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# An epigenetic memory – a phenomenon affecting adaptive traits in Norway spruce

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# A memory in *Picea abies*

**The temperature that prevails when the mother trees produce their seeds, affects the adaptive performance of the progenies when they grown in common garden experiments!**

**The timing of**

- dehardening and bud burst in the spring**
- leader shoot growth cessation in the summer**
- bud set and cold acclimation in the autumn**

**are all progeny traits that are changed**



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# The memory

**The timing is delayed**

**if seeds are produced under warmer conditions  
than normal**

**Timing is advanced**

**if seeds are produced colder than normal**

**The altered performance is persistent**

# A spin-off from breeding

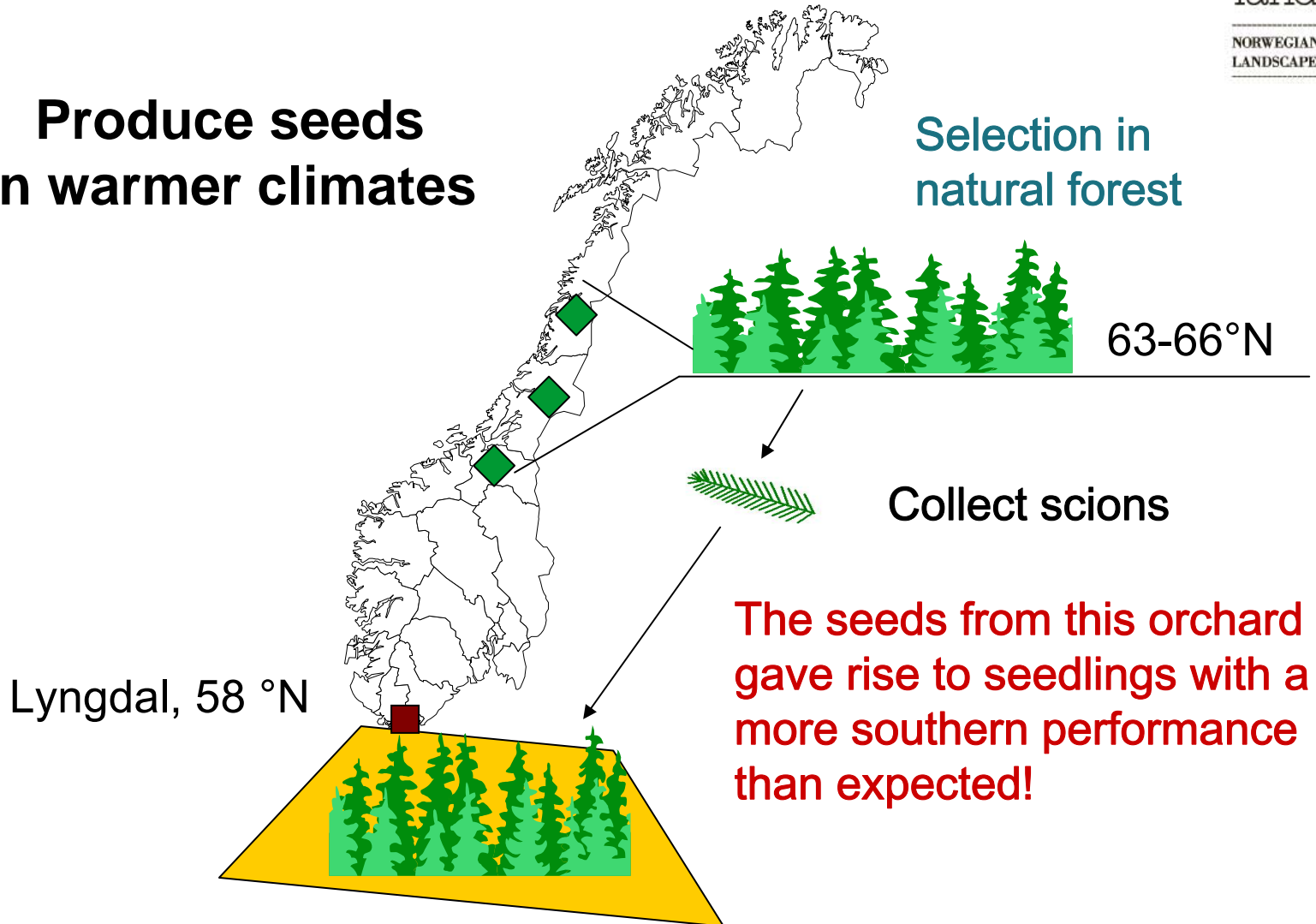


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**Produce seeds  
in warmer climates**

**Selection in  
natural forest**



# Kaupanger seed orchard

- Seeds for high elevation areas
- Produced at the sea level
- Change in timing of phenology and frost hardiness

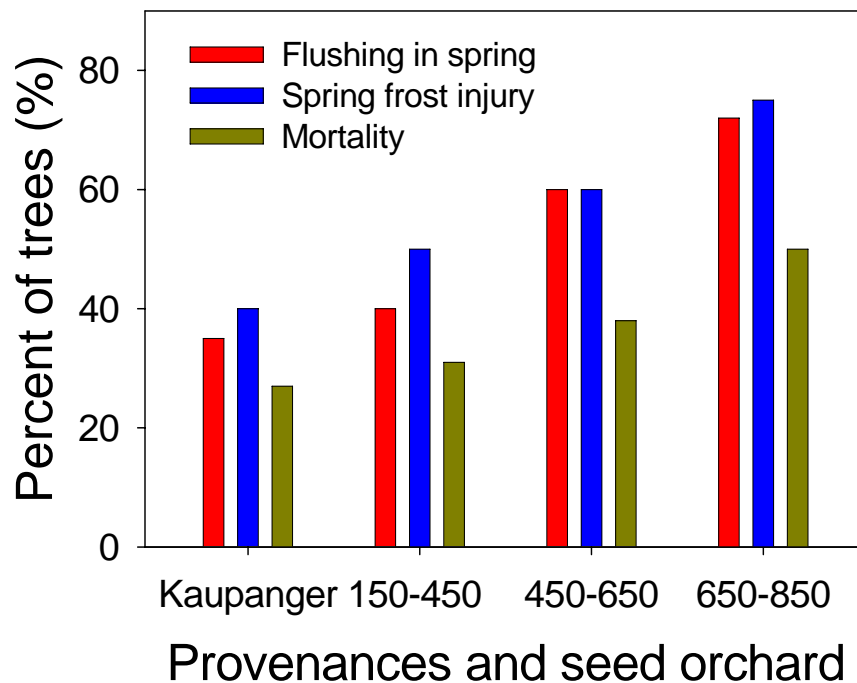
Skrøppa et al 2007



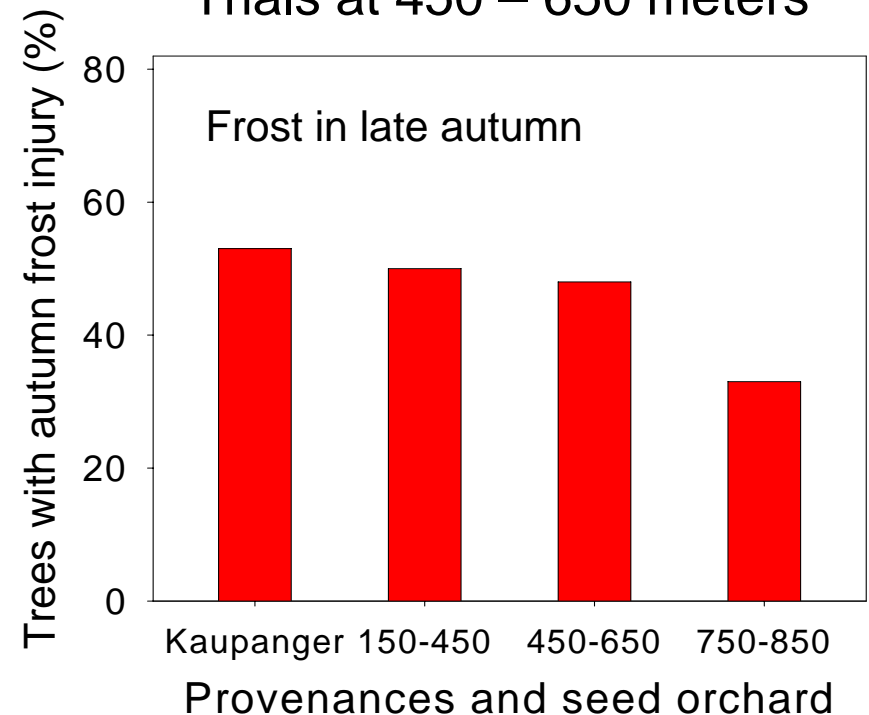
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## Trials at 650 – 940 meters

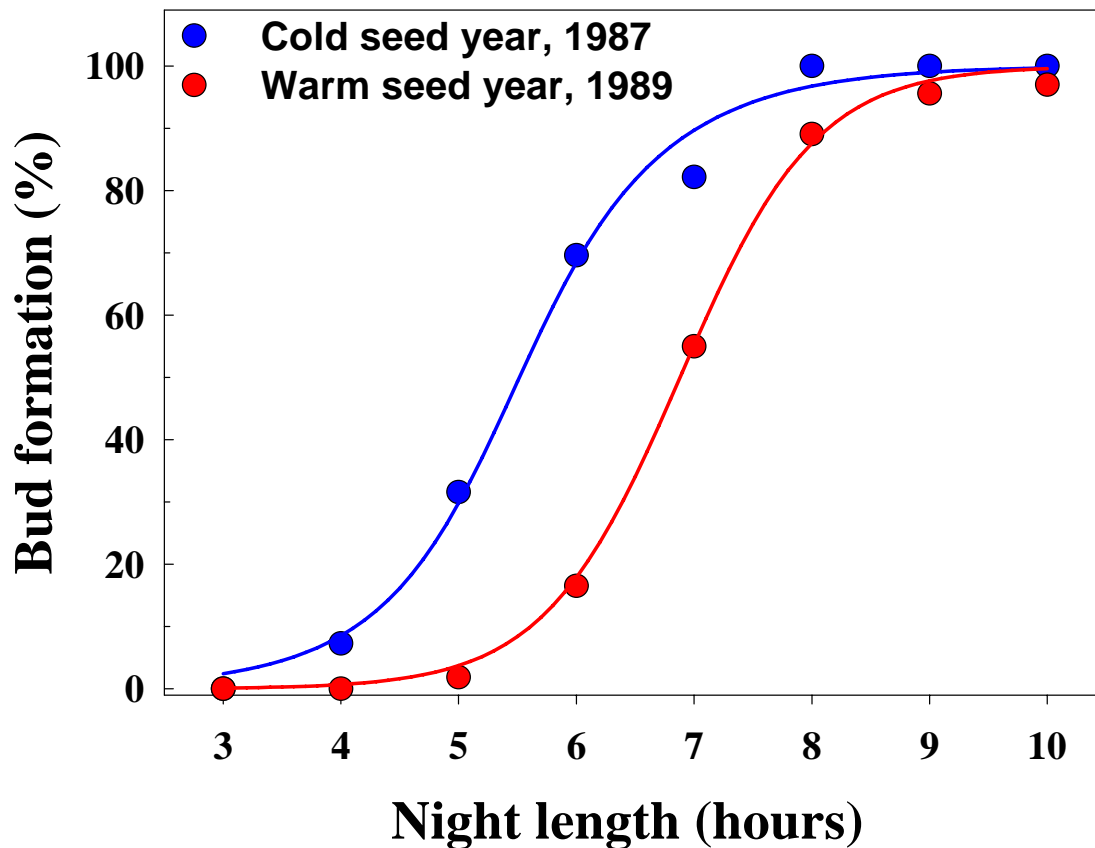


## Trials at 450 – 650 meters



# Warm and cold seed year at Lyngdal seed orchard

Kohmann and Johnsen 1994



- Growth chambers
- 8 weeks old

# Lyngdal material in field trials



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Lyngdal: Compartment and seed year	Growth start %	Growth cessation %	Injury %	Mortality %	Height 8 years cm
Dale 1987	8.5	83.9	30.2	50.8	101
Dale 1989	5.1	72.5	17.2	43.3	115

# German provenance, Harz

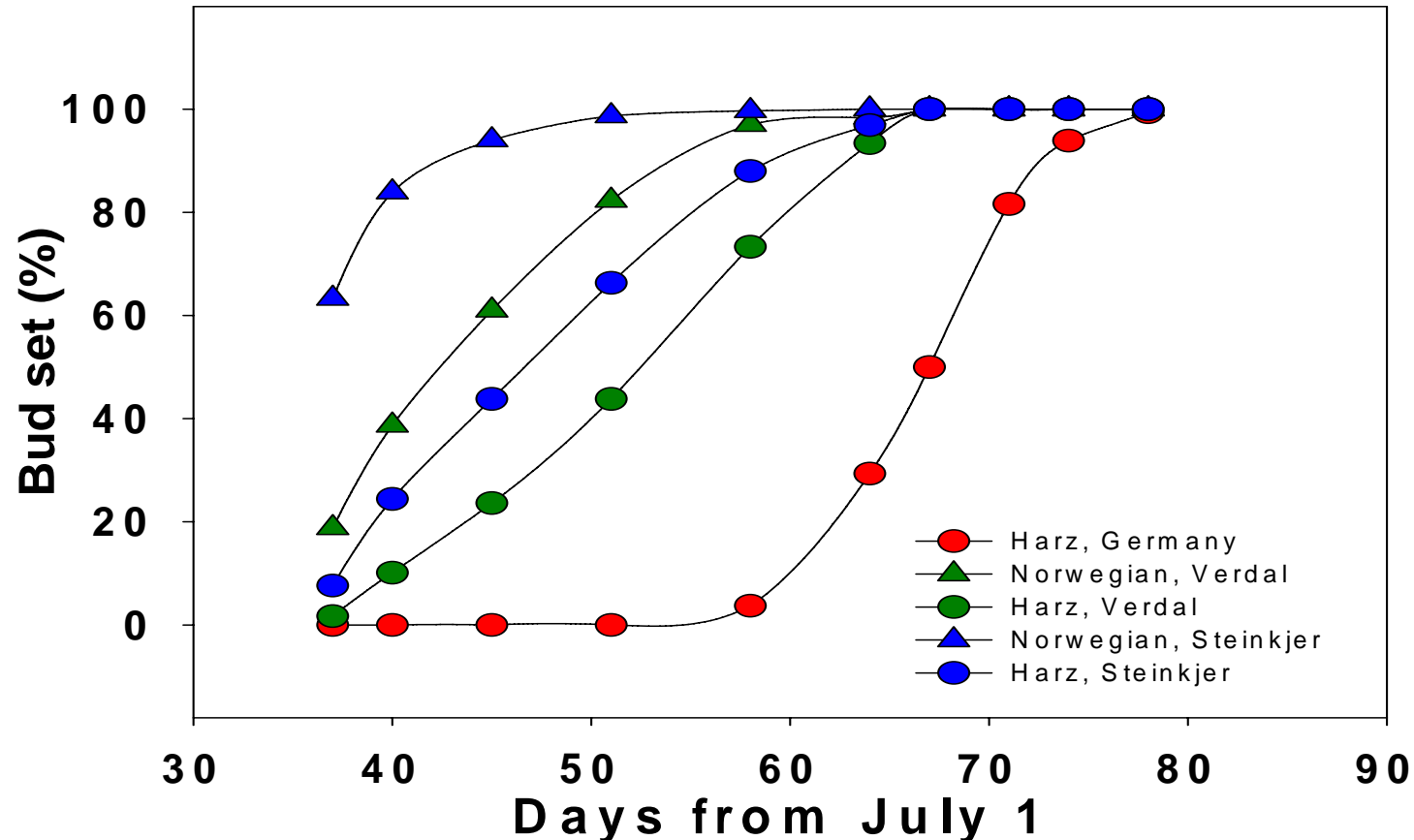
- If grown in Central Norway
- Produces progenies with a northern phenology



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Skrøppa et al 2009, in press (?)





# We make seeds in different temperatures



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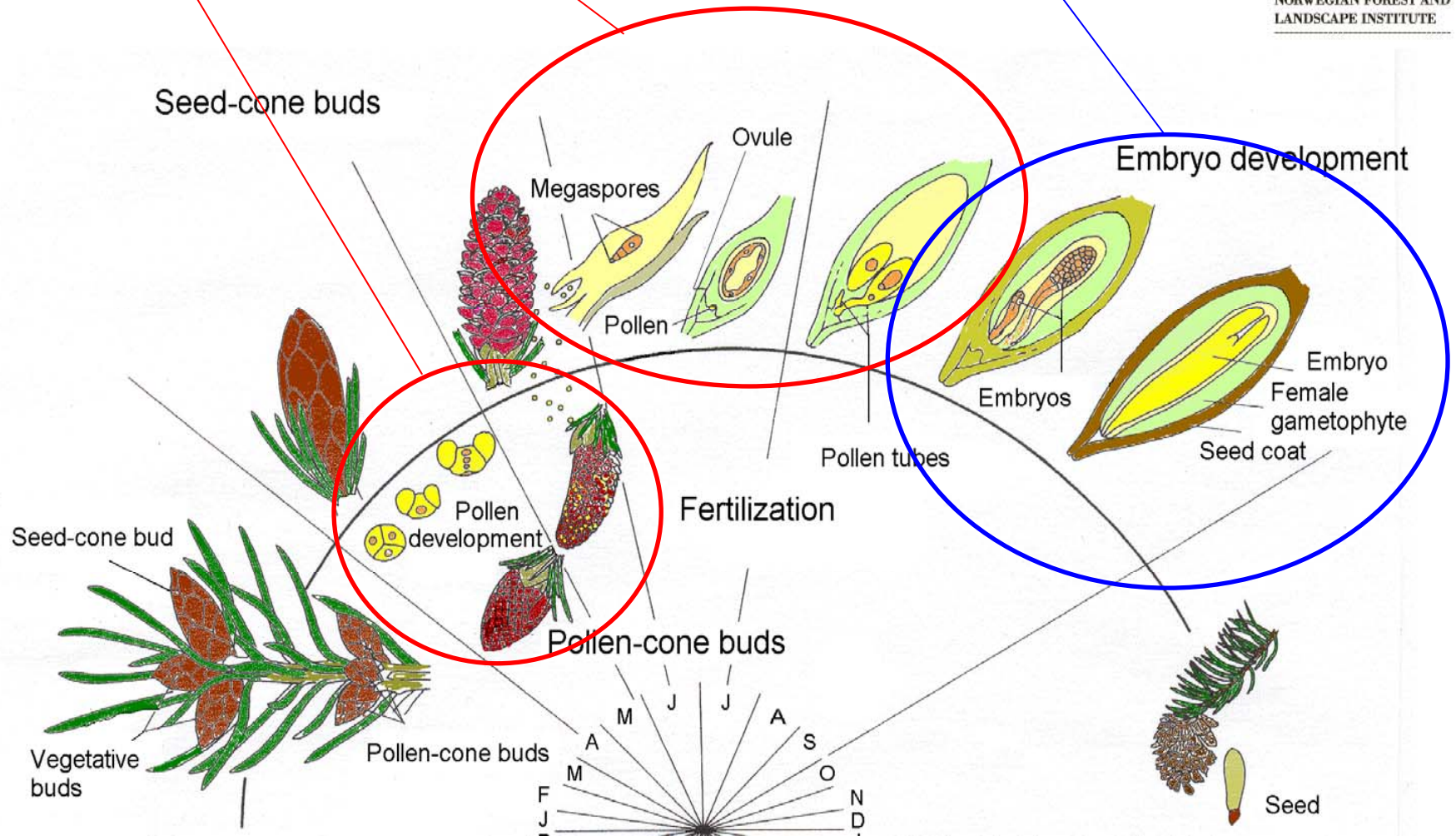
Observed no effect of treatments on the progeny

Memory is expressed



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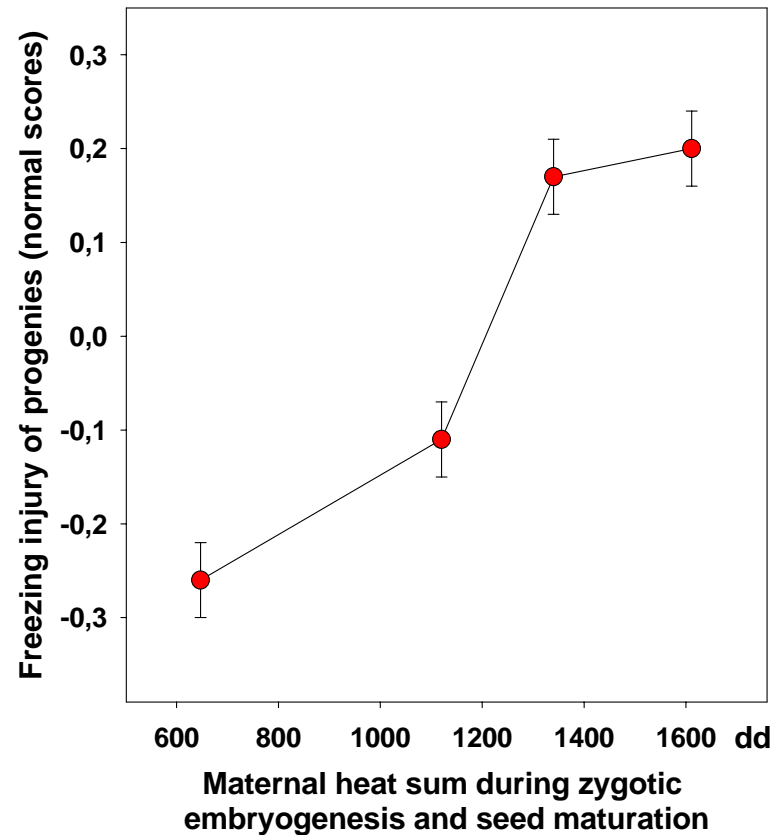
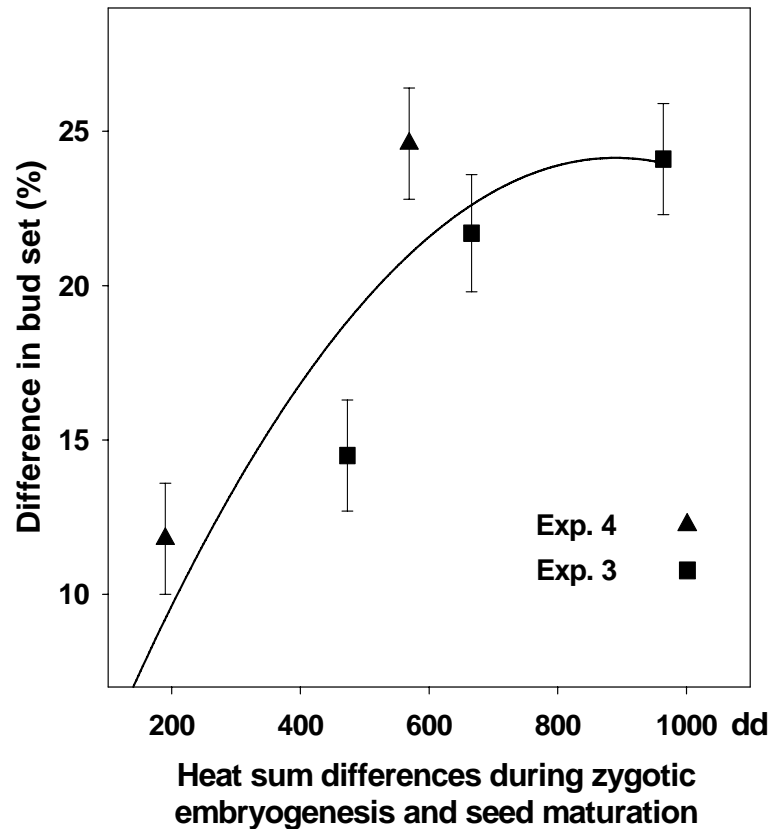


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# Embryogenesis is the stage!

Johnsen et al 2005



# Somatic embryogenesis



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is a good model!



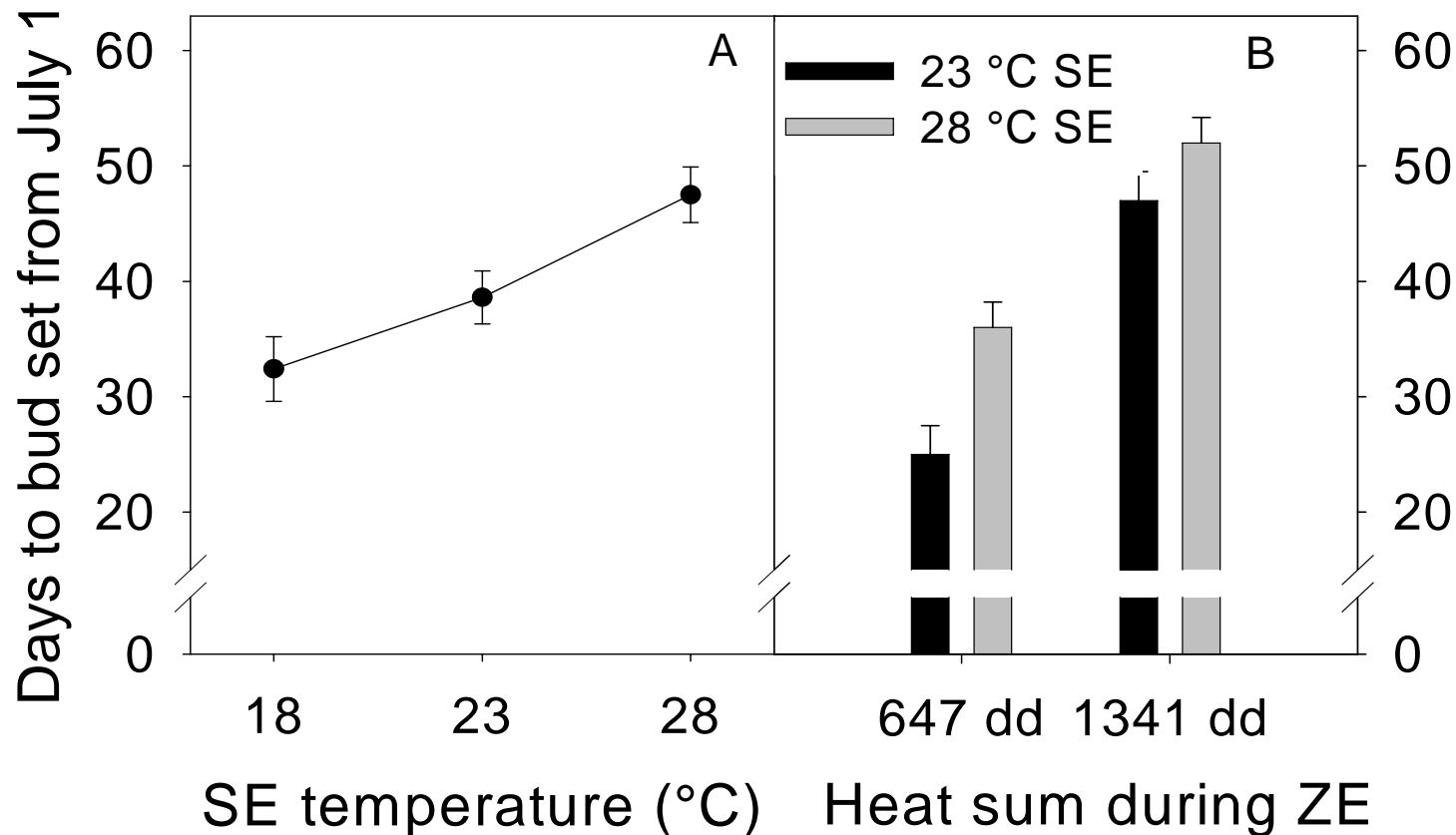
Applied temperature treatments during sub-culturing and maturation, and then tested the regenerated clones

Kvaalen and Johnsen 2008

# Memory from embryo development



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- > The memory of temperature is expressed the 2nd year
- > The difference in bud set is equal to provenance separation of six latitudinal degrees (from 58° - 64°N)

# Family variation in the memory

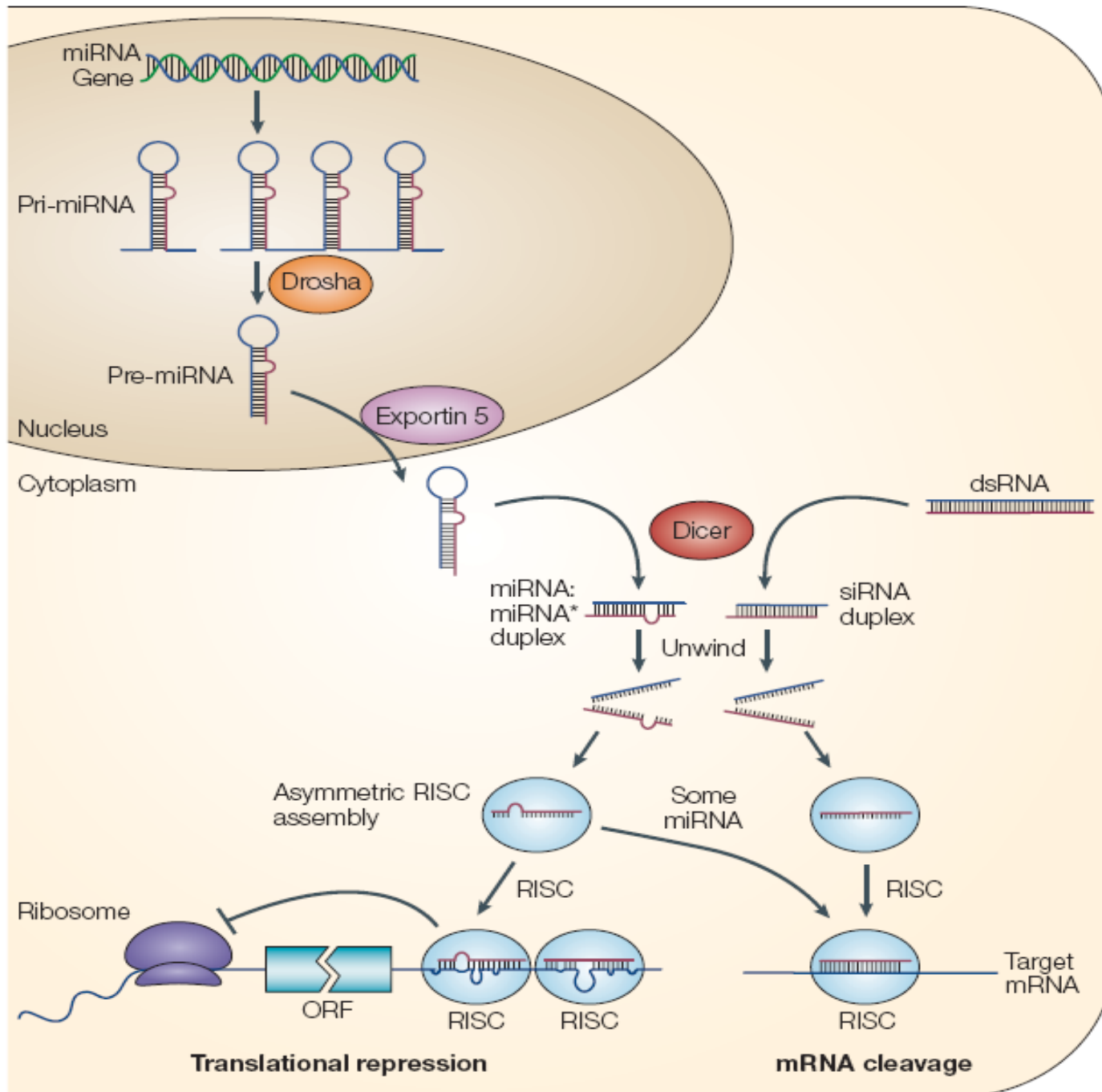
Made suppressive subtraction hybridization (SSH) cDNA libraries after 6 short days, and studied transcription of 34 of these ESTs

We found three unknown “genes” and two putative reverse transcriptases



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# microRNAs



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- > Either block the transcription of genomic regions by DNA methylation and/or histone methylation
- > Or block translation of the gene in the cytosol
- > The short sequences are templates of particular genes and could indicate the target genes on which they exert their regulation
- > If the sequence is known, one might find the (genomic) sequence of the gene and study if it is methylated



# miRNAs in Norway spruce

Sequenced **328** colonies

In total we obtained **199** different small RNAs,

**122** from warm embryonic library and **93** from cold embryonic library, **16** shared

Most of the small RNAs did have a dominating length of 21 nt



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# Summary for microRNAs



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- **We identified 24 new and 4 conserved miRNAs in Norway spruce**
- **Nearly all genes targeted by miRNA were unknown**
- **Most of the miRNAs were differentially expressed in relation to whether the siblings originates from cold crossing environment (CE) and warm crossing environment (WE)**
- **Some of the miRNAs are expressed differently in relation to the CE vs. WE in the epigenetically responding full-sib family compared to what we see in the non-responding full sib family**
- **Further studies of miRNAs will be exciting!**

# New project financed from June 2009

- > Cooperation with Prof. Jorunn Olsen, UMB
- > Using spruce micro-arrays to find more candidate genes in cooperation with Prof. Sally Aitken, UBC, Vancouver
- > Continue to study microRNAs
- > Find full length genomic sequences of the candidates
- > Study DNA methylation
- > We offer a post-doc position to work on these matters....  
do you know some good candidates?