Science Societies

Protecting Long Beans From Aphids and Nematodes

By Stacy Kish

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Hyun Park Kang, co-author of a recent article in the Journal of Plant Registrations, punching leaf samples for DNA marker genotyping. Photo by Bao-Lam Huynh.

While long beans are a staple of many Asian cultures, agricultural pests limit the legume's commercial viability. New germplasm lines may change the future of this important crop.

Commercial markets are embracing traditionally ethnic vegetable crops, adding diversity to the food system. Long beans (*Vigna unguiculata* subsp. *sesquipedalis*), which originated in Africa and have been refined through domestication in Southeast Asia, have an export value of $80 million. The edible pods are a symbol of luck and longevity, but more importantly, they pack a nutritious punch, offering a new food option, enriched in protein, vitamins, and minerals.

Despite these benefits, long beans have not broken into larger commercial markets because the current varieties require frequent applications of synthetic chemicals to manage aphids and root-knot nematodes, limiting marketability and opening the potential of pest resistance. Concerns with pesticide applications have affected consumer demand, and the acreage of long bean has been in decline.

A recent article in the *Journal of Plant Registrations* (https://doi.org/10.1002/plr2.20361) details the development by researchers at the University of California–Riverside of new long bean germplasm lines that resist aphids and nematodes.

“We are confident that once consumers become familiar with the new resistant varieties, the crop will gain momentum,” says Bao-Lam Huynh, assistant professor in the Department of Nematology at the University of California–Riverside and first author on the paper.

Meet the Long Bean

The long bean descended from the African cowpea. As people migrated from Africa to Asia, they probably carried the seeds on their travels. Over time, growers selected plants that could survive and thrive in the new environment, resulting in the pods that are common today.

Growing vertically several feet along trellises, the long bean produces pairs of large white or purple flowers that self-pollinate to produce green or purple pods that grow up to 3 ft long. The crop comes in three varieties—light green or purple pods mature mid-May to mid-June while the darker green pods are ready later in the summer. The first flush of immature pencil-thin pods are typically harvested about 60 days after seeding.

Long bean is a pretty hardy and tolerant crop. It can thrive in high heat and under drought stress. The plants grow in a variety of soil types from sandy loam to clay. These legumes also harbor nitrogen-fixing bacteria along the nodules in the root system that can pull nitrogen from the atmosphere and fix it, enriching the surrounding soil. While the Asian variety thrives under harsh conditions, it has lost the inherent resistance to many pests.

Chock Full of Nutrients

Unlike their legume cousins, long beans have a lower sugar concentration with a flavor profile similar to asparagus. The pods are denser than snap beans and hold up well to

various cooking processes. Southeast Asian dishes have harmonized the flavor profile of the legume with a variety of spicy, sour, and sweet ingredients.

Long beans offer a nutritious option. According to the USDA–ARS, one serving of long beans (100 g) is only 47 calories and contains 2.8 g of protein. This sample size is enriched in vitamin C (23% of the daily requirement), vitamin A (5% of daily requirement), thiamin (9% of daily requirement), folate (16% of the daily requirement), and riboflavin (9% daily requirement). The pods are also enriched in magnesium, phosphorus, potassium, iron, copper, and calcium. Beyond the nutrition, long beans are rich in fiber that has been associated with a healthy colon and reducing the level of "bad" LDL cholesterol.

A Favorite of Aphids and Nematodes

The long bean that farmers grow today is highly susceptible to aphids. These tiny, soft-bodied insects have a voracious appetite, especially for the pods of the plant. About the size of two poppy seeds, aphids have specially designed mouthparts that make them particularly effective at piercing the flesh of stems, flowers, and leaves. The tiny pests inject toxic saliva and suck sap from the plant. This assault stunts the plant's growth, and as the infestation continues, shoots die and leaves turn yellow and curl, leaving the plant susceptible to microbial infections.

Farm managers typically deal with aphids with applications of insecticides. In fact, Fresno County records the highest incidence of insecticide applications on long bean among specialty crops.

Long beans are also susceptible to root-knot nematodes, tiny worm-like, soil-dwelling pests that lay eggs in the roots of plants, producing a "knotty" appearance. Previous studies have found that nematodes hinder the timing of flowering in plants and reduce

seed pod production, resulting in lower seed yield. Nematodes also induce vascular disorders in plants that reduce root penetration in the soil, robbing the plant of moisture and nutrients. Root-knot nematodes also suppress the Rhizobium root nodulation, which affects the plant's nitrogen-fixing ability. The pests are commonly managed with soil fumigants.

Developing New Lines for Resistance

Knowing the potential of this crop, Huynh led a research team at University of California–Riverside to develop aphid- and nematode-resistant lines of long beans. They did not use genetic engineering in this pursuit. The team used conventional cross breeding but accelerated this approach by focusing on DNA markers, growing up to three generations in one year in the greenhouse.

"If we did [this work] in the field, it would take longer, up to four or five years," Huynh points out. "This approach saved time and saved labor to get new varieties into market as soon as possible."



After DNA marker genotyping, the research team retained desirable plants. Photo by Bao-Lam Huynh.

p>Huynh's team used marker‐assisted backcrossing (MABC) to expedite the breeding process. They drew from pure germplasm lines of native cowpea that possess known quantitative trait loci (QTL) that harbor resistance genes against aphids and/or root‐knot nematodes. In particular, the team crossed 'California Blackeye 77,' which carries two aphid‐resistance QTL from African cowpea, with three market‐preferred vine‐type germplasms. The team conducted three MABC cycles to create new resistant versions of the local varieties: Dark Green 1994 (Reg no. GP‐320, PI 702995), Light Green 2055 (Reg. no. GP‐321, PI 702996), and Purple 2056 (Reg. no. GP‐322, PI 702997).

“The Southeast Asian farmers of Fresno County deserve recognition as well for their contributions,” says Ruth M. Dahlquist‑Willard, interim director of the University of California Sustainable Agriculture Research & Education Program and co‑author on the study.

“The new varieties were a cross of the resistant cowpea lines and long bean seeds that local farmers contributed, that they had been selecting for several years on their own farms for the characteristics they preferred.”

Huynh and his team also crossed African landrace FN2‑9‑04, which carries broad resistance for root‑knot nematodes, with one bush‑type germplasm. They followed with two MABC cycles to develop a resistant bush‑type line: Bush 2074 (Reg. no. GP‑319, PI 702994).

A Field Study Proves Lines Effective

In 2022, the team compared three vine‑type lines resistant to their non‑resistant parent plants during a field trial planted at the University of California Kearney Agricultural Research Center, a site known for high aphid concentration. To ensure uniform aphid infestation during the study, the team planted Big Buff, a cowpea cultivar susceptible to aphids, in alternating rows. The trial consisted of two adjacent

fields. The protected trial received regular applications of insecticide. The unprotected trial did not receive insecticide. The researchers scored the severity of aphid infestation based on crop damage on a scale from 0 (no aphids) to 10 (plant death). In addition, fresh pods were harvested twice a week once the crop reached commercial maturity to obtain pod total fresh weight.

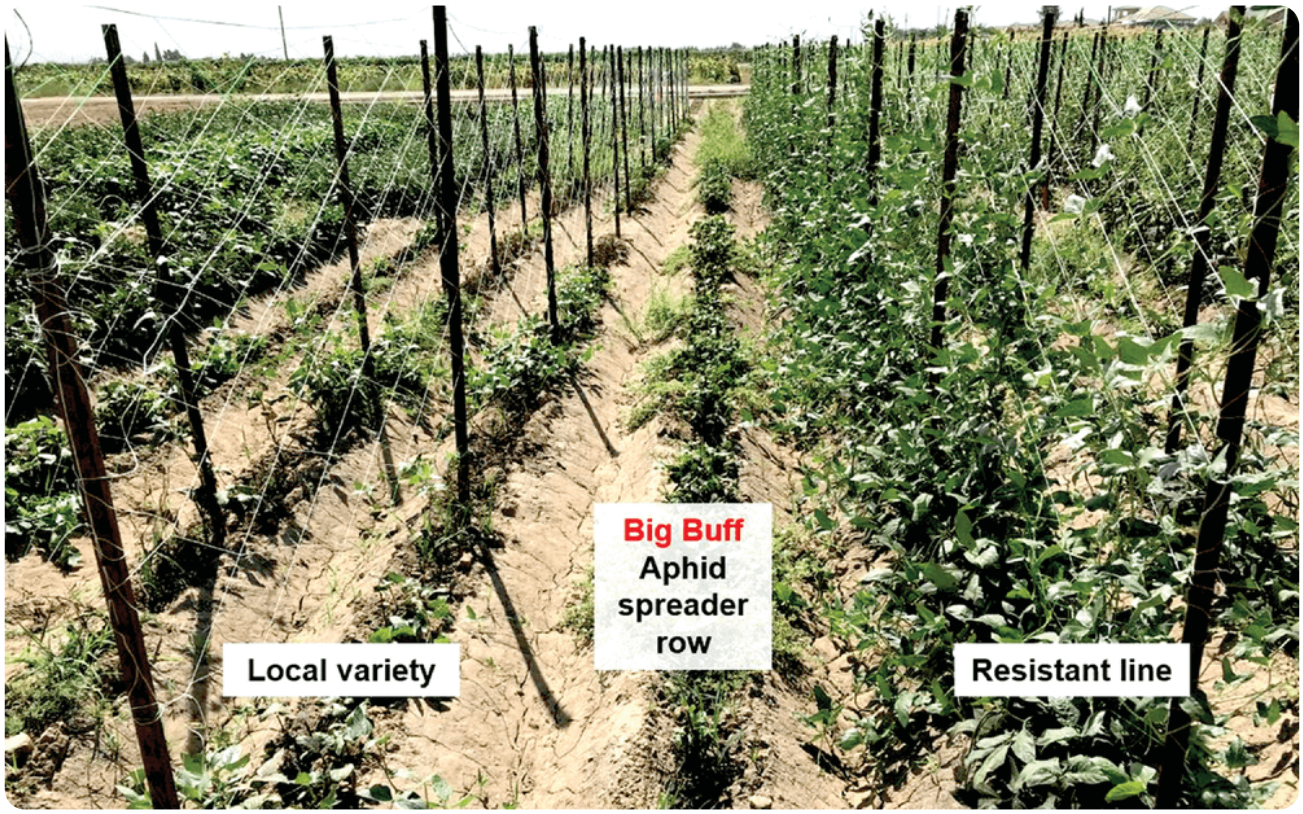
The following growing season, the team compared six bush-type lines resistant to their parent plant at the Coachella Valley Agricultural Research Station. The trial consisted of two adjacent fields similar to the aphid study. One plot was infested with nematode eggs (*Meloidogyne incognita* isolate 77) extracted from greenhouse tomatoes. As the trial progressed, the team dug up five root systems per plot and scored the severity of root-galling symptoms from 0 (no symptoms) to 9 (severe galling). They also gathered fresh pods when the plants reached commercial maturity to obtain pod total fresh weight.

The team found that the new plant lines scored better in both trials. For the aphid study, the three new vines experienced less damage from aphids (score 3–6) compared with the parents (score 7–9). The researchers also found that all of the vine-type aphid-resistant long beans (score of 2.3–3) outperformed their parents in the unprotected plots.

The nematode-resistant lines also experienced no root galling (score 0) compared with the susceptible check, which experienced severe galling (score 9). The nematode-resistant lines also showed near negligible amounts of egg masses (*Meloidogyne incognita* isolate 77 and *Meloidogyne javanica* isolate 811) based on seedling growth-pouch assays in a growth chamber.

“These nematode-resistant lines block the *M. javanica* nematodes from entering the root,” Huynh says. “If [the nematodes] do get into root, they cannot develop normally

and starve."

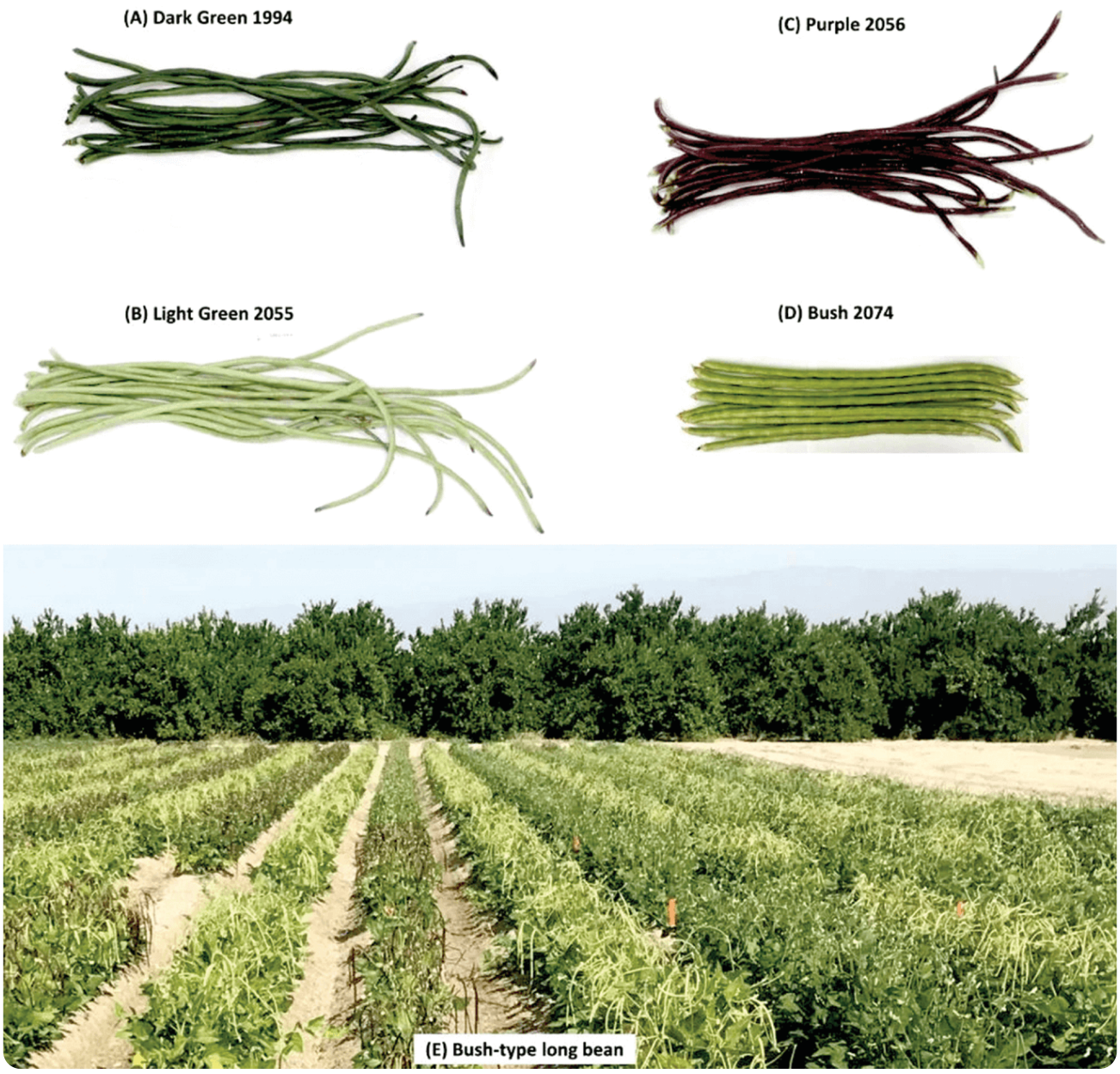


On Day 58, the team evaluates local varieties and resistant lines to aphids. In the middle is Big Buff, a cowpea cultivar susceptible to aphids. Photo by Bao-Lam Huynh.

The new germplasm lines also produced higher fresh pod yields than the parent lines in the unprotected plot with the highest yield differences obtained from the purple and dark green aphid resistant vine lines. Huynh's collaborators at the University of California Cooperative Extension in Fresno County conducted a taste test among the Hmong community to see if the pods from the new lines were approved.

"[Long bean] is a culturally important crop in broader southeast Asian cuisine," Dahlquist Willard says. "Farmers growing long bean are an essential part of the research process, and their feedback is essential in evaluating new varieties for characteristics such as taste, color, and texture to ensure that the new varieties will work for both farmers and consumers."

The field test showed that the pods had a similar flavor to pods from traditional plants, and some lines were rated better due to the higher sugar content. In addition, the pods from some of the new lines were more tender.



Pod samples of aphid-resistant vine-type long bean lines (A) Dark Green 1994, (B) Light Green 2055, (C) Purple 2056, and (D) nema- tode-resistant bush-type Bush 2074. The field trial (E) was conduct- ed in Thermal, CA in 2022. The images shows the determinate, erect growth habit of bush-type long bean plants. Photo by Lilian Thaox- aochay and Bao-Lam Huynh.

The Future Looks Bright for Long Bean Research

The new vine-type lines have the potential to replace local lines in commercial fields, which should greatly reduce the amount of synthetic chemicals needed to maintain a healthy crop. In addition, the routine rotation of the new bush-type line in cropping systems enables mechanical harvesting and could help suppress root-knot nematode populations to benefit future crops.

“The new lines could make it easier and more profitable to grow long beans for specialty markets,” Dahlquist-Willard says. “Host plant resistance is an important tool for integrated pest management, and adding resistance for multiple pests would be even more beneficial.”

At this time, researchers at University of California–Riverside have made breeder seed of the three vine-type long bean lines and the bush-type long bean line available to California growers. The trial packets provide enough seed for a farmer to grow half a row for evaluation. The plants self-pollinate, so the inbred line should remain genetically stable over generations unless impurity becomes introduced by outcrossing, such as by bees and/or seed handling contamination.

The seeds will be shared with the University of California–Davis Foundation Seed Program to produce foundation seed. Breeder seed of the four lines has also been deposited into USDA–ARS National Laboratory for Genetic Resources.

Despite the initial success, Huynh acknowledges that there is more work to be done. He would like to develop lines that include both aphid and nematode resistance by imparting nematode resistance in the new aphid-resistant varieties and vice-versa. He would also like to move upstream in the plant’s genomics to identify functional genes

that impart resistance to these common pests using the long bean/cowpea near isogenic lines and molecular techniques. Huynh believes this information could be incredibly beneficial not only for the long bean and cowpea, but also for a variety of valuable crop plants, such as soybean, corn, and cotton; however, turning on or inserting new genes could move outside the realm of conventional breeding and require genetic transformations in plants that would need additional scrutiny and evaluation.

Dig Deeper

Huynh, B.-L., Dahlquist-Willard, R.M., Ploeg, A.T., Yang, M., Thaoxaochay, L., … & Roberts, P.A. (2024). Registration of four pest-resistant long bean germplasm lines. *Journal of Plant Registrations*, 18(2), 415-425. https://doi.org/10.1002/plr2.20361

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