

## Research Note

### **TARO LEAF BLIGHT (PHYTOPHTHORA COLOCASIAE): A NEW DISEASE IN PUERTO RICO<sup>1</sup>**

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Taro leaf blight, caused by *Phytophthora colocasiae*, has been known of since the beginning of the last century. Raciborski, in Java (Indonesia) in 1990, first described this Oomycete as the causal agent of a leaf spot of taro, *Colocasia esculenta* (L.) Schott. During the early 1990s, an outbreak of this disease devastated the staple taro crop throughout the Pacific Basin (Sar et al., 1998). This pathogen is recognized as a major limiting factor for the production of taro throughout Southeast Asia and the Pacific Basin. Within the Caribbean Basin, this pathogen has been reported in the Dominican Republic (Erwin and Ribeiro, 1996).

During December 2004, a type of blight was observed affecting leaves of cultivated taro in the sector of Quebrada Arenas, within the municipality of San Lorenzo, Puerto Rico. In this area, the dasheen type of taro is produced under wetland conditions. The pathogen was identified by using selective media for *Phytophthora* along with the observation of symptoms in the field. The Plant Pathogen Identification Laboratory at North Carolina State University confirmed the species by molecular procedures. This is the first time taro leaf blight has been reported in Puerto Rico. The disease has been confirmed to have spread among farms located in the Yabucoa municipality.

In the field, symptoms are circular brown to olive green spots on the upper leaf surface; these appear to be darker, water-soaked or greasy spots on the underside. Infection commonly begins on the lobes and the sides of the leaves where water often collects. As the disease develops, the spots enlarge and become irregular in shape. Spots sometimes turn dark brown with a yellow margin and concentric circles (Figure 1A); there may be present a clear amber fluid that exudes from the center of the spots. The exudate becomes dark brown and hard after drying. White powdery masses of spores indicating the presence of *P. colocasiae* are often produced around the lesions, seen most clearly early in the morning. The leaf blades are sometimes completely rotted, but do not drop from petioles. In the infected petioles, rot spots are usually long, brown and occur anywhere in the petiole. As the rot expands, the petioles become soft and, unable to support the leaf, break. Pathogenicity tests confirmed *P. colocasiae* as the cause of the taro leaf blight (Figure 1B).

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FIGURE 1. A. Symptoms of taro leaf blight in the field. B. Symptoms of taro leaf blight caused by *P. colocasiae* after five days of inoculation.

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## Research Note

### **SOLUBMAX™ FEEDING TO DAIRY COWS IN CONFINEMENT AND EFFECTS ON MILK FAT CONJUGATED LINOLEIC ACID (CLA) CONCENTRATION<sup>1</sup>**

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SynerMax® is a liquid coproduct obtained from the microbial production of the antibiotic erythromycin by means of an industrial fermentation at the operations of Abbott Laboratories in Barceloneta, Puerto Rico. It consists of the spent culture medium and residues of the cultured organism (a filamentous bacterium) after extraction of the desired antibiotic. A typical chemical analysis of this coproduct shows dry matter (DM), 48.6%; crude protein, 13.8%; crude fat, 10.2%; and ash, 9.2% (Abbott, 2000).

Most of this coproduct generated in Puerto Rico is shipped to North America, where it is sold primarily for use in poultry feeds. However, local commercialization of the coproduct is mainly as a liquid supplement in mixture with cane molasses, sold under the trademark SolubMax, and used mostly for feeding bovines, especially dairy cattle.

Among the interesting properties of this high-lipid coproduct is the fact that it is rich in precursors of conjugated linoleic acid (CLA). This term designates a group of isomers that are produced in the rumen as stable intermediates in the microbial biohydrogenation of linoleic acid, and also in the mammary gland by the desaturation of the precursor, trans-11-18:1 (vaccenic acid; Parodi, 2002). It is of interest to ascertain whether the milk of cows consuming the coproduct might have an enhanced CLA content. If so, this finding could have implications of health benefits for human consumers. CLA has shown evidence of anti-carcinogenic potency and other health-promoting properties (Khanal, 2004). The present experiment seeks to contribute to knowledge of dietary effects on the concentration of CLA and other fatty acids in bovine milk fat under conditions of Puerto Rico.

Ten of the highest producing cows of the Lajas Substation herd (9 Holstein and one crossbred) were used in an experiment that lasted for 52 days during June and July. The first seven days were considered a preliminary period in which the animals got accustomed to the experimental routine. Thereafter, period 1 was of 16-day duration; and period 2, 29 days. The animals were maintained as one group in total confinement, in an unpaved corral with tree shade and watering trough available. They left the corral only at the two daily milkings. The diet consisted of tropical grass hay in long form and a bulky concentrate (BC), which contained ground hay and had the formulas in periods 1 and 2 shown in Table 1. The period 1 formula also applied to the preliminary period. In period 2 the BC incorporated 15% of SolubMax as a substitute for 8% maize, 5.3% wheat middlings, and 1.7% soybean meal. Samples of long hay and BC were taken periodically and oven dried to determine DM content.

Each cow received 9 kg daily of BC while stanchioned in the barn before or after the two daily milkings, in the amounts of 5 kg in the morning and 4 kg in the afternoon. Any

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TABLE 1. *Percentage formulas of the bulky concentrate fed in each period.*

Ingredient	Period 1	Period 2
	%	%
Ground maize grain	35.00	27.00
Wheat middlings, partially pelleted	23.30	18.00
Ground grass hay	20.00	20.00
Soybean meal	17.70	16.00
SolubMax™	—	15.00
Ground limestone	2.00	2.00
Urea	1.00	1.00
Salt	0.50	0.50
Commercial phosphorous supplement	0.25	0.25
Commercial vitamin-mineral supplement	0.25	0.25

feed left uneaten was recovered and reweighed. Additional BC, in daily amounts ranging from 60 to 65 kg in period 1 and 65 to 75 kg in period 2, together with 35 kg of long hay, was group-fed in a feeder located in the corral. Long hay was placed in the feed bunk first and BC spread on top of it. The aim of this procedure was to mitigate the problem of cows pushing or pulling bunches of long hay out of the feeder while seeking the concentrate. The BC-hay combination was available to the animals from the time they returned to the corral after the afternoon milking until the following day. The total feed offering permitted somewhat less than full ad libitum intake, as usually either none or only a small amount of uneaten feed was recovered; maximum recoveries were 0.25 and 1.95 kg/day in periods 1 and 2, respectively. Orts were presumed to be all long hay, except on one occasion in period 2 when 0.8 kg of BC was recovered.

Individual milk samples were taken at four consecutive milkings on days 9, 10 and 11 of period 1; and days 12, 13 and 14 and days 20, 21, and 22 of period 2. Milk from the four milkings was proportionally composited and frozen. On three occasions a single frozen sample from each cow was sent by parcel express to Abbott Laboratories in North Chicago, Ill. Later the 30 frozen samples were transferred to the laboratory of Dr. James A. Drackley at the University of Illinois, Urbana, where they were analyzed for fatty acid composition.

Liveweight (LW) of individual cows was estimated from thoracic perimeter by using a standard tape measure at the end of periods 1 and 2. Milk fatty acid data were subjected to paired "t" tests to compare the results obtained in period 1, when SolubMax™ was not included in the diet, vs. period 2 (average of two samplings), when SolubMax™ was fed.

Table 2 summarizes animal performance results. These data could not be analyzed statistically because of the lack of replication, since all the animals ate together as a single group. Daily intake of DM from BC and total DM intake were about half a kilogram more in period 2 than in period 1, whereas intake of DM from long hay remained constant. Daily intake of DM from SolubMax™ in period 2 was 1.22 kg, which represents 7.1% of total DM. Long forage supplied 18.3% and 17.8% of total DM intake in the two successive periods. Daily milk production was slightly above 20 kg and showed a minimal decrease between periods 1 and 2. The ratio of milk/DM intake indicates slightly better feed conversion efficiency in period 1. Mean liveweight was close to 550 kg and increased slightly in period 2. Daily DM consumption expressed as a percentage of LW was exactly 3.0 in period 1 and 3.08 in period 2. In general, the inclusion of SolubMax™ or not in the

TABLE 2. *Daily feed intake, milk production, liveweight and feed conversion efficiency.*

Criterion	Period 1	Period 2
Bulky concentrate DM intake (kg)	13.54	14.07
Long hay DM intake (kg)	3.04	3.05
Total DM intake (kg)	16.58	17.12
SolubMax™ DM intake (kg)	—	1.22
Proportion SolubMax™ in DM intake (%)	—	7.10
Proportion long hay in DM intake (%)	18.30	17.80
Milk production (kg)	20.70	20.50
Ratio milk production/DM intake	1.25	1.20
Liveweight (kg)	549	556
DM intake proportional to liveweight (%)	3.00	3.08

diet had little effect on these criteria of productive performance. This finding is in agreement with previous observations in this herd on the capacity of SolubMax™ DM to substitute for DM supplied by solid concentrate ingredients in rations for lactating cows (Randel and Moyá, 2003).

Milk fat composition results from both periods combined showed that the four acids, palmitic (16:0), oleic (18:1), stearic (18:0), and myristic (14:0), constituted 70.71% of the total fatty acids. The numerous other fatty acids present each contributed much less, but the addition, in descending order, of the acids 4:0, 12:0, 10:0, 18:2, 6:0, 16:1, and 8:0, brought the accumulated total to 88.15% of the fatty acids. These findings agree with the information presented by Taylor and MacGibbon (2002), that the main straight-chain saturated fatty acids with from 4 to 18 C account for 62 to 70% of the total fatty acids in bovine milk fat and that oleic acid constitutes about 20%. The latter compound owes its abundance to the fact that bovine mammary tissue has the capacity to desaturate 18:0 to 18:1.

In Table 3 the 16 most abundant fatty acids, of the total of 33 that were identified in the chemical analysis, are designated by systematic nomenclature, along with their concentration in the milk fat in periods 1 and 2, and the statistical significance of the difference between periods. Three of the four most abundant fatty acids (16:0, 18:1 and 18:0) also showed the largest absolute change between periods, whereas the fourth (14:0) changed much less. The principal effects of period 2 conditions, which included SolubMax™ in the diet, relative to those of period 1, without SolubMax™ feeding, were as follows: (1) Saturated fatty acid increases in 18:0 ( $P < 0.05$ ) and 17:0 ( $P < 0.01$ ); decreases ( $P < 0.01$ ) in 16:0, 6:0 and 4:0, and ( $P < 0.05$ ) in 8:0; non significant ( $P > 0.05$ ) decreases in 14:0, 12:0, 10:0, and 15:0. (2) Mono-unsaturated fatty acid increase in 18:1 ( $P < 0.05$ ) and 16:1 ( $P < 0.01$ ); no change in 14:1. (3) Di-unsaturated fatty acid decrease in 9-cis, 12-cis-18:2 (common isomer of linoleic acid) and increase ( $P < 0.05$ ) in 9-cis, 11-trans-18:2 (CLA isomer known as ruminic acid). (4) tri-unsaturated fatty acid decrease ( $P < 0.01$ ) in 18:3 (linolenic acid). Also, milk fat from period 2 gave higher ( $P < 0.01$ ) proportions of unidentified fatty acids, including both those with fewer than 17 C (4.23 vs. 3.37%) and those with more than 17 C (3.05 vs. 2.59%).

Milk fat content of CLA was of particular interest in this study. Concentration of the CLA isomer, 9-cis, 11-trans-18:2 increased from 0.70 to 0.77% between periods 1 and 2, equivalent to a relative increase of 11%. Another isomer of CLA, 10-trans, 12-cis-18:2 was found to be present at a concentration of only 0.01% in the milk fat of 28 of the 30 samples analyzed, whereas in the other two samples it was not detected. Perfield et al. (2004) also found milk fat concentrations of less than 0.01% of this fatty acid in Holstein cows fed a

TABLE 3. *Percentage fatty acid composition in milk fat.*

Fatty acid		Period 1 %	Period 2 <sup>1</sup> %	Difference between periods (2-1)	P difference
16:0 <sup>2</sup>	Hexadecanoic	28.55	25.80	-2.75	0.01
18:1-cis-9	Octadecenoic	21.42	22.58	1.16	0.05
18:0	Octadecanoic	10.76	11.40	0.64	0.05
14:0	Tetradecanoic	10.66	10.54	-0.12	NS
4:0	Butanoic	4.38	4.00	-0.38	0.01
12:0	Dodecanoic	3.18	3.11	-0.07	NS
10:0	Dedecanoic	2.66	2.53	-0.13	NS
18:2-cis-9, 12	Octadecadienoic	2.54	2.33	-0.21	0.05
6:0	Hexanoic	2.25	2.07	-0.18	0.01
16:1-cis-9	Hexadecenoic	1.51	2.10	0.59	0.01
8:0	Octanoic	1.28	1.13	-0.15	0.05
14:1-cis-9	Tetradecenoic	0.99	0.99	0.00	NS
15:0	Pentadecanoic	1.01	0.94	-0.07	NS
17:0	Septdecenoic	0.70	0.83	0.13	0.01
18:2-cis-9, trans-11	Octadecadienoic <sup>3</sup>	0.70	0.77	0.07	0.05
18:3-cis-9, 12, 15	Octadecatrienoic	0.22	0.19	-0.03	0.01

<sup>1</sup>Average of two samples.

<sup>2</sup>Number of carbon atoms: number of double bonds.

<sup>3</sup>Conjugated linoleic acid (CLA).

control diet of alfalfa hay and concentrates, whereas when two rumen-protected supplements, containing tran-10, cis-12 and cis-9, trans-11 CLA (protected by formation of fatty acyl amide bonds or by lipid encapsulation) were fed separately, the concentrations of trans-10, cis-12 CLA were 0.08 and 0.09%, respectively, whereas those of cis-9, trans-11 CLA increased from 0.57 (control) to 0.83 and 0.80% with the three respective treatments. The milk produced in this experiment contained CLA almost exclusively as rumenic acid. Percentage composition of this isomer in the milk fat was  $0.697 \pm 0.028$  (SE) in period 1; it increased ( $P < 0.01$ ) to  $0.808 \pm 0.053$  at first sampling in period 2, but then decreased to  $0.739 \pm 0.051$  at second sampling in period 2 (not significantly different  $P > 0.05$ ) from the mean of period 1. The combined mean  $\pm$  SD of both samplings in period 2 was  $0.774 \pm 0.164$ , which corresponds to a coefficient of variation (CV) of 21%. Variability among individual cows was less in period 1 (CV = 13%). The range of individual values in period 1 and at the two consecutive samplings in period 2 was 0.58 to 0.83, 0.61 to 1.22, and 0.55 to 1.09, respectively.

It might be questioned whether the 11% relative increase in concentration of the principal isomer of CLA, obtained with a diet containing 7.1% of SolubMax on the dry basis, would be of practical importance to the health of human consumers. The range in CLA concentration usually found in bulk milk supplies is from 2 to 37 mg/g of milk fat or 0.2 to 3.7% (Parodi, 2002). The values observed in this experiment were closer to the lower end than to the higher end of this range. Although dietary effects are the most important cause of variability in milk CLA concentration, results obtained elsewhere suggest that a greater increase in milk fat CLA content could be expected from the inclusion of green forage in the diet than that which was seen in the present study with addition of a high-lipid supplement to a diet containing only dry forage.

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