

Cryptic Invasion in a changed Climate -

Ecophysiology and Gene-expression
of *Phragmites australis* from the US
Gulf Coast

Franziska Eller & Hans Brix

- **Cryptic invasion**

- European lineage is displacing native stands
- Gulf Coast (GC) is a Hot Spot of *Phragmites* diversity

- **Delta-type** → African/Mediterranean origin
- **EU-type** → European invasive N-American population



Delta-type

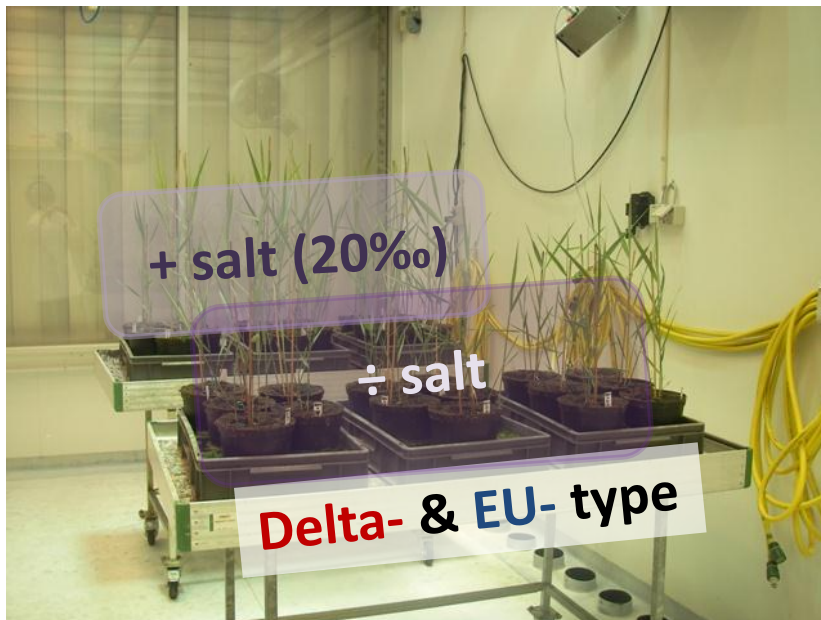
EU-type



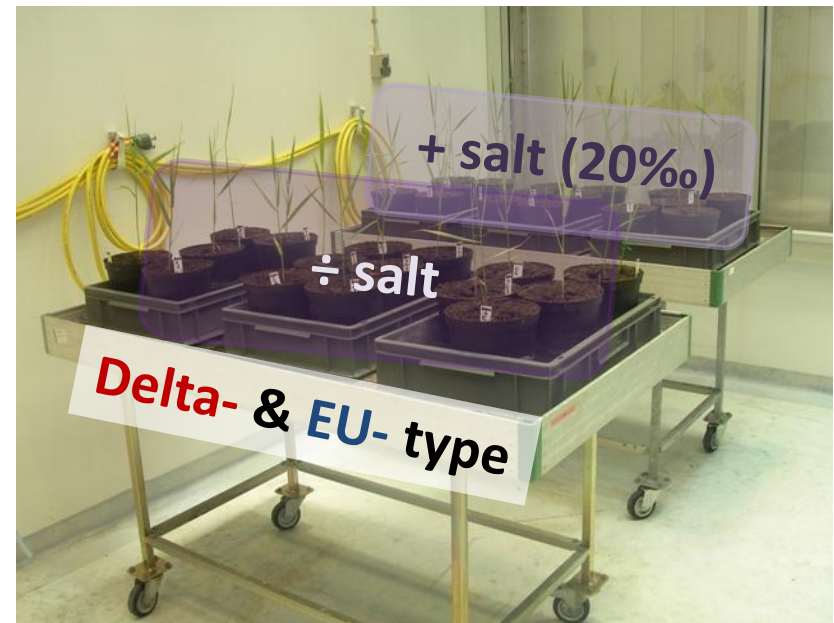
- **Investigation**

- Which of the two competing reed-types is ecophysiologicaly superior?
 - Salinity
 - Future climate
- Are there differences in gene-expression of genes related to stress-response and photosynthesis?

- **Experimental design**



“Future climate” treatment
(24/19°C; 700 ppm CO₂)



Ambient treatment
(19/14°C; 385 ppm CO₂)

- 11 weeks of growth
- weekly measurement of growth parameters
- Gas-exchange: CO₂-response
 Light-response



- Gene-expression analysis by RT-PCR
- Primers designed by alignment of homologue gene sequences of Poaceae



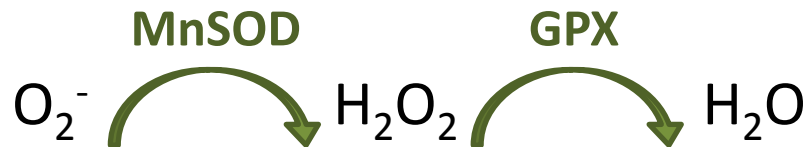
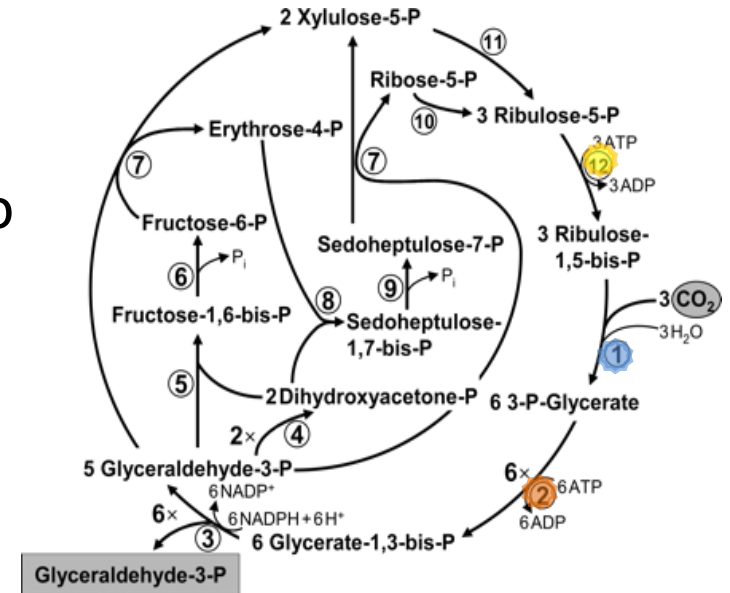
```

521 530 540 550 560 570 580 590 600 610 620 630 640 650
zi|1313150675|db.j|AB5      ATGGGCCCCACCGTGTATGCGCTCGT
zi|119909021|db.j|AB04     ACCAAGGAARACGACGCTAAGTGTCTGGCTATATATACCGTAGTGCACCCGCGCAATGGTGGCCCTACCTGTAGCCGGCATCTCCCTCTCCCTCCGATATACATACCATCCATGGCCCGCGTGTATGCGCTCGT
zi|1329782841|db.j|AK06    CAAATGGCTCCCGTGTATGCGCTCGT
zi|1379910671|db.j|AK12    ATGGGCCCCACCGTGTATGCGCTCGT
Consensus
.....
521 530 540 550 560 570 580 590 600 610 620 630 640 650
zi|1313150675|db.j|AB5      ATGGGCCCCACCGTGTATGCGCTCGT
zi|119909021|db.j|AB04     ACCAAGGAARACGACGCTAAGTGTCTGGCTATATATACCGTAGTGCACCCGCGCAATGGTGGCCCTACCTGTAGCCGGCATCTCCCTCTCCCTCCGATATACATACCATCCATGGCCCGCGTGTATGCGCTCGT
zi|1329782841|db.j|AK06    CAAATGGCTCCCGTGTATGCGCTCGT
zi|1379910671|db.j|AK12    ATGGGCCCCACCGTGTATGCGCTCGT
Consensus
.....
651 660 670 680 690 700 710 720 730 740 750 760 770 780
zi|1313150675|db.j|AB5      CCGCACCCCGCTCGCTCCGTTCCAGGGCTCAAGTCCACCGCCACCCCTCCCGCTCGCCGGCGTCCAGCA---CTAGCTTCGGCAGATCCAGCAGCCGCGCAGATCCAGTCCAGTCCAGT
zi|119909021|db.j|AB04     CCGCTACCCCGCTCGCTCCGTTCCAGGGCTCAAGTCCACCGCCACCCCTCCCGCTCGCCGGCGTCCAGCA---AGCCTCGCCGCTCCAGCAATGGCGAAT---GATATGCTCG
zi|1329782841|db.j|AK06    CCGCACCCCGCTCGCTCCGTTCCAGGGCTCAAGTCCACCGCCACCCCTCCCGCTCGCCGGCGTCCAGCAAGCTCAGCTTTGGCAGCTCCAGCAATGGCGCAGATCCAGTCCAGTCCAGT
zi|1379910671|db.j|AK12    CCGCACCCCGCTCGCTCCGTTCCAGGGCTCAAGTCCACCGCCACCCCTCCCGCTCGCCGGCGTCCAGCAAGCTCAGCTTTGGCAGCTCCAGCAATGGCGCAGATCCAGTCCAGTCCAGT
Consensus
CgGcCaCCaCCgTcGcTcCCTCCAGGGcCTCAAGTCCACCGCCcgcTcCCTcCaCCCGCCcTCCAGcAgc.c.c.aGcTcGgCaacGTCAGCAATGGCGCaAGGATCaGTCAGTCCAGGTT.....
.....
781 790 800 810 820 830 840 850 860 870 880 890 900 910
zi|1313150675|db.j|AB5      AGACTCGAGGACCAAAACTGATATGAGTTCTCTCTCTGTAGTACAAATTTACTAGCACCAGGTGTAGGCAAGTGTGTGCAATCCACAAATTCACATTCATTCAGGTTGGCCCGCTACCGGACCAAGA
zi|119909021|db.j|AB04     AGACTCGAGGACCAAAACTGATATGAGTTCTCTCTCTGTAGTACAAATTTACTAGCACCAGGTGTAGGCAAGTGTGTGCAATCCACAAATTCACATTCATTCAGGTTGGCCCGCTACCGGACCAAGA
zi|1329782841|db.j|AK06    AGACTCGAGGACCAAAACTGATATGAGTTCTCTCTCTGTAGTACAAATTTACTAGCACCAGGTGTAGGCAAGTGTGTGCAATCCACAAATTCACATTCATTCAGGTTGGCCCGCTACCGGACCAAGA
zi|1379910671|db.j|AK12    AGACTCGAGGACCAAAACTGATATGAGTTCTCTCTCTGTAGTACAAATTTACTAGCACCAGGTGTAGGCAAGTGTGTGCAATCCACAAATTCACATTCATTCAGGTTGGCCCGCTACCGGACCAAGA
Consensus
.....
911 920 930 940 950 960 970 980 990 1000 1010 1020 1030 1040
zi|1313150675|db.j|AB5      AGTTCGAGACCGTGTCTACTGCCCGCTCGACGAGGAGCAGCTCTGAGGACAGTGGACTACCTGCTGCGCAACTGGGGCCCTGCTCGAGTTCAGCAGGAGGCTTCGCTGTACCGCGAGAA
zi|119909021|db.j|AB04     AGTTCGAGACCGTGTCTACTGCCCGCTCGACGAGGAGCAGCTCTGAGGACAGTGGACTACCTGCTGCGCAACTGGGGCCCTGCTCGAGTTCAGCAGGAGGCTTCGCTGTACCGCGAGAA
zi|1329782841|db.j|AK06    AGTTCGAGACCGTGTCTACTGCCCGCTCGACGAGGAGCAGCTCTGAGGACAGTGGACTACCTGCTGCGCAACTGGGGCCCTGCTCGAGTTCAGCAGGAGGCTTCGCTGTACCGCGAGAA
zi|1379910671|db.j|AK12    AGTTCGAGACCGTGTCTACTGCCCGCTCGACGAGGAGCAGCTCTGAGGACAGTGGACTACCTGCTGCGCAACTGGGGCCCTGCTCGAGTTCAGCAGGAGGCTTCGCTGTACCGCGAGAA
Consensus
AGTTCGAGACCGTGTCTACTGCCCGCTCGACGAGGAGcAgCTcTGAAGCAAGTGGACTACCTGCTGCGCAACTGGGGCCcTcGCTCGAGTTCAGCAGGAGGCTTCGCTGTACCGCGAGAA
.....
1041 1050 1060 1070 1080 1090 1100 1110 1120 1130 1140 1150 1160 1170
zi|1313150675|db.j|AB5      CTCACCTCGCGTCTACTACGACGGCCGCTACTGGACATGTTGAAAGCTGCCCATGTTCCGGTGCACGACGCGCTCGCAGGCTCAAGGAGGCTCAGGAGGCGCATCGCTCTACCCGACCGCTAC
zi|119909021|db.j|AB04     CTCACCTCGCGTCTACTACGACGGCCGCTACTGGACATGTTGAAAGCTGCCCATGTTCCGGTGCACGACGCGCTCGCAGGCTCAAGGAGGCTCAGGAGGCGCATCGCTCTACCCGACCGCTAC
zi|1329782841|db.j|AK06    CTCACCTCGCGTCTACTACGACGGCCGCTACTGGACATGTTGAAAGCTGCCCATGTTCCGGTGCACGACGCGCTCGCAGGCTCAAGGAGGCTCAGGAGGCGCATCGCTCTACCCGACCGCTAC
zi|1379910671|db.j|AK12    CTCACCTCGCGTCTACTACGACGGCCGCTACTGGACATGTTGAAAGCTGCCCATGTTCCGGTGCACGACGCGCTCGCAGGCTCAAGGAGGCTCAGGAGGCGCATCGCTCTACCCGACCGCTAC
Consensus
cCaCaGATCcCcCgGaTACTAcGAcGCaGgTACTGGACATGTTGAAAGCTGCCCATGTTCCGGTGCACGAcGCaCaCcCaGgTgctCAAGGAGCTcAGGAGGcCaGagaagggcTACCCtGAcGcTAc
.....
1171 1180 1190 1200 1210 1220 1230 1240 1250 1260 1270 1280 1290 1300
zi|1313150675|db.j|AB5      GTCGGATCCTCGGCTTCACACACATCAGCAGACGCGAGTGCCTCAGCTTCATCGCTACAGCCCGCGGGCAGCGAGTGA
zi|119909021|db.j|AB04     GTCGGATCCTCGGCTTCACACACATCAGCAGACGCGAGTGCCTCAGCTTCATCGCTACAGCCCGCGGGTTCAGGAGGCTTCGGCAGGCGCTAAGCTACAGGTTAAACCAATGGCCCTATGATGATTG
zi|1329782841|db.j|AK06    GTCGGATCCTCGGCTTCACACACATCAGCAGACGCGAGTGCCTCAGCTTCATCGCTACAGCCCGCGGGTTCAGGAGGCTTCGGCAGGCGCTAAGCTACAGGTTAAACCAATGGCCCTATGATGATTG
zi|1379910671|db.j|AK12    GTCGGATCCTCGGCTTCACACACATCAGCAGACGCGAGTGCCTCAGCTTCATCGCTACAGCCCGCGGGTTCAGGAGGCTTCGGCAGGCGCTAAGCTACAGGTTAAACCAATGGCCCTATGATGATTG
Consensus
GTCGCaTcCaTcGGCTTCAGCACAcTcAGGcAGAcGCGAGTGCCTCAGCTTCATCGCTAcAGCCcCGGGcTTCAGGAGGcTTCGGCAGGcGcTcAGGAGgagctcggcgcaact.aagc.....

```

– Genes of interest

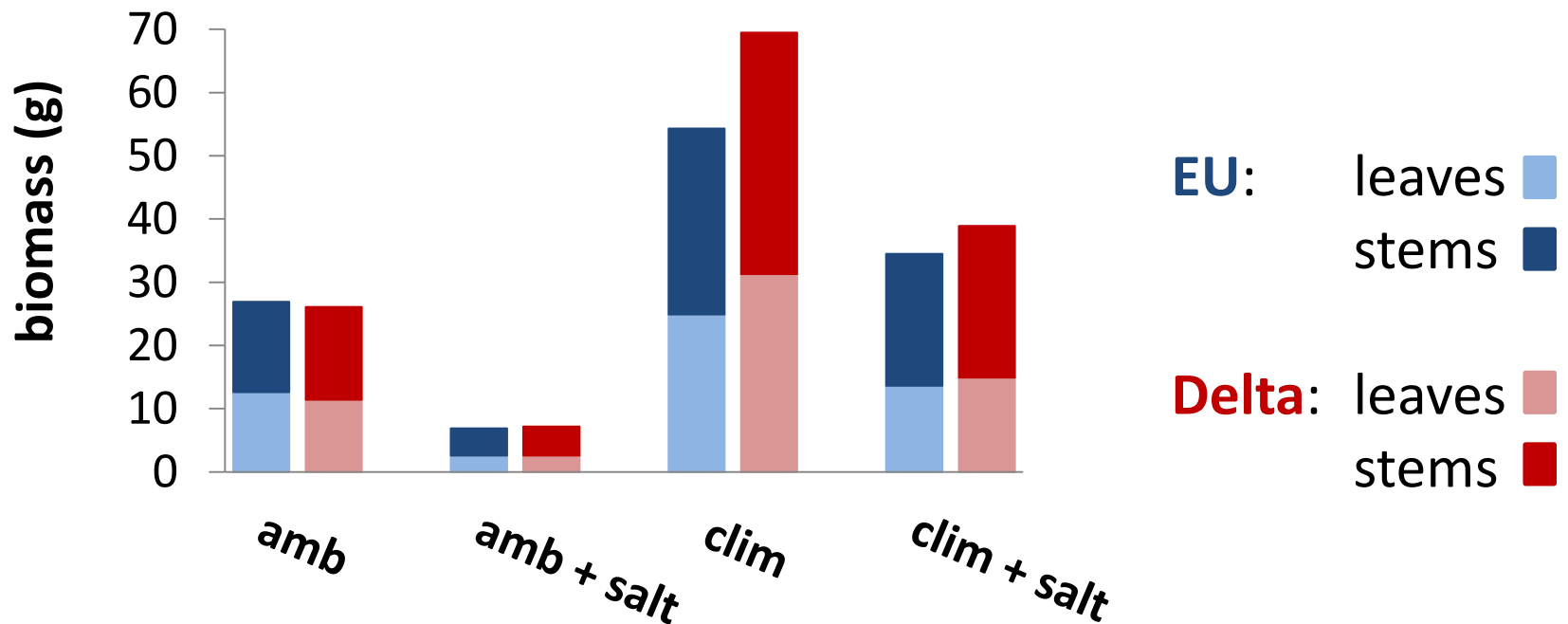
- $rbcS$ – small subunit of Rubisco
- Phosphoglycerate kinase
- Phosphoribulokinase
- Na^+/H^+ antiporter
- GPX – Glutathione peroxidase
- MnSOD – Mn Superoxide dismutase



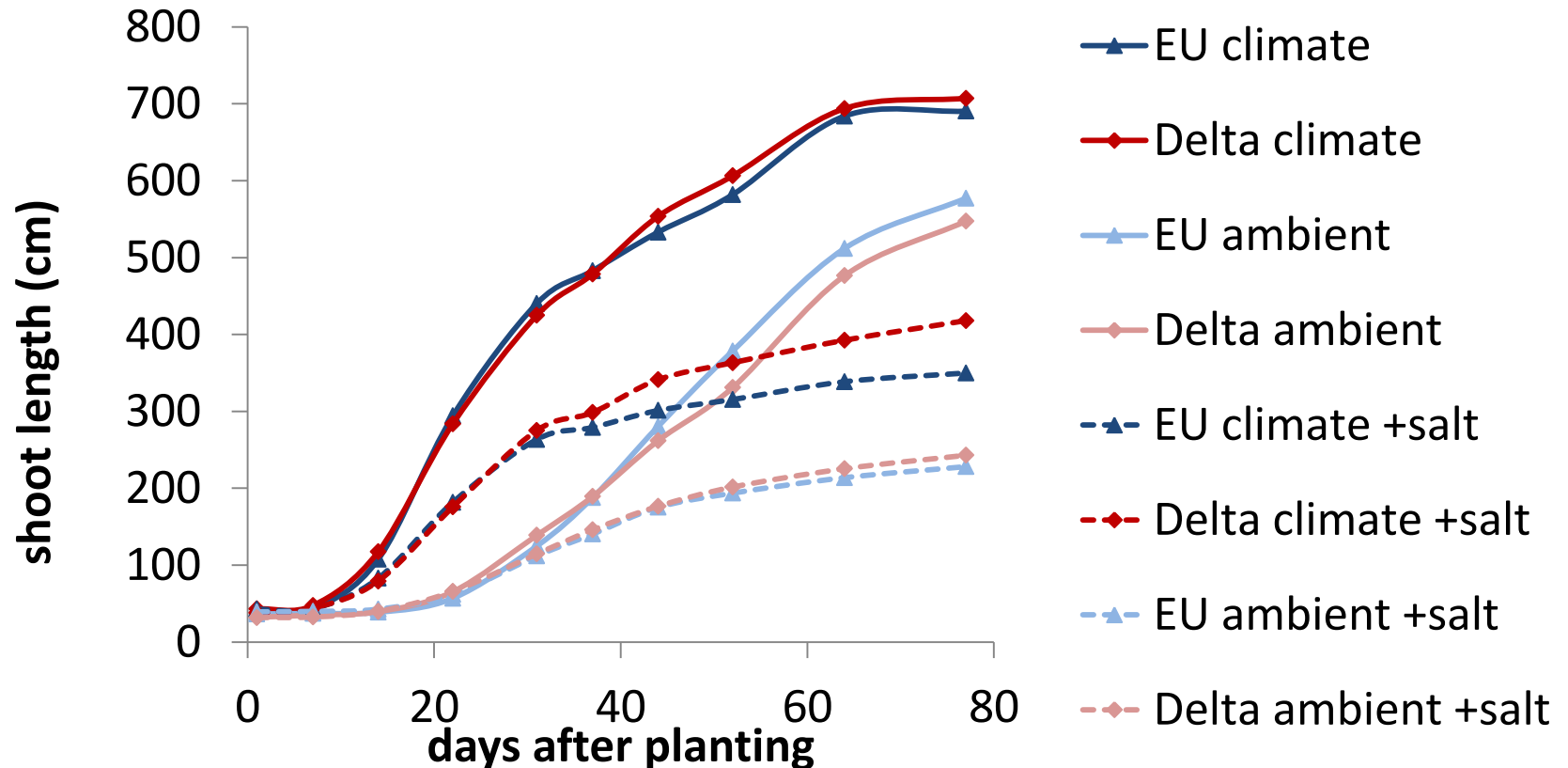
• Results

Parameter	Main factors			Interactions		
	Genotype	Salt	Climate	Genotype × Salt	Genotype × Climate	Salt × Climate
Final biomass	0.4	406.7***	566.5***	0.3	2.3	0.1
Leaf dry mass	1.5	77.7***	111***	0.5	2.7	2.8
Stem dry mass	3.1	36.1***	109.6***	0.7	2.4	0.1
Leaf production rate	0.6	66.1***	308.5***	0.0	0.0	11.7**
Shoot production rate	0.3	9.2**	162.9***	0.1	0.2	5.5*
Shoot elongation rate	4.2*	681.4***	152.8***	7.9**	1.2	19.3***
Quantum yield	3.9	0.6	38.1***	0.3	0.3	0.4
P_{max}	4.8*	6.5*	37.4***	0.2	0.1	21.9***
Light compensation pt.	0.2	0.3	60.5***	0.1	0.6	8.9**
Dark respiration	2.7	0.0	17.2***	0.0	0.0	8.9**
Light saturation pt.	2.3	9.0**	10.7**	0.2	0.3	40.5***
CO ₂ compensation pt.	0.5	11.7**	21.7***	0.1	0.9	3.1
V_{cmax} (carboxylation rate)	5.5*	2.1	38.2***	0.9	0.0	0.3
J (electron transport)	5.5*	0.0	5.9*	0.9	0.1	0.5
E (transpiration rate)	1.2	38.5***	10.6**	0.2	0.3	19.9***
Water use efficiency	2.5	9.5**	0.7	0.6	0.8	0.7

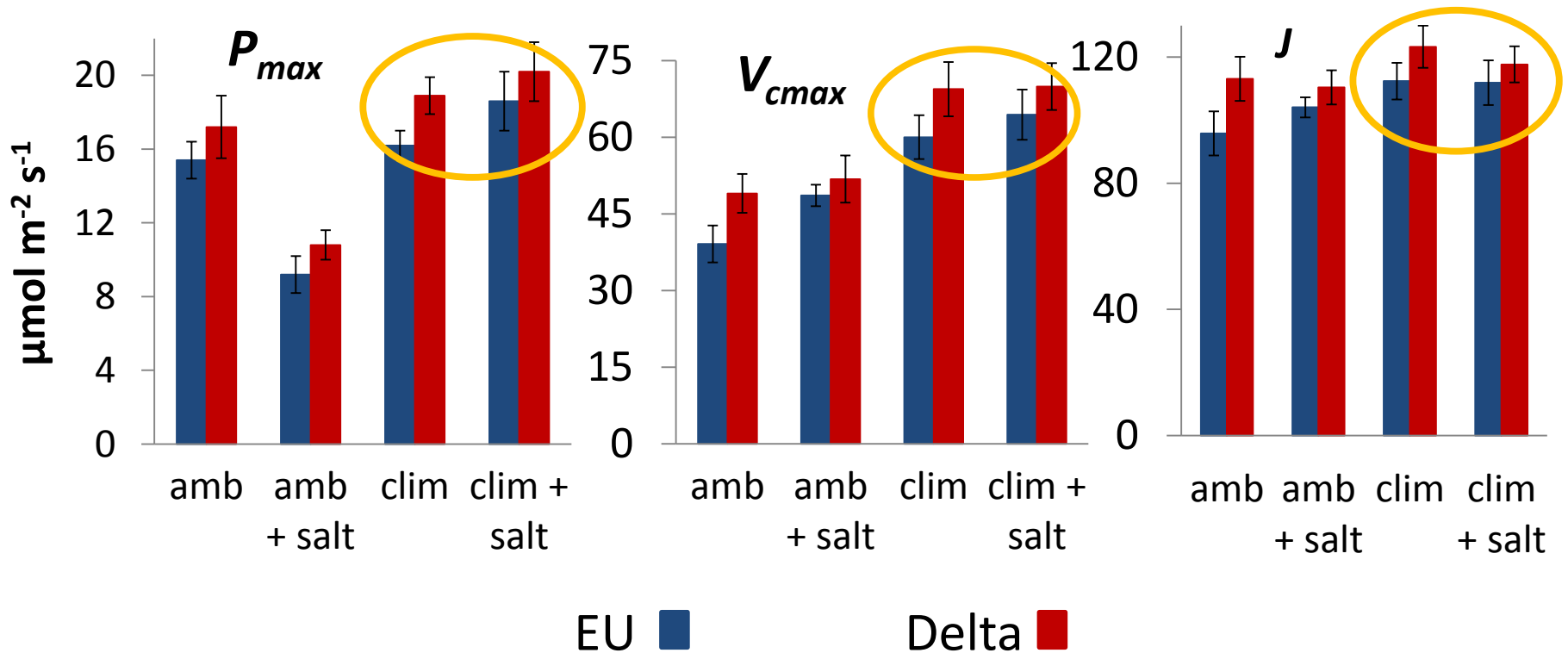
- Future climate enhanced growth & biomass
- Salt stress diminished growth **but**: less severe under future climate conditions



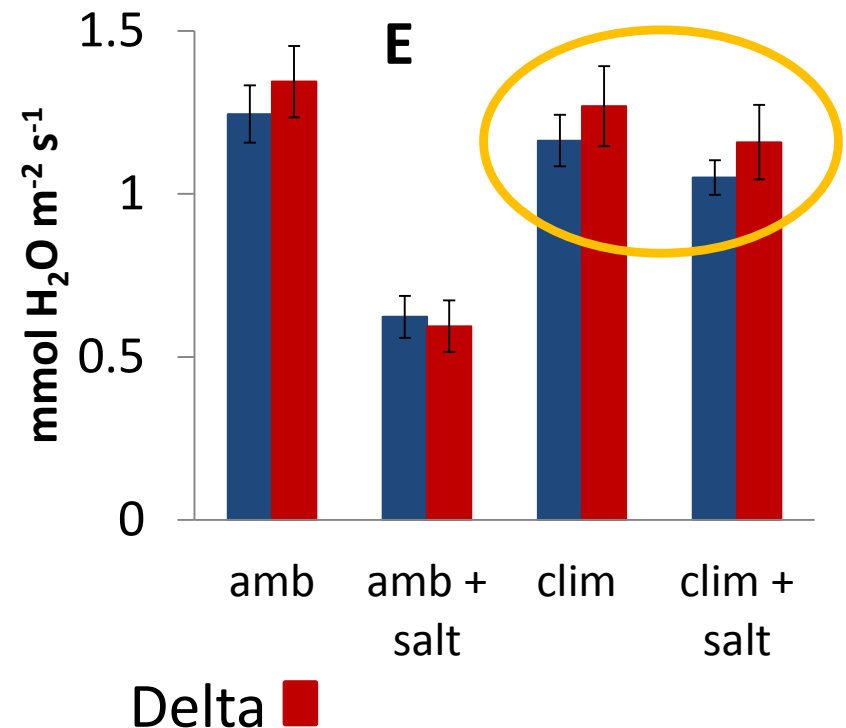
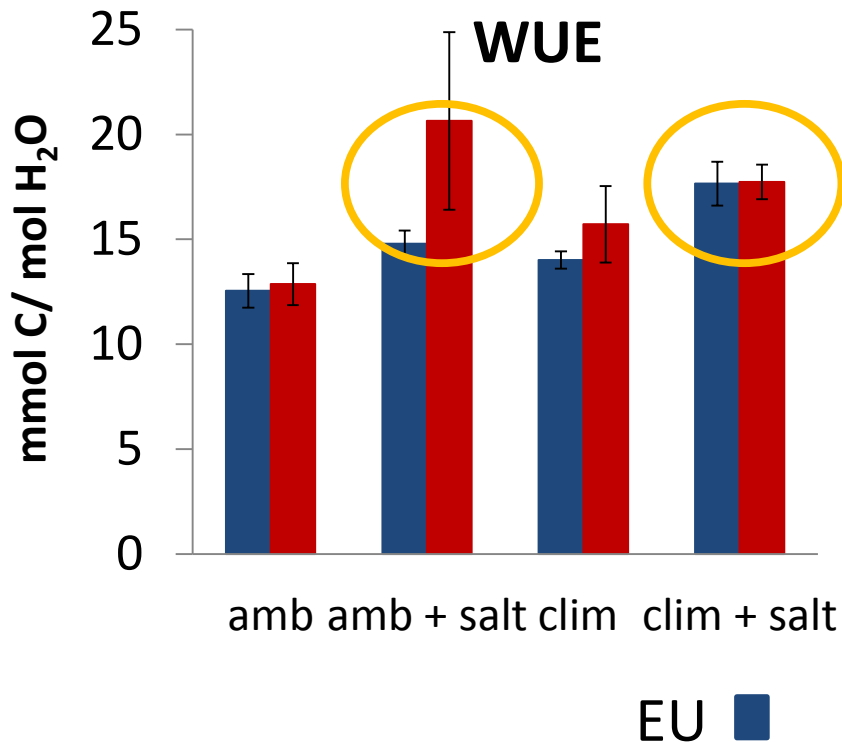
– **Shoot elongation rate** – only significant difference between of genotypes (growth)



– P_{max} , V_{cmax} , J : only significant difference between genotypes (physiology)



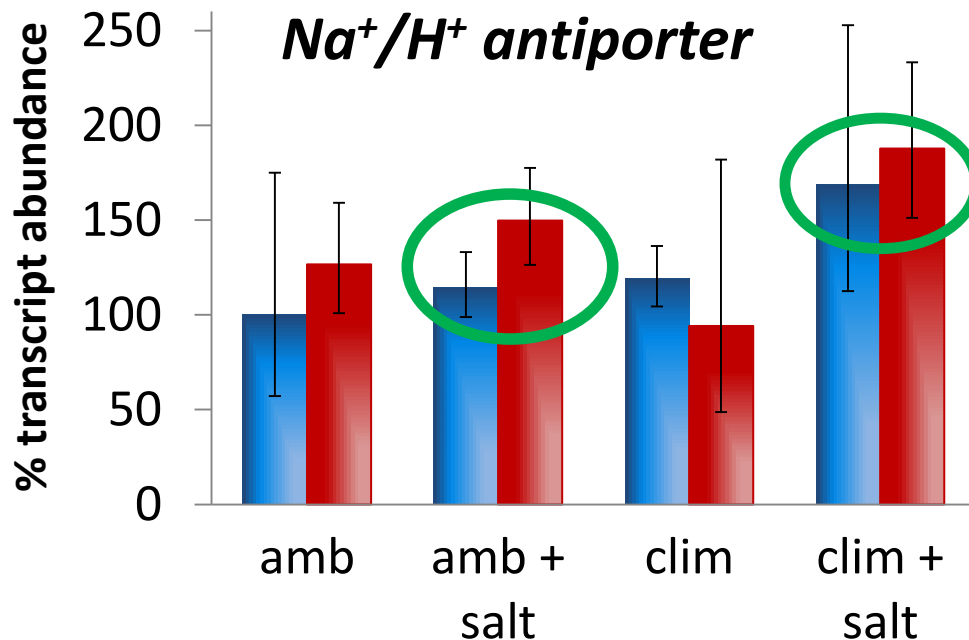
- **WUE (P_{\max}/E):** high under salt stress
- **E (transpiration rate):** little salt impact in future climate



– no significant differences in gene expression

→ *tendency*

– **Salt stress related genes:**

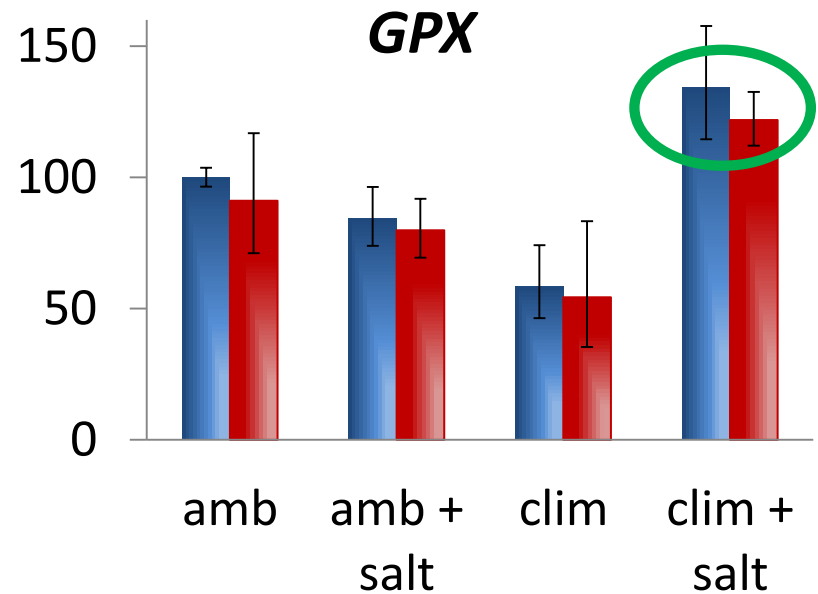
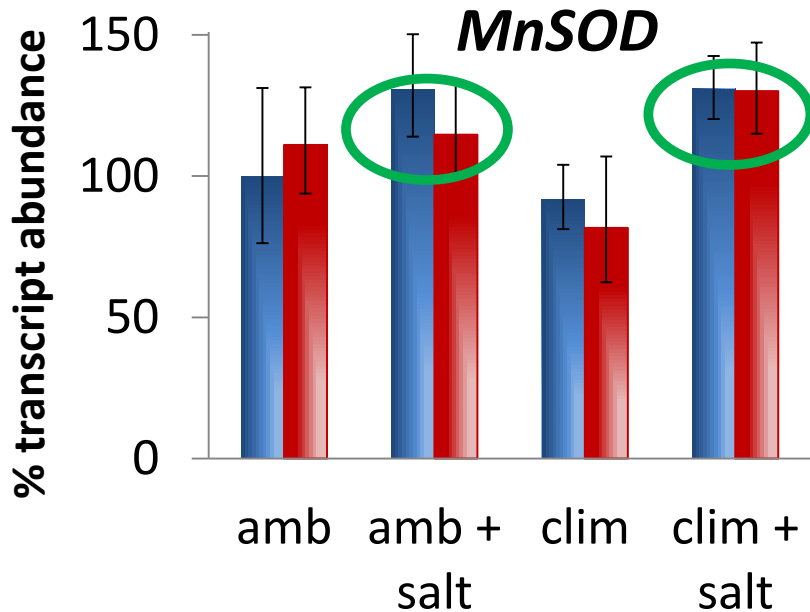


– Up-regulation under salt stress, especially in future climate

EU 

Delta 

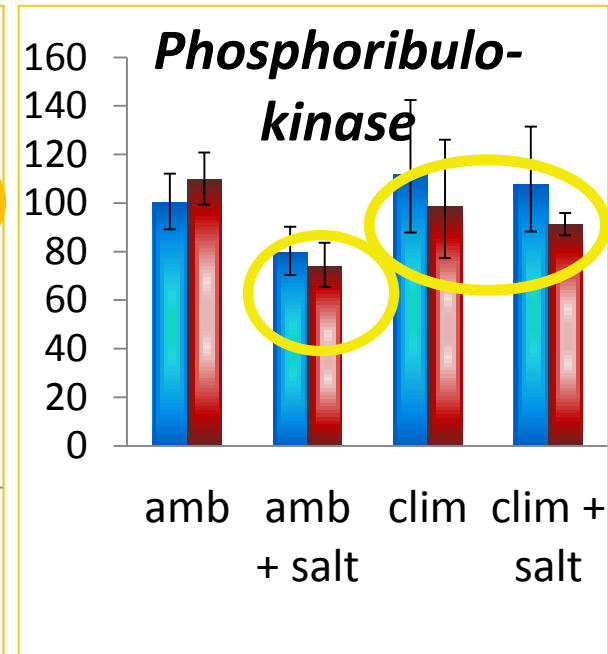
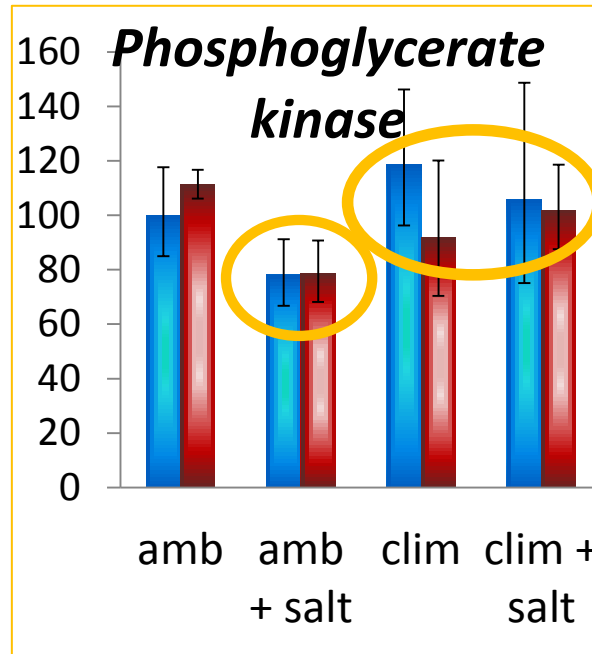
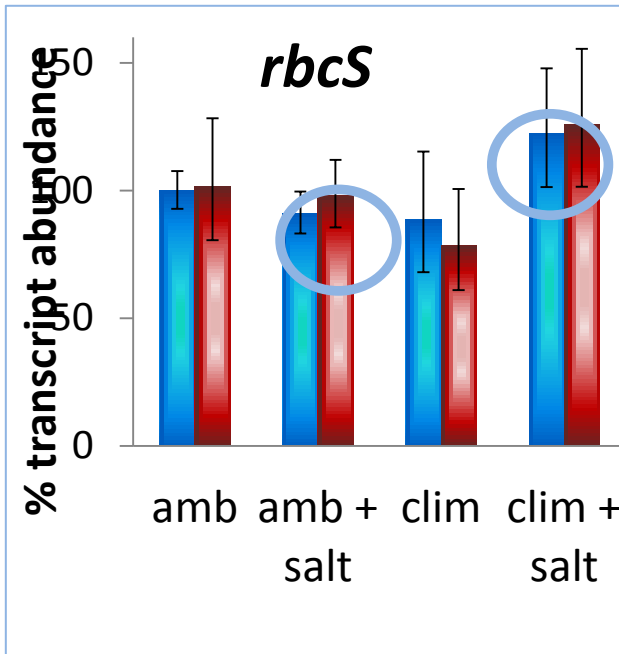
- General stress response (oxygen-scavenging enzymes)
- Up-regulation under salt stress



EU ■

Delta ■

- Calvin Cycle related genes:
- Down-regulation under salt-stress only in ambient climate



EU 

Delta 

– Amelioration of salt stress under \uparrow [CO₂]
and \uparrow temperature:

- \uparrow WUE, \uparrow C-uptake
- \uparrow assimilates for osmotic adjustment
- Osmotic adjustment by ion uptake
- Induction of specific salt stress genes in future climate (GPX)

- **In conclusion**

- Which of the two competing reed-types is ecophysiologicaly superior?

- **Delta-type:** superior (SER) and few physiological traits
- **Salinity:** unfavorable but not severely
- **Future climate:** favors growth and withstanding salt stress



→ *Will the Delta-type outcompete the EU-type in the field?*

- Are there differences in gene-expression of genes related to stress-response and photosynthesis?
- **Are those differences reflected in parameters?**
 - In "future climate + salt" treatment:
 - high transcript abundance
 - high photosynthesis
 - large biomass

→ ***Yes! (largely)***



Thanks to:

Hans Brix

Carla Lambertini

E. Simona Radutoiu

Camilla Håkansson

the staff at Plant Biology

(Bioscience AU)

the staff at Risoe Biosystems (DTU)

