Survival, Growth, and Reproduction of red mangroves (*Rhizophora mangle* L.) in Restoration: Importance and Interaction of Genetic and Environmental factors

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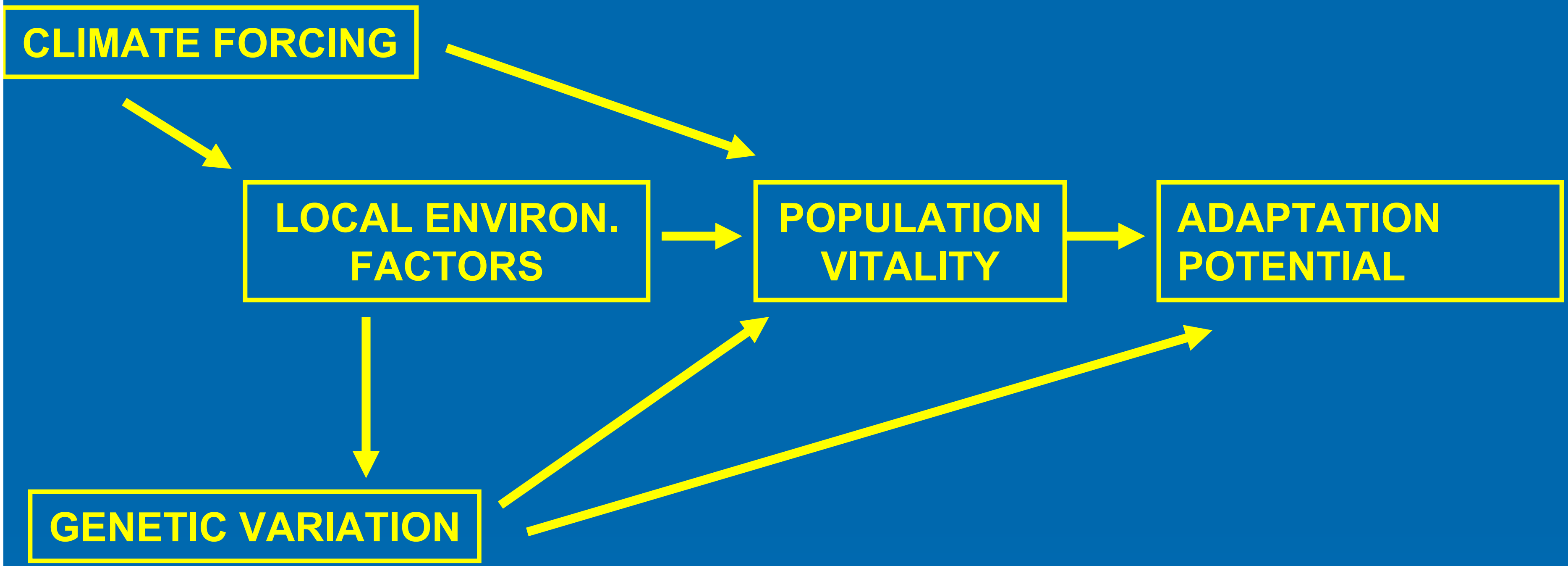
Foundation Species

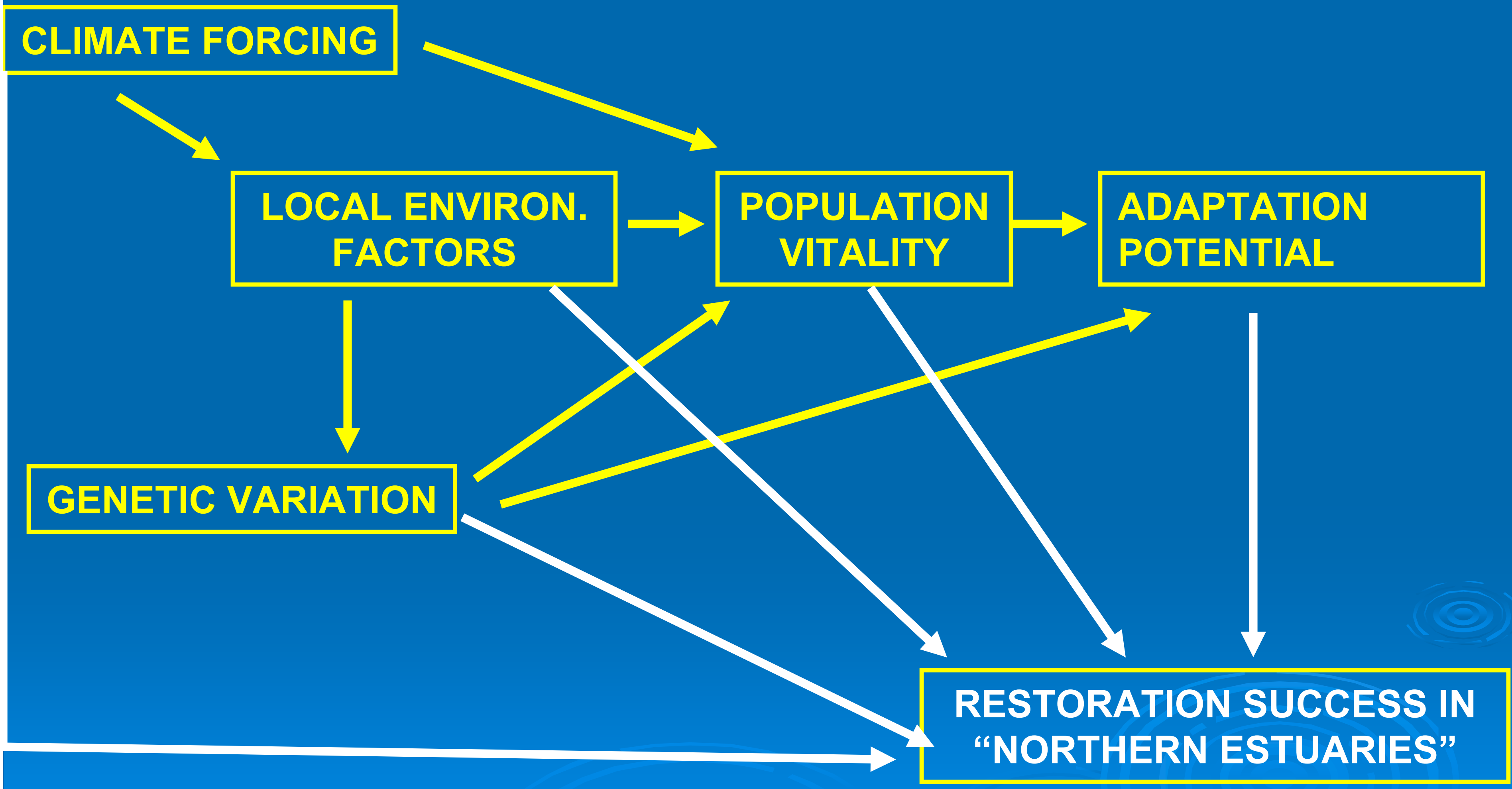
- Presence forms habitat structure and productivity base for the entire system
  - Mangrove trees
  - Salt marsh grasses
  - Reef-building corals
  - Seagrass
- Will interact
  - Intermed. Latitudes
  - Varying strength

Mangrove “islands” in a salt marsh “matrix”
Merritt Island National Wildlife Refuge

HYPOTHESIZED PATH DIAGRAM: ADAPTATION TO CHANGING CLIMATE



HYPOTHESIZED PATH DIAGRAM: RESTORATION



Field Experiment: Is there evidence for ecologically important genetic effects in red mangroves?

Address using:

- **Full sibling seedling families from individual maternal trees**
- **Planted in a common garden experiment with an elevation stress gradient**
- **Evaluate with statistical models and visualize with Norms of Reaction plots**

Specific Factors

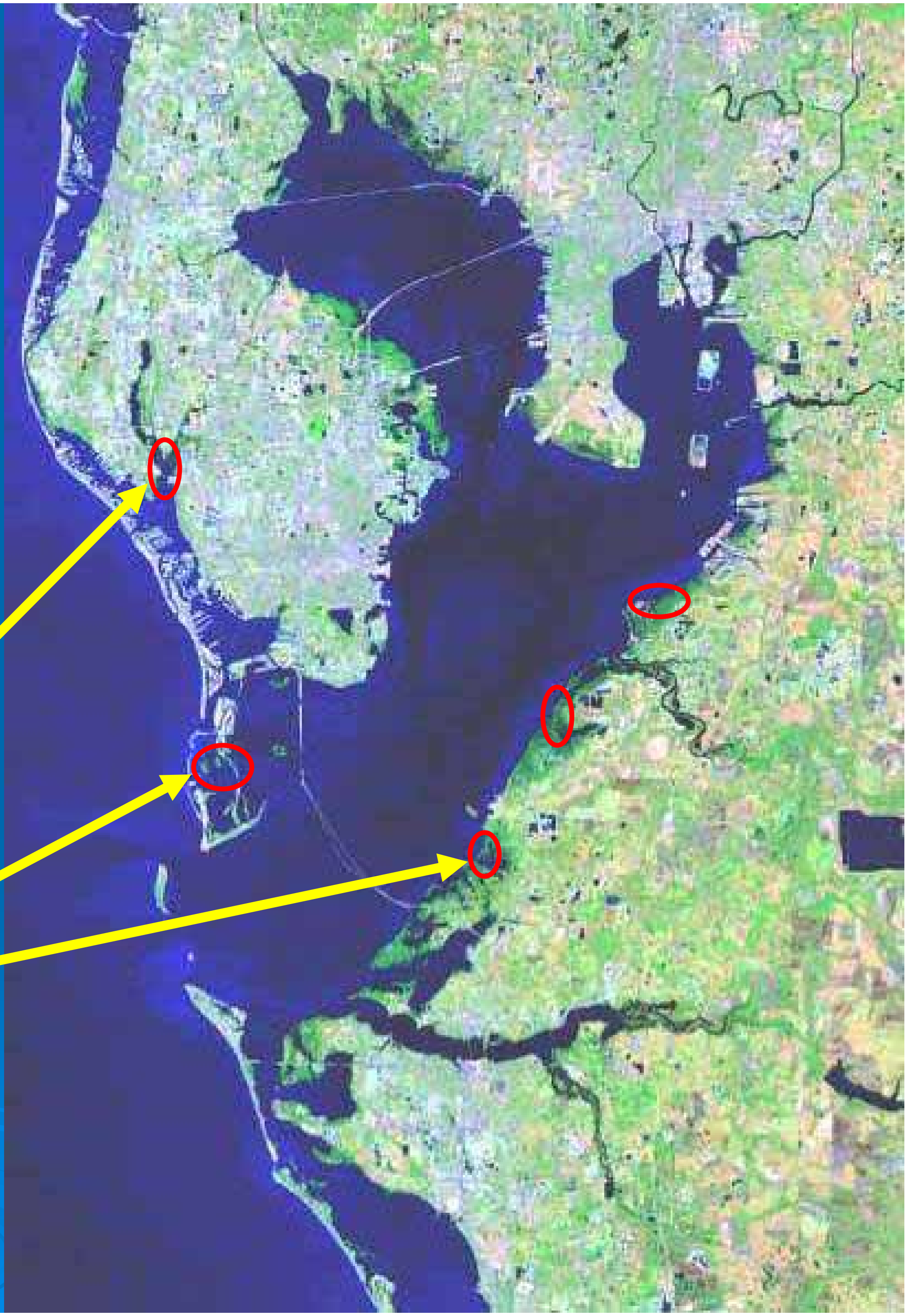
- Embayments within Tampa Bay……...5
- Donor trees from embayments………86
  - Sibling seedlings from each donor……40
- Islands Planted at Pt Redwing……….5
- Elevations w/in each Island……………2
  - Low: Less desiccation stress
  - High: More stress

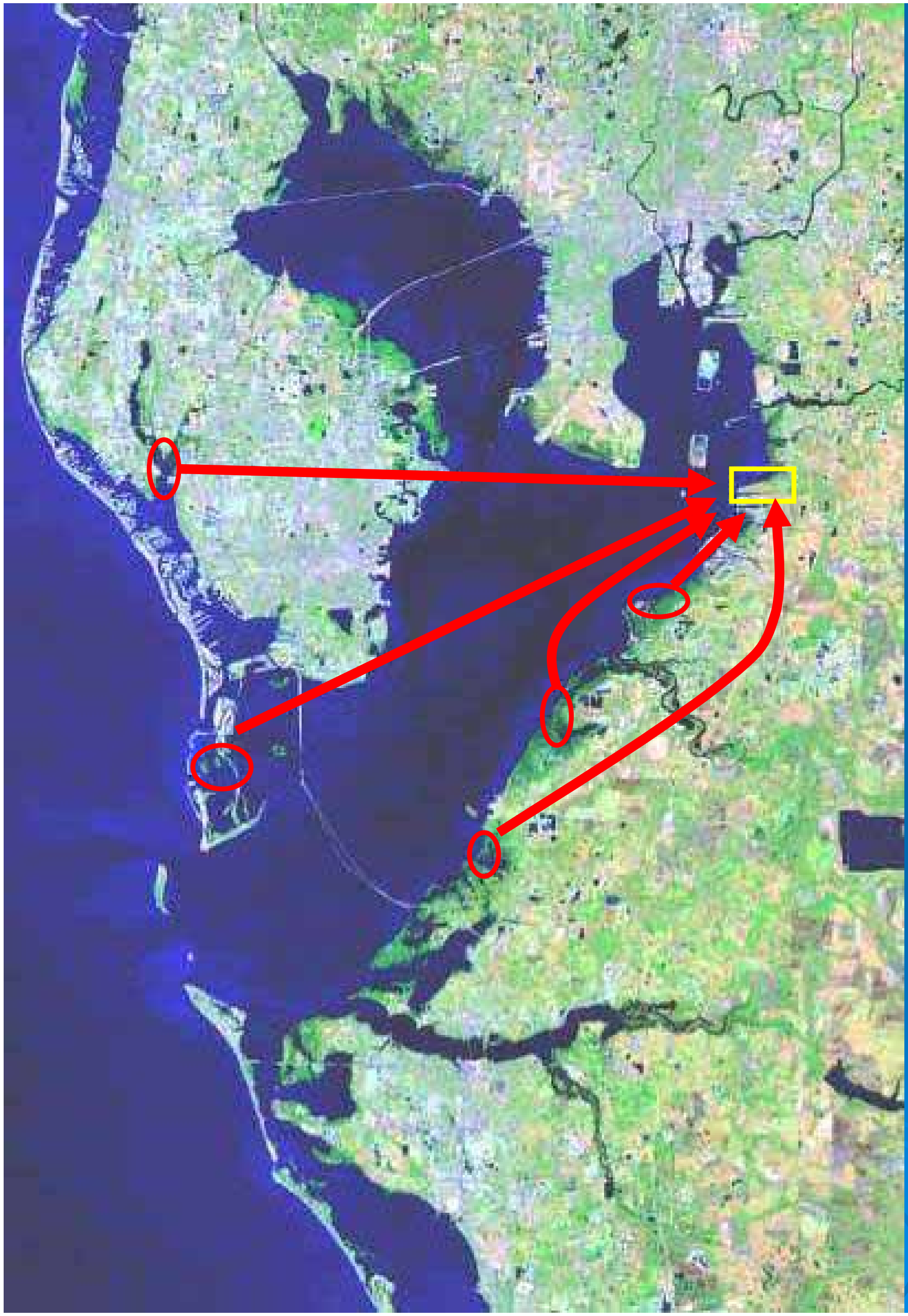
Why use Donor Trees from Different Embayments?

- Embaym. may differ in environmental growing conditions for the maternal trees (that might become maternal effects in the seedlings)

More urban area

Historically contaminated→high mutation rates





Embayments: Regions of Tampa Bay where DONOR trees were located (ie, Maternal Families taken from)

PLANTING LOCATION: (5 Islands at Schultz Family Park [Port Redwing])

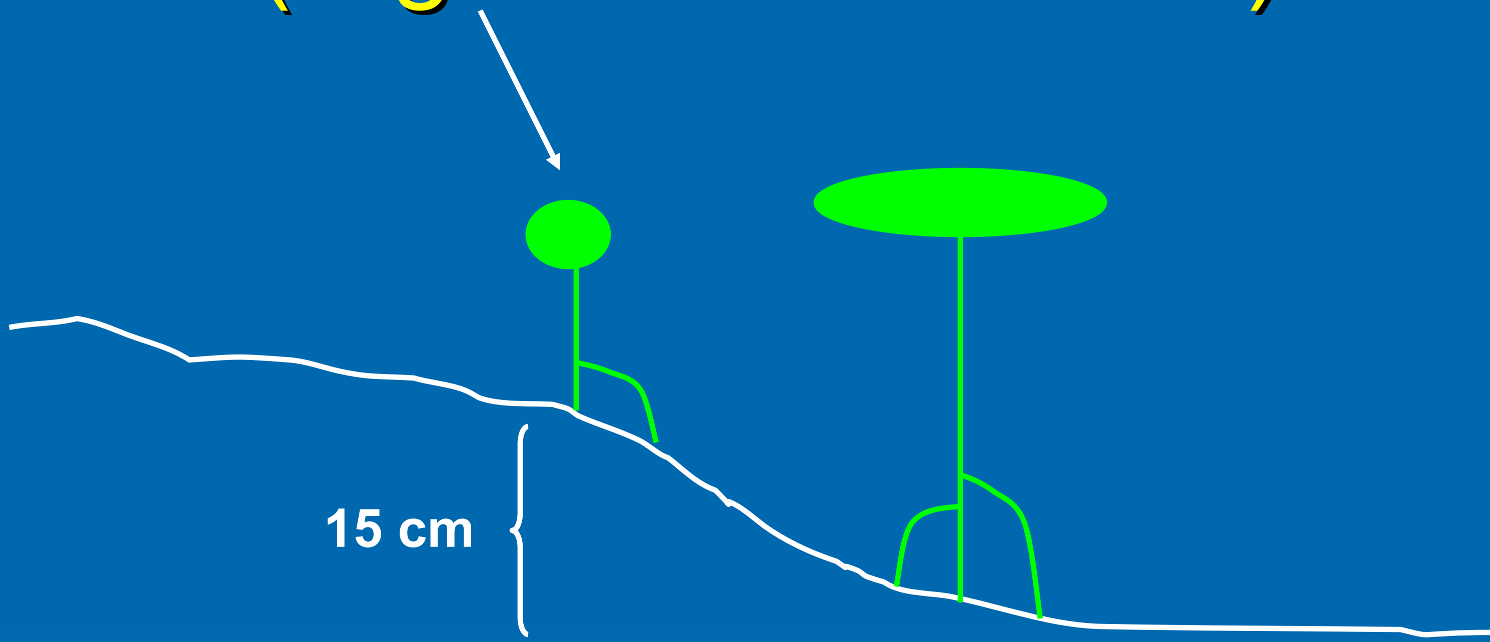
Maternal Families: Donor Trees (within embayment)

- Seedlings from a donor tree comprise a *sibling cohort* or *Maternal Family*
- Differences among Maternal Families reflects
  - Maternal tree environment that might be incorporated into propagules (e.g., nutrients; stressors like pollutants)
  - Maternal tree genotype

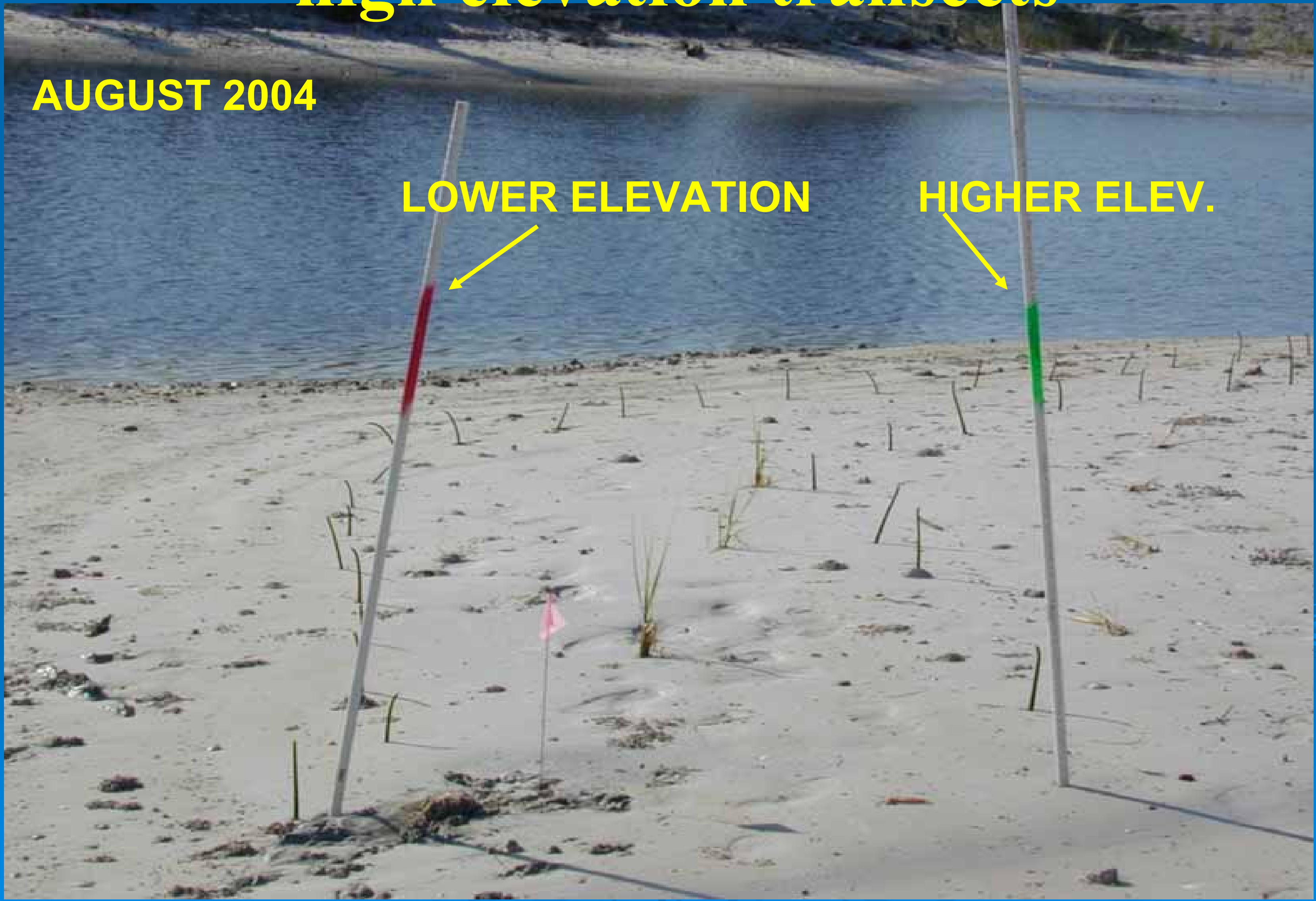
Elevation (stress) Effects

- Planted HIGH & LOW transects on each island separated by 15 cm elevation gradient

Hypothesized Elevation Effects (High=more stressful)



R. mangle experiment showing low & high elevation transects

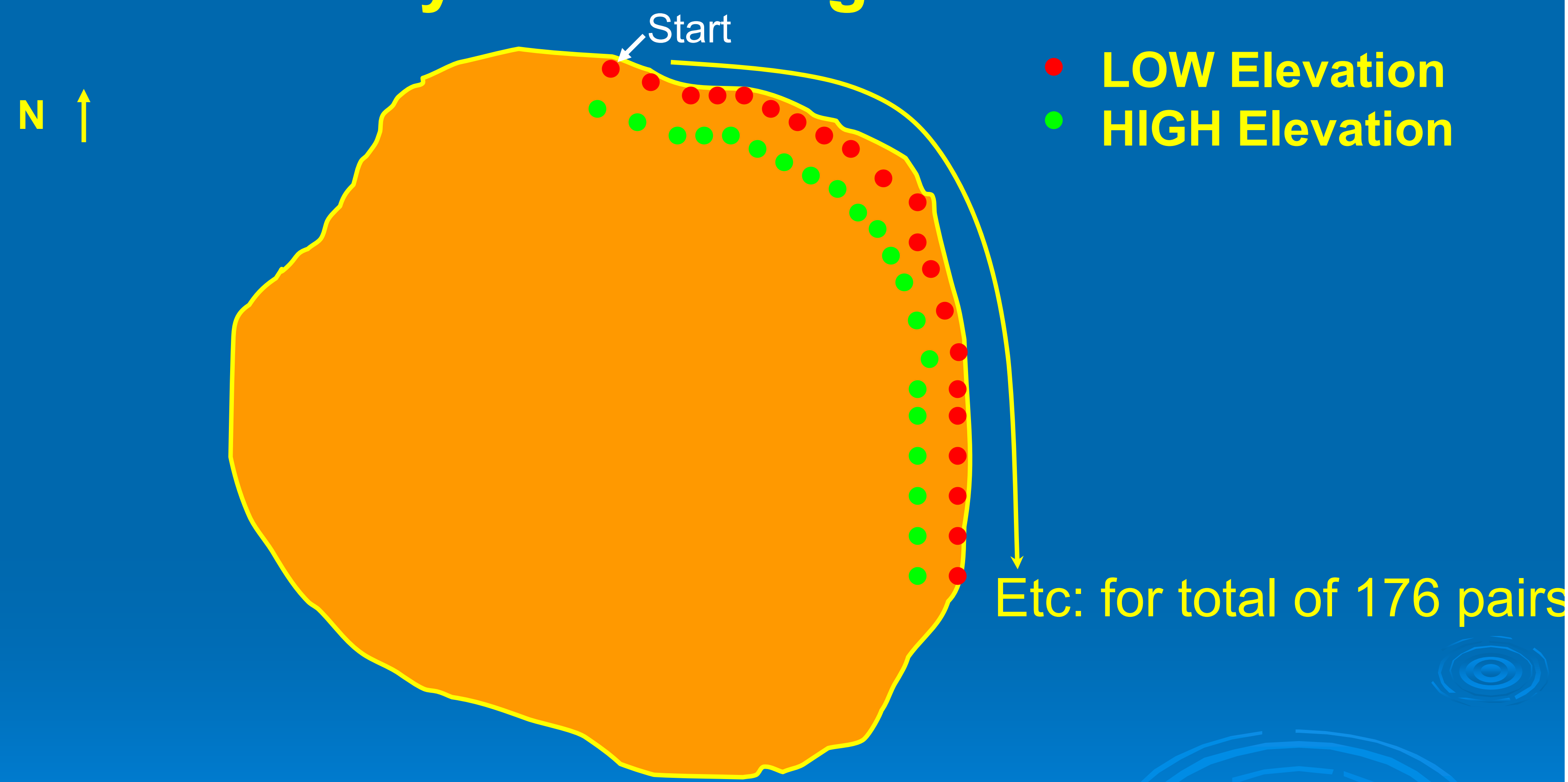


Planting 5 Different Islands

- **Effect of seedling environment over a spatial scale of 10s of m**

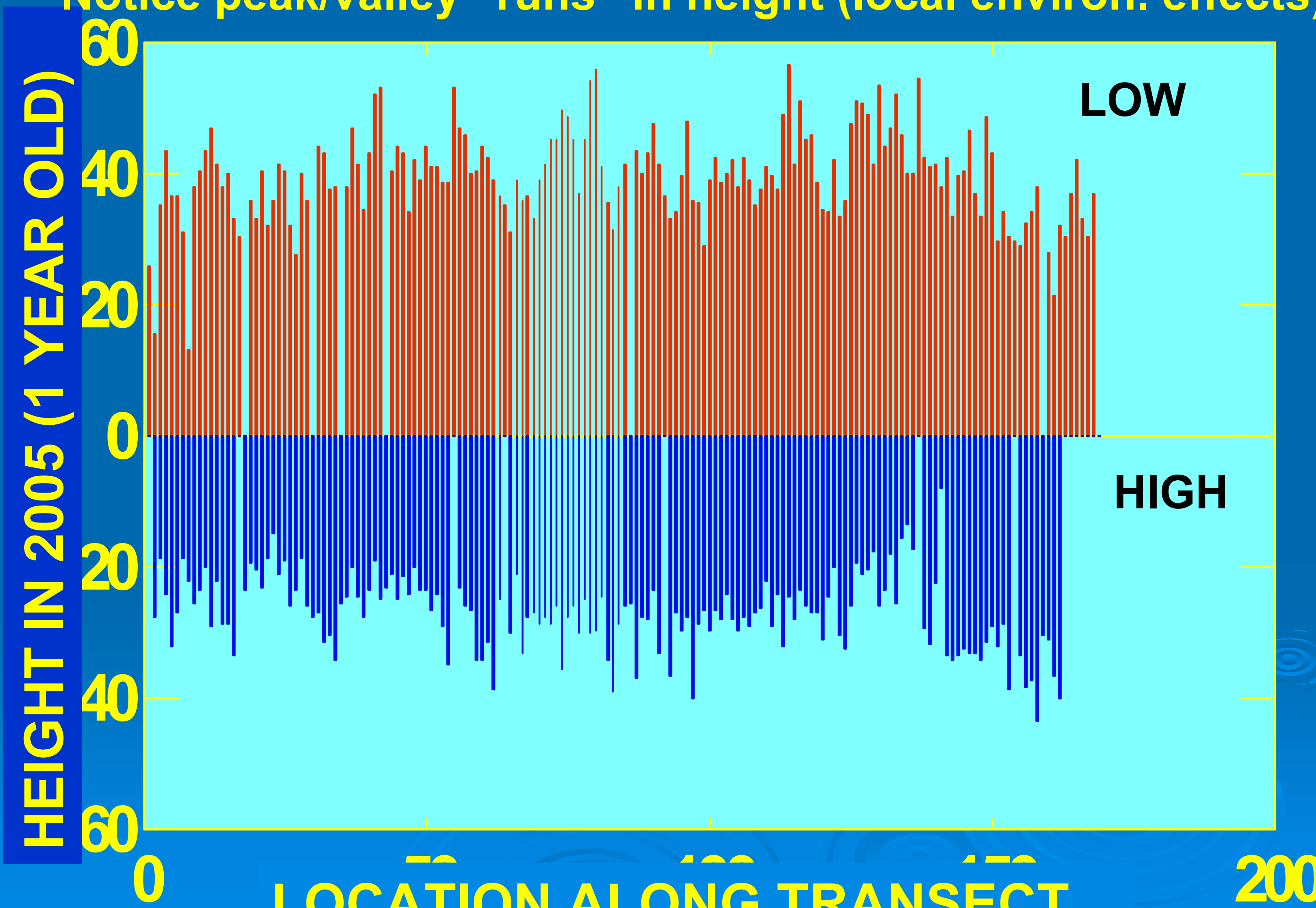
Photo courtesy of B. Henningsen, SWFWMD

Smallest Spatial Scale: different planting units every 0.5 m along Elev. transects



Island 4: Illustration of the LOW & HIGH elevation transects

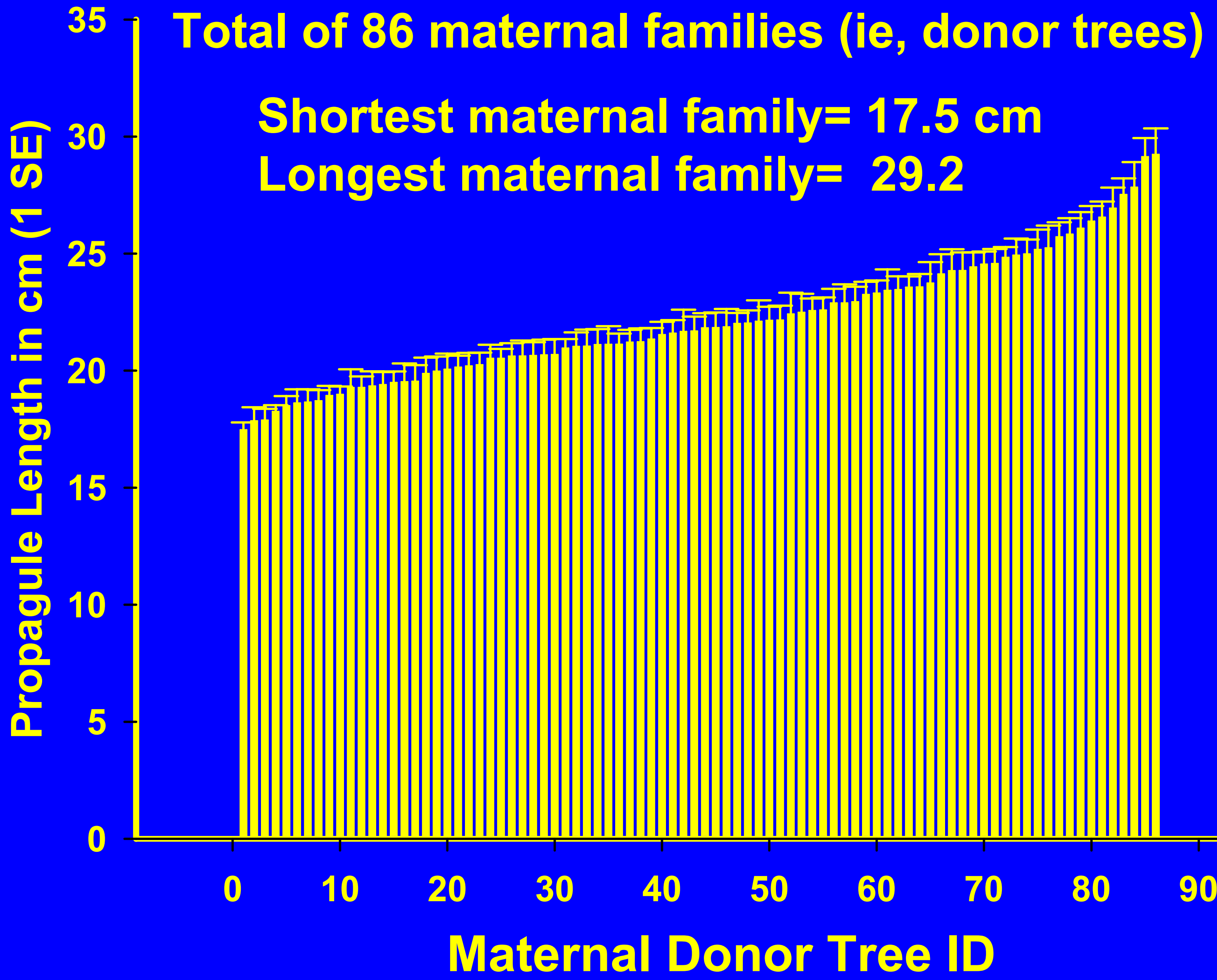
ISLAND 4: Heights along transects
– Notice peak/valley “runs” in height (local environ. effects)



Propagule Size

- Covariate in analyses.
- May reflect degrees of maternal stores

Rhizophora propagule length (mean & SE by donor tree)

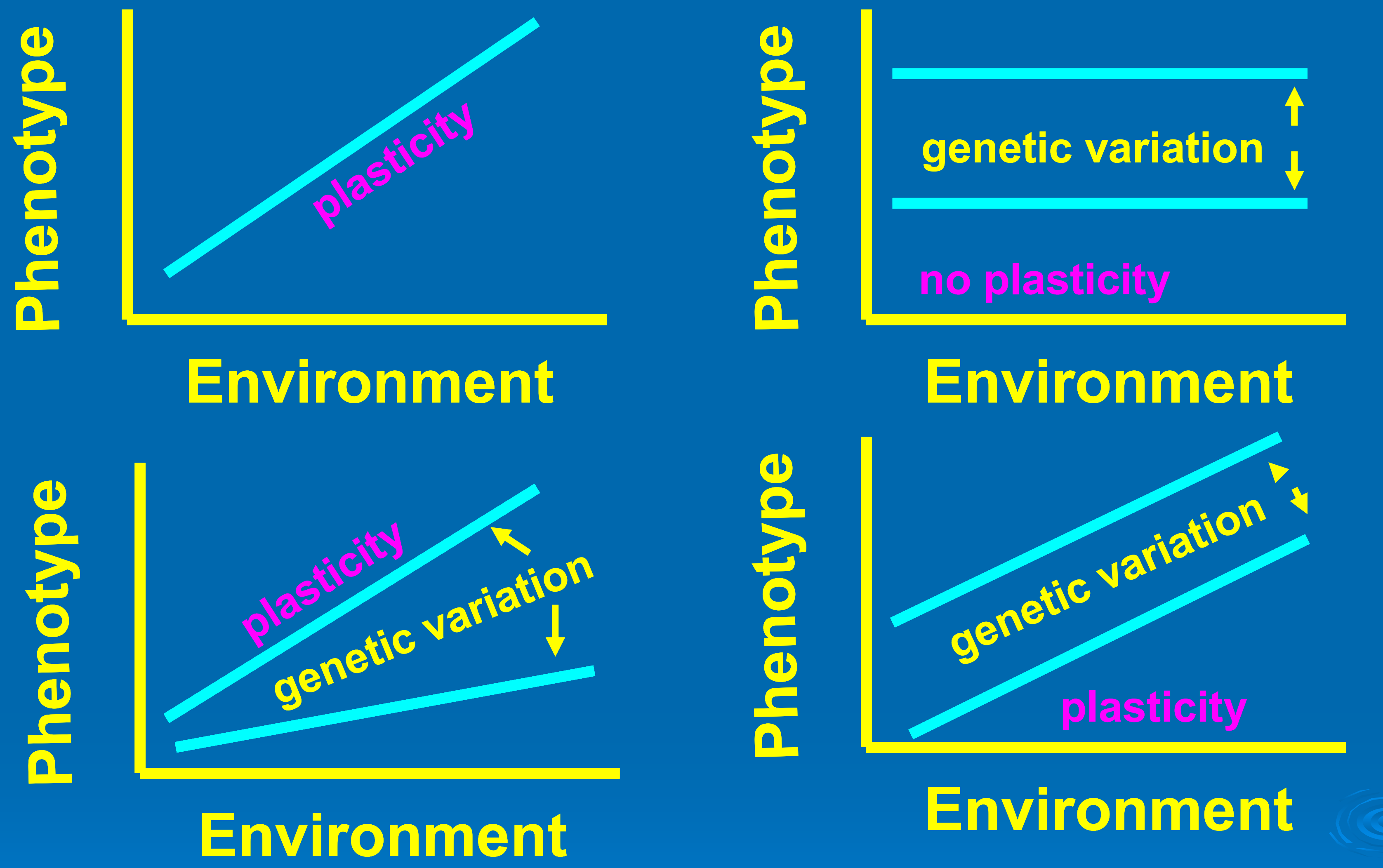


Determining Genetic Effects (from maternal families)

- This Experiment: Norms of Reaction for different maternal families of seedlings over the elevation stress gradient
- Next Experiment: confirm using F_1 offspring from these seedlings

How separate genetic, maternal, plastic, and environmental effects?

- **Field Experiment:**
  - ***Norms of reaction*** **over an environmental gradient (*Elevation*) of a series of seedling sibling families (ie, from different donor trees)**
  - **Assess presumed *maternal effects* by:**
    - **Use *propagule size* as a covariate**
  - **Determine *local environmental* effects –**
    - **Diff's among maternal tree environments (*embayments within Tampa Bay*)**
    - **Diff's. among *islands planted***
    - **Diff's *along transects* on each island planted**



NORMS OF REACTION

See D. Devlin, PhD dissertation, Univ. of Louisiana at Lafayette, 2004

RESULTS

Experimental Design: Reminder

Statistical Effects (fixed)

- **ELEVATION : (2 Levels)**
- **ISLAND PLANTED: (5 Levels)**
- **Maternal Tree Home Embayment or EMBAYMENT: (5 Levels)**
- **MATERNAL FAMILY: (86 Levels)**

PROPAGULE LENGTH: Covariate

Total Propagules Planted = 1,685

Response Variables

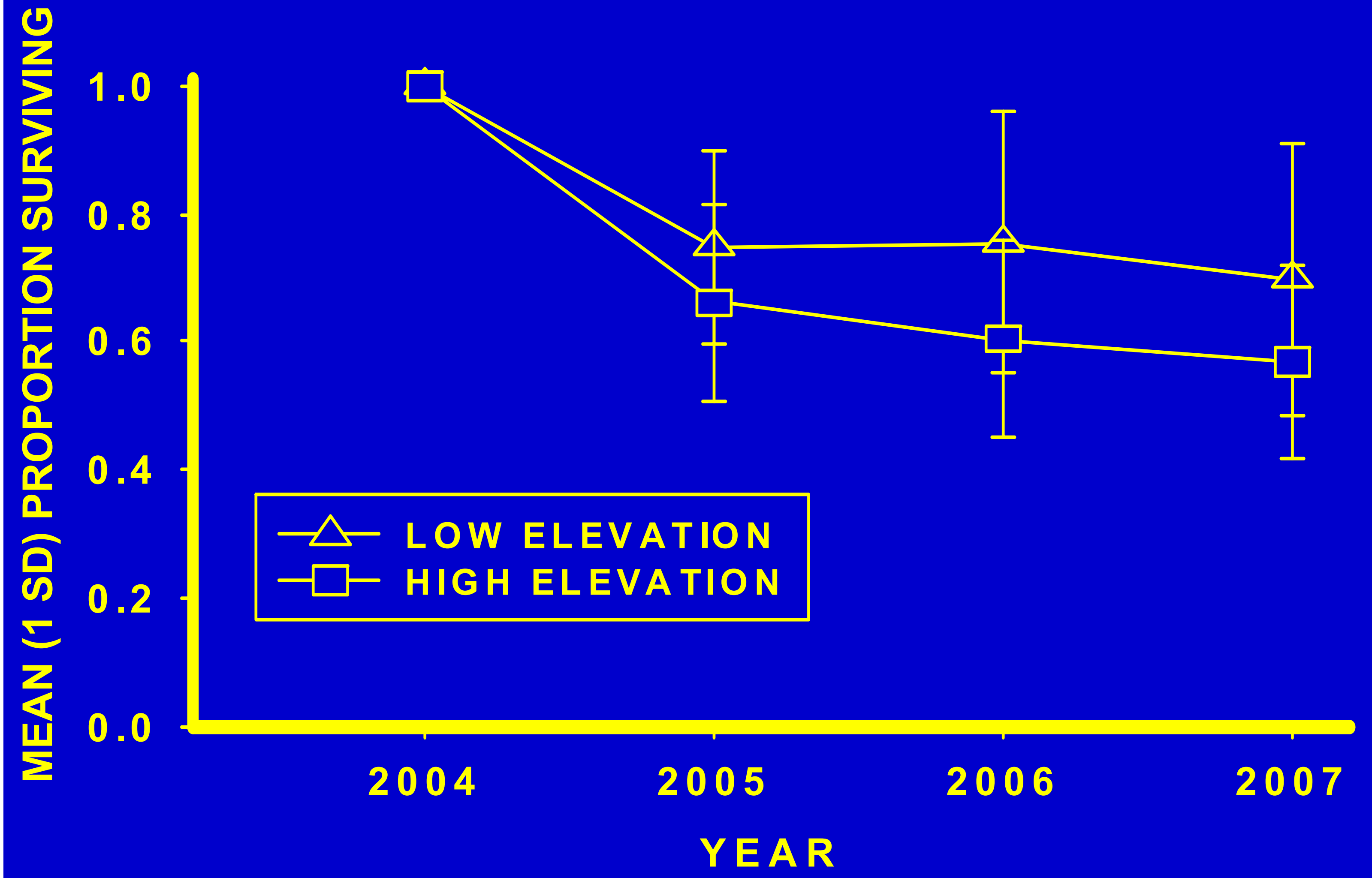
- Survival
- Plant Height (annually for 3 years)
- Trunk Diameter (annual for 3 years)
- Annual Incremental *Growth* in Height
- Number of Stems
- Canopy Area (based on diameters of major & minor axes of “ellipse”)
- Ratio of Sq. Rt(Canopy Area) : Height
- Reproductive output (propagules produced at year 3)

Main Research Hypotheses

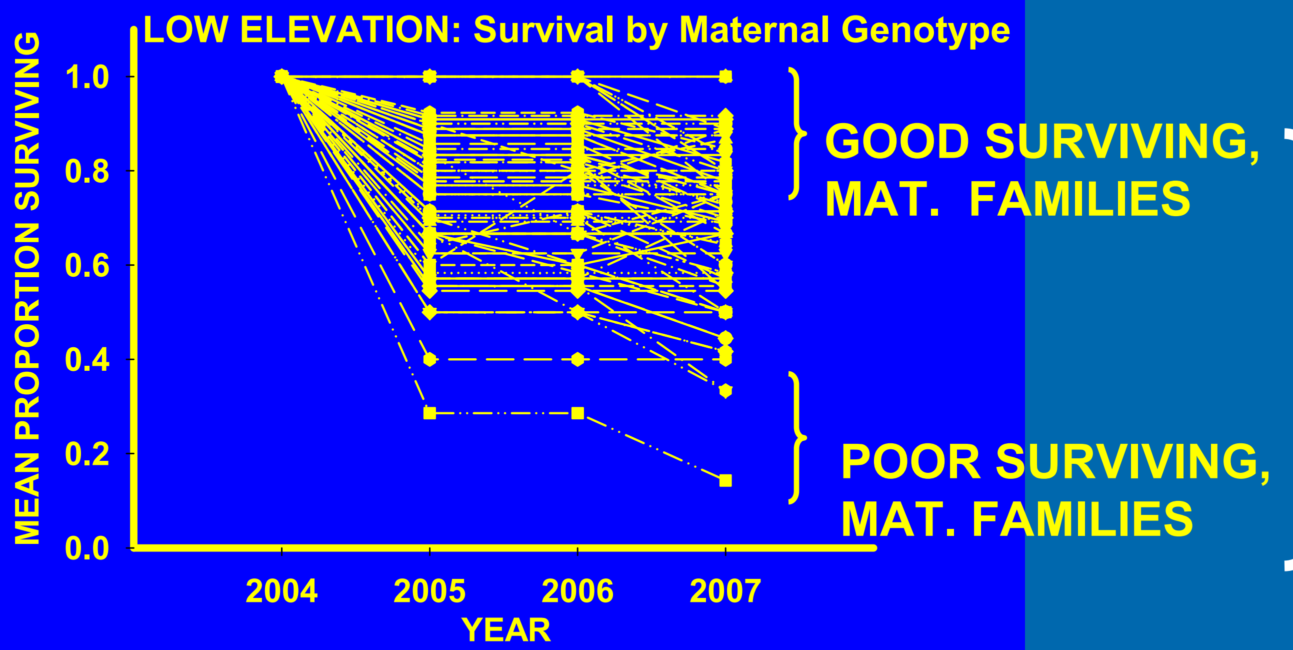
- Does plant performance differ with *seedling maternal family ?*
- Do seedlings from a maternal family respond differently to *Low & High elevation*?

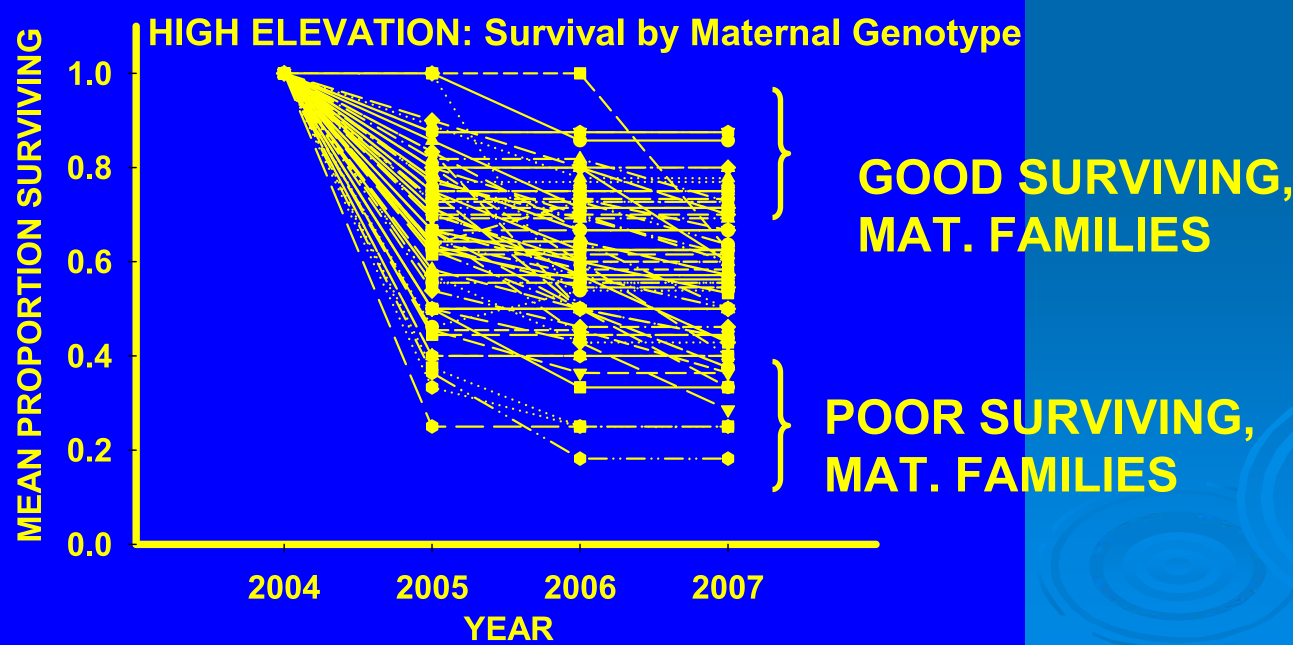
SEEDLING SURVIVAL

SURVIVAL AT LOW & HIGH ELEVATIONS



SURVIVAL BY MATERNAL FAMILY



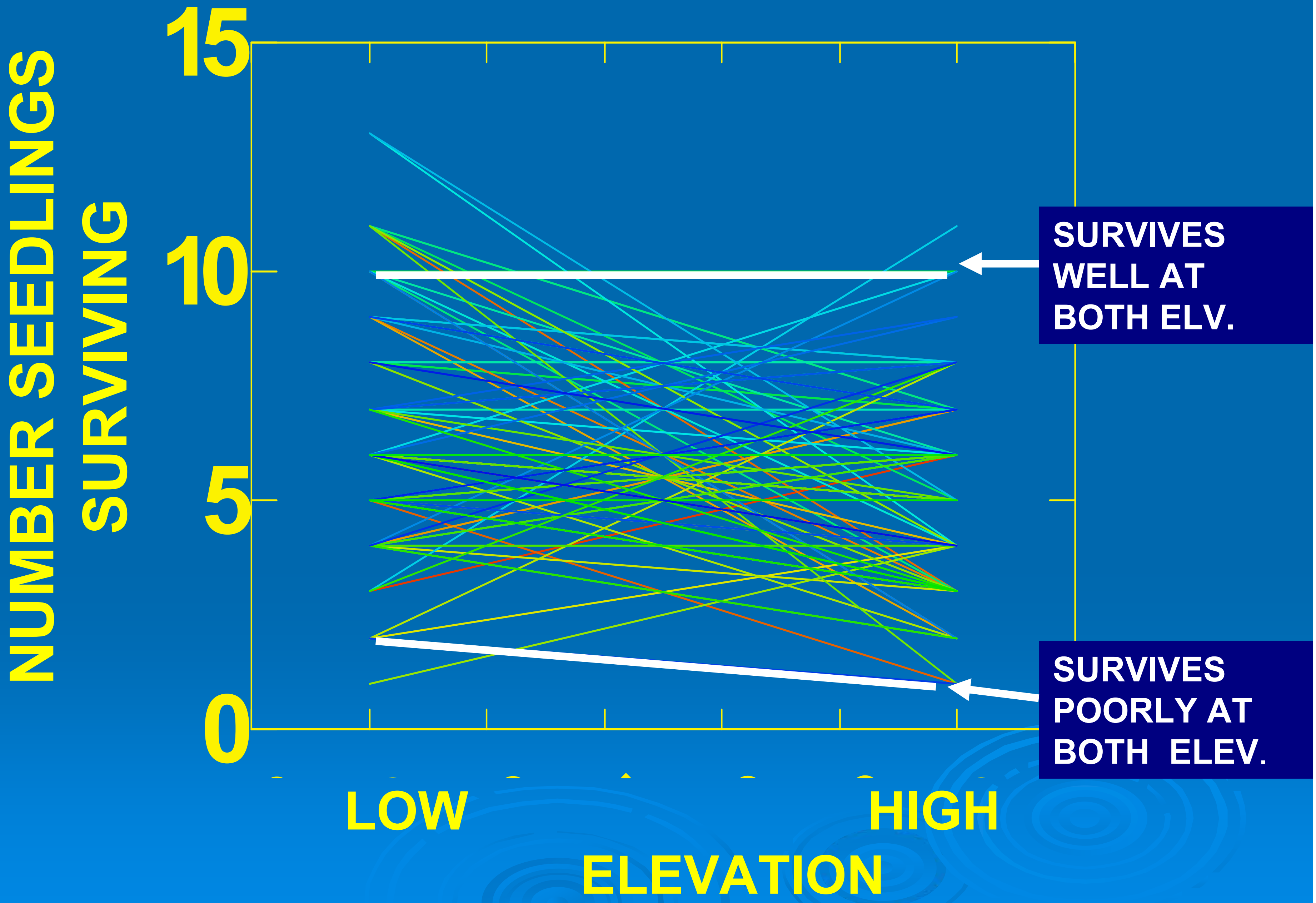


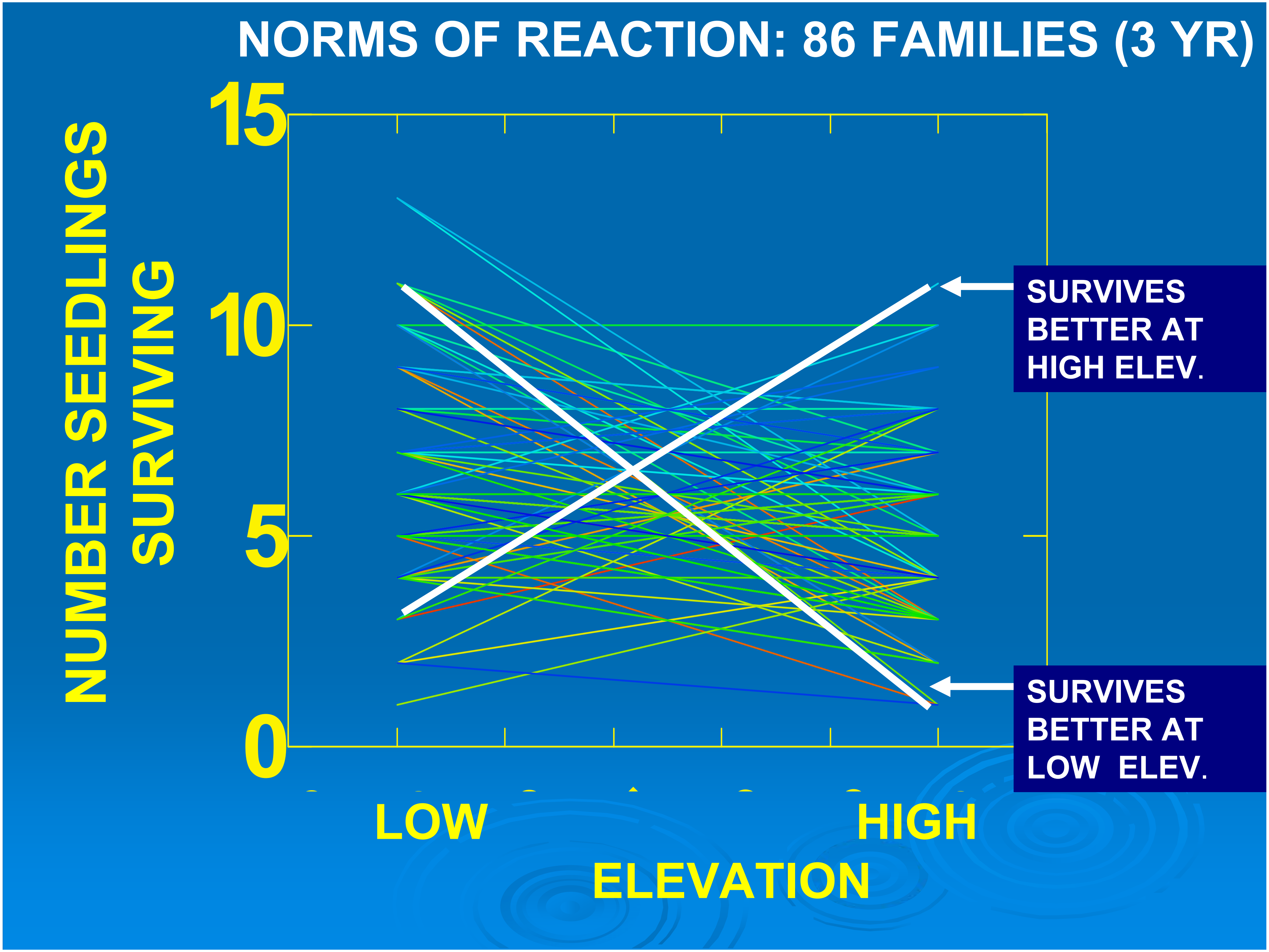
SURVIVAL

LOW ELEV.

HIGH ELEV.

NORMS OF REACTION: 86 FAMILIES (3 YR)




NORMS OF REACTION: 86 FAMILIES (3 YR)
NUMBER SEEDLINGS SURVIVING
15
10
5
0
SURVIVES BETTER AT HIGH ELEV.
SURVIVES BETTER AT LOW ELEV.
LOW
HIGH
ELEVATION

SUMMARY: NORMS OF REACTION

- Plasticity exists within maternal families (with environmental stress)
  - Independent of propagule size
- Genetic differences among maternal families
  - Assumption that aspects of survival are heritable

Logit modeling Results : Survival at 3 years

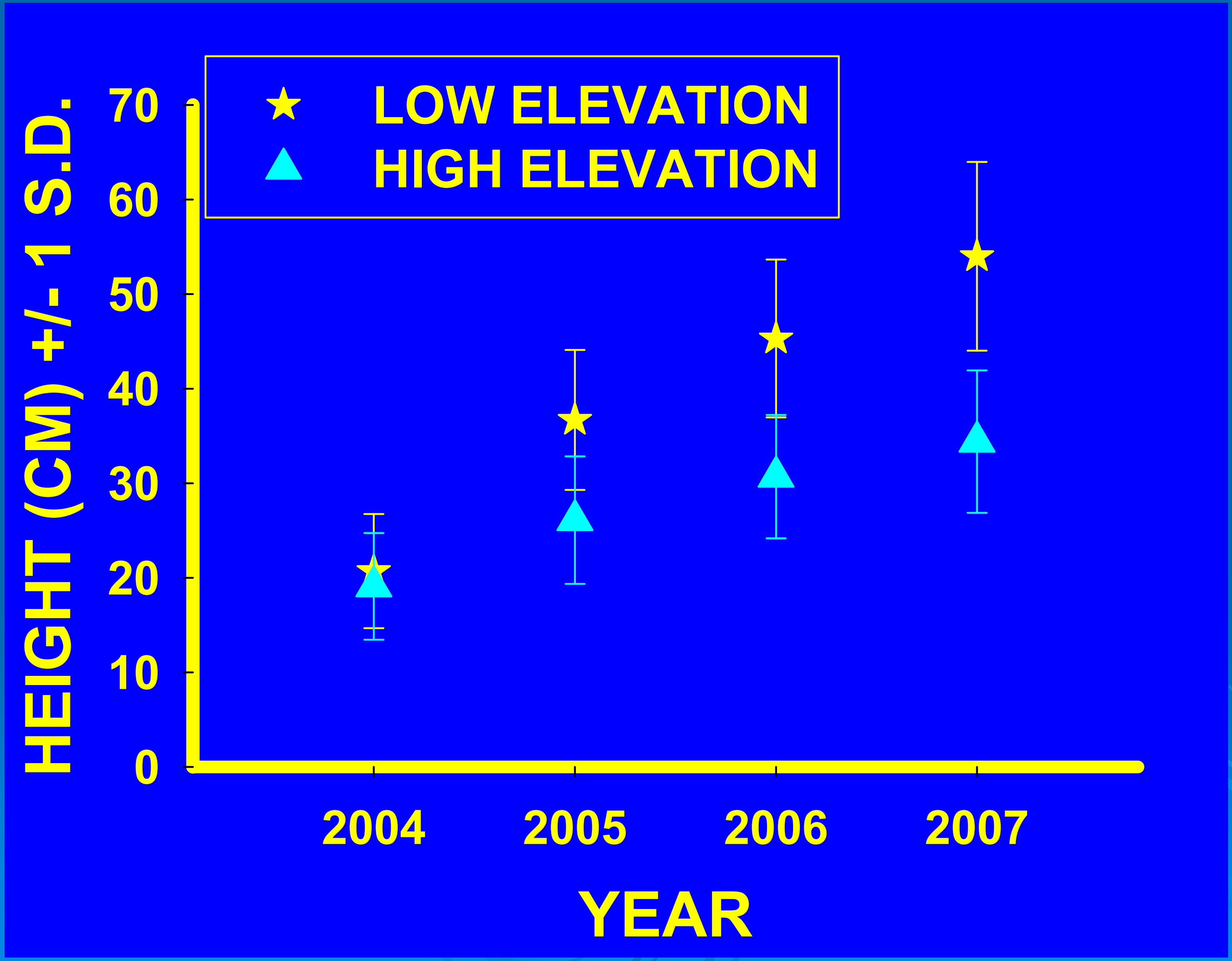
- **Response Variable (modeled alive = 1, dead =0), p < 0.0005, McFadden's Rho Squared = 0.29,**
- **Significant Explanatory Variables were (best model selected by AIC):**
  - **Elevation – Odds of surviving 3.1x greater at LOW Elv. When maternal family not considered**
  - **Maternal family – Odds ratios ranged from**
    - **3.2 : 1 (greater survival at LOW elv. For one seedling family**
    - **0.06:1 (greater surv. At HIGH elv. For another seedling family**
  - **Island Planted (no sig. effect of "local" seedling environment)**
  - **Propagule size (no sig. effect)**

Growth in Height

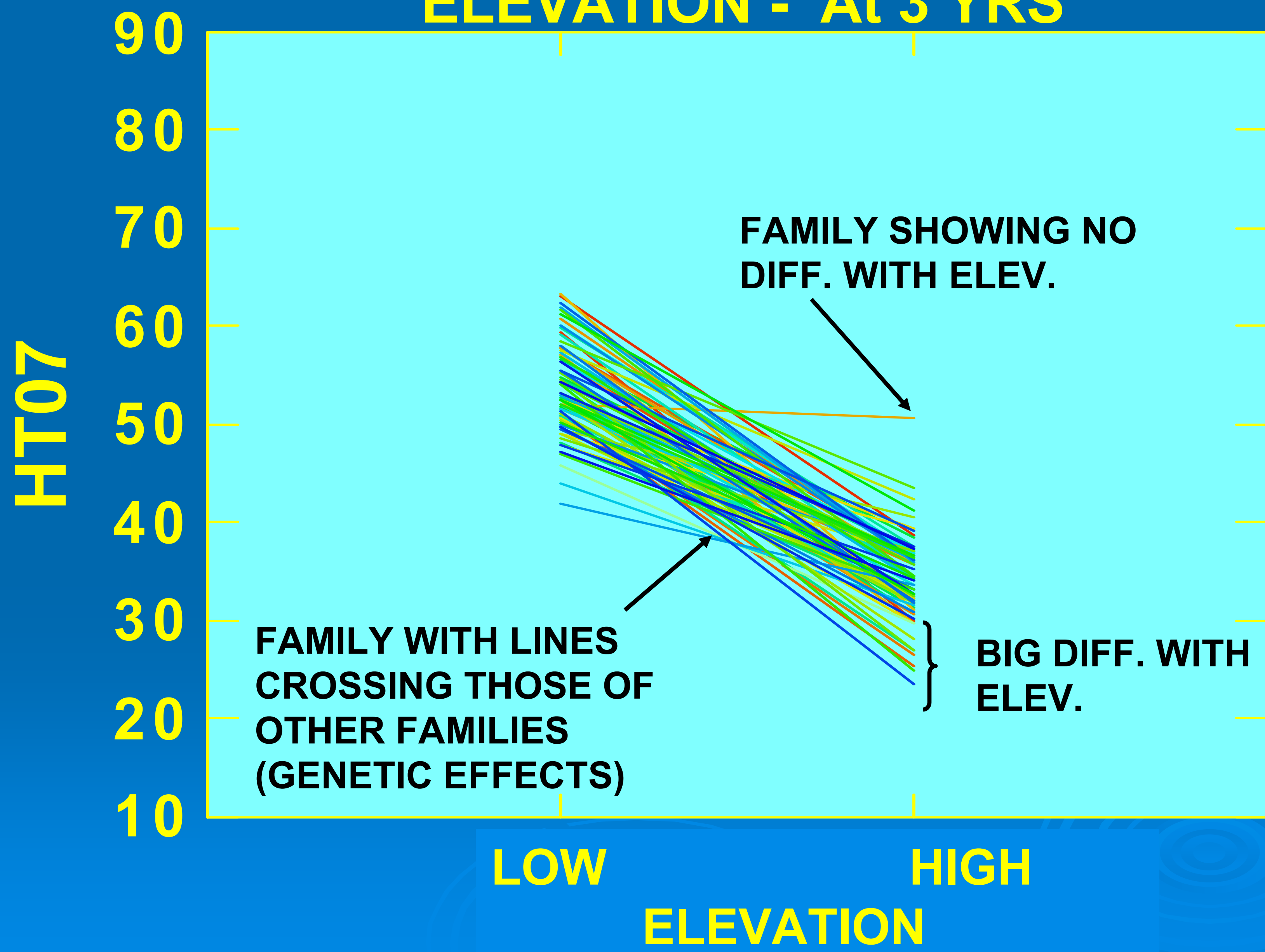
HEIGHT (2005): 1 year

2007 (3 yrs old)

GROWTH IN HEIGHT SINCE EXPERIMENT INITIATION IN 2004



HEIGHTS BY MATERNAL FAMILY & ELEVATION - At 3 YRS



HEIGHT (Repeated-Measures ANOVA over 3 years)

Repeated Measures Analysis of Variance

Between-Subjects Effects

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Embayment	4	103.3598	25.8399	0.23	0.9236
Elevation	1	169264.1010	169264.1010	1483.66	<.0001
Island Plntd	4	17010.2821	4252.5705	37.28	<.0001
MatFam(Embay)	81	14224.8712	175.6157	1.54	0.0022
Propag. Len.	1	6890.1448	6890.1448	60.39	<.0001
Error	951	108495.1138	114.0853		

All significant EXCEPT environmental influences among *embayments-within-Tampa Bay* (large spatial scale; locations of maternal trees)

Height con't: effects over time

Within subjects over time (Univariate effects)

Source	DF	MS	F Value	Pr > F	G - G
height	2	1225.22	73.33	<.0001	<.0001
height*Embaym	8	19.85263	1.19	0.3022	0.3096
height*Elev.	2	5127.79451	306.89	<.0001	<.0001
height*ISLAND	8	70.26781	4.21	<.0001	0.0003
ht*MatFam(Emby)	162	20.00410	1.20	0.0519	0.0773
height*prop Leng.	2	162.39419	9.72	<.0001	0.0003
Error(height)	1902	16.70889			

The effects on height of:

- elevation
- island planted
- propagule length

} *Changed over time*

The effects on height of:

- Maternal family

} *Constant over time*

Height: Independent Variables Effect Sizes

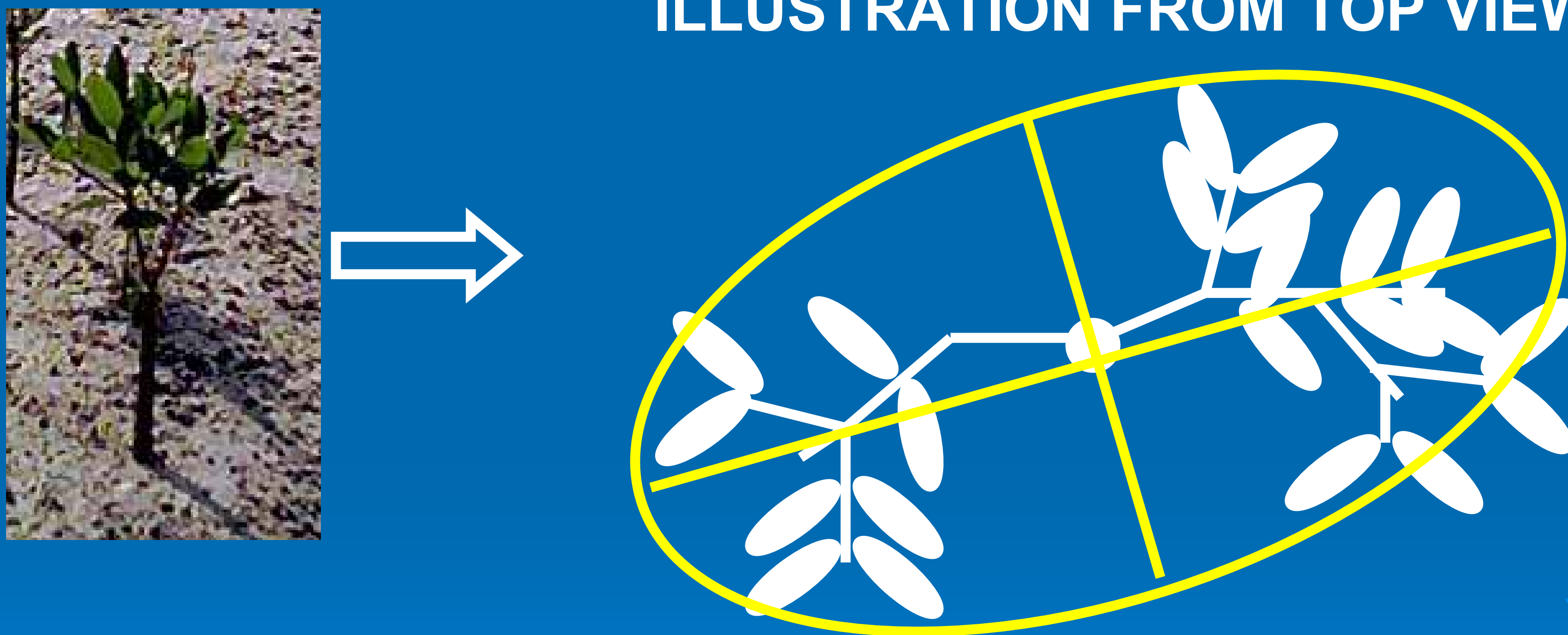
- Calculate at year 3 (cumulative effect over 3 years)

HEIGHT AT 3 YRS (2007) ANOVA & EFFECT SIZES

SOURCE	*SIG.*	*EFFECT SIZE*
MATERNAL TREE EMBAYMENT	**NS**	
ELEVATION (LOW or HIGH)	**0.0005**	**9.94**
PLANTING ISLAND (1…5)	**0.0005**	**+3.99 TO -3.47**
MATERNAL FAMILY	**0.011**	**+6.40 TO -7.14**
PROPAGULE LENGTH (COVAR.)	**0.0005**	**0.64**
ELEV. X ISLAND	**0.0005**	**+3.46 TO -1.99**

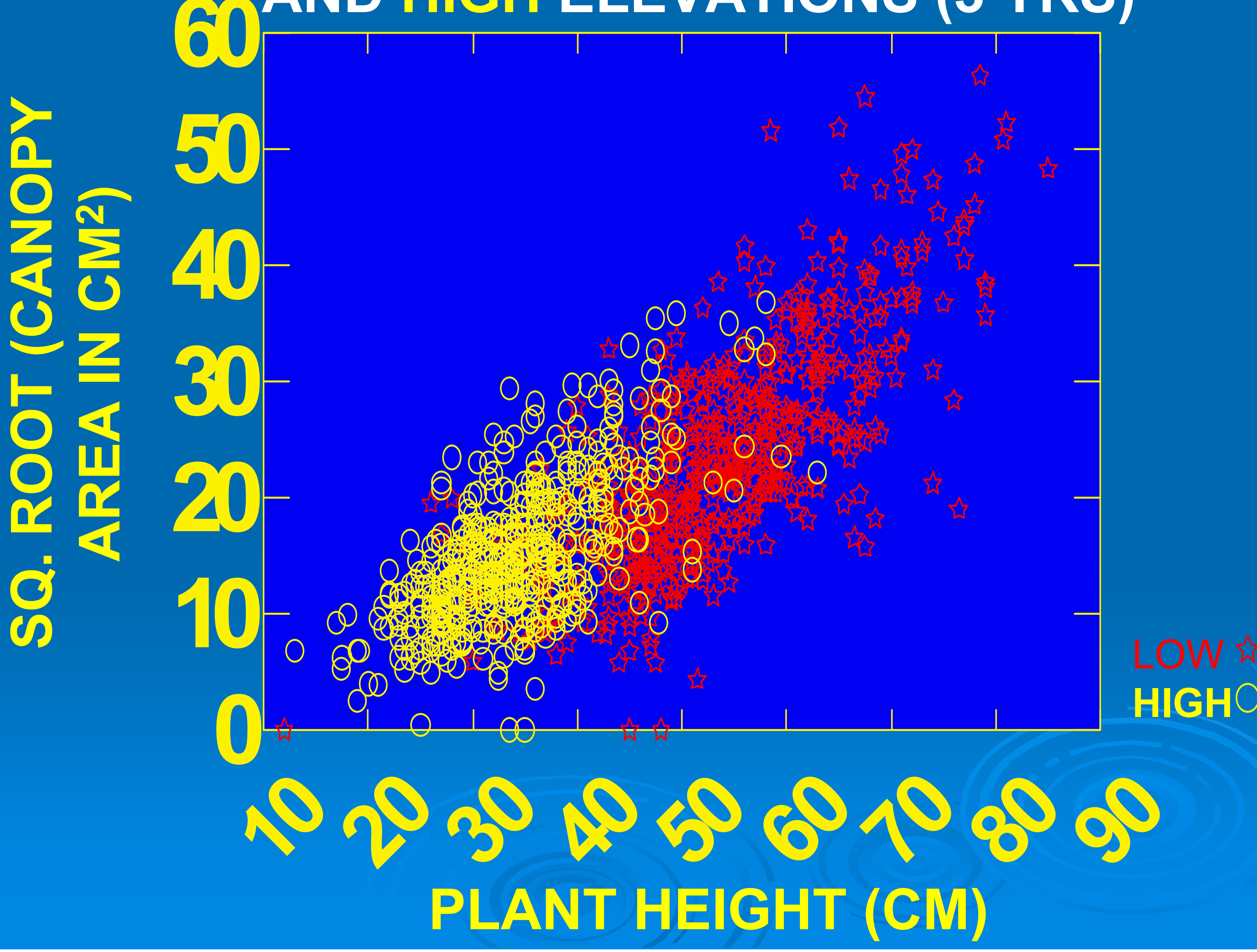
CANOPY AREA (AT 3 YEARS)

ILLUSTRATION FROM TOP VIEW

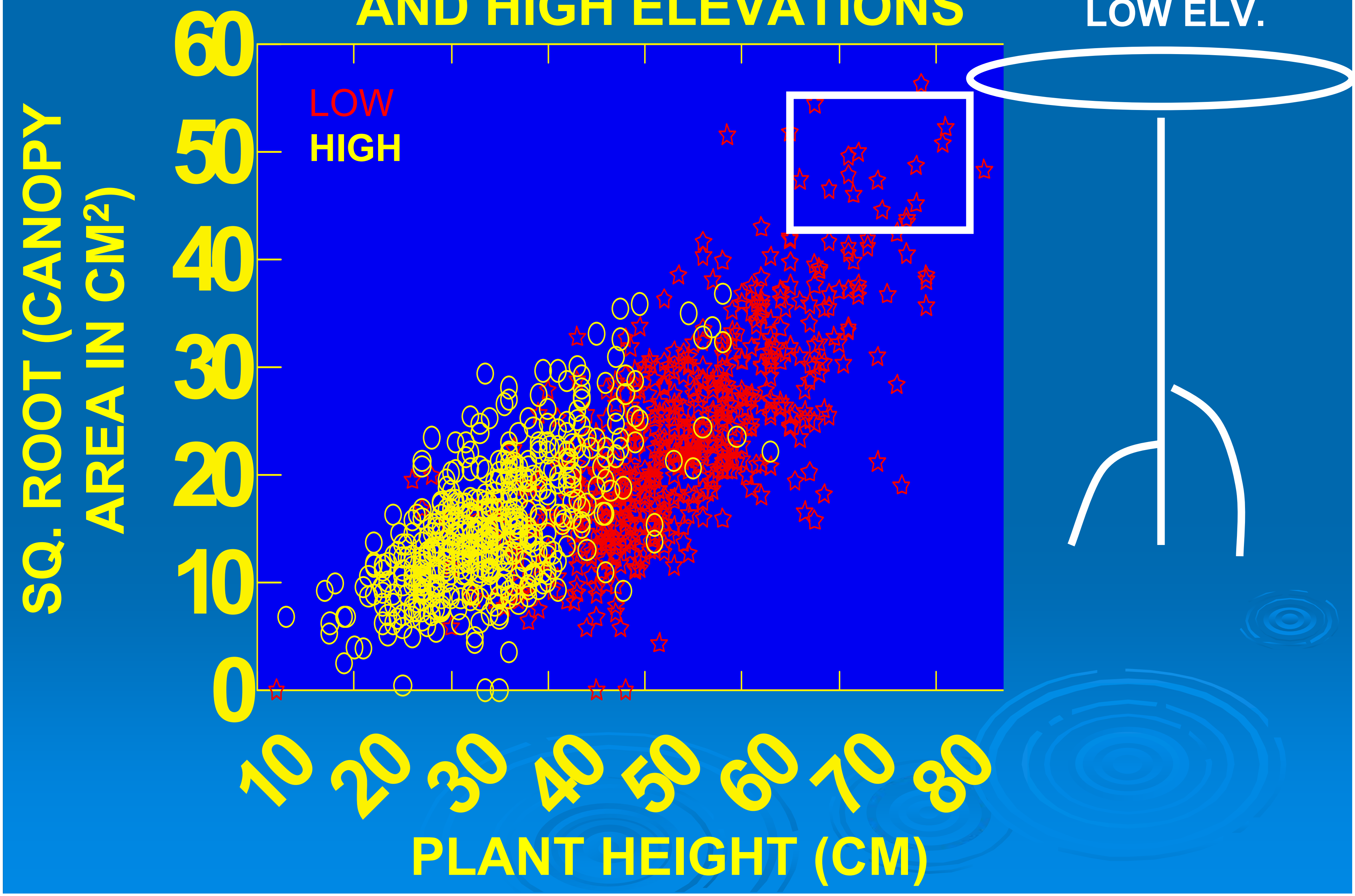


- MEASURE LENGTH OF MAJOR & MINOR AXES
- ESTIMATE AREA FROM EQUATION FOR ELIPSE

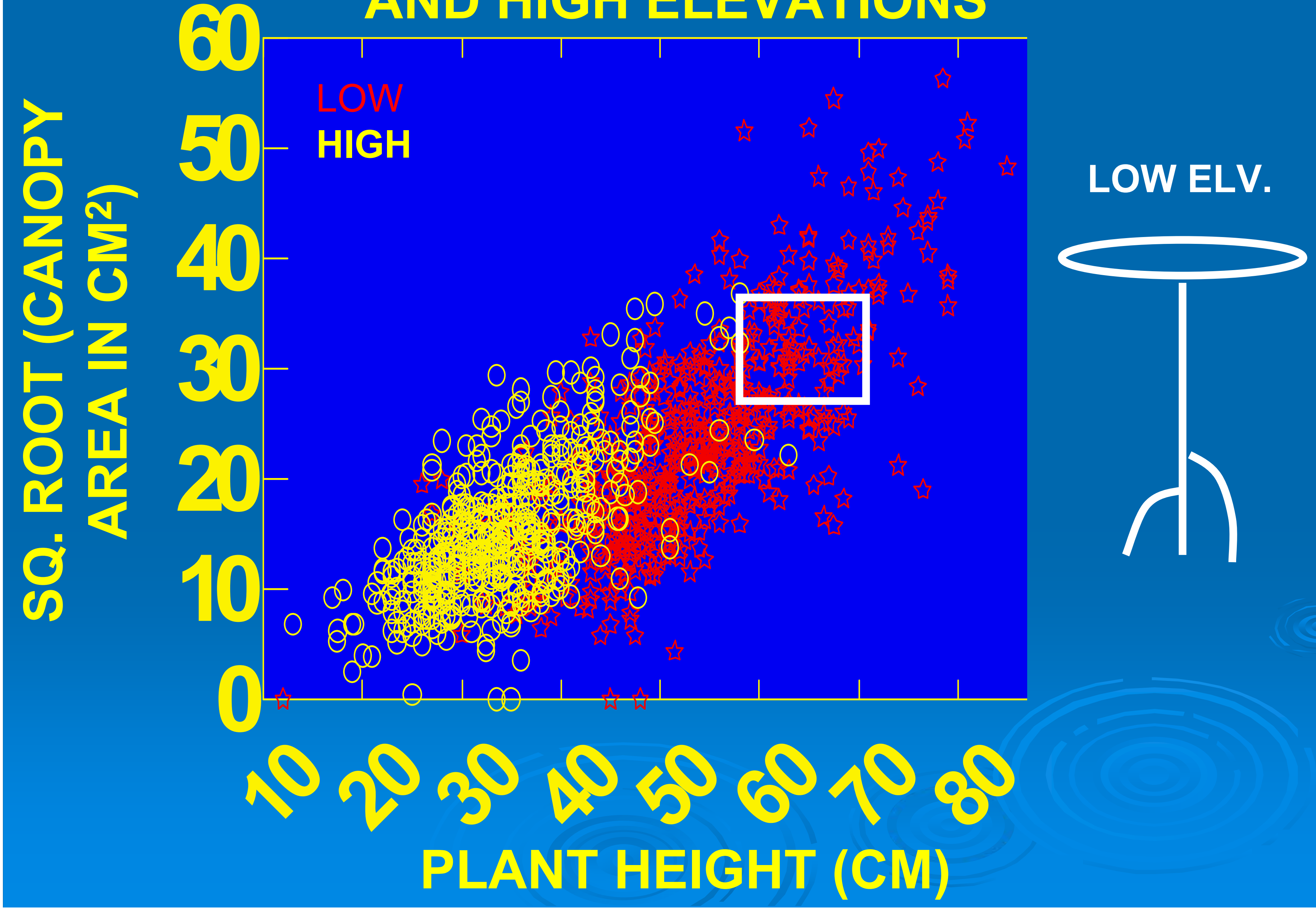
CANOPY AREA VS HEIGHT AT LOW AND HIGH ELEVATIONS (3 YRS)



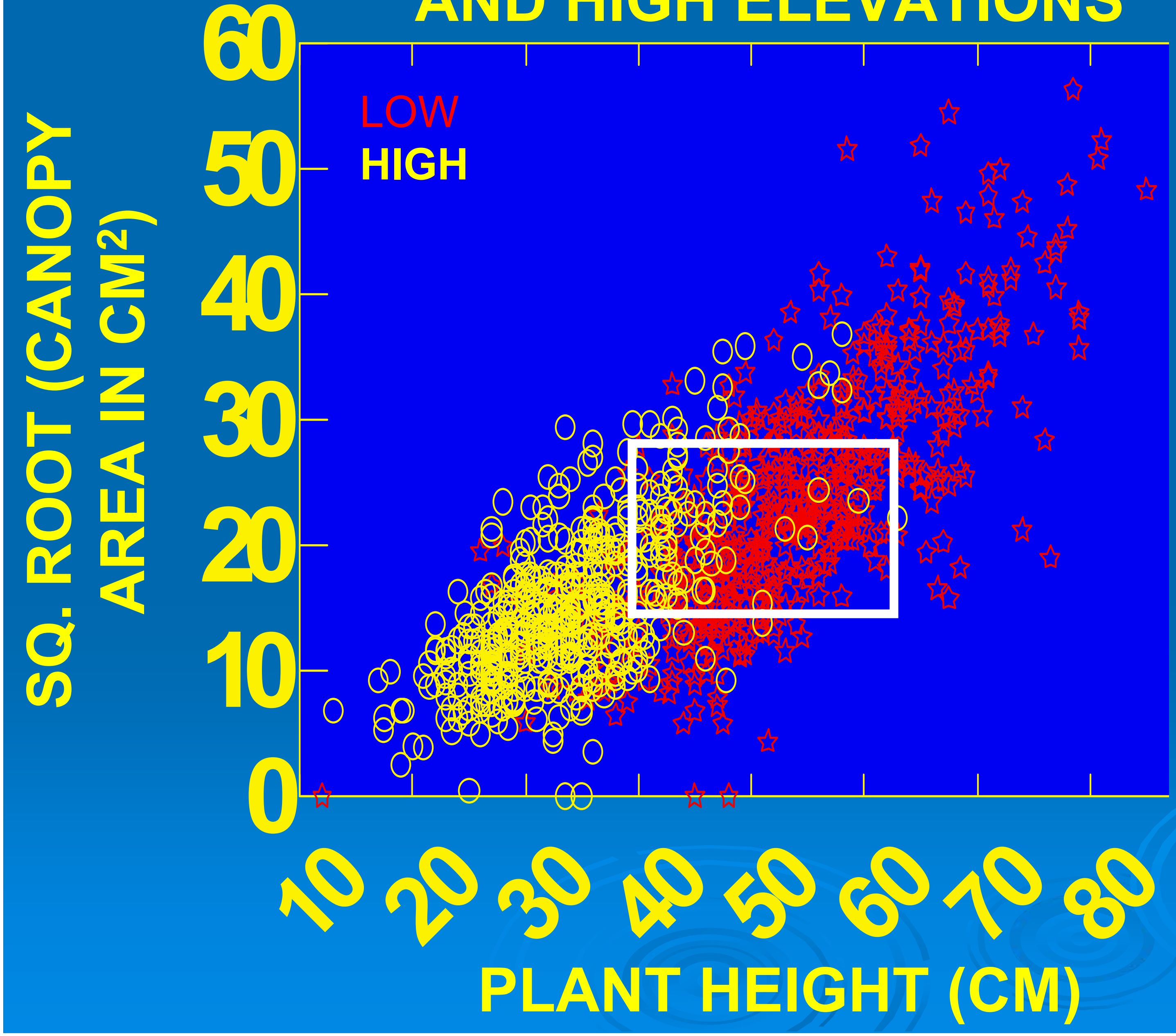
CANOPY AREA VS HEIGHT AT LOW AND HIGH ELEVATIONS



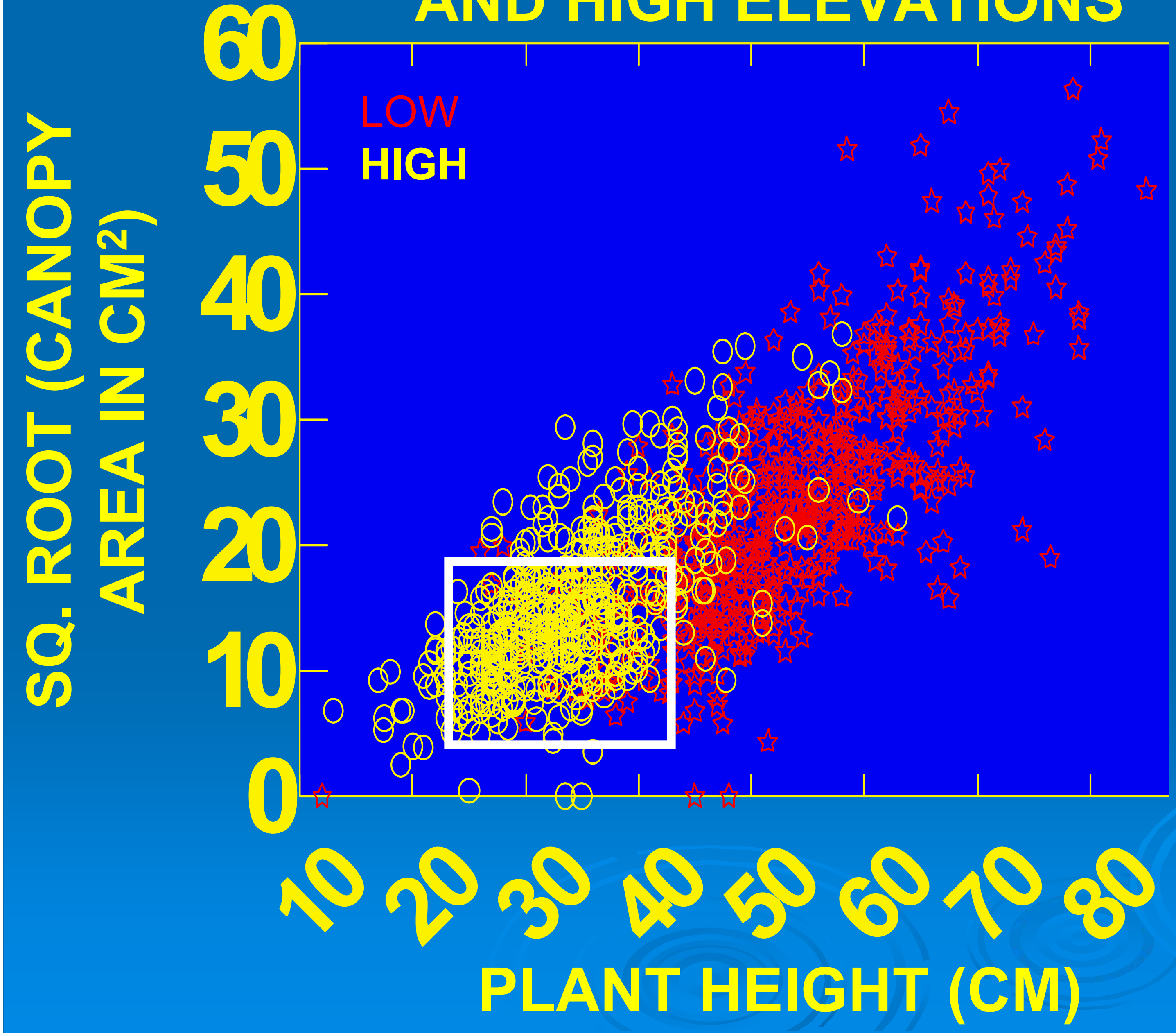
CANOPY AREA VS HEIGHT AT LOW AND HIGH ELEVATIONS

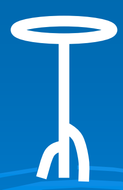


CANOPY AREA VS HEIGHT AT LOW AND HIGH ELEVATIONS

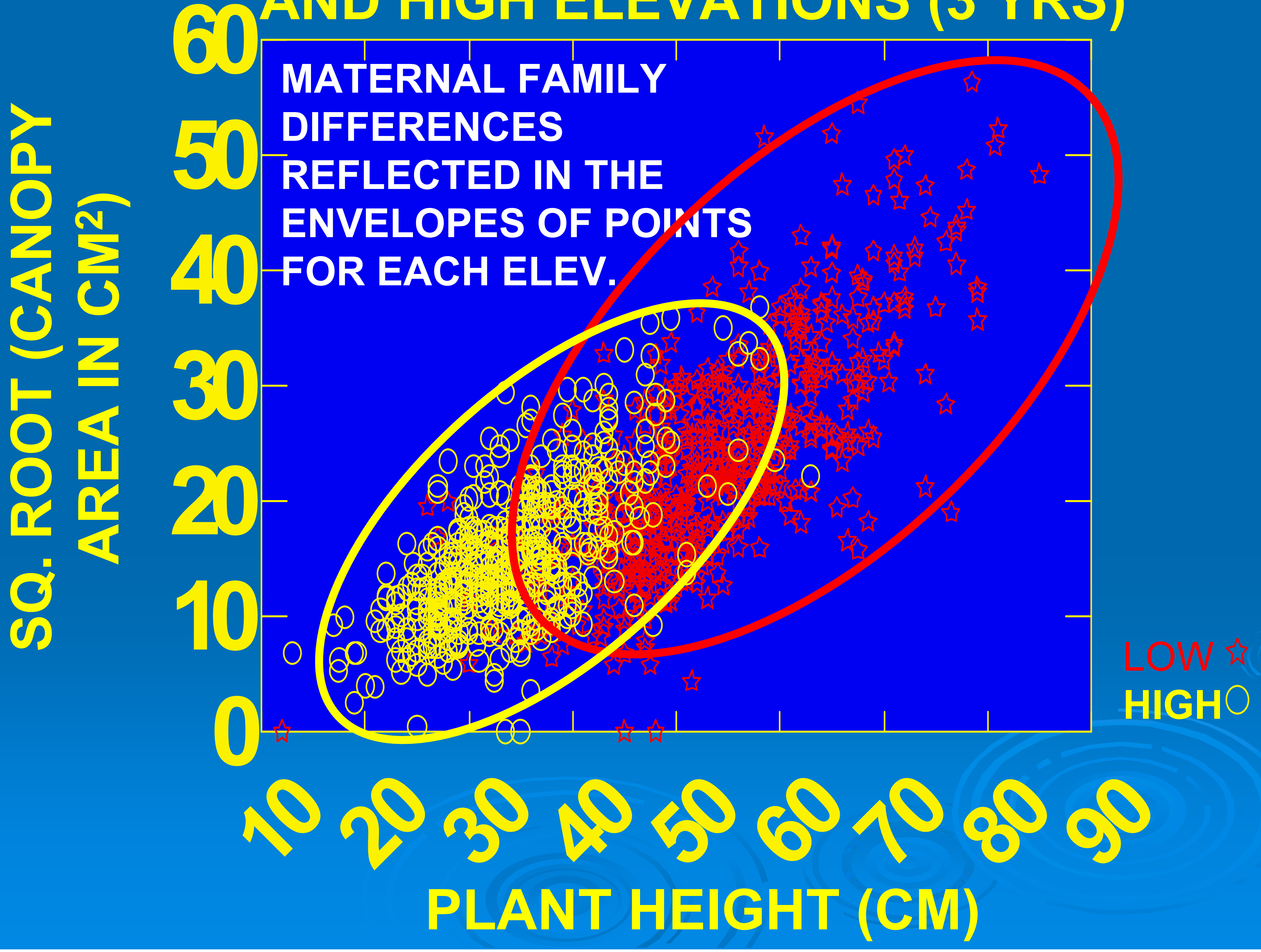


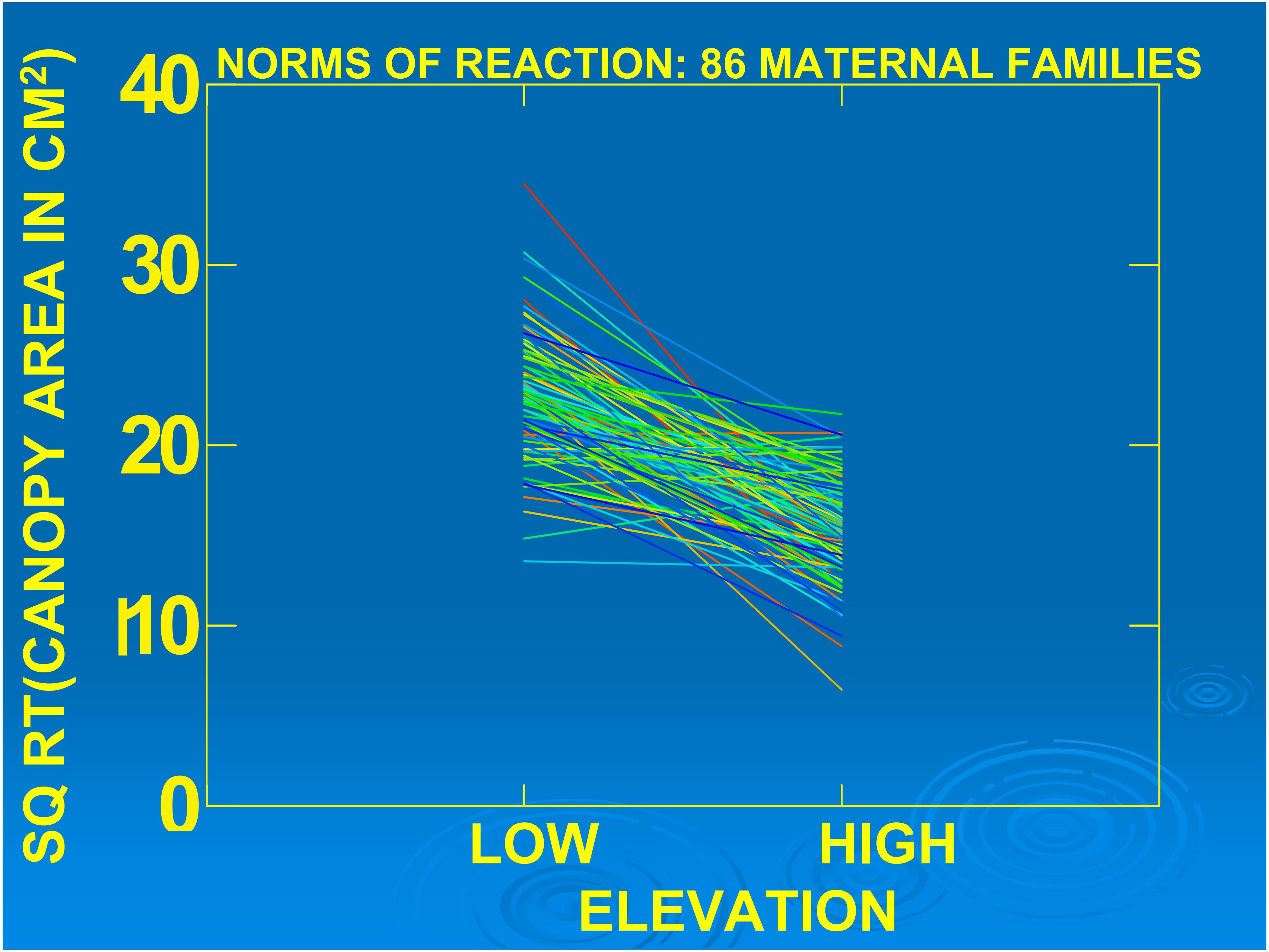
CANOPY AREA VS HEIGHT AT LOW AND HIGH ELEVATIONS



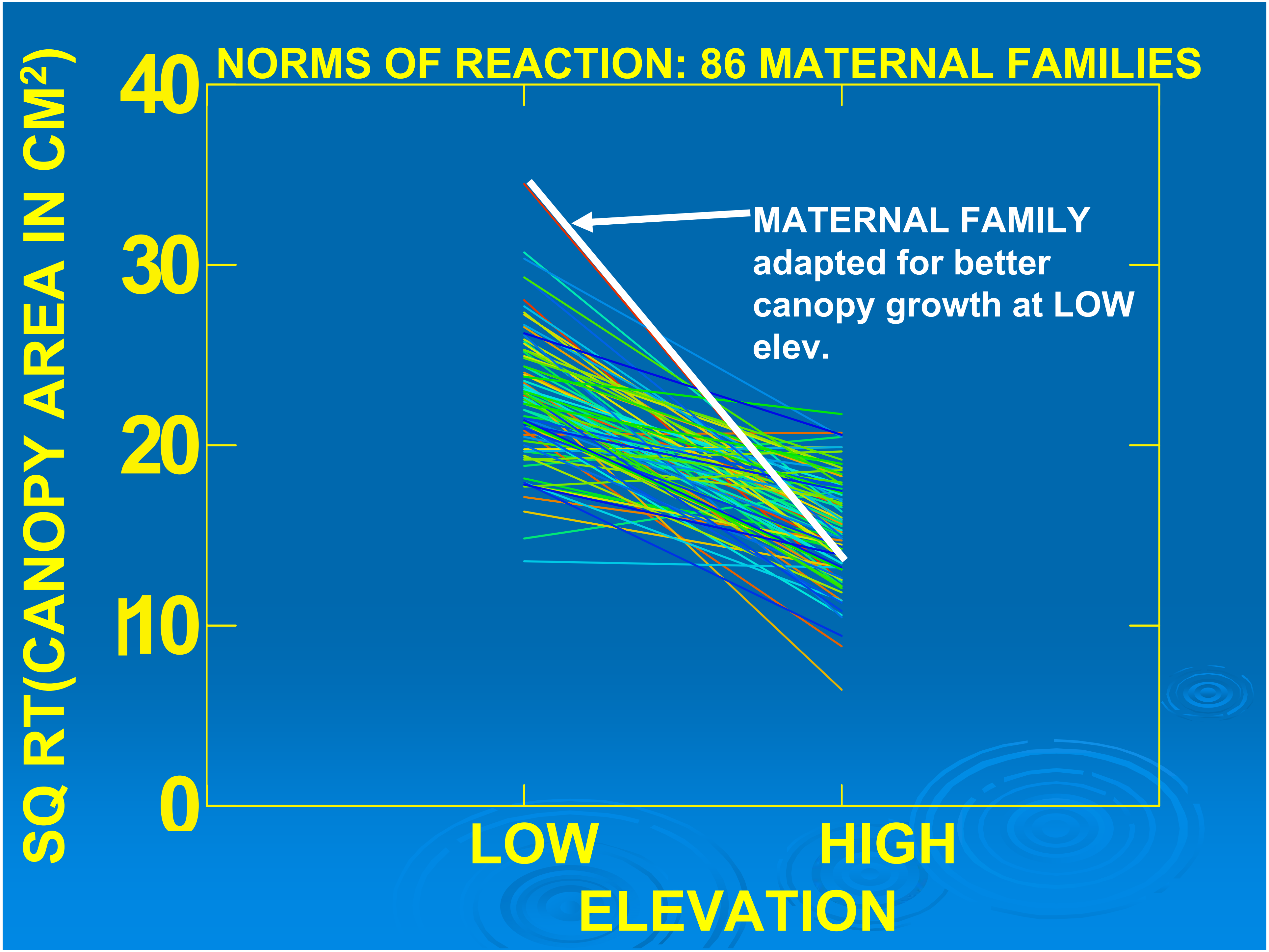


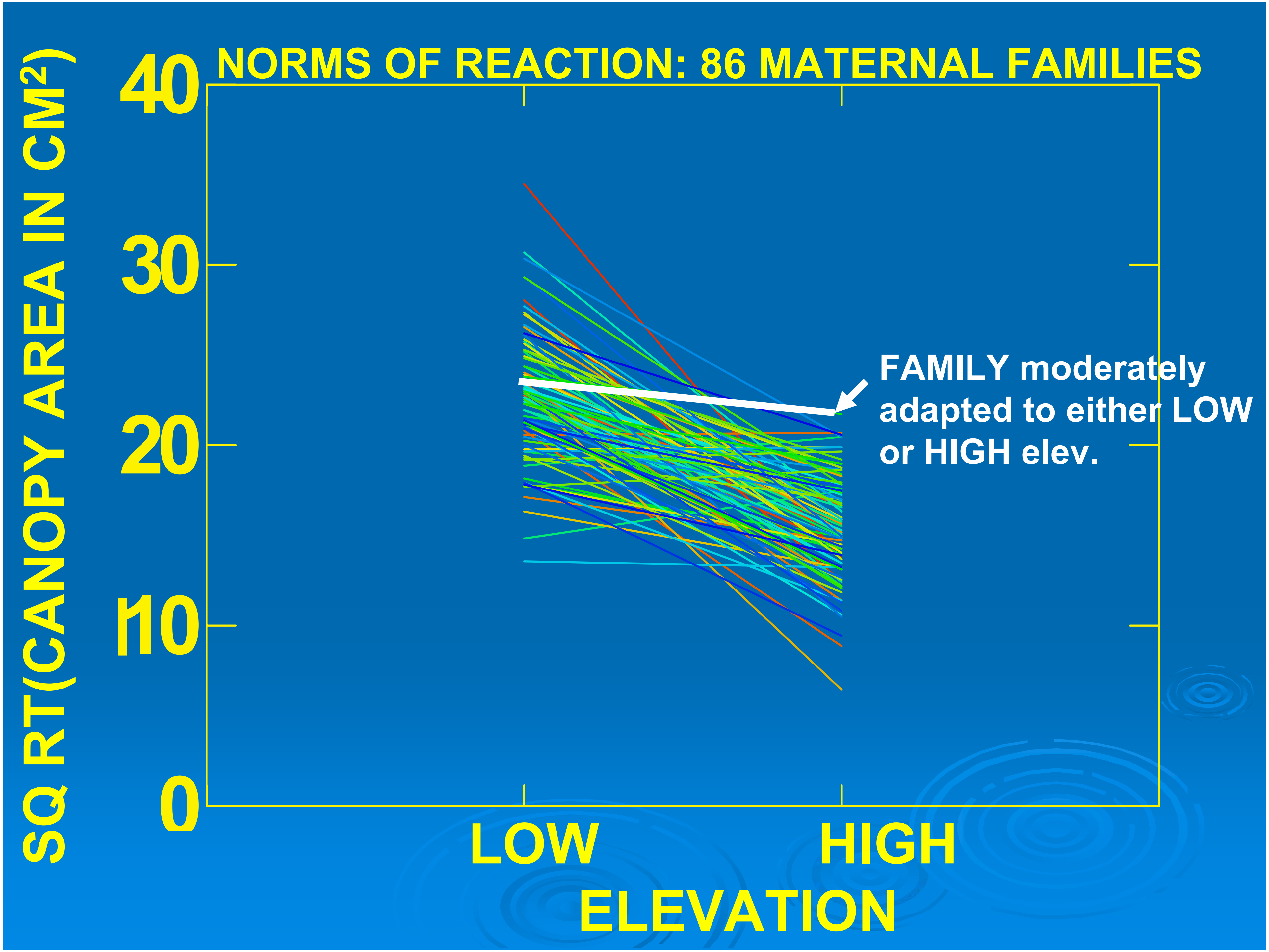
CANOPY AREA VS HEIGHT AT LOW AND HIGH ELEVATIONS (3 YRS)



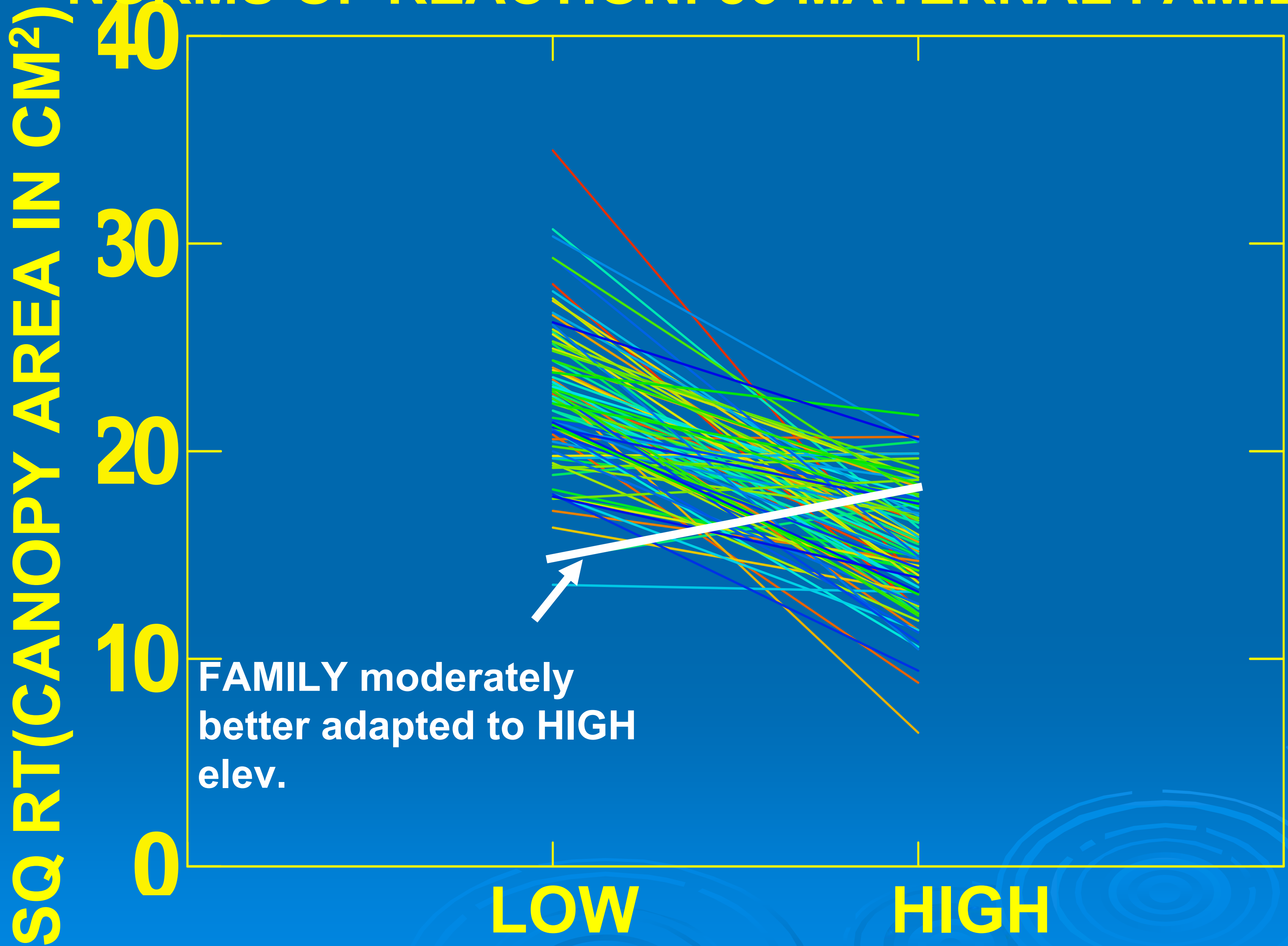


NORMS OF REACTION: 86 MATERNAL FAMILIES
SQ RT(CANOPY AREA IN CM2)
40
30
20
10
0
LOW
HIGH
ELEVATION


NORMS OF REACTION: 86 MATERNAL FAMILIES
SQ RT(CANOPY AREA IN CM²)
40
30
20
10
0
MATERNAL FAMILY
adapted for better
canopy growth at LOW
elev.
LOW
HIGH
ELEVATION

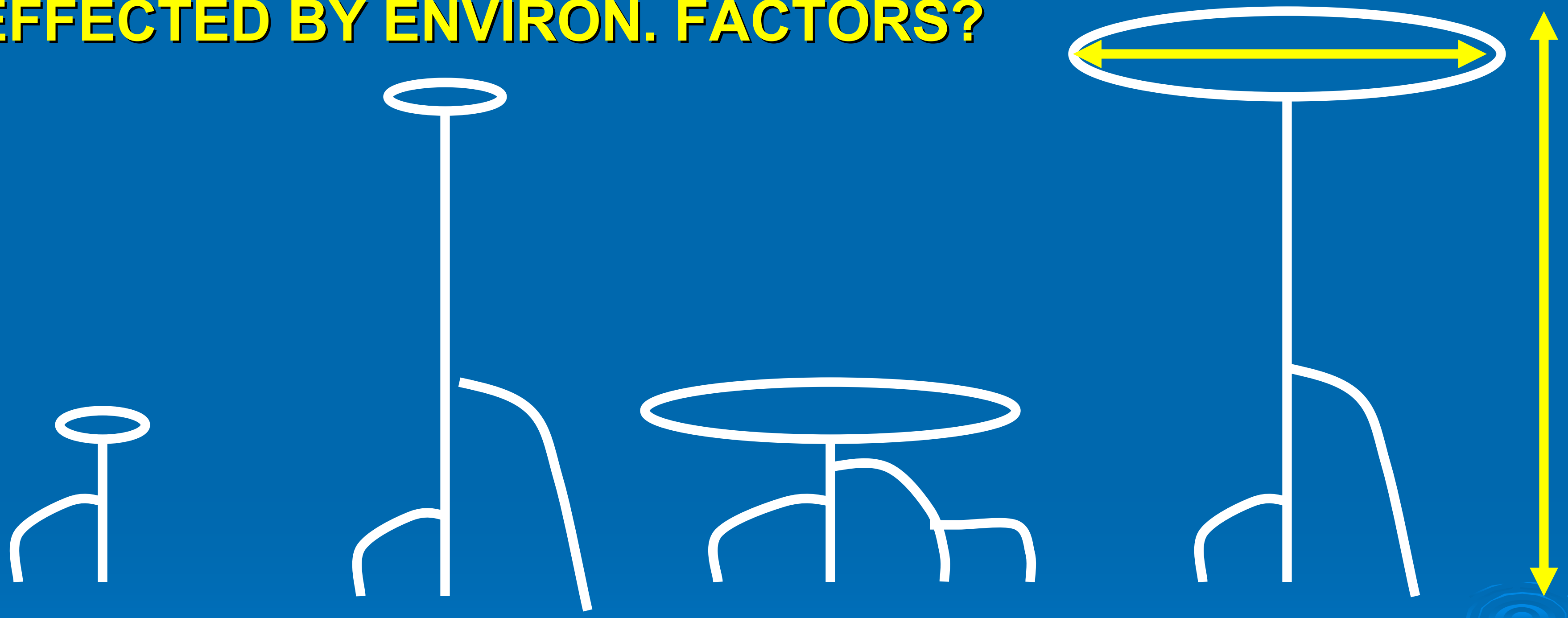

NORMS OF REACTION: 86 MATERNAL FAMILIES
SQ RT(CANOPY AREA IN CM²)
40
30
20
10
0
FAMILY moderately adapted to either LOW or HIGH elev.
LOW
HIGH
ELEVATION

NORMS OF REACTION: 86 MATERNAL FAMILIES



CANOPY AREA / HEIGHT RATIO

- **ARE THERE DIFFERENT PLANT ARCHITECTURES?**
- **DOES IT VARY BY MATERNAL GENOTYPE?**
- **EFFECTED BY ENVIRON. FACTORS?**



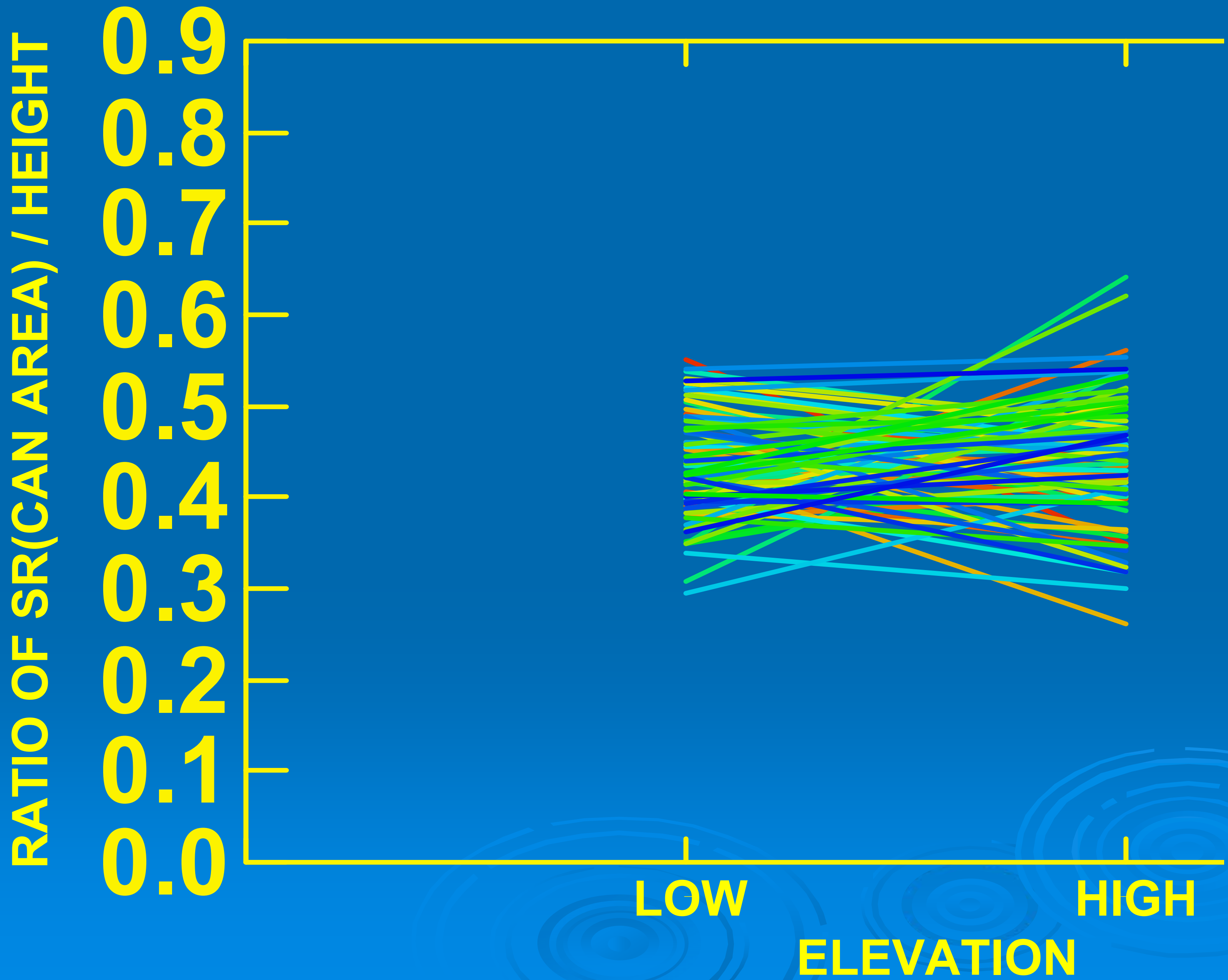
R. MANGLE PLANT ARCHITECTURE (3 YR OLD SEEDLINGS)

ANOVA SUMMARY: SR(CANOPY AREA) / HEIGHT: R^2=0.34

Source	DF	F Value	P
ELEVATION	1	2.90	0.0890
EMBAYMENT	4	0.31	0.8687
ISLAND PLANTED	4	29.84	0.0001
MAT. FAM(EMBAY.)	81	1.51	0.0036
ELV*FAM.(EMBAY.)	85	1.06	0.3518
ELEV.*ISLAND	4	15.89	0.0001
PROPLEN	1	0.15	0.6976

- EFFECT OF MATERNAL FAMILY
- EFFECT OF LOCAL ISLAND PLANTED ENVIRONMENT
- NO EFFECT OF ELEVATION
- NO COVARIANCE WITH PROPAGULE SIZE

TREE ARCHITECTURE (RATIO OF CANOPY AREA/HEIGHT)
86 FAMILIES NORMS OF REACTIONS AT LOW & HIGH ELV.



PLANT ARCHITECTURE

- MATERNAL FAMILY HAS A LARGE, SIGNIFICANT EFFECT
- ELEVATION DOES NOT
- LOCAL SEEDLING ENVIRONMENT DOES

REPRODUCTION

- BEGAN PRODUCING PROPAGULES
  - A FEW PLANTS AT YEAR 2
  - A NUMBER OF PLANTS AT YEAR 3

2007 (3 yrs old): Reproduction

Reproduction: Propagules YR 3

	Elevation	
	Low	High
% Plantings Producing Propagules	4.9%	4.6%

Reproduction: Propagules

% of Maternal Families whose seedlings Produced Propagules in 2007	**40%**
% Families. Repro. at LOW Elv. Only	**25.6%**
% Families. Repro. at HIGH Elv. Only	**20.9%**
% Families. Repro. at BOTH Elvs.	**7%**

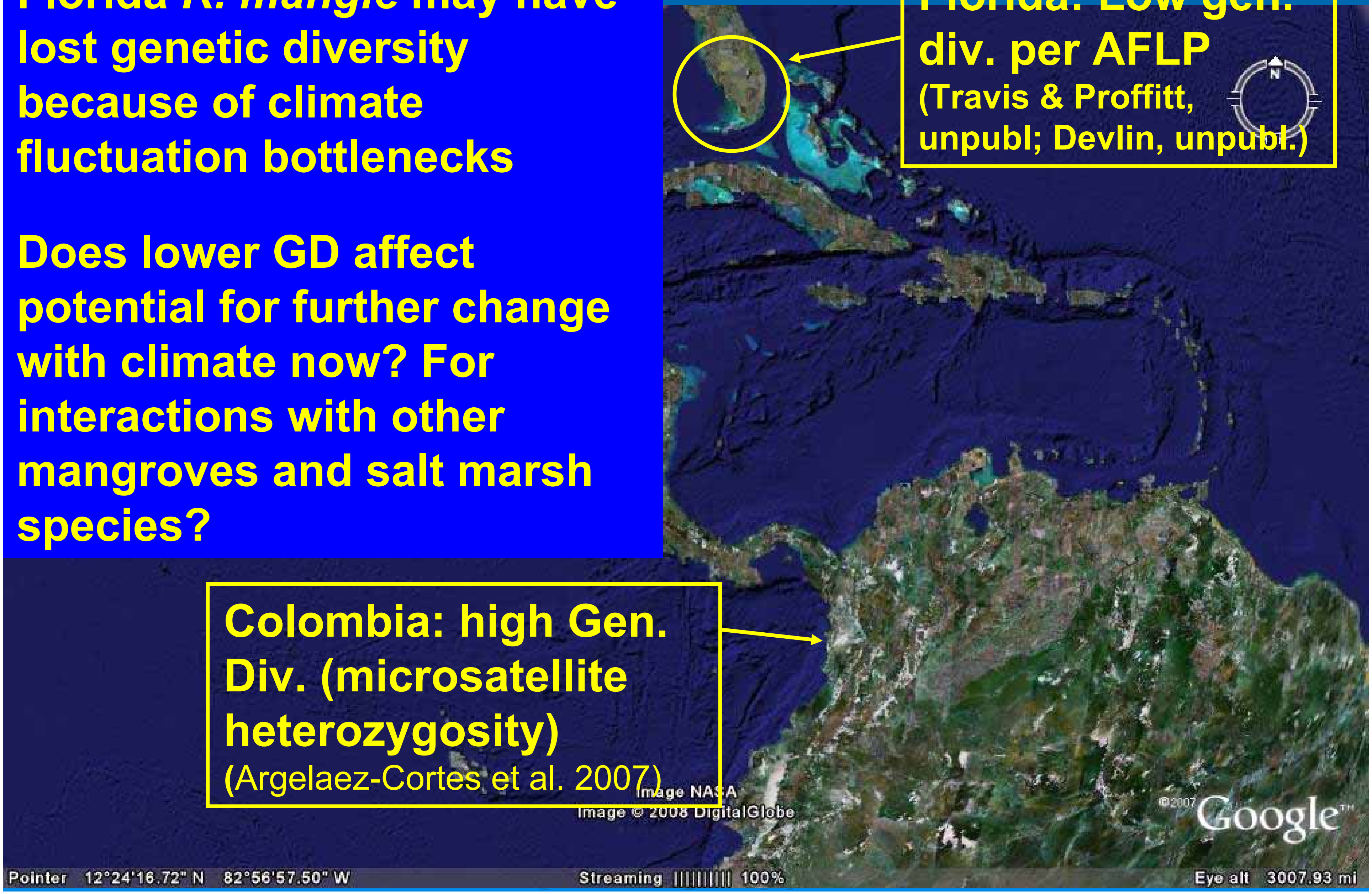
Potential for Local Adaptation Exists

- May affect the rate at which *R. mangle* (relative to other species) moves with global change
  - Interactions among species affected
- Can influence restoration success
  - Diversity of seedling parentage may be very important
- Confirm with next experiment using F_1 generation of this Tampa Bay study

Next Step 2: Red Mangrove Genetic Diversity

Florida *R. mangle* may have lost genetic diversity because of climate fluctuation bottlenecks

Does lower GD affect potential for further change with climate now? For interactions with other mangroves and salt marsh species?



Thanks

- Jordan Sanford, Katie Tiling, and Glenn Coldren for all the help in the field
- Brandt Henninsgen for promoting use of the Schultz Family Park restoration site
- Randy Runnells for help in all phases of the Tampa Bay study
- USGS for funding of the first year and for logistic help

END

Field Experiment: SUMMARY

- **Seedling height, canopy area, and "architecture" (can. Area / ht) affected by**
  - **Maternal Sibling Family and probably genotype (NEXT F_1 experiment)**
  - **Seedling environment**
    - **Island scale**
    - **Several m within island (transect) scale**
  - **Propagule size (maternal effects + genotype)**
  - **NOT maternal tree environment (embayment)**

END

MANOVA OF GROWTH AT 2007

Correlations among the 3 “growth” response variables (2007): height, trunk diameter (dbh) and canopy area

	dbh07	SR_carea
ht07	0.554507 <.0001	0.620549 <.0001
dbh07		0.490476 <.0001

MANOVA: response variables Height, Trunk Diameter, and Canopy Area all measured at 2007 (3 years)
Sig. = from Wilk's Lambda multivariate analysis

SOURCE	*SIG.*	*Wilk's Lambda*
MATERNAL TREE EMBAYMENT	NS	
ELEVATION (LOW or HIGH)	0.0001	0.427
PLANTING ISLAND (1…5)	0.0001	0.740
MATERNAL TREE GENOTYPE	0.0001	0.307
PROPAGULE LENGTH (COVAR.)	0.0001	0.955
ELEV. X ISLAND	0.0001	0.910
ELEV. X GENOTYPE	0.0002	0.577
ISLAND x GENOTYPE	0.0001	0.096
ISL X GENO X ELEV	0.9991	0.573

UNIVARIATE ANOVAS (P VALUES)

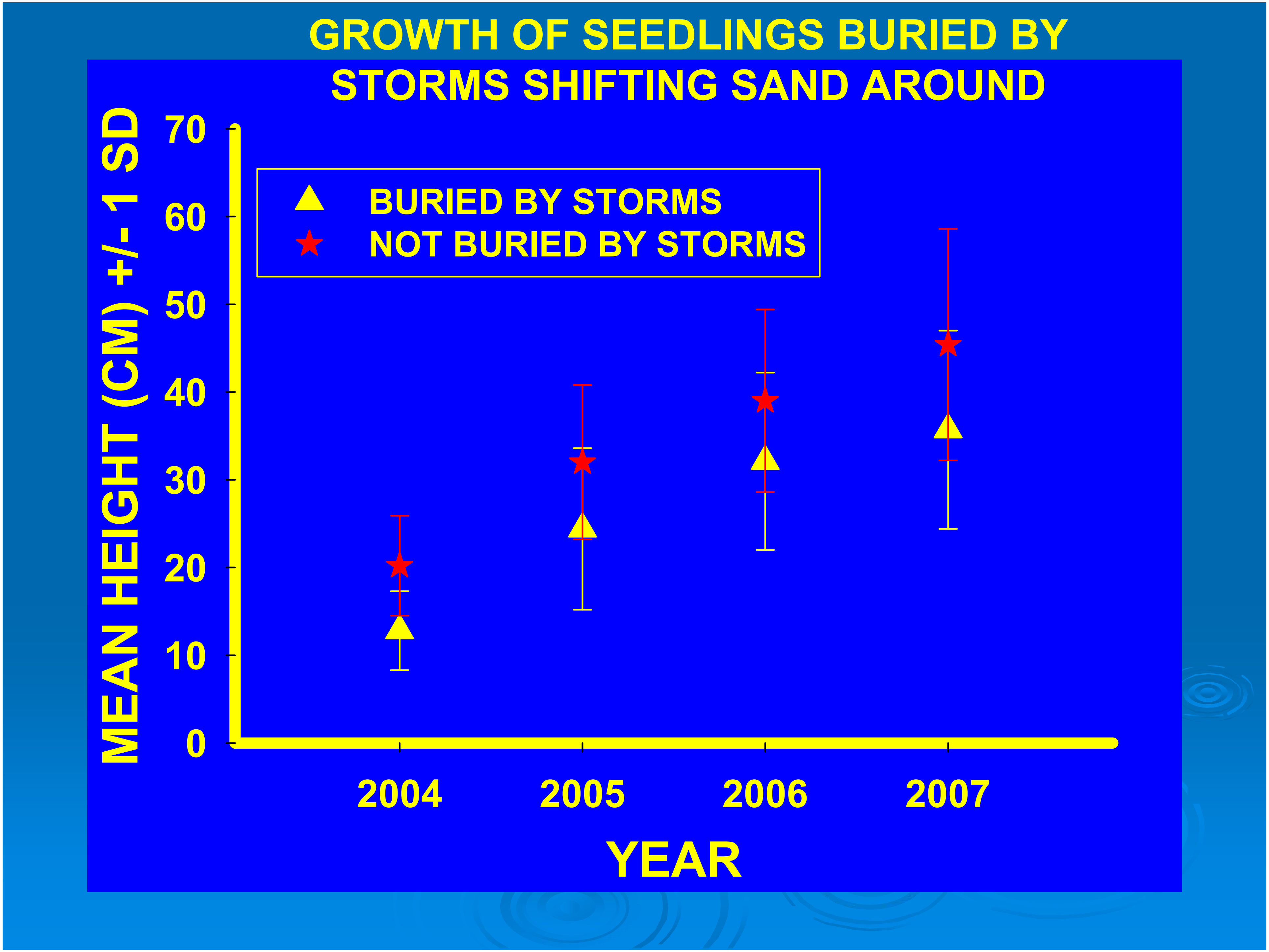
SOURCE	HT	DBH	CAN. AREA
R^2	0.863	0.822	0.691
ELEVATION	0.0001	0.0001	0.0001
PLANTING ISLAND	0.0001	0.0001	0.0001
MATERNAL GENOTYPE	0.0050	0.0580	0.0200
PROPAGULE LENGTH	0.0001	0.0002	0.1320
ELEV. X ISLAND	0.0001	0.0043	0.0001
ELEV. X GENOTYPE	0.0001	0.9954	0.9698
ISLAND x GENOTYPE	0.0001	0.0001	0.9989
ISL X GENO X ELEV.	0.2934	0.9973	0.9486

2004 STORM EFFECTS (BURIAL)

Buried

Seriously Buried

(PHOTOS 6 MONTHS POST 2004 STORMS)


GROWTH OF SEEDLINGS BURIED BY
STORMS SHIFTING SAND AROUND
MEAN HEIGHT (CM) +/- 1 SD
70
60
50
40
30
20
10
0
BURIED BY STORMS
NOT BURIED BY STORMS
2004
2005
2006
2007
YEAR

Spartina alterniflora (smooth cordgrass) and *Rhizophora mangle* (red mangrove)

- Both are foundation species at diff. latitudes
- Interact with one another at transition latitudes (in part of Florida peninsula)

Spartina alterniflora salt marsh

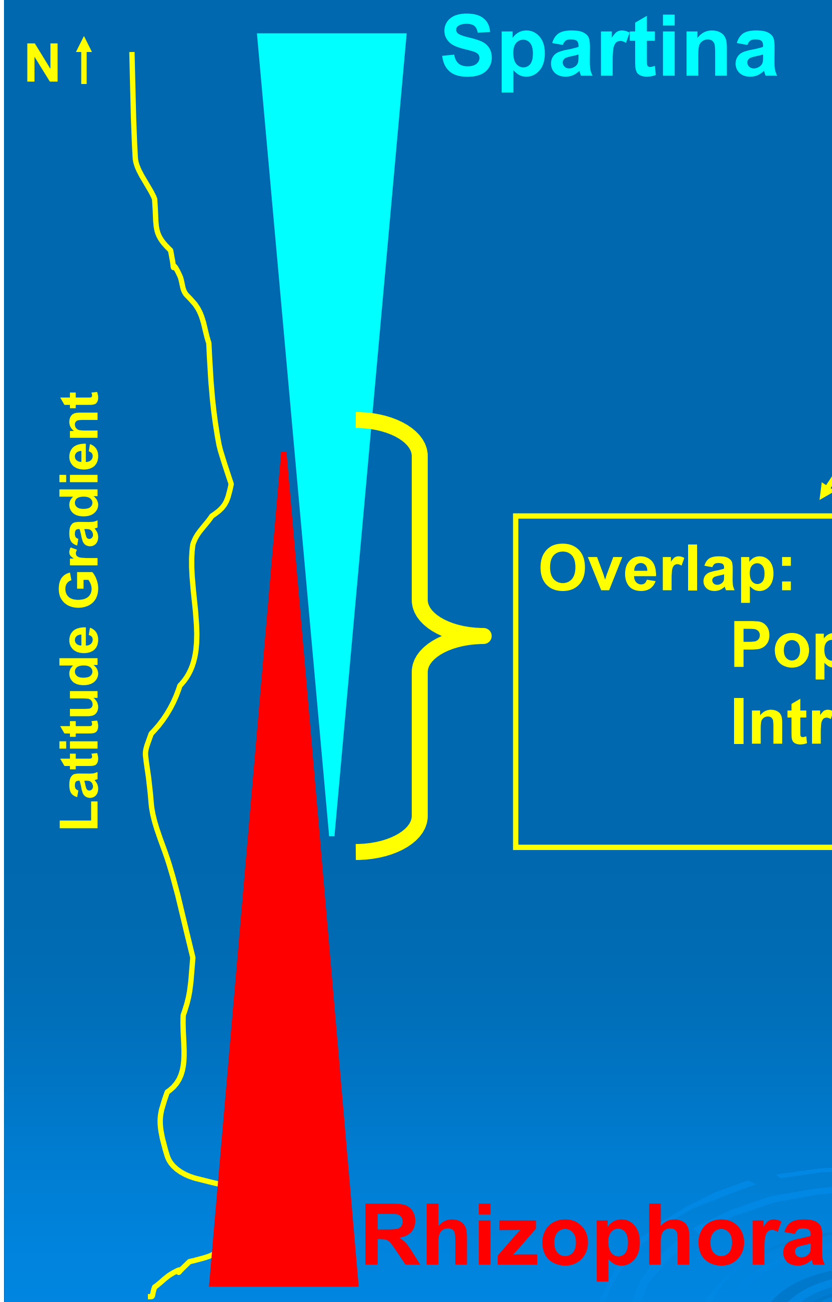
Rhizophora mangle (red mangrove)

- Foundation species in tropics & subtropics
- Low – mid intertidal
- Viviparous (seedling propagules live on maternal trees for 4-6 mo.)
- Evergreen – continuous growth & leaf production

species comparisons

- *Spartina*
- clonal grass
- clonal genets may be very long-lived but individual ramets are not
- sexual: near-obligate outcrosser
- Not shade tolerant

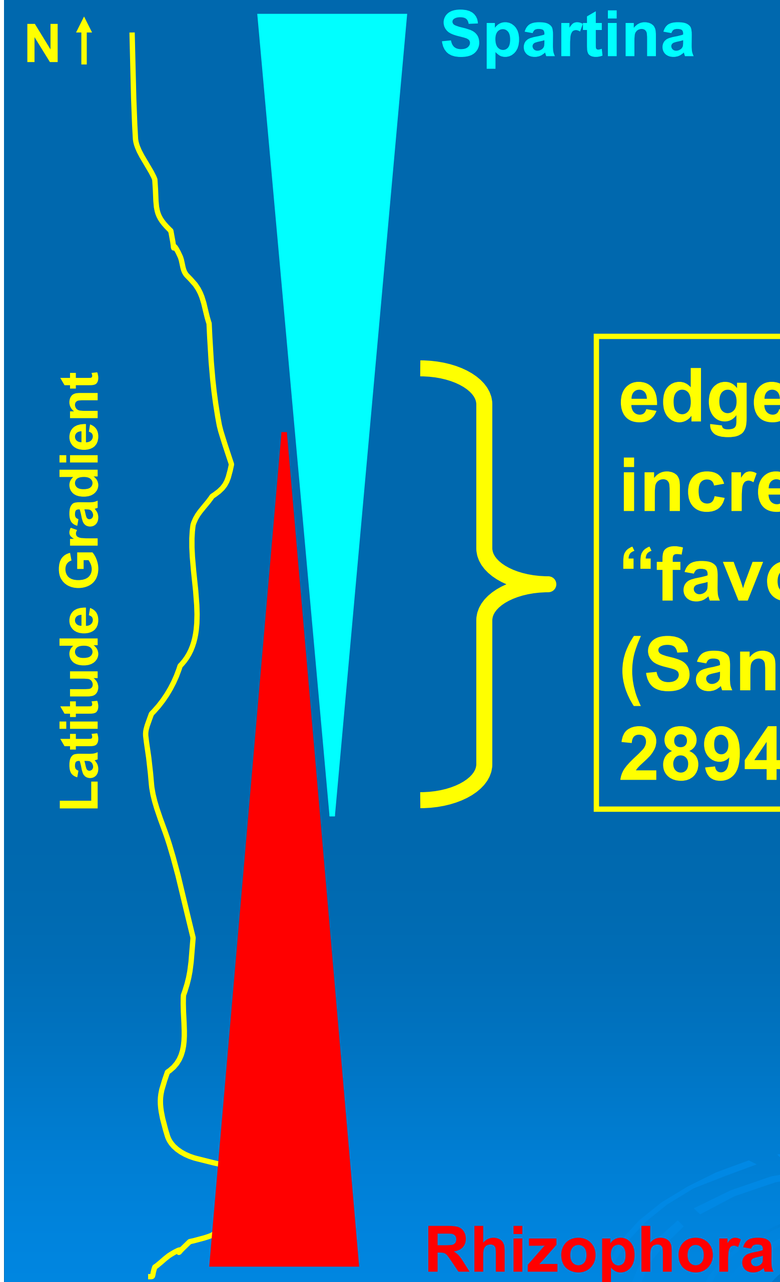
- *Rhizophora*
- tree (non-clonal although does make new trunks & canopy by iteration
- fairly long lived (maybe 70-100 years?)
- sexual: highly selfing in many estuaries; but, this varies from 0-33% outcrossing
- some degree of shade tolerance



Influenced strongly by global change
- Position
- Size ?

Overlap:
- Population process of both species
- Intra-and-inter-species interactions

Ecological processes (productivity, etc.) & habitat use affected



edge of species' ranges is area of increased accumulation of "favorable adaptations" at the (Sanford et al. 2006. Ecol. 87:2882-2894 for *Uca*)

salt marsh & mangrove at latitudes where they dominate

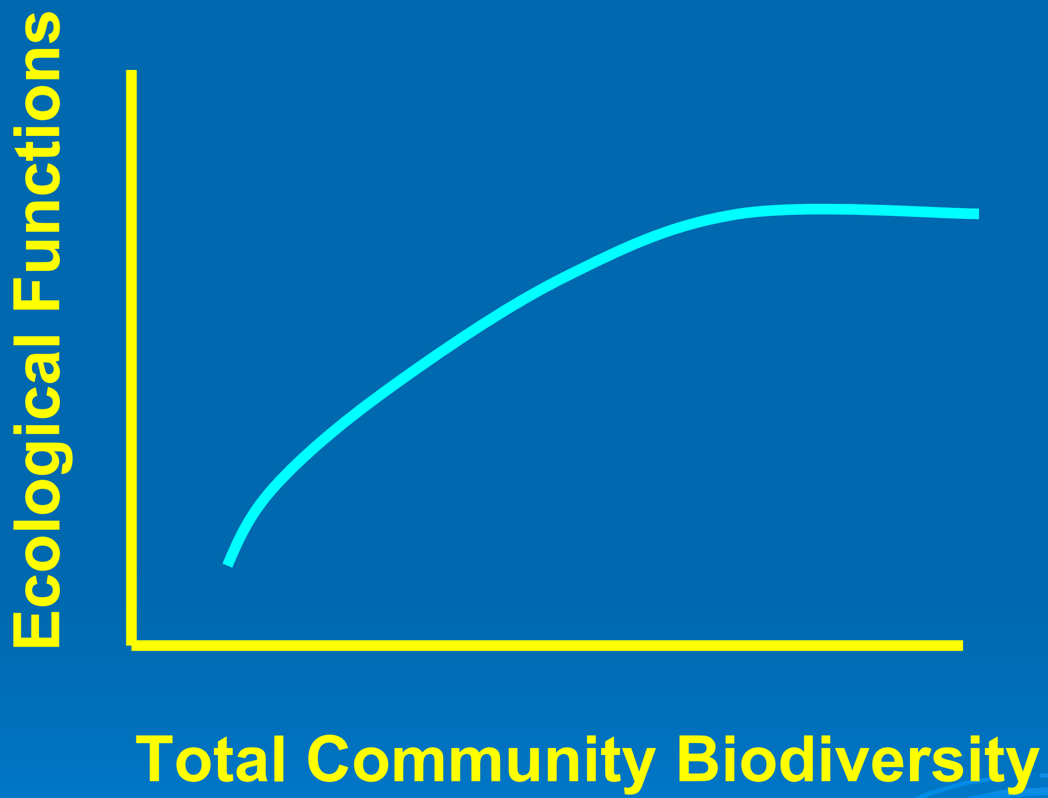
Mangrove “islands” within a salt marsh “matrix”
Merritt Island National Wildlife Refuge

Merritt Island National Wildlife Refuge Banana River (Kennedy Space Center)

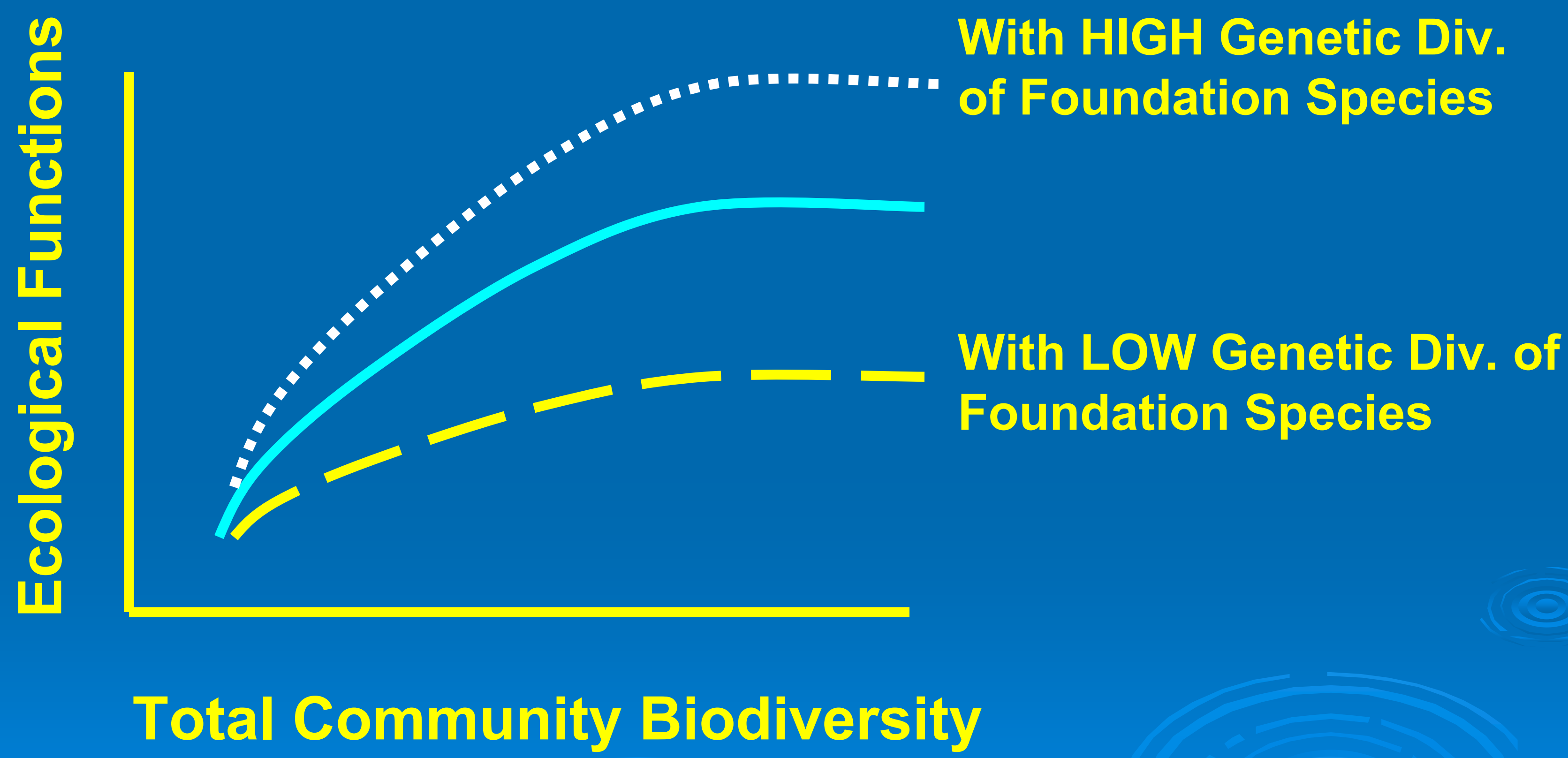
Population & Genetic Ecology

- Essential to understanding change with climate and change due to human manipulations
- Critical to understand in Foundation Species because affects so many other species

WHY IS DIVERSITY OF FOUNDATION SPECIES IMPORTANT?



WHY IS DIVERSITY OF FOUNDATION SPECIES IMPORTANT?



Knowledge Bases

- ***Spartina alterniflora*: many studies**
- ***Rhizophora mangle*: few studies**

Field Experiment
Spartina alterniflora
genetic ecology

***Spartina alterniflora* SALT MARSH FOUNDATION SPECIES GENETIC ECOLOGY: Genotypes vary:**

- **In morphology and architecture (Proffitt et al. 2003)**
- **In ecological effects**
  - **On other species (competition and facilitation). Proffitt et al. 2005)**
  - **Intra-specific competition w/ other genets of *Spartina* (Proffitt & Travis in review)**
- **In genetic and genotypic structure among marshes and over latitudinal gradient (Travis et al. 2003).**

Premises: With Climate Change

- Mangroves will have to adapt to changing
  - Physical environmental conditions
  - Biotic interactions
- Overlap of temperate & tropic foundation species may be area of enhanced selection for traits needed with climate change
- Requires
  - Phenotypic Plasticity
  - Genetic variability
  - Genetic component of plasticity x environment changes (evolution *for* plasticity)

Related Question

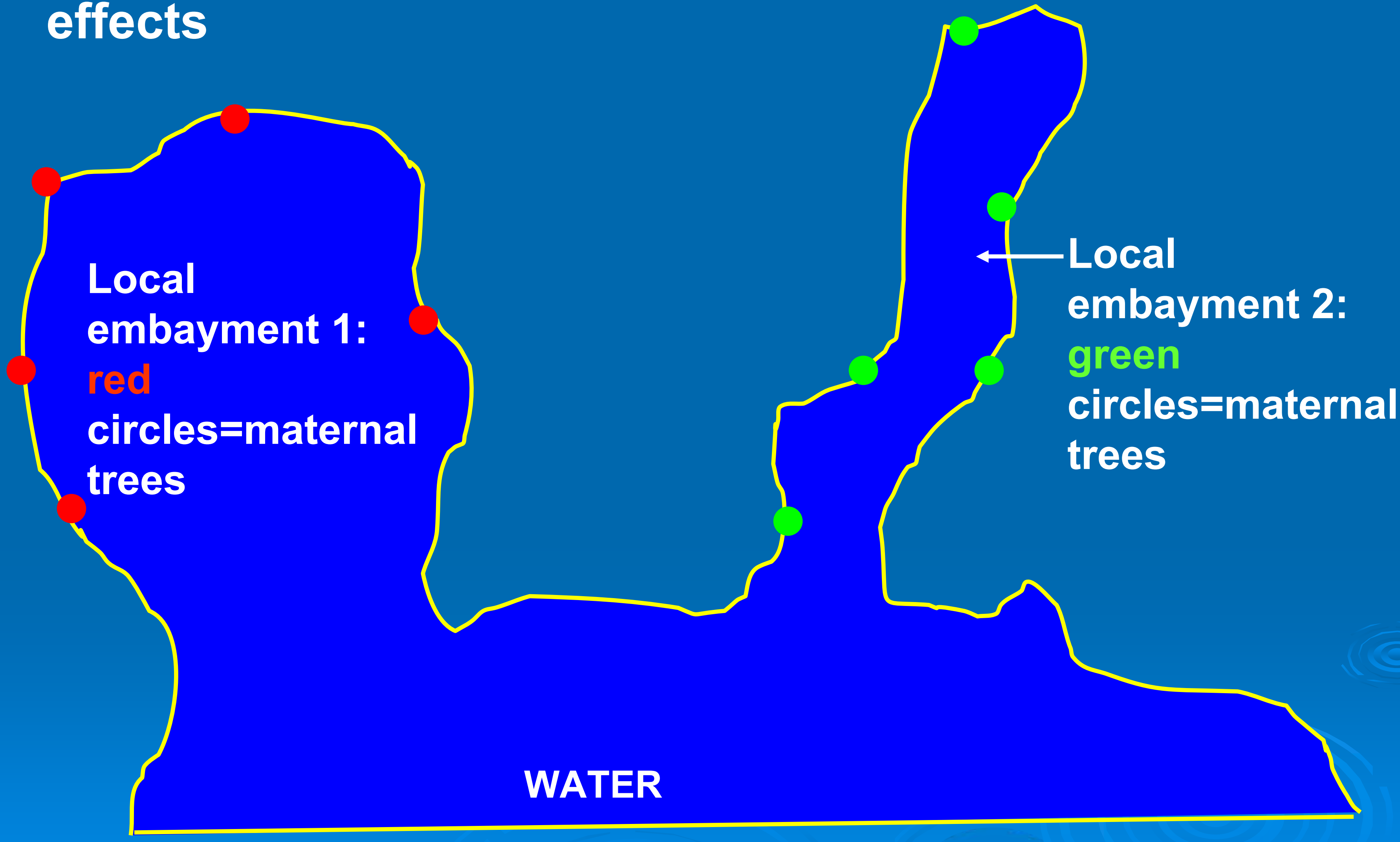
- Restoration (i.e., the "Northern Estuaries" of Everglades Restoration)
  - Are there different requirements because of
    - Proximity of subtropical / temperate biogeographical limits
    - Changes in climate (and factors related to climate, like hurricane frequency, etc.)

Needed Studies

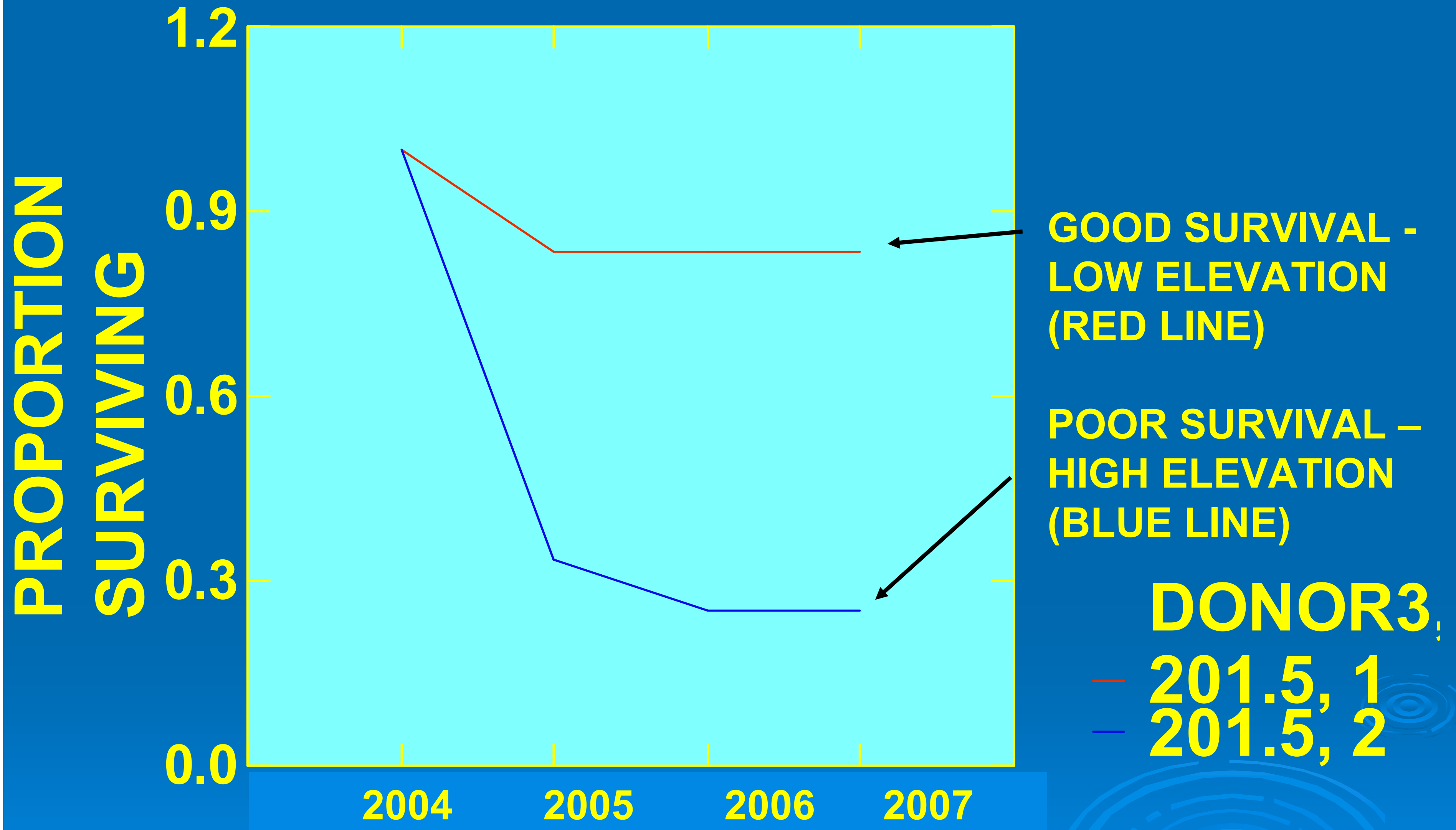
- **Genetic variability and gene flow**
- **Outcrossing and inbreeding rates**
- **Experiments addressing:**
  - **What are the important physical and biotic stressors (and resources)?**
  - **Is there potential for local adaptation?**
  - **What affects colonization, dispersal, and recruitment of seedlings/saplings into canopy?**
  - **Interactions with other foundation species (e.g., salt marsh *Spartina alterniflora*)**

R. mangle genetic diversity ?

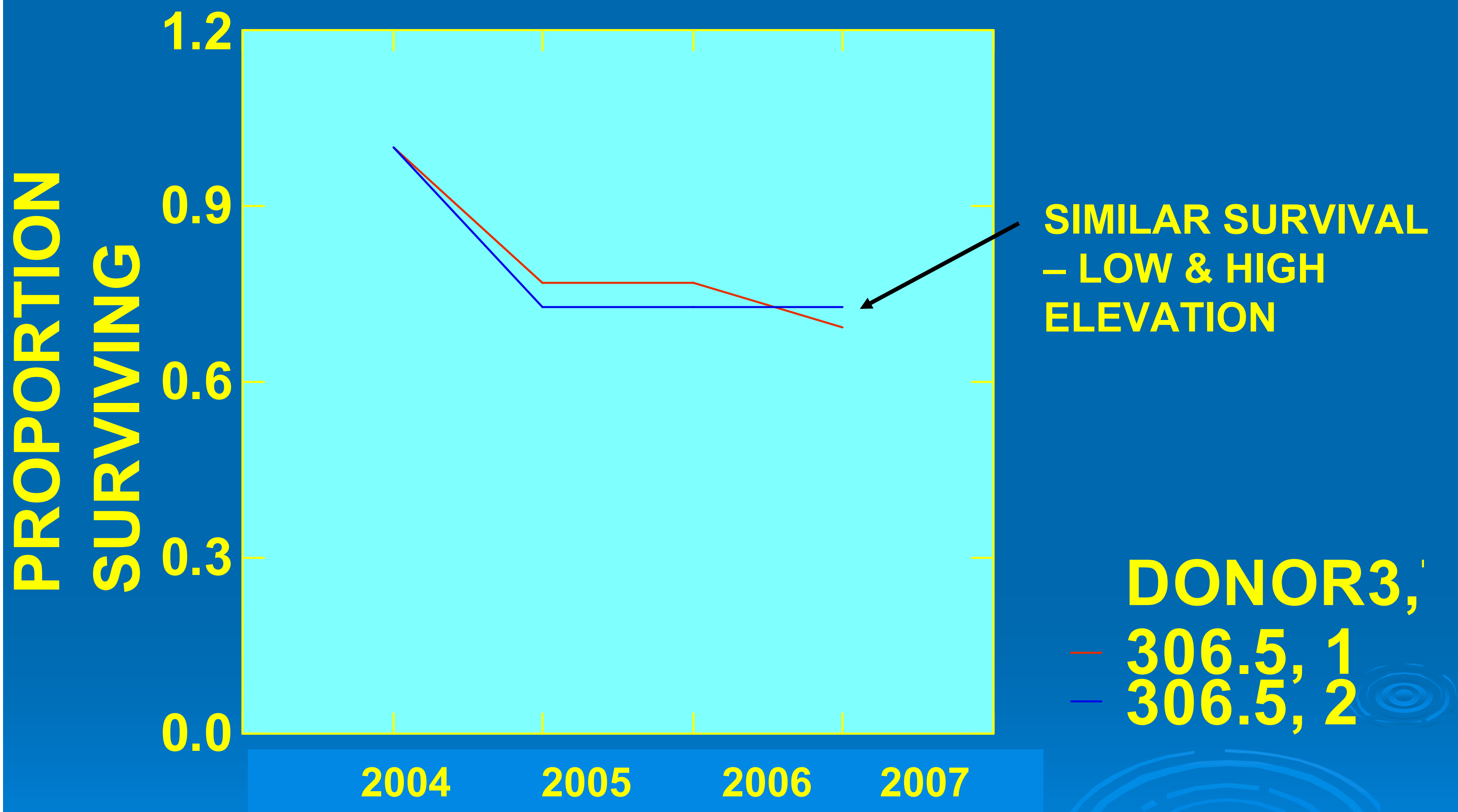
Account for SITE effects (local embayment environmental conditions) DONOR TREE (maternal families) WITHIN SITE effects



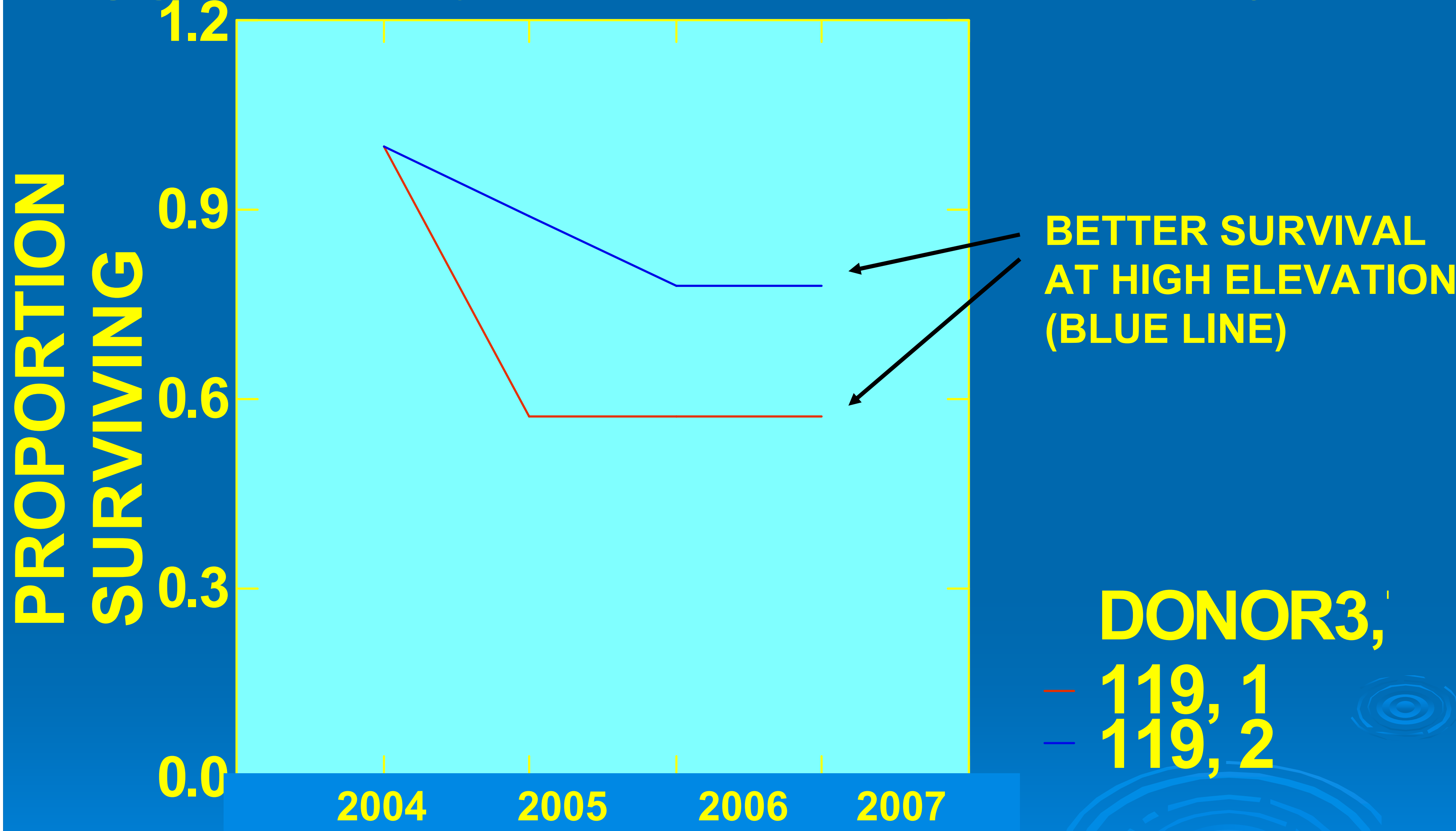
SURVIVAL of MATERNAL FAMILY 201.5



SURVIVAL of MATERNAL FAMILY 306.5



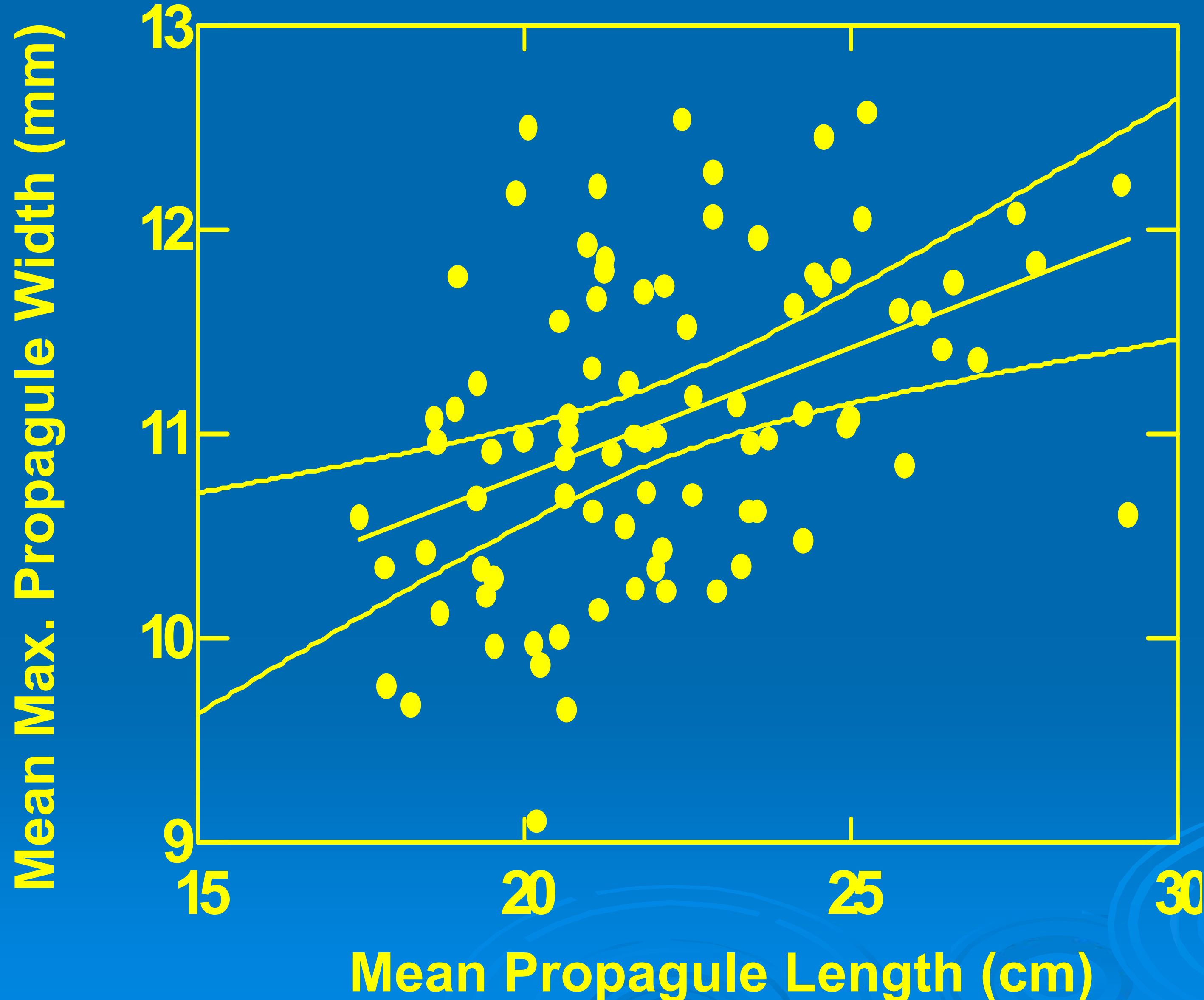
SURVIVAL of MATERNAL FAMILY 119



Propagule Length vs Max. Width

- Are propagule size measurements correlated?

Propagule Length vs Width (Family Means)



Correlation:
$r = 0.432$
$P < 0.0001$

Does Propagule Size Differ

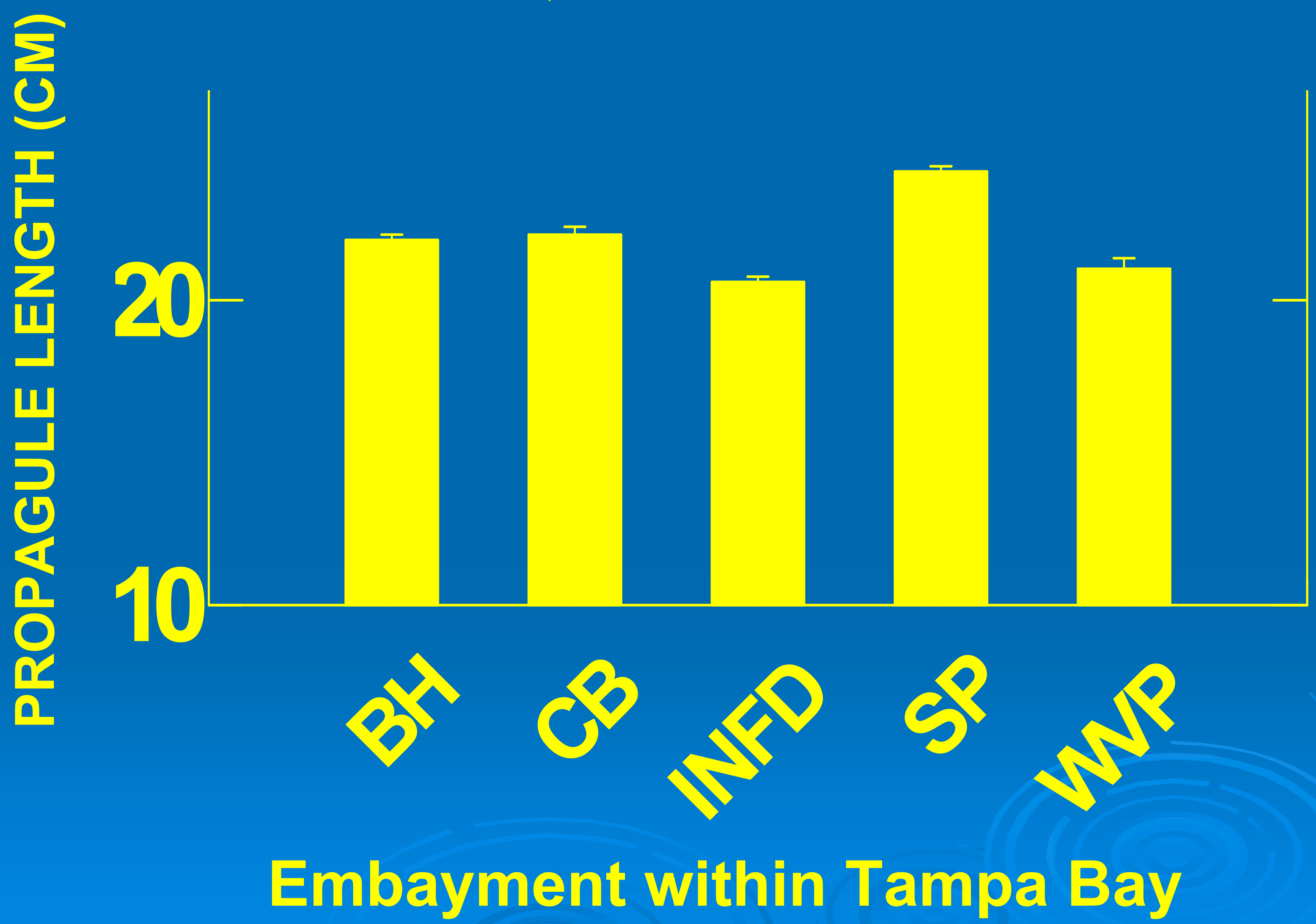
- Among Embayments (sets of maternal trees from same general location)
- Among maternal trees within embayment

ANOVA Response variable: Propagule Length

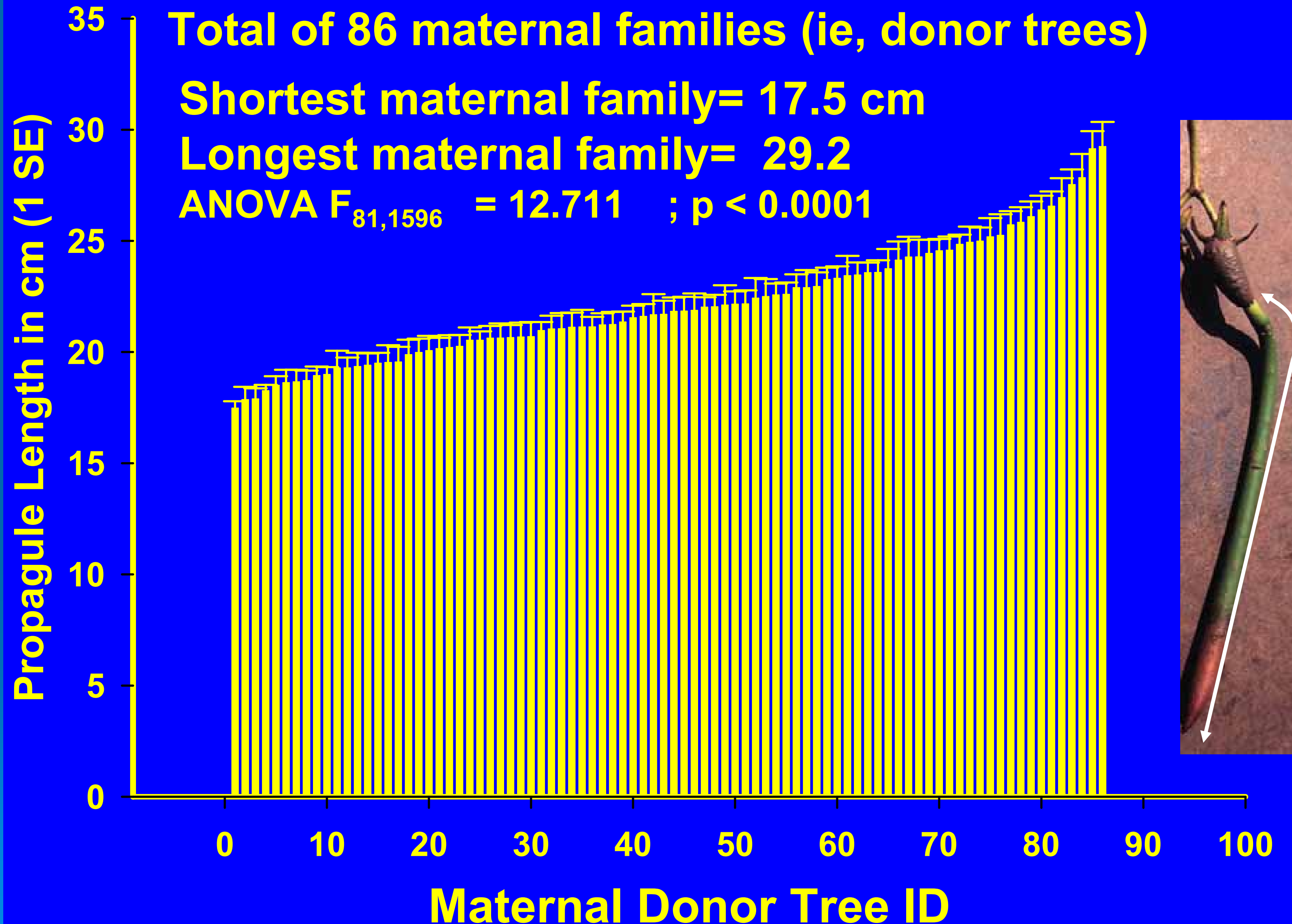
$R^2 = 0.46$

Source	*p*
Embayment	0.0001
Maternal Family(Embay.)	0.0001

PROPAGULE LENGTH by Embayment
ANOVA $F_{4,1596} = 85.411$; $p<0.0001$



Rhizophora propagule length (mean & SE by donor tree)

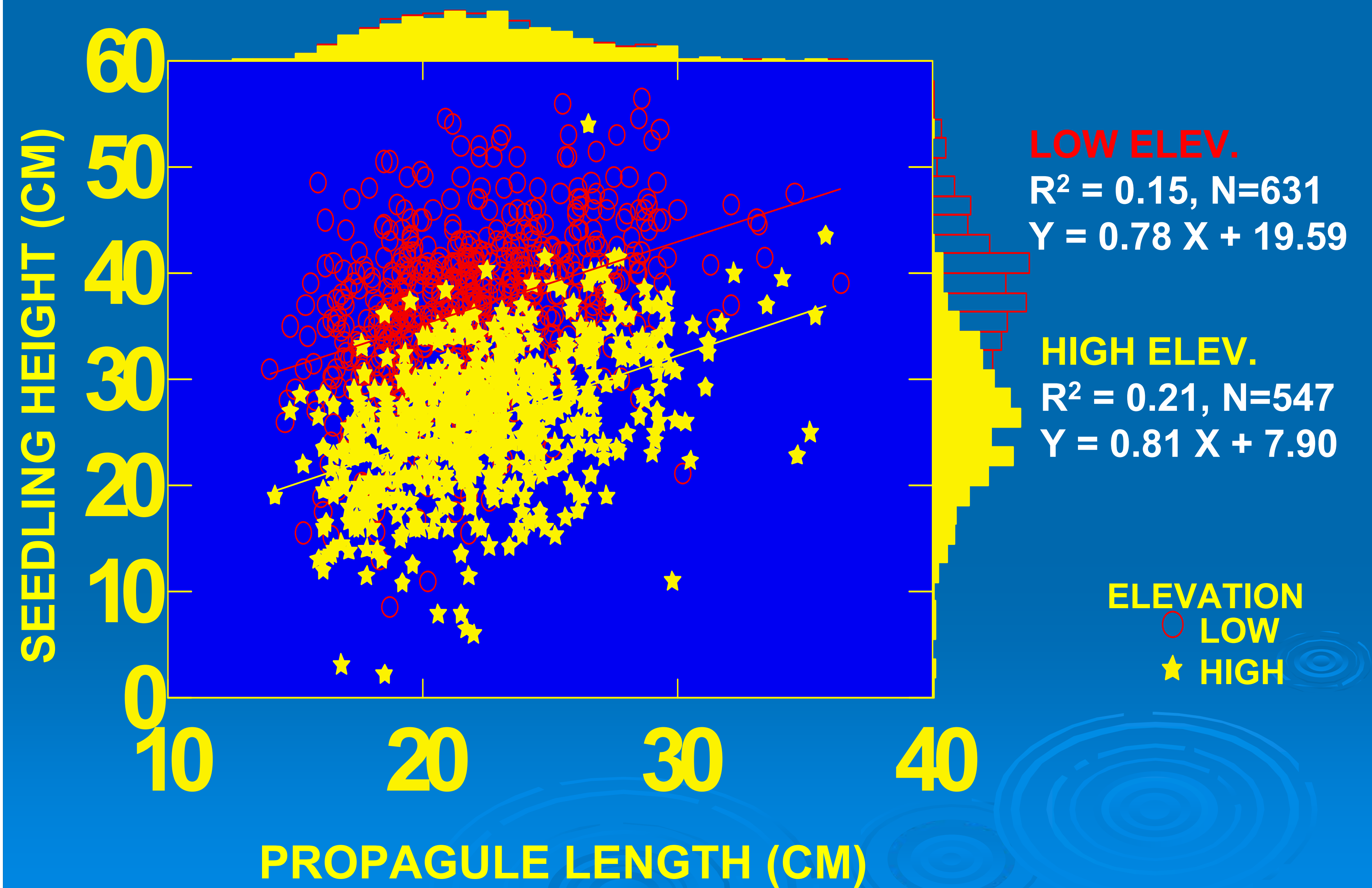


IS SEEDLING HEIGHT AFFECTED BY PROPAGULE SIZE?

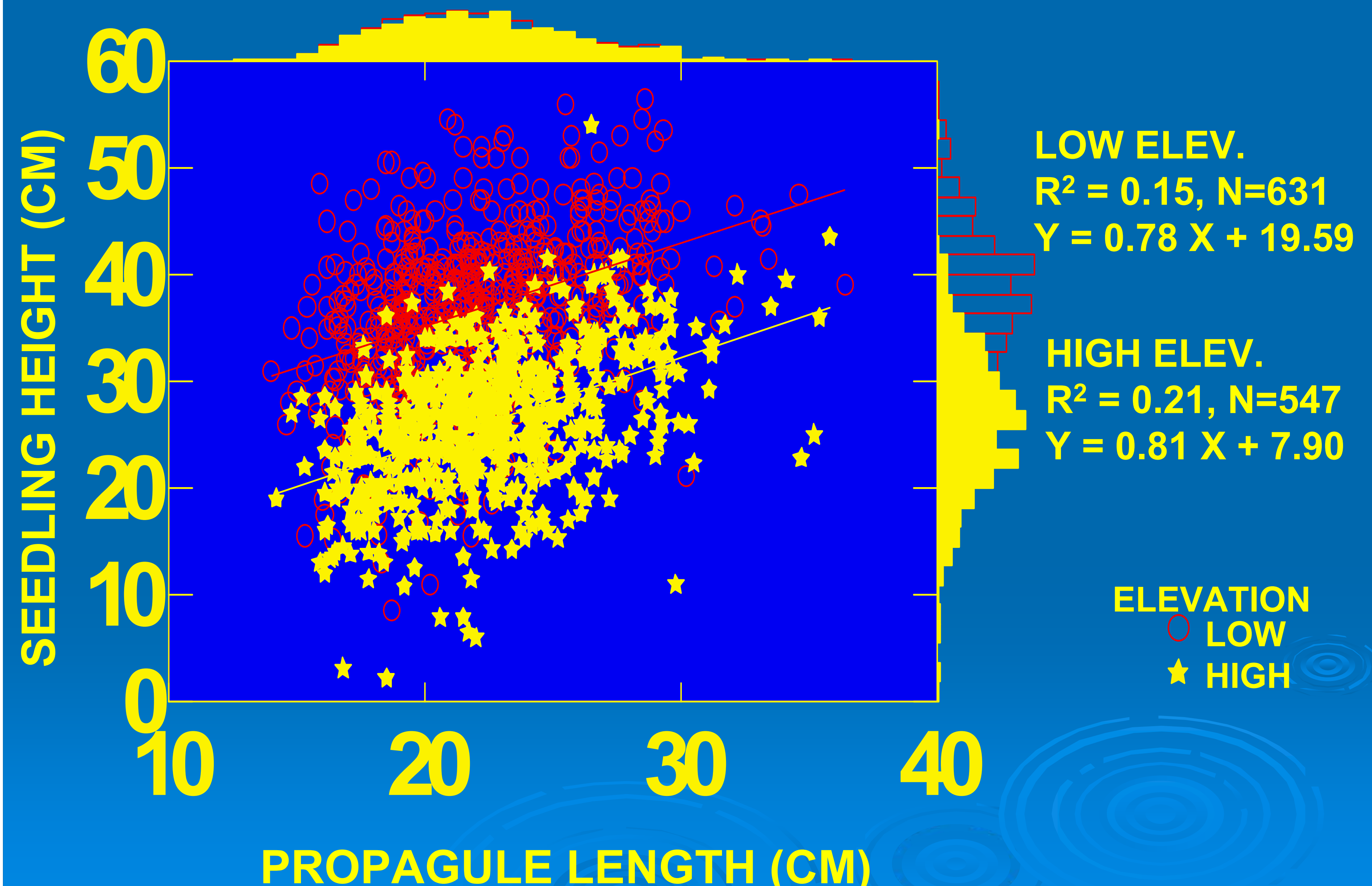
Summay of Linear Regression Results:

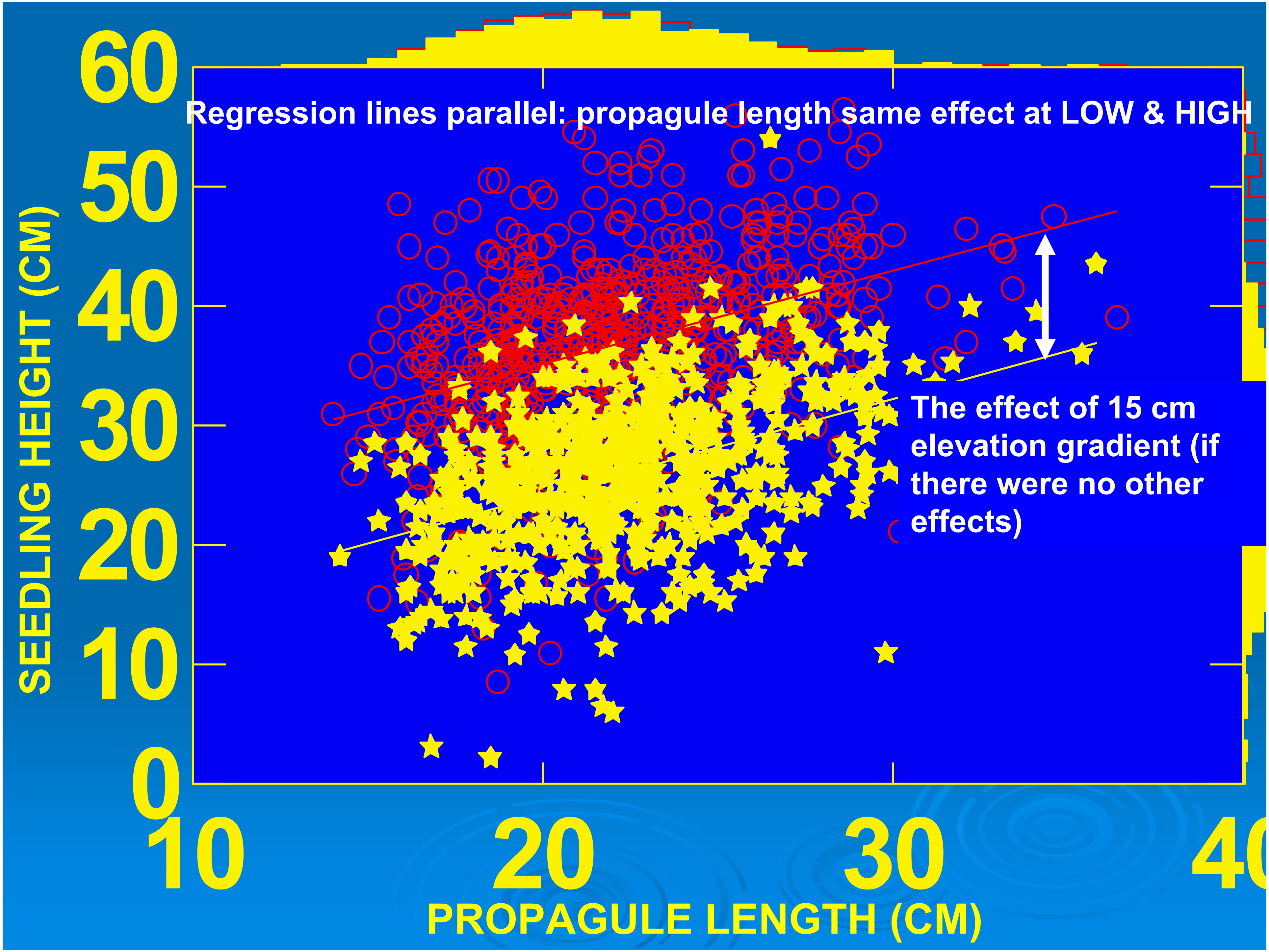
- **Weak, but significant + relationship**
- **Occurs in first year**

REGRESSION: HEIGHT AT 1 YEAR ON PROPAGULE LENGTH



REGRESSION: HEIGHT AT 1 YEAR ON PROPAGULE LENGTH




Regression lines parallel: propagule length same effect at LOW & HIGH
The effect of 15 cm elevation gradient (if there were no other effects)
SEEDLING HEIGHT (CM)
60
50
40
30
20
10
0
10
20
30
40
PROPAGULE LENGTH (CM)

Reproduction: Propagules

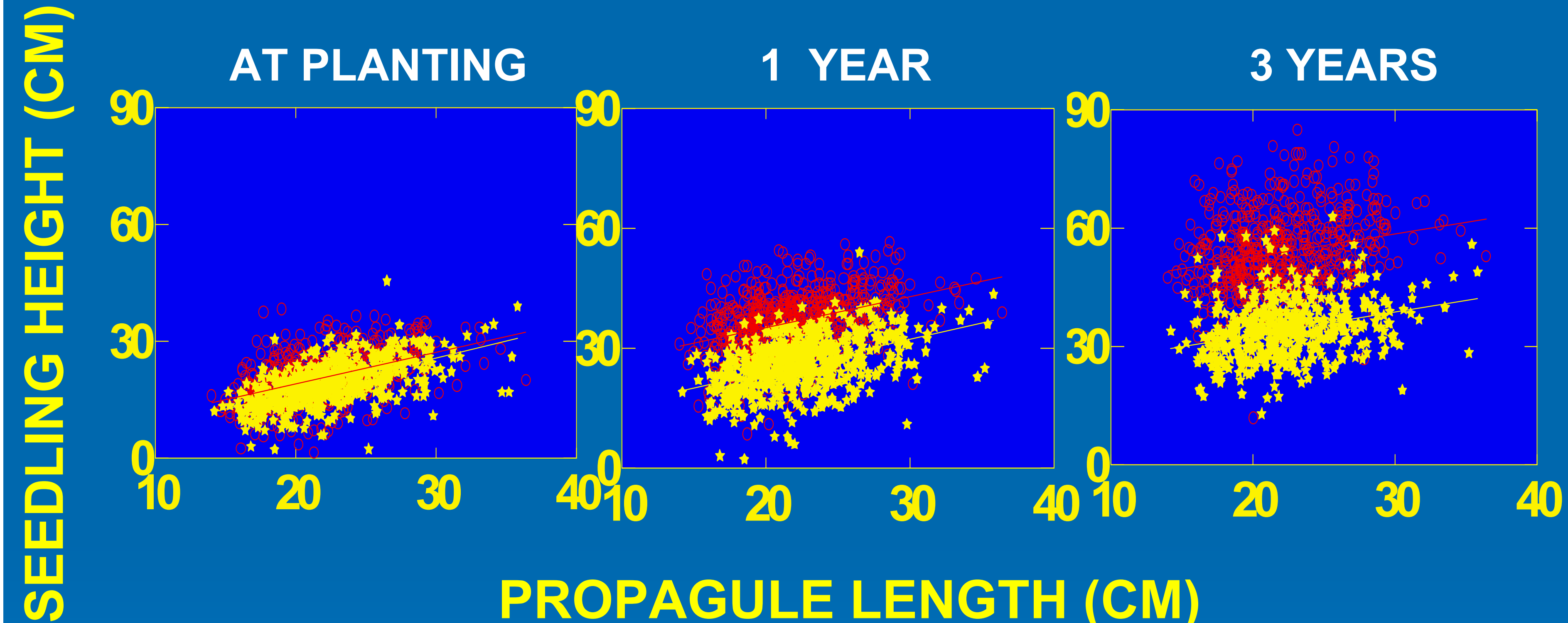
For Logistic Regression: 0 = Produced 1 propagule
1 = Produced >1 propagule

Logistic Regression Results:

Significant variation in propagule production (0 or 1) with CANOPY AREA *BUT, not a big biological diff*. (odds ratio: 1.1), variance explained: McFadden's Rho-Sq=10.6%)

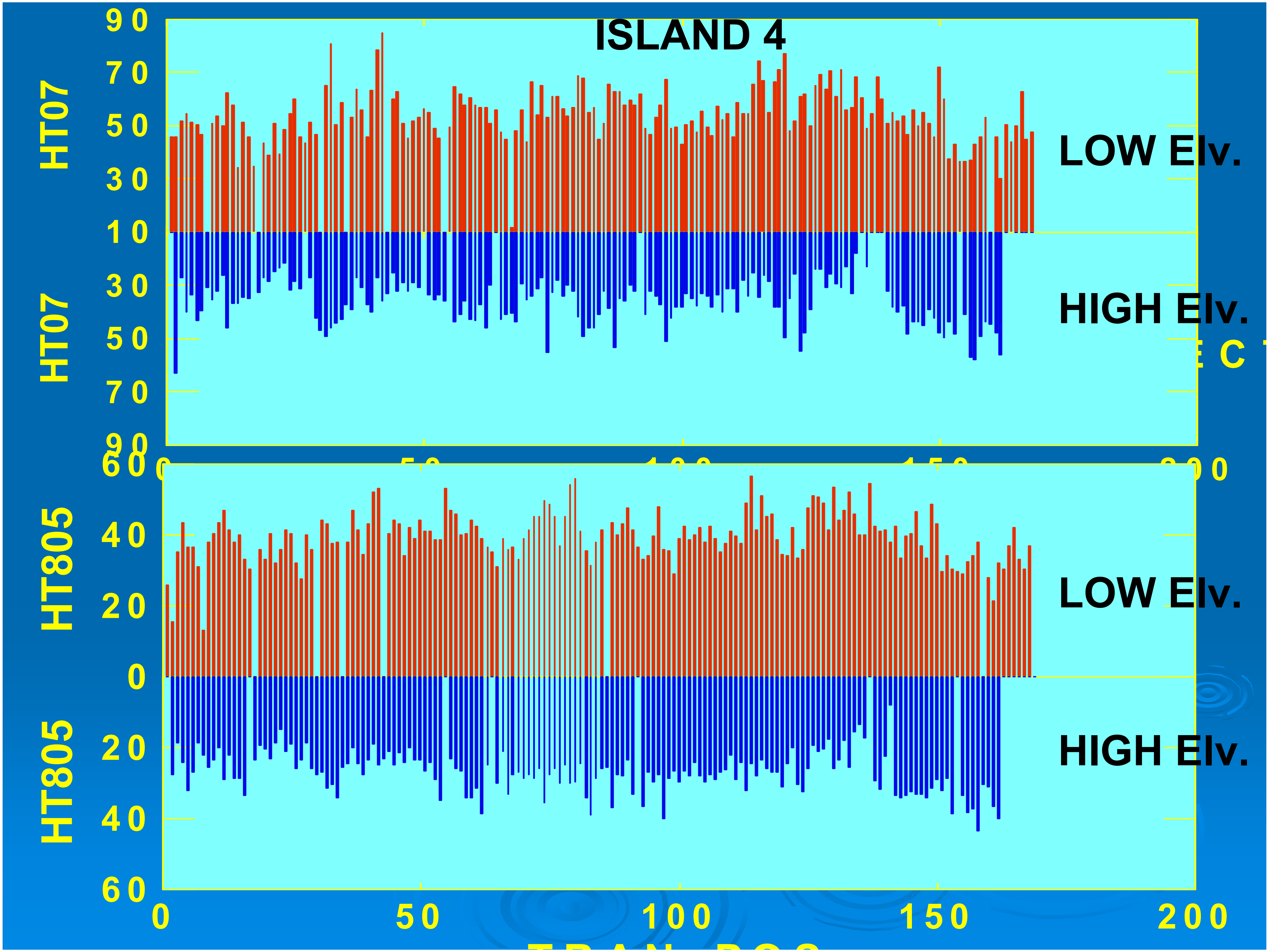
No diff. at either ELEVATION or COMBINED ELEV's:

PLANT HEIGHT VS PROPAGULE LENGTH: LOW & HIGH ELEVATIONS



- PROPAGULE SIZE EFFECT IN YR 1 (SAME AT LOW & HIGH)

- SCATTER INCR. IN LATER YRS AS OTHER FACTORS BEC. INCREASINGLY MORE IMPORTANT


ISLAND 4
HT07
HT07
LOW Elv.
HIGH Elv.
90
70
50
30
10
30
50
70
90
HT805
HT805
LOW Elv.
HIGH Elv.
60
40
20
0
20
40
60
0
50
100
150
200

R-M ANOVA for Height (summary)

- Height varied with 15 cm elevation gradient
- Height varied with island planted (ie, spatial scale of seedlings over 10's of meters)
- Height varied with maternal tree family (genotype?) BUT NOT with maternal tree location (ie., the embayment)
- Effects varied over the 3 years
  - Island Planted
  - Elevation
- Effects stable over 3 years
  - Maternal family (maternal genotype)