Biopharmaceutical Production under Controlled Environments: Photosynthetic Rate, Soluble Protein Concentration and Growth of Transgenic Tomato Plants Expressing a *Yersinia pestis* F1-V Antigen Fusion Protein

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CEAC
Controlled Environment Agriculture Center

Plant-Made Pharmaceuticals (PMP)

Transgenic plants that express high-value pharmaceutical protein have great potential for inexpensive and scalable protein production and delivery system.

Our Final Goal

To establish an efficient PMP production system under controlled environments

EuroFresh Farms, Willcox, AZ

- **Fruits of transgenic tomato plants transformed with a gene encoding vaccine protein can be used as 'edible' vaccine.**
- **Greenhouse tomato production has advantages over open-field tomato production in terms of high productivity and high containment.**

Plague

is a deadly infectious disease caused by the bacterium *Yersinia pestis*.
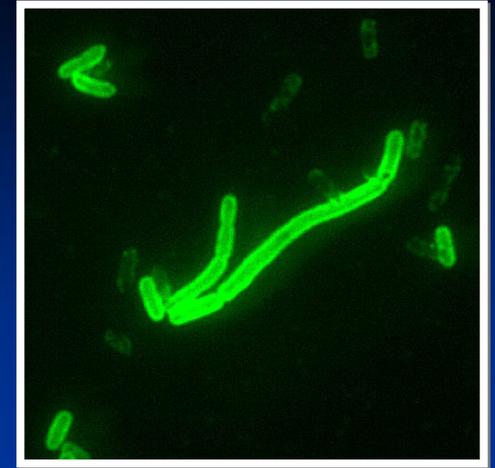
Y. pestis

F1-V protein

is a predominant antigen fusion protein against plague.

f1-v transgenic tomato plants were produced, which can accumulate F1-V protein in fruits (Alvarez et al. 2006).

Objectives

- **To characterize growth and development of the *f1-v* transgenic tomato plants under the environmentally-controlled greenhouse conditions**

- **To evaluate fruit and protein productivity of the *f1-v* transgenic plants by comparing with that of a commercial greenhouse cultivar**

Plant Materials

Tomato (*Solanum lycopersicum* L.)

- Transgenic lines (background: TA234)

  'F1-V'　　　　With *f1-v* gene

  'F1-V/P19'　　With *f1-v* and *p19* genes

  　　　　　　　(Alvarez et al. submitted)

- Non-transgenic cultivars

  'TA234'　　　Wild type

  'Durinta'　　Commercial GH cultivar

Experimental GH (BSL-2)

Floor space: 66.9 m^2

Peak height: 5.2 m

Plant density: 2.4 m^{-2}

196-d-old plants

Rockwool substrate

Transplanting

Germination

Fruit harvesting

8/15   9/20   11/26   2/27

Date

Environmental Conditions inside GH

(from Sep 29, 2007 to Feb 26, 2008)

Mean daytime temp.:   **19-22°C**

Mean nighttime temp.:   **18-19°C**

Daily PPFD integral: **15-25 mol m^{-2} d^{-1}**

Mean relative humidity: **60-90%**

Measurement Items

- Stem length
- Number of leaves
- Light-saturated rate of photosynthesis in leaves
- Weekly Fruit yield
- Total soluble-protein (TSP) concentration in fruits
- F1-V concentration in fruits

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Growth & Photosynthesis

- **Stem Length**

  'Durinta' > 'F1-V' = 'TA234' = 'F1-V/P19'

- **Number of leaves**

  'Durinta' = 'F1-V' = 'TA234' = 'F1-V/P19'

- **Light-saturated rate of photosynthesis**

  'Durinta' ≥ 'F1-V' = 'TA234' = 'F1-V/P19'

'Durinta':	**Commercial GH cv.**
'TA234':	**Wild type**
'F1-V':	***f1-v* transformant**
'F1-V/P19':	***f1-v* & *p19* supertransformant**

Fruit Yield
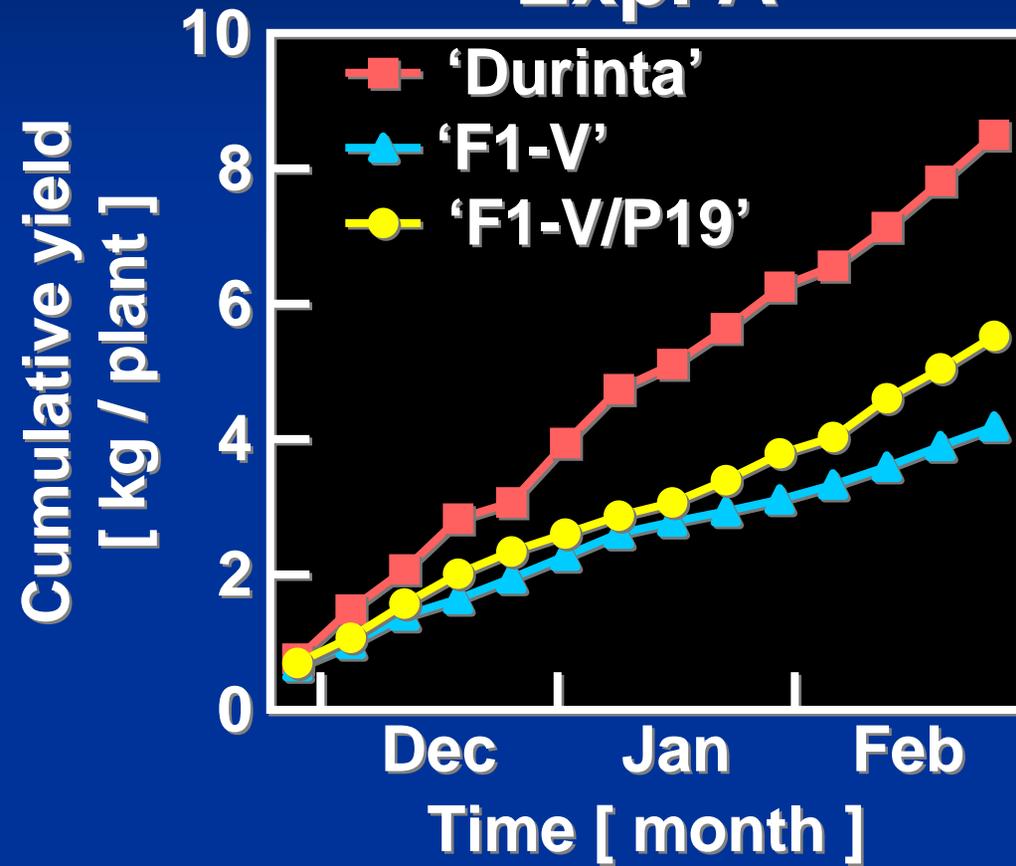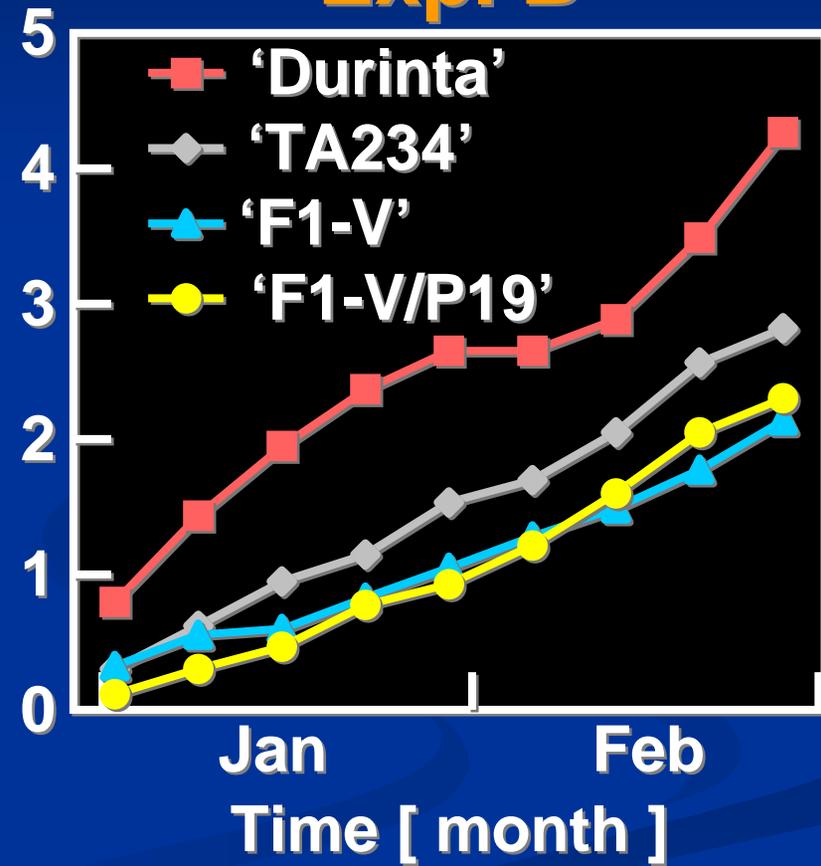
Total Soluble Protein (TSP) in Green Fruits
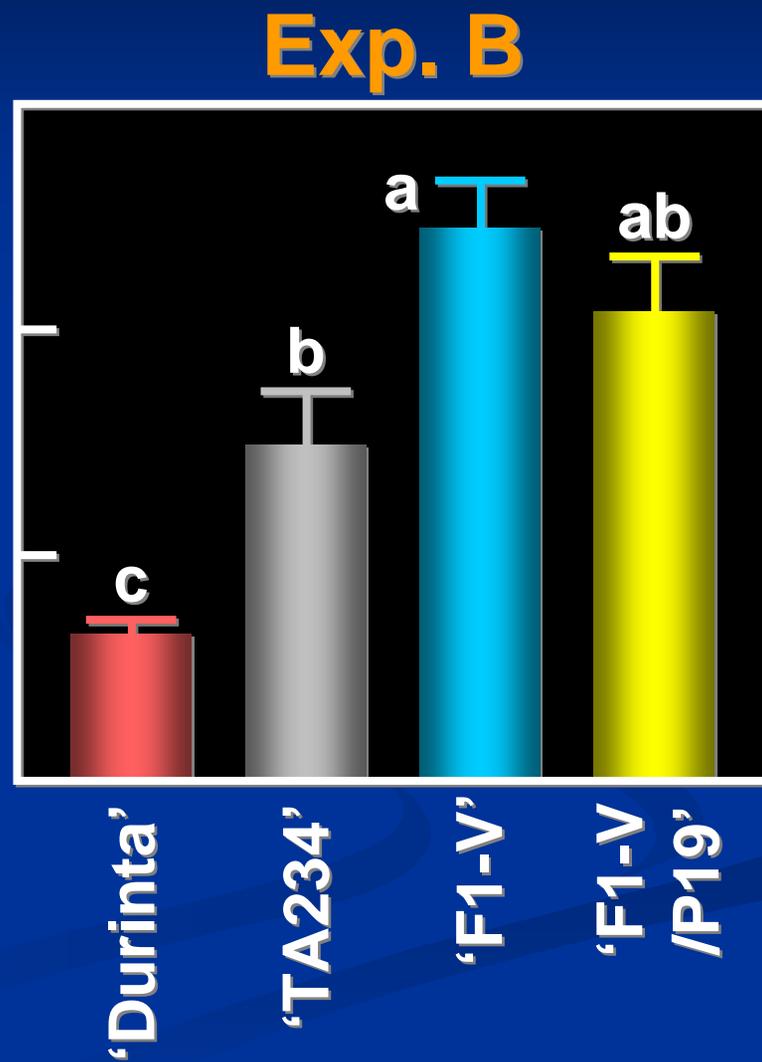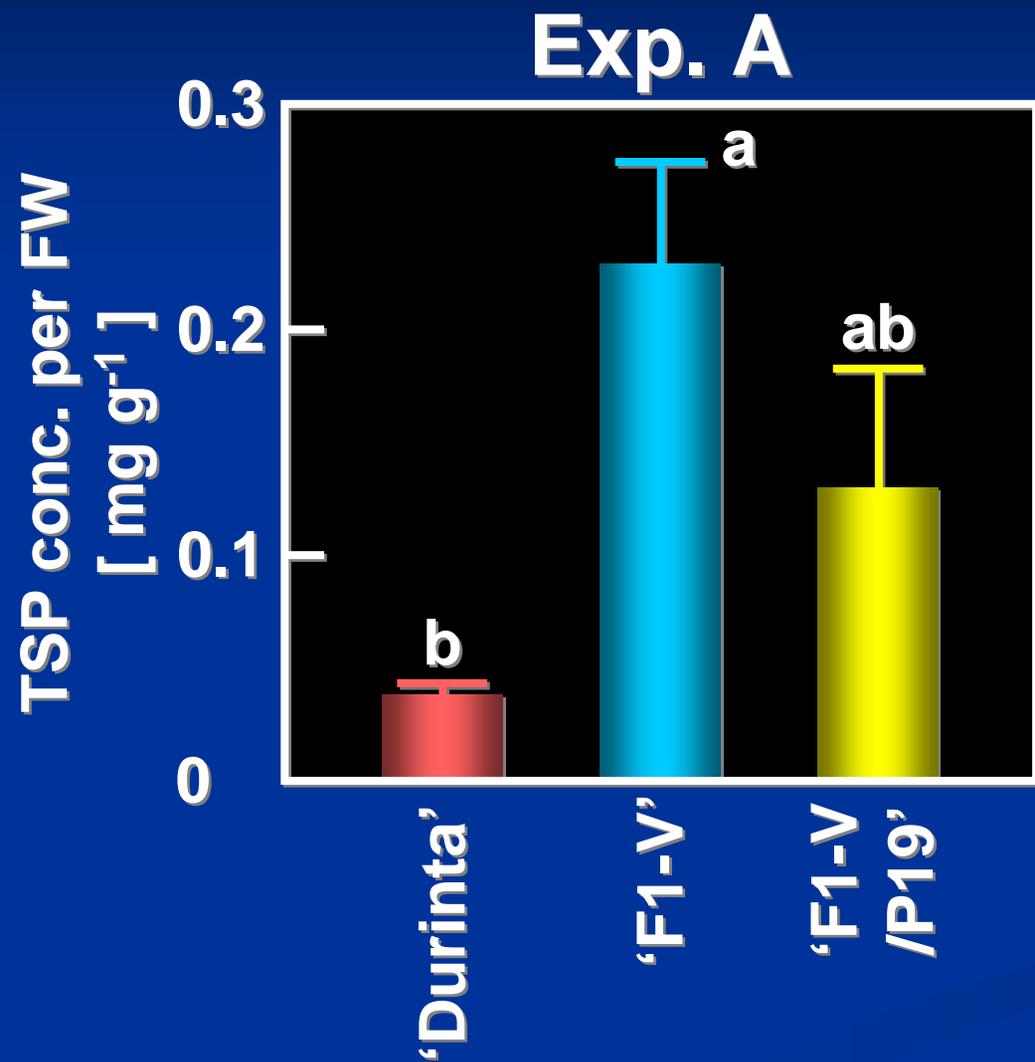
Estimated Protein Productivity
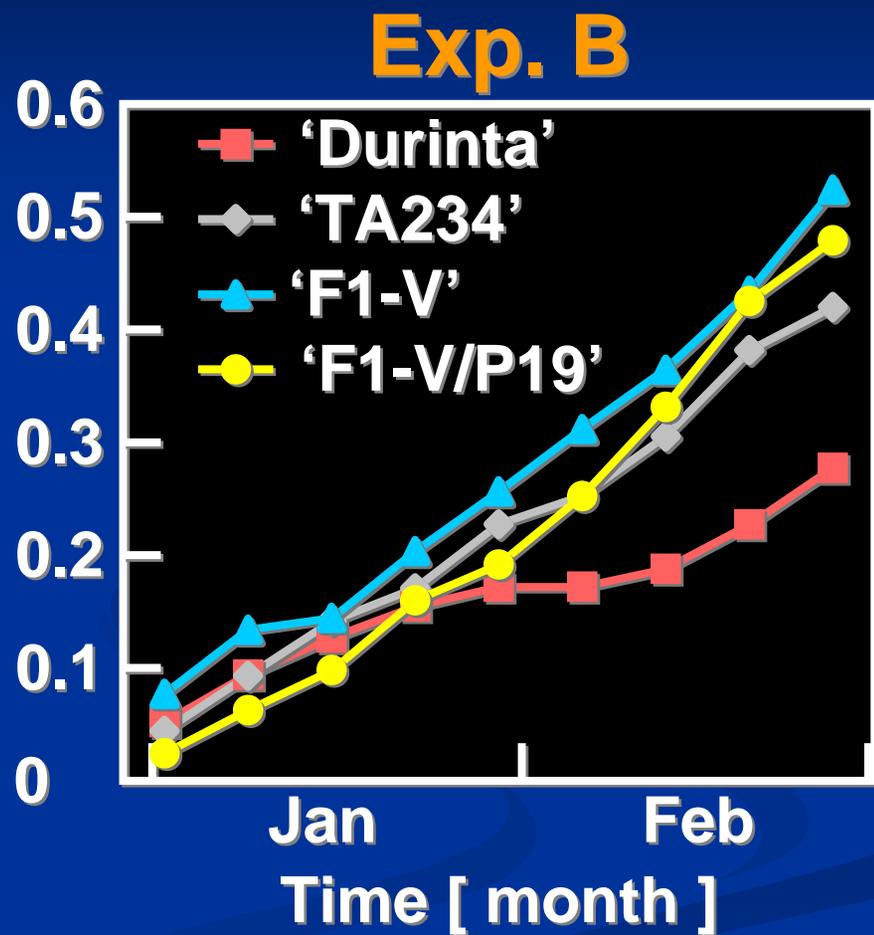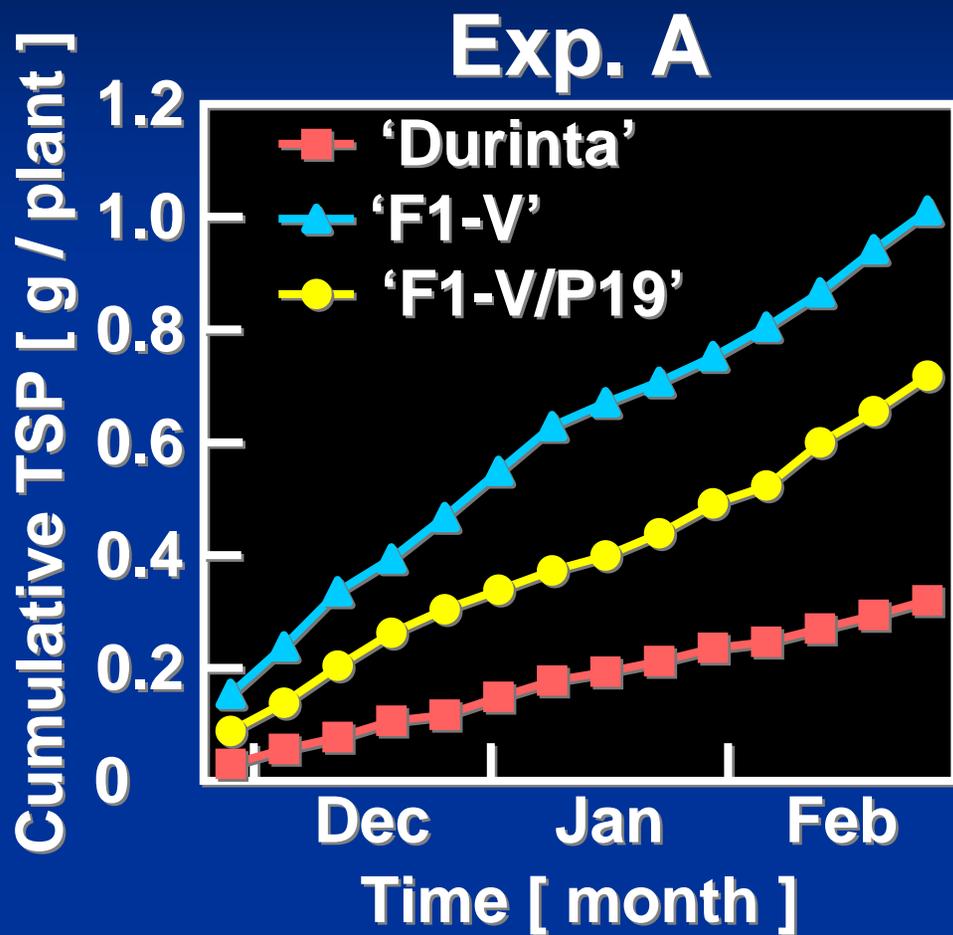
Conclusions

- **Transgenic tomato plants expressing plague vaccine protein showed lower yield but higher protein production in fruits than 'Durinta', a commercial greenhouse cultivar.**

- **A tomato cultivar that has high fruit productivity is not necessarily a suitable cultivar for biopharmaceutical protein production.**

Future Perspectives

- Analysis of F1-V vaccine protein productivity of the two transgenic lines is now in progress.

- In future studies, effects of environ-mental conditions (e.g., light intensity, temperature) and effects of nutritional conditions (e.g., N concentration) on TSP and F1-V production will be examined.

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