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## ► To cite this version:

Katri Kärkkäinen, Luc Pâques. Impact of high throughput phenotyping on estimation of genetic parameters. Designing Trees for the Future:Data are the keystone, Natural Resources Institute Finland (Luke). FIN., Apr 2016, Bruxelles, Belgium. hal-02797852

**HAL Id: hal-02797852**

**<https://hal.inrae.fr/hal-02797852>**

Submitted on 5 Jun 2020

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## Impact of high throughput phenotyping on estimation of genetic parameters

Katri Kärkkäinen, Luke  
Luc Paques, INRA



## Impact of high throughput phenotyping on estimation of genetic parameters

- High throughput phenotyping?
- Why?
- What?
- How?
- When?

## High throughput phenotyping?

Forest Research:

**phenology,**

Sex reproduction,

**Frost and drought resistance**

**Pest resistance**

Annual radial growth

Root characteristics

Stem architecture

**Wood properties**

(some examples from bolded ones)



## WHY?

### Aim:

- Tree breeding
  - Assessing optimal deployment of forest regeneration material
  - Assessing evolution of tree populations
- Estimation of genetic parameters
- Estimation of genetic basis of phenotypes

## Estimation of genetic parameters

Phenotypic variance,

$$V_P = V_A + V_D + V_I + V_E$$

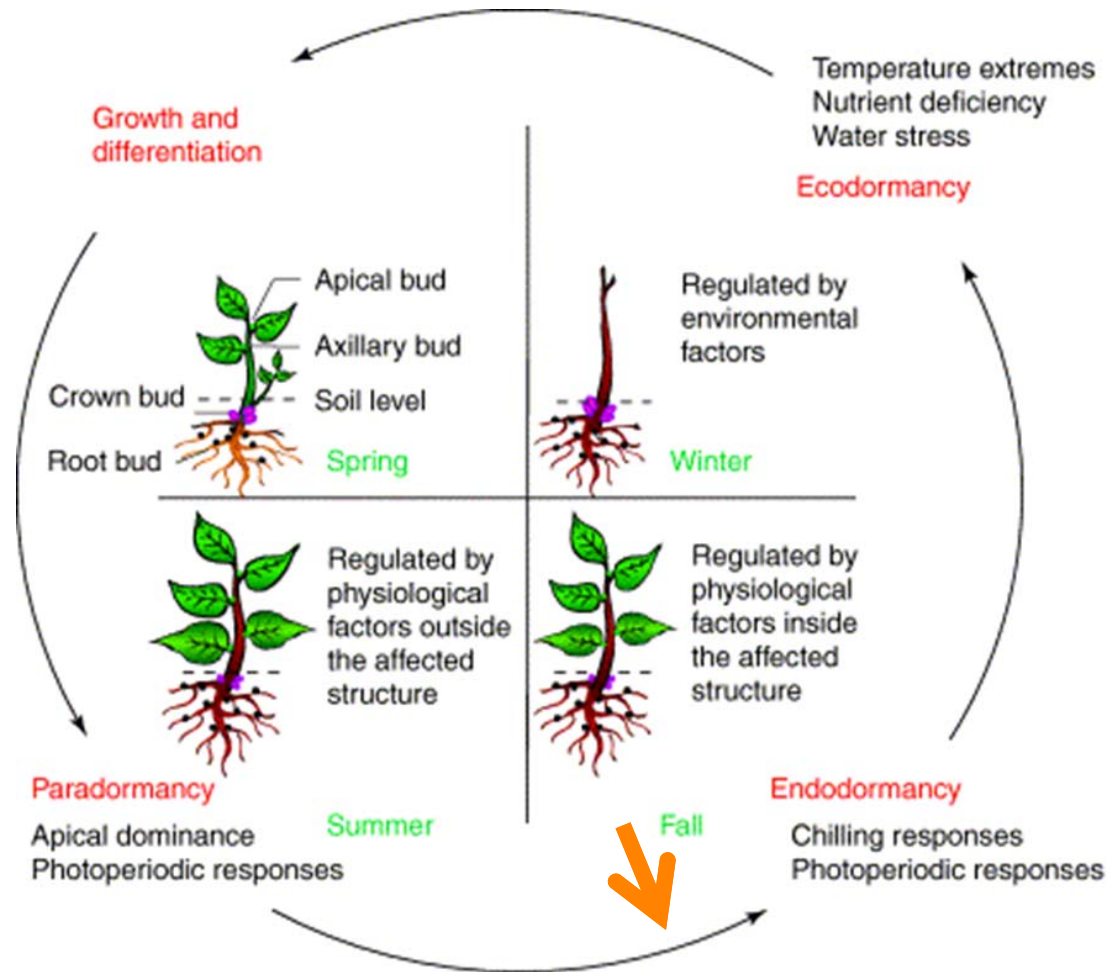
Additive value

Environmental deviation

"true" environmental variation  
+ measurement errors

$$h^2 = V_A/V_P$$

## WHAT? Aim: How to identify the most suitable trait? Case 1: Phenology – selection on growth rhythm



TRENDS in Plant Science

## Aim: How to identify the most suitable trait?

Case 1: Phenology – selection on growth rhythm:  
Cessation of growth (95% of final height) vs. bud-set,  
greenhouse study





## Cessation of growth (when 95% of final height reached) and timing of budset

### Variance components and genetic parameters for cessation of growth

		Variance components				Genetic parameters					
Population	Seedlings	Mean	$\widehat{\sigma}_f^2$	$\widehat{\sigma}_i^2$	$\widehat{\sigma}_e^2$	$V_A$	$CV_A$	$V_P$	$CV_P$	$h^2$	SD $h^2$
Punkaharju	11919/12000	96.04	2.71	0.00	66.27	10.83	0.03	68.98	0.09	0.16	0.02

### Variance components and genetic parameters for budset

		Variance components				Genetic parameters					
Population	Seedlings	Mean	$\widehat{\sigma}_f^2$	$\widehat{\sigma}_i^2$	$\widehat{\sigma}_e^2$	$V_A$	$CV_A$	$V_P$	$CV_P$	$h^2$	SD $h^2$
Punkaharju	11884/12000	102.99	10.00	0.71	73.33	39.99	0.06	84.03	0.09	0.48	0.04

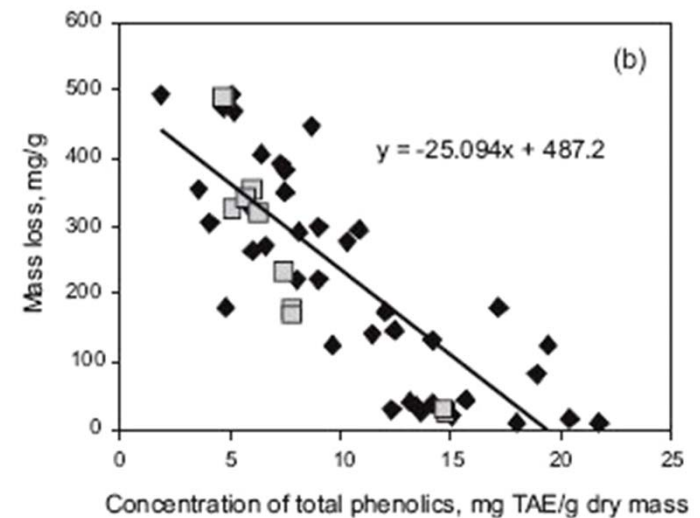
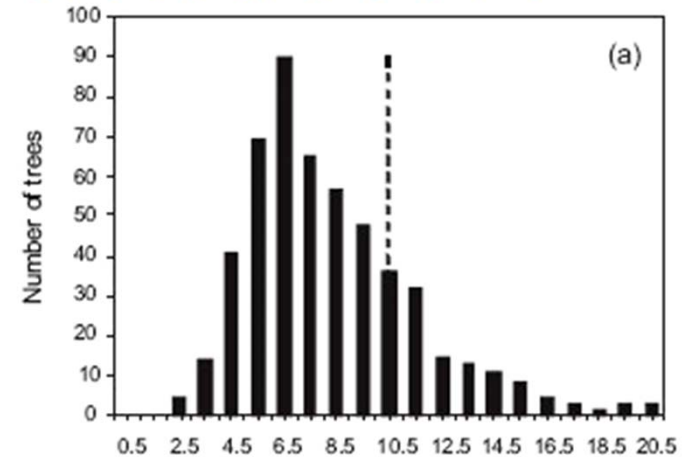
## WHAT? Aim: How to identify the most suitable trait?

Case 2: Decay resistance of timber

Decay tests: correlation between decay resistance and concentration of phenolics

### Conclusions:

Decay tests necessary, but phenotyping may be more efficient using chemical traits



Total phenolics



## How?

Aim: High-throughput phenotyping

- Non-destructive
- Low cost, fast
- Optical analyses



## How? Aim: High-throughput phenotyping

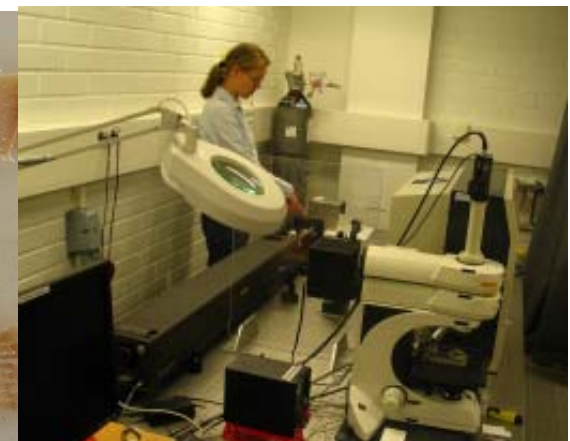
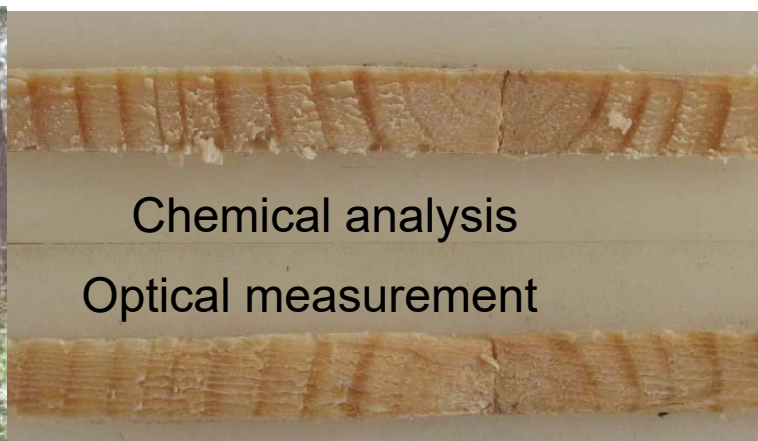
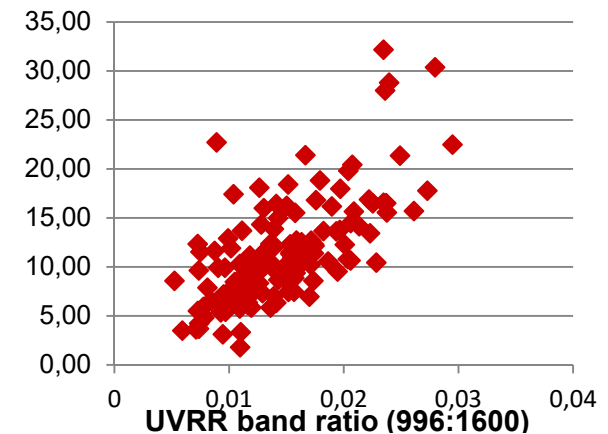
Case 1: Heartwood extractives in Scots pine: wet chemistry vs. two optical methods

Parallel samples for

GC-MS (wet chemistry; Luke)

UVRRaman (Aalto Univ)

NIRS (INRA, Innventia)



## Aim: High-throughput phenotyping

Case 1: Heartwood extractives in Scots pine: wet chemistry vs. two optical methods

Method	Va	h2	Mean	CVa	SD(h2)
GC	14,15	0,85	11,30	0,33	0,30
UVRaman	6,94	0,33	10,14	0,26	0,21
NIRS	12,93	0,84	11,39	0,32	0,29



## Aim: High-throughput phenotyping

Case 2: Heartwood extractives in larch:

### Objective:

Evaluate variability of extractives content in relation to natural durability

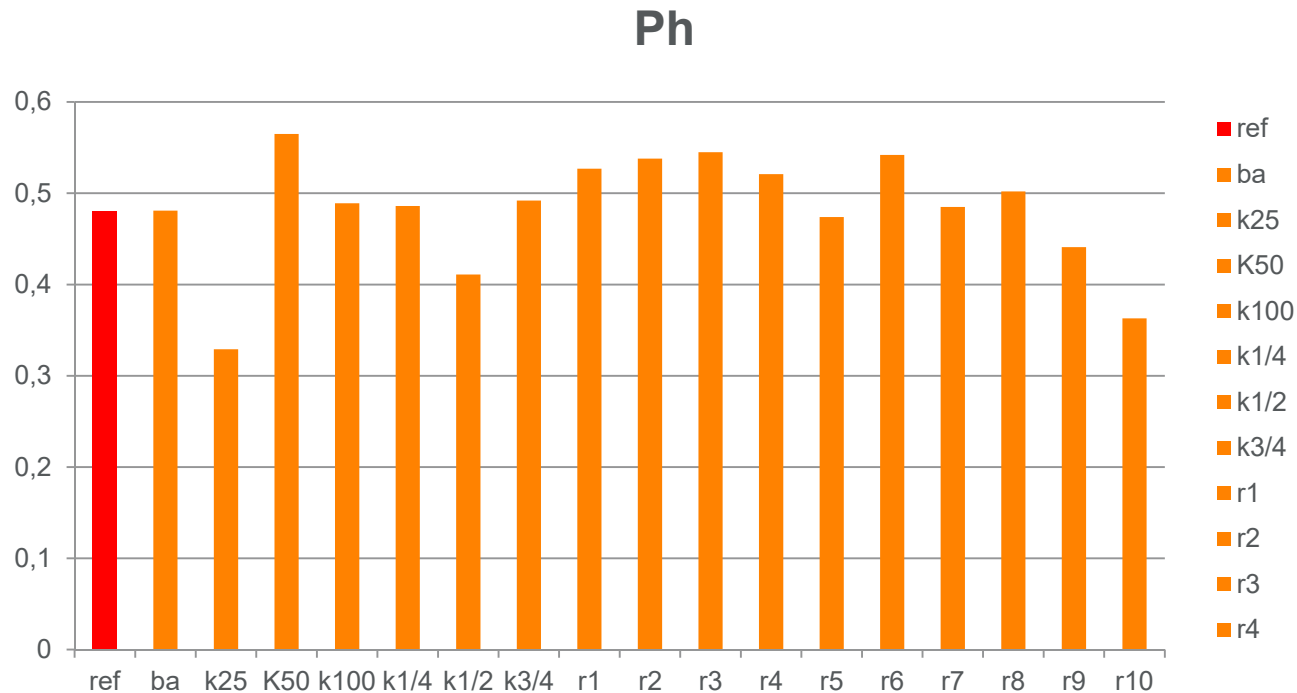
Develop a NIRS calibration model for both extractives and natural durability

**Material:** 42 FS progenies from 12 EL x 8 JL

355 individuals (out 1302 increment cores collected), +/- 8.4 sibs/family

**NIRS spectra and calibration models**

## Family heritability variation: wet chemistry (red) vs. optical analyses: impact of different calibration models



Ref: Reference HPLC (wet chemistry)

1) batch:  $\frac{1}{4}$  used to build up the model, prediction of the next  $\frac{3}{4}$ .

2-7) sets of calibration made of 25, 50, 100,  $\frac{1}{4}$ ,  $\frac{1}{2}$  and  $\frac{3}{4}$  of individuals

8-17) 10 random samples of 100 individuals in the training set

## How? Aim: High-throughput phenotyping

Case 3: Physical wood characteristics in larch: towards high-throughput phenotyping

Material:

- 25 FS hybrid larch progenies (~10 trees/family)

Measurements:

- Pilodyn: 2 directions; in 1993 (age 11); mm
- Density (from microdensitometry): ring 1989-1995 (age 13); kg/m<sup>3</sup>
- MOE (from rigidimeter on standing trees): in 1998 (age 16); MPa



## Case 3: Physical wood characteristics in larch: towards high-throughput phenotyping

Level of throughput (trees around 20-25 yrs)

Technique	Operation	Timing	Throughput Nber of samples per M/D	'efficiency' towards MOE
MOE (rigidimeter)	Pruning & measure	20-25 trees/D Need 2 persons	12	1
Pilodyn	Measure	250 trees/M/D	250	21
Infradensity (cores)	Collect Measure	100 cores/M/D 200 cores/M/D	70	6
Microdensitometry	Collect Sawing RX Windendro	100 core/M/D 50 cores/M/D 150 /M/D 50/M/D	18	1.5

M = person

D = day

## Estimation of heritability (family level) and of direct and indirect genetic gains from family selection

Heritability, absolute and relative gains for direct FS family selection ( $i=1$ )

	Mean	$\sigma_{pFS}$	$H^2_{FS}$	$\Delta G$ abs	$\Delta G$ rel (%)
MOE (MPa)	7169.827	819.378	0.706 (0.500-0.851)	578.48	8.1
Density (Kg/m <sup>3</sup> )	411.764	19.658	0.726 (0.534-0.861)	14.27	3.5
Pilodyn (mm)	14.312	1.325	0.678 (0.452-0.837)	0.898	6.3

## Case 3: Physical wood characteristics in larch: towards high-throughput phenotyping

Correlated response

Target trait: MOE

	Correlated gain with MOE (MPa) (i=1)	Selection efficiency	i Due to throughput level	Correlated gain with MOE (MPa)	Selection efficiency
Pilodyn	394.612	0.682	2.459	970.351	1.677
Infradensity	375.786*		1.96	736.541	1.273
Density	375.786	0.650	1.271	477.624	0.826

\* Supposed gain equal to that from microdensity

## WHEN?

→ Phenotypic variation in different life stages

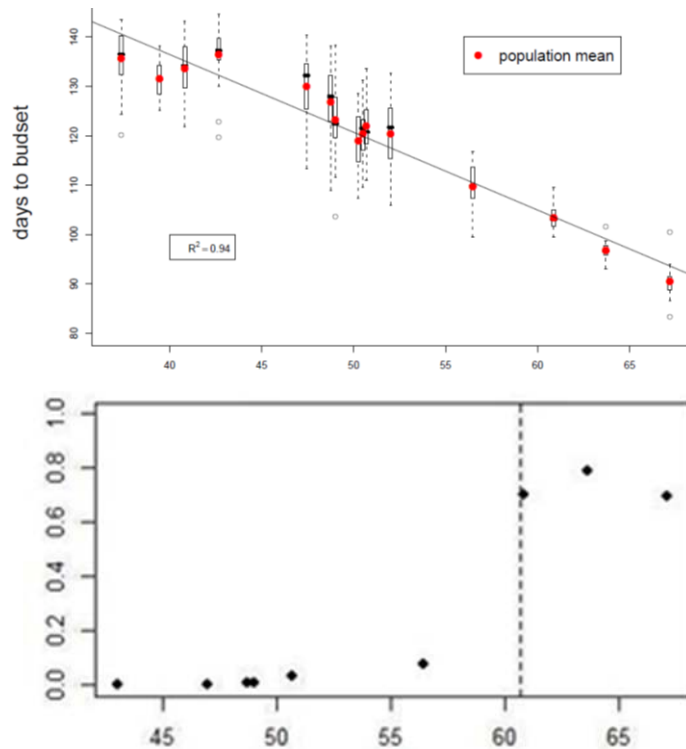


→ Phenotyping when it matters

## When? Correlation between phenotypic variation in different life stages

Case 1: Growth rhythm in different life stages

Bud-set in greenhouse of 1<sup>st</sup> yr seedlings v. survival (10 yrs) in field



Apuli et al, MS

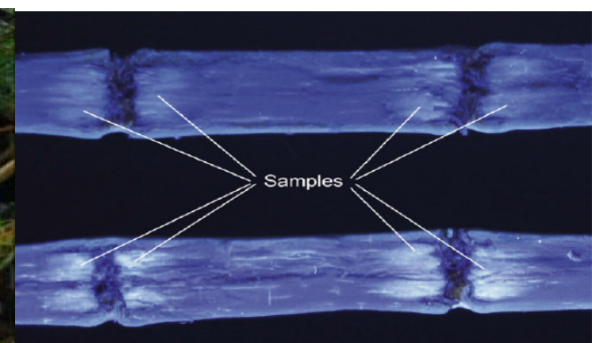
## When? Correlation between phenotypic variation in different life stages

Case 2: Can induced defense of seedlings used to assess constitutive heartwood extractives?



Extractive (mg g <sup>-1</sup> d.w.)	$h_{OP}^2$
PS	0.31
PSM	0.09
Sum of stilbenes	0.20
Sum of RAC	0.13

Harju et al 2008. Tree Physiol.



## Conclusions

High-throughput phenotyping

Why?

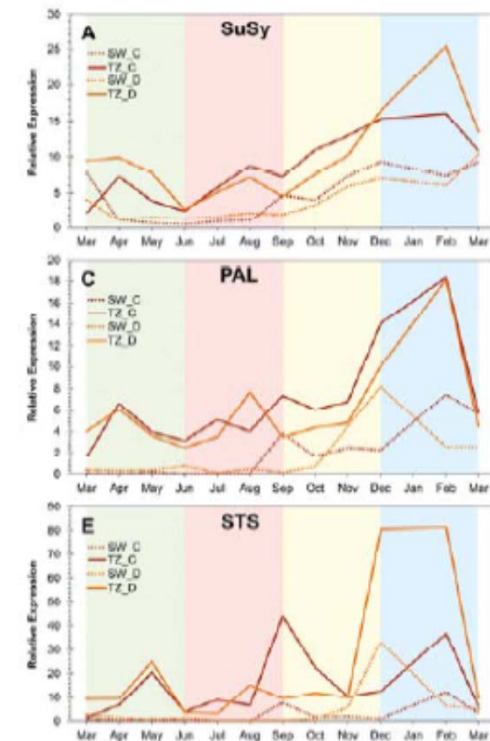
What?

How?

When?

Important to plan well beforehand

Future demands: more detailed phenotypes for genomic selection



A photograph of a forest with tall, thin trees and a stack of logs in the foreground. The text "Thank you" is overlaid in the center.

Thank you