

March 26, 2001

To: Chairman and Committee Members
Varietal Release Committee
California Crop Improvement Association
University of California, Davis

From
Applicants:

Steven R. Temple
Extension Agronomist
Dept. of Agronomy
and Range Science
Univ. of California
Davis, CA. 95616

Philip A. Roberts
Professor of Nematology
Dept. of Nematology
Univ. of California
Riverside, CA. 92521

Donald M. Helms
Staff Research Associate
Dept. of Agronomy
and Range Science
Univ. of California
Davis, CA. 95616

William C. Matthews
Staff Research Associate
Dept. of Nematology
Univ. of California
Riverside, CA. 92521

RE: We are submitting a proposal, for committee consideration, for the release of a nematode resistant baby lima variety. We believe this variety would benefit many growers throughout most of the baby lima growing areas of California.

Variety Candidate V8:

We have developed baby lima plant resistance to two species of nematodes (*Meloidogyne incognita* race 1 and race 3, and *Meloidogyne javanica*) and combined them onto the same genetic background. The varietal release of line V-8 will be useful to growers who need a resistant baby lima variety because of soil root-knot nematode infestation. Resistant varieties will avoid yield loss from root galling, limit nematode egg production on roots, and diminish soil nematode population build up, which has become a problem for many Central Valley crop rotations.

planted in a known and previously mapped nematode-limiting field near Mendota (Fresno County, Table 4).

Table 2: Meridian Replicated Strip Test

Variety	Cwt./Acre	Statistical Significance
Mezcla	25.28	a
Wilbur	25.14	a
V-8	17.66	b
LSD @ 5% = 5.25	CV % = 13.37	--

Weights are means of two row replicated yield plots.

Results in Table 2 show why V-8 will not be recommended for Northern Sutter County, nor other sub-irrigated production areas, unless nematode infestation is severe and growers insist on planting limas. Those areas have soil and environmental conditions very different than the Northern San Joaquin Valley production region. The Meridian grower reported that, following a late-season attack of spider mites, V-8 matured nearly a week earlier than Mezcla and Wilbur, the long-established and dominant varieties produced there. Some V-8 yield loss appeared to be a result of relatively late cutting and threshing.

Table 3: Tracy Strip Test

Variety	Lbs./Plot	Lbs./Acre
Mezcla	220	2904
V-8	240	3168

Weights are from large, non-replicated, measured plots, and harvested by a commercial harvester. This test was conducted entirely as part of a commercial operation from planting to harvest. While this data has statistical limits, it is important to evaluate new variety candidates outside the UC field station environment.

Table 4: Mendota Replicated Strip Test

Variety	Mean Cwt./Acre	Mean Grams/100 Seeds
Mezcla	22.41 b	32.5
V-8	40.28 a	38.5
LSD (p=0.05) = 5.09	CV% = 7.12	---

Weights are means of replicated two row yield plots. Each plot was confirmed for nematode activity (galling of Mezcla). Confirmation of resistance in a commercial field is very difficult because of crop rotations, and field infestation patterns are inconsistent and unpredictable. After observing and mapping this field for several years we were able to utilize this site when our trial coincided with a baby lima planting. The results from this trial are representative of the stress Mezcla shows under nematode pressure, in both yield reduction and a smaller seed size.

**APPLICATION FOR VARIETY CERTIFICATION
FOR THE BABY LIMA VARIETY
CARIBLANCO N**

B. Origin and Breeding History of the Variety

1. Genealogy. 'Cariblanco N' is the variety name proposed for breeding line V-8 of the University of California Davis (UCD) Dry Bean Breeding Program. Cariblanco N was increased from a single plant selection (9399-6), which is a nematode resistant, indeterminate (vine) baby lima, derived from a cross of UCD Selection-144 x UCD Accession L-136.

2. Male parent, UCD Accession L-136. L-136 carries an introduced source of root-knot nematode resistance for baby limas. L-136 has an indeterminate growth habit producing bright red seed, with a very late maturity. L-136 carries two independently inherited resistance traits. It resists root galling and reproduction by *Meloidogyne incognita* and resists root galling (but not nematode reproduction) by *Meloidogyne javanica*. L-136 has proven to be a superior root-knot nematode resistant parent.

3. Female parent, UCD Selection-144. Selection-144 has a determinate (bush) growth habit, matures in about 110 days and during the growing season is very similar in appearance to commercial bush varieties Henderson bush and UC-Luna. Selection-144 was derived from a simple cross between commercial varieties 'Bridgeton' and 'Mezcla'. In the F₂ and F₃ generations, single plants were selected for white seed coat, white cotyledon, determinate bush types, and maturity. Selection-144 was one of several F₄ families bulked from a total of 215 F₃ progeny rows. In an F₅ observation/seed increase nursery, Selection-144 and 20 other white seeded bush lines were selected for yield testing. Selection-144 has small flat white seeds similar in appearance to those of commercial bush varieties Henderson Bush and UC-Luna. Selection-144 was tested in yield trials at UC Davis, West Side Field Station, Chico, and Stockton. Selection-144 grows well and produces good quality seeds, with yields ranging from 31 cwt./acre to 39 cwt./acre. Selection-144 emerged as an elite candidate from several years of preliminary and uniform testing. Like all commercial baby limas, Selection-144, is susceptible to all prevalent races of root-knot nematode.

4. Breeding selection and multiplication. In 1989 a breeding program was initiated to incorporate root-knot nematode resistance into both bush and vine baby lima types. Greenhouse crossing and an F₁ generation advance produced 411 F₂ seeds, which were planted in a UCD field in May 1990. Selections were made using genetic markers to confirm hybrids, and for identifying other important agronomic characters. From the F₂ population, 66 white seeded bush and vine F₂ single plants were selected (Table 1). The F₃ seed from these single plants was expected to segregate for nematode resistance. In 1992 F₃ progeny rows (24 F₃ baby lima

families) were planted at the Kearney Agriculture Center (KAC) *M. javanica* and *M. incognita* fields (Table 2). University of California Riverside (UCR) nematologists collected root samples for laboratory evaluation and confirmation of nematode resistance. During 1993 and 1994 we continued bush and vine agronomic selections starting with F3 reserve seed from the most resistant families tested at KAC. Individual seeds were planted in the greenhouse and evaluated for morphological marker traits, seed coat and cotyledon color, and seed quality (shape, size, and eye pattern). F4 seed was harvested from individually selected F3 plants (Table 3). The seed from each F4 plant was then split between a controlled greenhouse pot test at UCR for assessing resistance to both *M. incognita* and *M. javanica* and a seed increase planted in a 1994 UCD summer field nursery. Based on nematode resistance data from UCR nematologists, F5 seed from UCD F4 families was bulk-harvested, or in the case of families that continued to segregate for nematode resistance, individual single plants were harvested to reselect homozygous resistance to *M. incognita* and *M. javanica* (Table 4).

5. Commercial Stability. A vigorous early generation agronomic character selection and nematode screening schedule in the greenhouse and fields, under stringent criteria, allowed only the most resistant and environmentally adapted selections to be advanced. Baby lima F5 vine families that demonstrated a consistent positive reaction to challenges by the two nematode species were advanced to multi-location yield testing in commercial fields during 1995, while simultaneously being tested to confirm nematode resistance through year 2000 (Table 5).

C. 1. Botanical Description of the Variety

Leguminosae, Lima Bean, *Phaseolus lunatus*

C. 2. Objective Description of the Variety

The Cariblanco N plant type has an indeterminate (vine) growth habit and during the growing season is very similar in appearance to the commercial variety Mezcla. Cariblanco N matures in about 110 to 130 days depending on planting date, growing season temperature, soil type, and environmental conditions of the production location. In all test years and locations the maturity was equal to Mezcla except two occasions. In Tracy in 1996 maturity differences were noted based on a comparison with certified Mezcla seed and commercial grower 'Mezcla B' seed (Table 6). A maturity later than 'Mezcla A' is preferred by most growers. In 2000, V-8 matured several days earlier than Mezcla and Wilbur at the Meridian test site.

Cariblanco N has small flat white seeds similar in appearance to those of Mezcla, Pat and Wilbur vine babylimas. In 1996 seed weight averaged 40 to 50 grams/100 seeds, depending on the growing location. However, to compare the seed size range

within Cariblanco N and Mezcla in different growing locations/environments, seeds were sized using round hole sieve screens (Table 7).

D. Evidence Supporting Identity of the Variety

1) Background and basis for development. In the San Joaquin Valley, there are two predominant species of root-knot nematode that attack limas, *Meloidogyne incognita* race 1 and race 3, and *Meloidogyne javanica*. These species have very wide host ranges among crops commonly grown in rotation with limas. Many host crops do not have resistant varieties available. Thus nematode populations build up on susceptible plantings, injuring roots of both the current crop and the next susceptible crop in the rotation. Some specific differences in nematode-plant associations occur that make prediction of injury difficult. For example, the distribution of these species is associated to some extent with the acreage and rotation intervals of tomatoes and cotton. Cotton is a non-host to *M. javanica* and this species will not multiply on cotton. However, *M. incognita* race 3 will attack cotton and multiply on roots. Susceptible tomato varieties will host *M. incognita* and *M. javanica*, allowing root-knot nematode reproduction and soil population build-up. Nematodes reproduce on the roots of susceptible varieties, including lima beans, resulting in high levels of egg production and root-swelling or 'galling.' This leads to diminished root function, root-rotting and yield reduction. Root systems are protected from these reactions in resistant varieties. Thus, root-knot nematode resistance in dry bean production is important and useful in two respects; protection of the current dry bean lima crop, and protection of subsequent crops in the rotation. The high nematode reproduction levels found on roots of commercial large lima varieties, and the lack of resistance in baby lima varieties, indicated a clear need for improving the resistance in all current lima varieties. 'Cariblanco N' would provide growers with a resistant baby lima variety for the first time.

2) Nematode resistance. To date, the only commercial lima varieties available with nematode resistance are 'White Ventura N' (WVN) (a large seeded vine grown in the central valley), 'Maria' (a large seeded bush variety adapted to coastal production), and UC-92 (a large seeded bush type grown in the San Joaquin Valley). The resistance of these varieties is based on low galling and partial suppression of nematode reproduction, which affects only plant health and yield. This resistance is effective against *M. incognita* but not against *M. javanica*. None of the current commercial varieties of baby limas have resistance to either of these root-knot species. The 'Cariblanco N' resistance provides an important improvement in protection against root-knot nematodes in California production areas, because galling to both species is suppressed and reproduction by *M. incognita* is also suppressed by its resistance. The introduced L-76 source of nematode resistance used in WVN and Maria in the early 1960's (and a sister line used as a donor for UC-92) is limited and was discontinued in breeding activities in 1986 in favor of the superior L-136 resistance.

3) Nematode screening and confirmation. Selection in baby lima lines for resistance to both species of root-knot nematode (*M. incognita* and *M. javanica*) was

accomplished by screening in greenhouse pots and in field plots on two sites at the UC Kearney Agricultural Center. Each field site is infested with either of the two species of root-knot nematode. In greenhouse pot screening, enough plants (generally 6 to 10 per family) were tested to confirm that a family was fixed for resistance to both nematode species. Individual resistant plants in segregating families were identified and harvested for progeny testing. Progeny testing was then either again conducted in greenhouse pot tests or in field plots.

In pot tests, 2-3 week old test plants, along with susceptible checks (usually plants of baby lima cultivar 'Mezcla'), were inoculated with nematode eggs (around 50,000 eggs/pot). After about 8 weeks, plants were harvested and roots were washed free of soil for scoring. Because root galling was the primary criterion for determining resistance, a simple +/- system was employed, with + used for roots with significant galling and - for roots relatively free of galls. In all pot tests, the susceptible check plants were always significantly galled, indicating the test was strong/virulent enough that test plants identified as being resistant were most likely truly resistant, and not escapes. Resistance was then confirmed through progeny testing.

When scoring plants in field plots, either the +/- system or a scale (usually 0 to 4) was used. In the scale, 0 was used for clean plants and 4 for the most heavily galled plants. Again, a requirement in all field screens, the susceptible adjacent checks (usually plants of baby lima cultivar 'Henderson'), were uniformly heavily galled, giving us confidence that resistant test plants were truly resistant. Through this sequence of testing and progeny testing in both the greenhouse and field, most lines were fixed for resistance to both species of root-knot nematode by F5. Line V-8 was fixed for resistance to *M. javanica* at a relatively early stage, and through additional selection, was fixed for resistance to *M. incognita* by F7.

4) Nematode resistance in relation to yield. During the past four years, a number of selections were yield tested at nine sites throughout the state: Meridan, Davis, Farmington, Tracy, Denair, Mendota (two locations), Parlier (KAC), and Irvine's SCFS (Table 8). Several lines were evaluated in field plot or strip trials on nematode infested field sites. The 1997 Mendota nematode field strip trial did not give a good nematode reaction, while the South Coast Field Station (SCFS) site gave a reaction that lowered the yield on the susceptible check. The Denair nematode field gave a non-significant reaction mainly because it is not a lima growing environment. In 1998, yield tests were conducted on the KAC *M. incognita* and *M. javanica* fields. In the KAC *M. javanica* field the nematode reaction was significant between each entry (Table 9). The main problem for getting yield data at KAC was the inability to control lygus bug. The 2000 Mendota trial had high nematode pressure and provided an excellent nematode test. Growth of susceptible 'Mezcla' was severely impacted by nematode infection, resulting in a yield reduction of about 50 percent and a smaller, more variable seed size compared to resistant 'Cariblanco N' (tested as V-8). In all 'Cariblanco N' plots, its roots were free of root-galling.

5) Canning quality. Cariblanco N has very satisfactory canning quality (1 or 2 rating) acceptable and equal in quality and color when compared to the commercial varieties Mezcla, Pat, and Wilbur. The canning evaluations were based on the following categories: seed size variation, seed color (gold, blue, green, or gray), seed splitting, and excretion of starch. Lines were rated #1 for excellent (passing all categories), #2 for good (failing only one category), and #3 for reject (failing in two or more categories). Replicated samples were scored in a "blind" (coded) test.

E. Area of Adaptation.

Cariblanco N is well adapted to the climate and soils of baby lima growing areas along the west side of the upper San Joaquin Valley of California. Cariblanco N has been yield tested and performs well in San Joaquin, Stanislaus, and Fresno Counties. It also performs well in the Western Sacramento Valley which includes Yolo and Solano Counties.

This variety is not recommended for the Northern California regions of Sutter, Yuba, Glenn, Butte, and Colusa Counties nor other sub-irrigated areas, which would also include the Sacramento/Stockton Delta. These areas have a different soil and temporal environment than Northern San Joaquin Valley regions, and appear more suited to Mezcla and Wilbur, the most widely grown varieties of vine baby lima beans under those conditions.

There has been no wide spread testing of Cariblanco N in Northern California Counties and sub-irrigated areas. Where nematode infestations are identified in these areas, Cariblanco N should be evaluated if nematode resistance is needed for crop rotation programs.

F. Procedure for Maintaining Seed Stock.

The genetic purity of this new baby lima variety and all lima varieties will be compromised by outcrossing. Outcrossing occurs at high rates among limas when isolation distances are less than one mile, which is about the average distance a honey bee travels from its hive. Outcrossing is becoming increasingly important as new varieties have "special" characters of resistance, or maturity. It is therefore very important that Breeder, Foundation, and Registered seed be grown with adequate isolation.

Foundation Seed will be maintained and distributed by the UCD Foundation Seed and Certification Service, University of California, Davis. Certification will be available under the supervision of the California Crop Improvement Association. Classes/Generations of seed will be Breeder, Foundation, Registered, and Certified.

Approximately 3,000 pounds of Foundation Seed is available for 2001.

Table 1.

Selections from Field Experiment (6)90

F2 Single Plants	Field Row	Pedigree	Seed Gen. (6)90
19	7	L-136 x 144	F3 (wht)
28	9	144 x L-136	F3 (wht)
19	10	144 x L-136	F3 (wht)
66 Field selected F2 white seeded single plants			

Table 2.

**White Bush/Vine Baby Limas Selections
Tested for Resistance at KAC 1992**

Line	Pedigree	Row/Plt	F3 Seed Source (6)90
9399-V	Sel-144 x L-136	9-14	

Resistant to *M. incognita* race 3; - Field test 1992 KAC.

Resistant low galling score *M. javanica*; - Field test 1992 KAC.

F3 seed planted in 1993 Gh increase, 12 pots each. Planted GH 4/30/93

V=vine

Table 3.

**White Bush/Vine Baby Limas Selections
Greenhouse 1993 seed increase UCD**

Selection	# Sgle Plants	Pedigree	F4 seed-Source
9399-V	10	Sel-144 x L-136	Gh 1993

F4 seed to UCR for *M. javanica* and *M. incognita* pot testing of single plants 3/94.
Planted UCD field Increase (30)94 and (70)94 in single plant row/families.

Table 4.

White Vine Baby Limas Selections

Line/ Selection	Plant # & Yield I.D.	F5 Seed Source	Test (B) <i>M. javanica</i>		Test (A) <i>M. incognita</i>	
9391-4	V-1	14(70)94	Good-5R	R	Good-5R	R
9391-5	V-2	15(70)94	Good-5R	R	Good-5R	R
+9391-7	V-3	17(70)94	Good-4R	R	Good-5R	R
+9392-5	V-4	25(70)94	Good-5R	R	Good-5R	MG
+9395-8	V-6	38(70)94	Good-5R	R	Good-5R	R
+9399-3	V-7	43(70)94	Good-5R	R	Good-5R	R
+9399-6	V-8	46(70)94	Good-4R	R	Good-6R	MG
93100-6	V-9	56(70)94	Good-5R	R	Good-4R	MG
93101-9	V-11	69(70)94	Good-5R	R	Good-5R	MG
+Mezcla	V-12	Check	+---KAC-8/29/95---+			

R = Resistant.

MG = Minor galling or segregation of some plants in the test.

The MG *M. incognita* lines were candidates for future pot screening.

Seed size sieves 21/64 round hole and 11/64 x 3/4 slotted bottom screen.

Lines V-5 and V-10 eliminated on KAC data 8/29/95.

+ = Continued in 1996.

Table 5.

V-8 Yearly Breeding/Testing Summary

Yearly Summary:

Source	Event	Advance of seed Gen. to:
(6)90	F2 plant selection	F3
1992	KAC, nema fields (<i>M. i</i> & <i>M. j</i>) 24 lines tested (bush & vine)	none
1993	Gh selection & increase of lines 9388-93103	F4
1994	UCR pot nema test (<i>M. i</i> & <i>M. j</i>)	none
1994	UCD (30) & (70)94 field increase	F5
1995	KAC, nema fields (<i>M. i</i> & <i>M. j</i>)	none
1995	Vine yield test (WSFS & Tracy)	none
1996	Vine yield test (four locations)	none
2/97	Line V-8 sent to UCR for reselection for Rk <i>M. i</i> .	F6
1997	Vine yield test (three locations)	none
1998	Yield test KAC, nema fields (<i>M. i</i> & <i>M. j</i>)	none
1988	UCR nematode reselection increase	F7
1999	Isolated head row field increase	F8
2000	Breeder seed increase Chico	F9

Table 6.

**Maturity Data from 1996 Tracy Yield Test
Percent Yellow and Dry Pods**

Line	Rep I	Rep II	Rep III	Rep IV	Mean
Mezcla	50	50	30	25	38.8
V-8	20	30	30	10	22.5
V-7	20	20	20	15	18.8
Mezcla B	20	15	20	15	17.5

Earliest maturing treatment is Mezcla (certified seed)

Mezcla B is a grower managed seed lot

No maturity differences on 9/6/96 @ 98 days

Maturity notes taken on 9/16/96 @ 108 days

Field cutting date 10/9/96 @ 131 days

Table 7.

**1996 LIMA
Seed Size Distribution**

Location	Line	% Seed Through 20/64	% Seed Above 20/64	% Seed Above 24/64	% Seed Above 26/64	Mean Above 24/64 & 26/64
Tracy	V-8	4	18	22	56	78
	Mezcla	4	44	36	16	52
Davis	V-8	2	49	41	8	49
	Mezcla	1	50	38	11	49
Meridian	V-8	2	35	33	30	63
	Mezcla	3	48	38	11	49
Denair	V-8	4	59	32	5	37
	Mezcla	7	72	17	4	21

Mezcla is the commercial check variety.

Lines ranked using mean of 24/64 and 26/64 column.

Seeds sized using round hole sieve screens.

Standard commercial bottom screen size

would be # 18/64 through # 20/64.

Table 8.

**Baby Lima
1995 to 1998
Multi-Site Yield Data**

Line	1995		1996		1996		1996		1997		1997		KAC-1998		KAC-1998		All Site	
	Tracy	Davis	Meridian	Denair	Tracy	Mendota	SCFS	Farming	M. incog	M. javan	Mean							
V-8	35.9	40.2	22.4	8.7	42.1	50.1	34.8	17.4	12.6	16.0	28.0							
V-7	36.1	37.7	18.4	9.2	43.6	48.3	42.1	15.7	10.6	12.6	27.4							
Mezcla	33.8	40.0	26.6	6.1	47.8	51.9	27.5	14.8	9.6	10.1	26.8							
LSD @ 5%	4.19	ns	ns	ns	2.68	2.16	10.15	ns	ns	0.85								
CV %	8.51	8.06	20.12	37.37	4.23	3.35	27.57	n/a	18.90	8.50								

Weights given in cwt/a

ns = Trial not significant

Weights are means of two row replicated yield plots

Mendota harvest plot size (two 40" beds) 6.7' x 40' x six reps

Table 9.

**Baby Lima
Yield Data**

Line	Nematode Infested Field Sites						Mean
	1996 * Denair	1997 SCFS	* KAC-1998 M. incog	* KAC-1998 M. javan	2000 Mendota	2000 Mendota	
V-8	8.7	34.8	12.6	16.0 a	40.3	a	22.5
V-7	9.2	42.1	10.6	12.6 b	n/a		18.6
Mezcla	6.1	27.5	9.6	10.1 c	22.4	b	15.1
LSD @ 5%	ns	10.15	ns	0.85	5.09		
CV %	37.37	27.57	18.90	8.50	7.12		

* Not a lima growing area, nematode test site only.

Line	Non-Infested Nematode Field Sites						Mean
	1995 Tracy	1996 Davis	1996 Meridian	1996 Tracy	1997 Mendota	1997 Farming	
V-8	35.9	40.2	22.4	42.1	50.1	17.4	34.7
V-7	36.1	37.7	18.4	43.6	48.3	15.7	33.3
Mezcla	33.8	40.0	26.6	47.8	51.9	14.8	35.8
LSD @ 5%	4.19	ns	ns	2.68	2.16	ns	
CV %	8.51	8.06	20.12	4.23	3.35	n/a	

Weights given in cwt/a

ns = Trial not significant

Weights are means of two row replicated yield plots

Mendota 1997 harvest plot size (two 40" beds) 6.7' x 40' x six reps