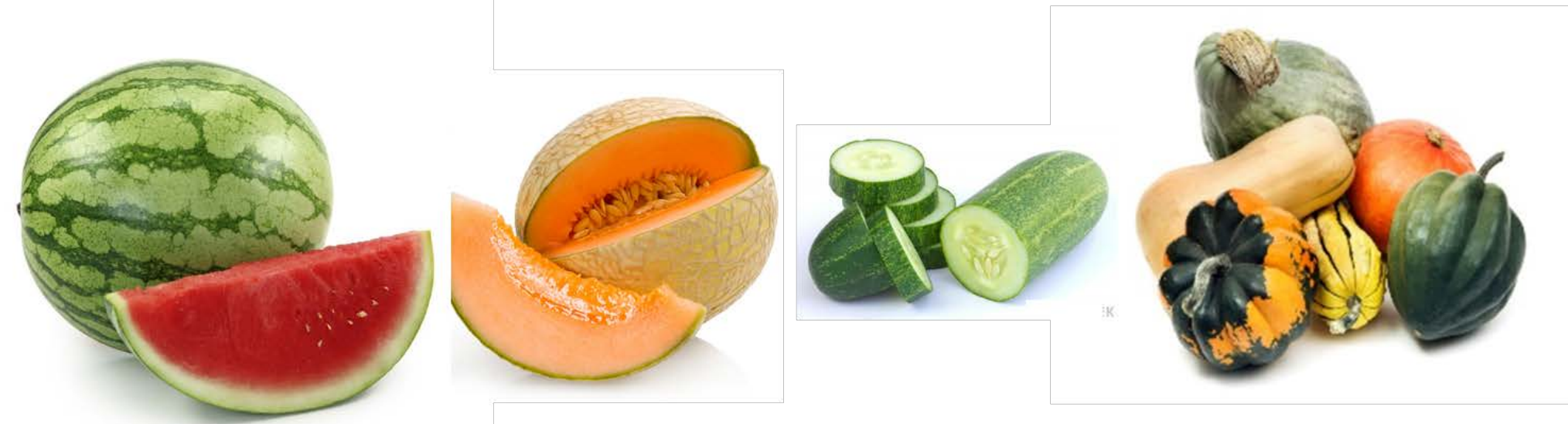


# CucCAP - Developing Genomic Resources for the Cucurbit Community

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## Introduction and Objectives

U.S. watermelon, melon, and cucumber growers, shippers, and processors identify losses due to diseases as their primary production challenges (Table 1). To develop resistant cultivars, breeders must: locate sources of resistance, identify genetic loci conferring resistance, efficiently incorporate resistances into elite cultivars, and combine resistances to multiple diseases, while at the same time, retaining superior productivity and fruit quality. Severity of the diseases and complexity of the breeding challenges requires use of the most effective and efficient technologies.

Recent assemblies of draft genome sequences for cucumber, *Cucumis sativus* L. (1-3), melon (*Cucumis melo* L.) (4), watermelon (*Citrullus lanatus* (Thunb.) Matsum. & Nakai) (5), and pumpkin and squash (*Cucurbita pepo* L., *C. maxima* Duchesne, and *C. moschata* Duchesne) (6,7) make it feasible to use advanced genomic approaches that were not possible even a few years ago. The diploid genomes and small genome sizes of cucurbit species, and close genetic relationships among the species make them highly amenable to genomic approaches.

These needs and opportunities led to establishment of an integrated project for cucurbit genomics "CucCAP: Leveraging applied genomics to improve disease resistance in cucurbit crops". Our goals are:

- I. Develop genomic and bioinformatic breeding tool kits for accelerated crop improvement across the Cucurbitaceae;
- II. Use these tools to facilitate efficient introgression of disease resistance into commercially valuable cucurbit cultivars; and
- III. Provide readily accessible information to facilitate disease control and perform economic impact analyses with respect to cost of production and disease control.

**Table 1. Major disease threats to cucurbit crop production as identified by cucurbit industry stakeholders.**

Disease	Industry priority	Also affects:
Downy mildew	cucumber	melon, watermelon, squash/pumpkin
<i>Fusarium</i> wilt	watermelon	melon, cucumber
Gummy stem blight	watermelon	melon, cucumber, squash/pumpkin
<i>Phytophthora</i> rot	cucumber, watermelon, squash/pumpkin	melon
Powdery mildew	melon, watermelon, squash/pumpkin	cucumber
Viruses (CMV <sup>2</sup> ; CYSDV <sup>3</sup> ; PRSV-W <sup>4</sup> ; CGMMV <sup>5</sup> )	watermelon <sup>4,5</sup>	cucumber <sup>3,5</sup> , squash/pumpkin <sup>2,4</sup>

Although the most critical diseases vary for each crop, there are shared disease problems

## I. Expand the genomic toolkit for cucurbits

### i. Develop communal sequence and phenotype databases and bioinformatics tools for watermelon, melon, cucumber and squash

Genotyping by sequencing (GBS) technology for identification of SNPs, has revolutionized the quantity of data and number of markers that can be deployed for genetic and physical mapping of QTLs. GBS procedures have been optimized for watermelon (8-10), melon (11), and squash (12), and have been successfully applied to identify QTLs in watermelon for fruit quality and resistance to *Fusarium* race 1 (8,11).

We will develop tool kits for GBS data processing and analysis including: a **SNP calling pipeline, genome-wide association study (GWAS) analysis package, and tools for gene mapping and QTL identification.**

We will also develop breeder-friendly **databases and visualization tools for sequences, maps, syntenic relationships, markers, QTLs, population and accession phenotypic information, and their interrelationships.**

### ii. Perform GBS analysis of cucurbit PI collections and develop functional arrays for disease resistance GWAS

The U.S. National Plant Germplasm System maintains:  
1,314 cucumber  
2,043 melon  
1,311 watermelon  
1,580 squash (*Cucurbita pepo*, *C. moschata* and *C. maxima*)

**Diversity in the collection will be genotyped by GBS for 1000-1500 accessions/crop.**

**Functional arrays for resequencing will be developed for each crop using key disease resistance resources (PIs and breeding lines) and representation of diversity as defined by GBS of the PI collections.**

**The associated sequence data, SNP datasets, and genetic maps will be available for future phenotypic and GWAS analysis of any traits of interest.**

### iii. Provide access to cucurbit genomics tools and databases

The current **International Cucurbit Genome Initiative (ICUGI) website** <http://www.icugi.org> database which stores **genome sequences and gene annotations for watermelon** and cucumber will be expanded to include **genome sequences of melon and *Cucurbita* species.**

The tools and data developed for (i) and (ii) above will be included along with transcriptome sequence, annotation information, marker data, and genetic maps for cucurbits.

**We will also develop and deliver genomics and bioinformatics workshops and webinars open to all members of the cucurbit scientific and breeding communities.**

### iv. Establishment of community-standardized gene and trait descriptors and nomenclature

Cucurbit genomic resources will have greater value if information is more readily transferrable across research teams and crop systems. Publications commonly have the same gene/QTL names for different traits, or different names for the same traits. To avoid further confusion and provide an important common language for comparative genetic analysis, **we seek to establish nomenclature guidelines across the cucurbit crops.** We hope that this project can provide a platform to develop guidelines to standardize and simplify description of genetic entities across the cucurbit species.

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## II. Introgress resistances into advanced breeding lines



Breeding strategies for each commodity and disease problem are based on knowledge of the pathogens, sources of resistance, cross compatibility between sources of resistance and cultivated materials, inheritance of resistance, availability of breeding populations, and state of breeding progress with regard to introgression of resistance, identification of QTLs, and marker development.

**Table 2. Current status of resistance breeding for the priority cucurbit diseases.**

Crop and disease	Sources of resistance	Resistant parental line	Phenotypic data for GWAS	Segregating populations	Analysis of inheritance	QTL analysis	Segregating populations	Marker	development into cultivated types	Advanced breeding lines for release	Cultivars for release to farmer
<b>Watermelon</b>											
<i>Fusarium</i> race2 (Fus)	PI 482246-USVL246 <sup>FR2</sup> ; PI 482252-USVL252 <sup>FR2</sup>	X	X	X						X	
<i>Fusarium</i> race 1	Calhoun Gray	X	X	X	X*	X				X	
Gummy stem blight (GSB)	PI 482276-UGA1081; PI 526223-UGA157	X	X	X						X	
<i>Phytophthora</i> (Phyt)	PI 494531-	X	X	X						X	
Powdery mildew (PM)	USVL531MDR; PI 560003-USVL003MDR	X		X	X*					X	
CGMMV	Currently evaluating										
PRSV-W	PI 595203	X	X	X	X						
<b>Melon</b>											
Powdery (PM)	MR-1	X		X	X*						
<i>Fusarium</i> (Fus)	MR-1	X		X	X*					X	X
CYSDV	PI 313970; TGR1551	X	X	X	X						
CMV	PI 161375; Freeman cucumber				X*						
<b>Cucumber</b>											
Downy mildew (DM)	PI 197088; PI 330628	X		X	X	X				X	X
<i>Phytophthora</i> (Phyt)	PI 109483			X	X						
<b>Squash</b>											
<i>Phytophthora</i> (Phyt)	PI 211996; PI 483347; PI 634693	X			X						
Powdery (PM)	<i>C. martenezii</i>	X			X	X	X	X	X	X	X
PRSV-W	Menina, Nigerian Local	X			X				X	X	X
CMV	Menina, Nigerian Local	X			X				X	X	X

Resistances to several of the cucurbit diseases, *Fusarium* wilt race 1 (8), powdery mildew (13) and PRSV in watermelon (14), and *Fusarium* wilt (15), powdery mildew (16), and CMV (17) in melon, and powdery mildew and PRSV in squash (18, 19) are simply inherited indicating potential to identify major QTL.

### GWAS and bi-parental populations will be used to identify resistance-associated quantitative trait loci (QTL) and initiate marker development.

Breeding for resistances will be performed in parallel with genomic analyses and tool development.

The genomic resources for the cucurbit species will be integrated to anchor consensus physical and genetic maps, allowing us to search for regions of synteny and orthologous disease resistance regions or gene islands across the sister species.

## III. Web-based cucurbit disease information and economic analysis

Genomic and breeding efforts will be complemented by a **centralized cucurbit disease website** with content in English and Spanish providing **diagnostic information, disease control recommendations, disease alerts, and links to disease clinics and forecasting tools.**

**"One stop shopping for cucurbit diseases"**

Economic analysis of the costs of production and disease control will be performed using risk simulation models developed from representative farm panels.



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