Creating sustainable markets with solid loofah genetics:
WorldVeg's leaf curl virus and downy mildew disease-
resistant lines of different market segments essential
to develop breakthrough hybrids

Research proposal
for
APSA-WorldVeg Vegetable Breeding Consortium

By
World Vegetable Center
Proposal Summary

<table>
<thead>
<tr>
<th>Project title</th>
<th>Creating sustainable markets with solid loofah genetics: WorldVeg’s leaf curl virus and downy mildew disease-resistant lines of different market segments essential to develop breakthrough hybrids</th>
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<tbody>
<tr>
<td>Submitted to</td>
<td>APSA-WorldVeg Vegetable Breeding Consortium Members</td>
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<tr>
<td>Main WorldVeg contact person</td>
<td>Mandy Lin (<a href="mailto:mandy.lin@worldveg.org">mandy.lin@worldveg.org</a>)</td>
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<tr>
<td>Main WorldVeg scientists and designation</td>
<td>Dr. Narinder Dhillon, Principal Plant Breeder – Cucurbits &amp; Okra</td>
</tr>
<tr>
<td>Project duration</td>
<td>2 years (1 March 2024 - 28 February 2026)</td>
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<tr>
<td>Project fees (US$)*</td>
<td>305,000 Minimum of 20 companies at 15,250 USD per company</td>
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*The participating seed companies will equally share Project fees.

Background

Ridge gourd (*Luffa acutangula*) and sponge gourd (*L. cylindrica; syn. L. aegyptica*) are the two cultivated species of the genus *Luffa*. Loofah fruit is a rich source of calcium (20 mg/100 g fresh wt.), magnesium (14 mg/100 g fresh wt.), potassium (139 mg/100 g fresh wt.), and vitamin A (410 IU). A 200 g serving covers 5 to 16% of the daily recommended intake of the above nutrients. Immature fruit is eaten as a vegetable, boiled, peeled, fried, and used in curries. These nutritious gourds are important sources of livelihood for resource-poor farmers in Asia and can be grown in various agro-climates. These cucurbits are critical components of home and community gardens in the tropics and rank high in the cucurbit portfolio of seed companies.

In India alone, the total loofah seed market is 490 MT (ridge gourd: F1 hybrid seed = 80 tons, OP = 80 tons; sponge gourd: F1 hybrid seed = 200 tons, OP = 130 tons) (Dhillon et al., 2020). The seed market of ridge and sponge gourds in Bangladesh is 16 and 22 MT, respectively. There is a fast conversion from OP to F1 seed at farmers’ fields. Most loofah breeding research is concentrated in India. Commercial cultivars released by seed companies are susceptible to begomovirus ‘Tomato leaf curl New Delhi virus (ToLCNDV)’, and controlling this disease is a major challenge for loofah growers (Dhillon et al., 2020). Downy mildew (DM) caused by

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*Pseudoperonospora cubensis* is another production constraint in the tropics. (Dhillon et al., 2020). Land-race-derived cultivars are rapidly being replaced by modern F1 hybrid cultivars, which has narrowed the genetic basis of the crop. Breeders recycle the loofah genetic material via repeated use of elite F1 hybrids to derive inbred lines for developing new hybrids (Wehner et al., 2019). Loofah genetic resources resistant to ToLCNDV and DM are hardly available.

Loofah germplasm of both the *Luffa* species used by seed companies is extremely limited. Heterogeneous and heterozygous landraces of loofah originating from different Asian countries and conserved in the WorldVeg genebank were used to develop loofah lines of ridge gourd and sponge gourd resistant to ToLCNDV and DM (using field screening, based on visual symptoms) with improved fruit quality traits (non-bitter taste with color and size compatible with market segments such as short, medium and long segments of ridge gourd and green and light green segments of sponge gourd), through pedigree selection method. WorldVeg’s global cucurbit breeding team has developed more than 200 loofah lines (100 each of ridge gourd and sponge gourd) of diversified origins with inbuilt ToLCNDV and DM resistance and improved fruit traits. These lines are an attractive genetic source for seed companies. This elite material will be showcased during the Loofah Open Field Day on 14 December 2023 in Thailand.

**Objectives**

The overall objective is to enable members of the APSA-WorldVeg Vegetable Breeding Consortium to access the WorldVeg elite ridge gourd and sponge gourd lines and F1 hybrids for developing market competitive F1 hybrids resulting in sustainable breeding gains and enhanced profitability of loofah production for smallholder farmers. The specific objectives are:

1. To test the performance (agronomic and disease resistance) of advanced WorldVeg loofah breeding lines of both *Luffa* species
2. To test the performance of loofah F1 hybrids developed by using WorldVeg elite lines and new sources of resistance to ToLCNDV and DM.
3. Organize annual Loofah Open Field Days to showcase the specific horticultural traits of WorldVeg’s unique breeding lines and F1 hybrids and make available the seed of entries selected by the project member seed companies.
4. Understand the genetic diversity of WorldVeg loofah lines along with the commercial checks, based on single nucleotide polymorphism (SNP) analysis.
5. Develop SNP markers associated with genetic loci conferring resistance to ToLCNDV in ridge gourd.
Methods/Activities

Activity 1. Conduct trials of WorldVeg loofah lines developed from landraces through pedigree selection and produce breeder seed of these entries

Evaluate 200 elite loofah lines (100 per year each of ridge gourd and sponge gourd) at the WorldVeg East and Southeast Asia Research and Training Station, Kasetsart University, Kamphaeng Saen, Thailand. Ten plants of each line will be transplanted into single 9.6 m² plots. The following horticultural traits will be recorded: (1) days to 50% pistillate flowering after transplanting; (2) days to 50% staminate flowering after transplanting; (3) fruit length; (4) fruit breadth; (5) fruit weight; (6) fruit number/vine; (8) fruit skin color; (9) fruit taste – bitter/non-bitter; and (10) field resistance to ToLCNDV and DM. A fruit picture database of lines will be created.

Activity 2. Evaluate WorldVeg experimental F1 hybrids of loofah

Evaluate 40-50 experimental WorldVeg F1 hybrids of loofah at various stages of testing of Observational Yield Trial (OYT), Preliminary Yield Trial (PYT) each year at the WorldVeg East and Southeast Asia Research and Training Station, Kasetsart University, Kamphaeng Saen, Thailand. Ten plants of each entry will be transplanted into single 9.6 m² plots and there will be a single plot for OYT and two replications for PYT. Horticultural traits as mentioned in activity 1 will be recorded.

Activity 3. Genetic diversity analysis of WorldVeg loofah lines along with the commercial checks

The genetic structure of WorldVeg breeding populations of both ridge gourd and sponge gourd along with commercial checks will be determined using SNP markers for a phylogenetic analysis.

Activity 4. Mapping of genetic loci conferring resistance to ToLCNDV in ridge gourd

QTLs associated with ToLCNDV resistance will be identified and validated in segregating populations. Detailed Information on SNP markers associated with these QTLs and resistance loci will be provided to the member companies.

Activity 5. Organize Loofah Open Field Day

WorldVeg Loofah Open Field Day will be held each year in December at the WorldVeg East and Southeast Asia Research and Training Station, Kasetsart University, Kamphaeng Saen, Thailand. Seed companies contributing to this project will be invited to observe the
performance of loofah lines and F1 hybrids along with commercial checks and select the candidate lines and hybrids for their breeding program.

**Deliverables**

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<th>#</th>
<th>Description</th>
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<tr>
<td>1</td>
<td>200 new elite lines of various market segments (100 each of ridge gourd and sponge gourd), resistant to ToLCNDV and DM. This also includes 4-5 white seeded diversified background sponge gourd lines</td>
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<tr>
<td>2</td>
<td>40-50 WorldVeg’s experimental F1 hybrids in observational Yield Trial (OYT) and Preliminary Yield Trial (PYT) each year for participants’ observation and selection</td>
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<td>3</td>
<td>Information about the genetic diversity of WorldVeg loofah (both ridge gourd and sponge gourd) lines and commercial checks based on SNP marker analysis</td>
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<td>4</td>
<td>SNP markers associated with QTLs conferring resistance to ToLCNDV in ridge gourd provided for marker-assisted selection</td>
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**Duration and budget**

The project will be conducted from 1 March 2024 to 28 February 2026 with the budget of US$ 305,000 which will be equally shared by the participating seed companies.

**Timeline of activities**

<table>
<thead>
<tr>
<th>Activity</th>
<th>Year 1</th>
<th>Year 2</th>
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<tr>
<td>Field trials conducted for activity 1, 2, and 5</td>
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<tr>
<td>Access to SNP based genetic diversity analysis of WorldVeg lines and commercial checks. Mapping of QTLs associated with resistance to ToLCNDV in ridge gourd (activity 3 &amp; 4)</td>
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**References**

