (46-49). Additions of 0.05 and 0.25% methionine to soya flour containing milk replacers did not improve calf performance (50). No effect on calf performance or nutrient digestibility was noted with addition of 0.10% methionine to a soya protein concentrate containing milk replacer (15). Addition of 0.60% methionine reduced the performance of calves fed milk replacers containing toasted soya flour (51), possibly because of amino acid imbalance.

The literature available does not indicate there are benefits from amino acid supplementation. If soya protein replaced 50% of milk protein in milk replacers, it would probably contain adequate amounts of essential amino acids (48).

## Enzyme Supplementation

Addition of enzymes to soya protein sources has been suggested as a means to improve calf performance. Enzymatic predigestion of soya flour did not stimulate calf growth even though the protein and carbohydrate were extensively damaged (5) (Fig. 8). Apparent improvement from one preparation (Diet C) was determined to be from the acidification of soya flour, not enzymatic digestion. Addition of 0.5% pepsin to milk replacers containing graded levels of soya flour did not prove to be beneficial (2). Calves fed milk replacers containing soya flour were not benefited by the addition of 0.5% pepsin (52).

Failure of a response to enzymatic treatment cited in the literature may be a result of only partial degradation of protein. The low correlation of calf growth and total trypsin and chymotrypsin content of the pancreas (3) suggests this practice may be futile.

#### Heat Treatment

Improved utilization of raw soya protein sources by calves has been noted as a result of heat treatment. Calf performance and digestibility improved significantly through heat treatment; however, a fully heated soya protein did not provide utilization to the extent of that from milk protein (9) (Fig. 9). In that study, soya protein digestibility was improved from 73 to 89% through heat treatment. Other reports in literature support the value of heat treatment in improving soya protein utilization (5,21,22,27). Overheating of soya protein may be undesirable (12).

Heating soya protein sources results in inactivation of trypsin inhibitor activity (12,26) (Fig. 10). Other potential benefits of heat treatment may be results of the destruction of other inhibitory factor(s), or improved susceptibility of protein to enzymatic degradation.

## Acid Treatment

The treatment of soya protein sources with acid has improved their utilization by calves. Calf performance im-



FIG. 10. Effect of heating on the anti-tryptic activity of soya flour (16).

8

6

4

ΰ

£

a – Caseinate

SPC 5

SPC 0

PC 0.25



FIG. 11. Mean cumulative changes in body weight of calves fed acidified and nonacidified fully cooked soya flour (modified from 5).

proved significantly when fully cooked soya flour was treated at pH 4.0 vs pH 6.4 (5) (Fig. 11). A response was also noted in calves to acid treatment of uncooked soy flour, but negative weight gain was experienced even for the acid-treated group. This indicated that additional detrimental factors were present that acid treatment did not eliminate. Further research with acid treatment revealed a positive effect on calf performance (53, 54), with a possible exception (16).

Acid treatment of soya protein sources reduced trypsin inhibitor activity (16,25,26). Reversal of this inactivation may occur with a return to alkaline solutions (16,25). Beneficial effects may also result from destruction of other inhibitory factors or from improved nutrient availability. Lack of satisfactory gains of calves from acidification of uncooked soya flour indicates an incomplete improvement from acid treatment.

## Alkali Treatment

Soya protein sources treated with alkali have yielded improved calf performance. A combination of thermal and alkali treatments improved calf performance significantly (12) (Fig. 12). Performance was optimum at 5 min of processing, longer duration yielded poorer results, indicating the potential to overprocess soy proteins (Fig. 13). The optimum treatment did not yield performance equivalent to a milk protein replacer, however. Thermoalkali treatment may decrease the availability of methionine (55) or produce lysinoalanine, a poorly absorbed amino derivative (56). Other workers have shown that alkali treatment of fully cooked soya flour has increased calf performance significantly (53) (Fig. 14).

The mechanism involved in the positive effects attributed to alkali treatment of soya protein sources has not been elucidated. Inactivation of inhibitory factors or improved nutrient availability remain plausible explanations.



Treatment of soya protein sources with alcohol has been reported to improve utilization of soy protein sources by calves. Extraction of soya protein sources with hot aqueous ethanol renders them nonimmunogenic and eliminates digestive disturbances normally found with soya protein use (18,19,31,57). According to researchers, alcohol treatment denatures specific antigenic proteins, glycinin and  $\beta$ -conglycinin, and soya protein sources containing these proteins yield the ill effects noted (31). Other researchers, however, indicate that heated-ethanol-extracted soybean meal does not render the soya protein antigens nonfunctional (28,29,58). Differences in treatment conditions may be responsible for this conflicting literature. The literature is consistent, however, in indicating a gastrointestinal allergic response of calves fed conventional soya protein sources.

Removing the antigenic properties of soya protein sources appears to enhance their utilization by calves; however, no direct growth data has been evaluated. Variations in sensitivity of calves to antigenic proteins may explain considerable variation seen within groups of calves fed various soya protein sources compared to those fed milk protein. It is puzzling that the literature cited as well as field experience indicate that calves fed milk replacers containing up to 50% replacement protein using nonalcohol-extracted soya protein sources do not generally show the severe symptoms noted. Perhaps further research will explain these observations.

### **CURRENT APPLICATIONS**

Approximately 70% of the dairy herd replacement calves in the United States are fed milk replacers and 60-65 % of this contains soya protein. This indicates that 3-5 million calves consume soya protein containing milk replacers



FIG. 13. Average cumulative body weight changes of calves fed replacers containing Na-Caseinate or TA-SPC processed for 5, 30, or 60 min (5).

yearly. Both processed soy flour and soy protein concentrates are used extensively.

Soya protein sources in calf milk replacer have been successful probably because of three major factors. First, soya protein sources used are "processed;" that is, they have been subjected to conditions from simple heating to ethanol extraction. Raw soya protein is not used. Second, the vast majority of replacers have 50% or less of the milk protein replaced by soya protein. This is undoubtedly a result of the unclear influences of the questionable characteristics that have been discussed in this paper. Third, 75% of the growth of calves during the first 4-6 weeks of life can be explained by intake of high-quality calf starters. Obviously, additional nutrients are supplied, supporting the requirements of the calf.

It is generally accepted that soya protein sources will



FIG. 14. Mean cumulative changes in body weight of calves fed milk replacers containing soya flour (modified from 54).

not provide calf performance and health equivalent to that from milk protein. Further research is needed to develop processing methods that will eliminate the influence of the questionable characteristics cited and will enhance utilization of soy protein by the calf.

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