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Introduction

Poplar wood is a renewable resource of interest for producing **second generation biofuels**. However, current varieties have not been bred for this purpose. Poplar **wood chemical properties** need thus to be studied in order to optimize its conversion into sugars (**saccharification**). **IR spectroscopy** is relevant for this purpose since it allows a **high-throughput** indirect characterization of the physical and chemical properties of the samples of interest whose number is typically very large in breeding programs.

In this context, this study aimed at defining a relevant way of collecting IR spectra for calibrating poplar **wood chemical composition** and **saccharification potential**.

Material & Methods

Four different conditions of acquisition of spectra have been tested on a calibration set of 116 *Populus nigra* samples using a spectrum 400 Perkin Elmer spectrophotometer: **1) NIR spectra** (8000-4000 cm⁻¹) averaged over 64 scans collected with a **rotating system in a quartz cup**; **2) NIR spectra** (8000-4000 cm⁻¹) averaged over **3 independent collections of scans in glass vial**; **3) NIR spectra** (8000-4000 cm⁻¹) from a **single collection of scans in glass vial**; and **4) MIR spectra** (1800-900 cm⁻¹) from a single scan.

These 116 samples from the calibration set have also been phenotyped for **12 quantitative traits** related to their chemical composition and saccharification potential.

For each condition of acquisition and each phenotype, the calibration procedure was carried out in 2 steps: **1) selection of relevant spectral bands** from the IR spectra matrix in order to decrease its complexity, using the Competitive Adaptive Reweighted Sampling (CARS) approach (1); and **2) on the resulting dataset, calibration model construction** using partial least squares (PLS) regression in a 4-fold cross-validation scheme repeated 500 times (**Monte Carlo Cross-Validation, MCCV**), where the optimal number of latent variables was determined using a Wold's R criterion equal to one (2). The best model among all 8 modalities was determined for each combination of phenotype and acquisition condition on the basis of both the **MCCV statistics** and the **number of latent variables**. Afterwards, for each phenotype the best models of each acquisition condition were **compared** and **ranked** according to the same criteria (MCCV statistics and number of latent variables).

All analyses were carried out in R (R Development Core Team, 2012).

Results and Discussion

The best calibration models for each acquisition condition are presented in **Figure 1** for the proportion of glucose in solubilized sugars, and in **Table 1** for all phenotypes related to wood chemical composition and to saccharification potential.

For each phenotype, models have been compared between acquisition conditions on the basis of the calibration statistics. The resulting ranks are graphically presented for all phenotypes in **Figure 2**. This ranking suggested that an average spectrum over **3 independent acquisitions** in a **glass vial** was the **most promising approach** for calibrating our phenotypes. In a contrast, a **single collection in a glass vial** gave the **worst performances**, while NIR spectra from the **rotating system** and **MIR spectra** had **intermediary performances**.

These comparisons might nevertheless be taken with care because the phenotypes used in the present study were not completely independent, with significant correlations between some of them (data not shown). It would thus be interesting to extend and confirm the present results on other phenotypes that would not be correlated with the ones used in the present study.

The present results allow us to conclude that, despite the fact it allows analyzing a small amount of material, a spectra acquisition from a single scan in a glass vial does not seem satisfactory for our purpose and **spectra averaged over multiple collections of scans** either in a rotating cup or preferably in a glass vial should be advised.

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Figure 1. Best calibration models for the proportion of glucose in solubilized sugars for each spectra acquisition condition

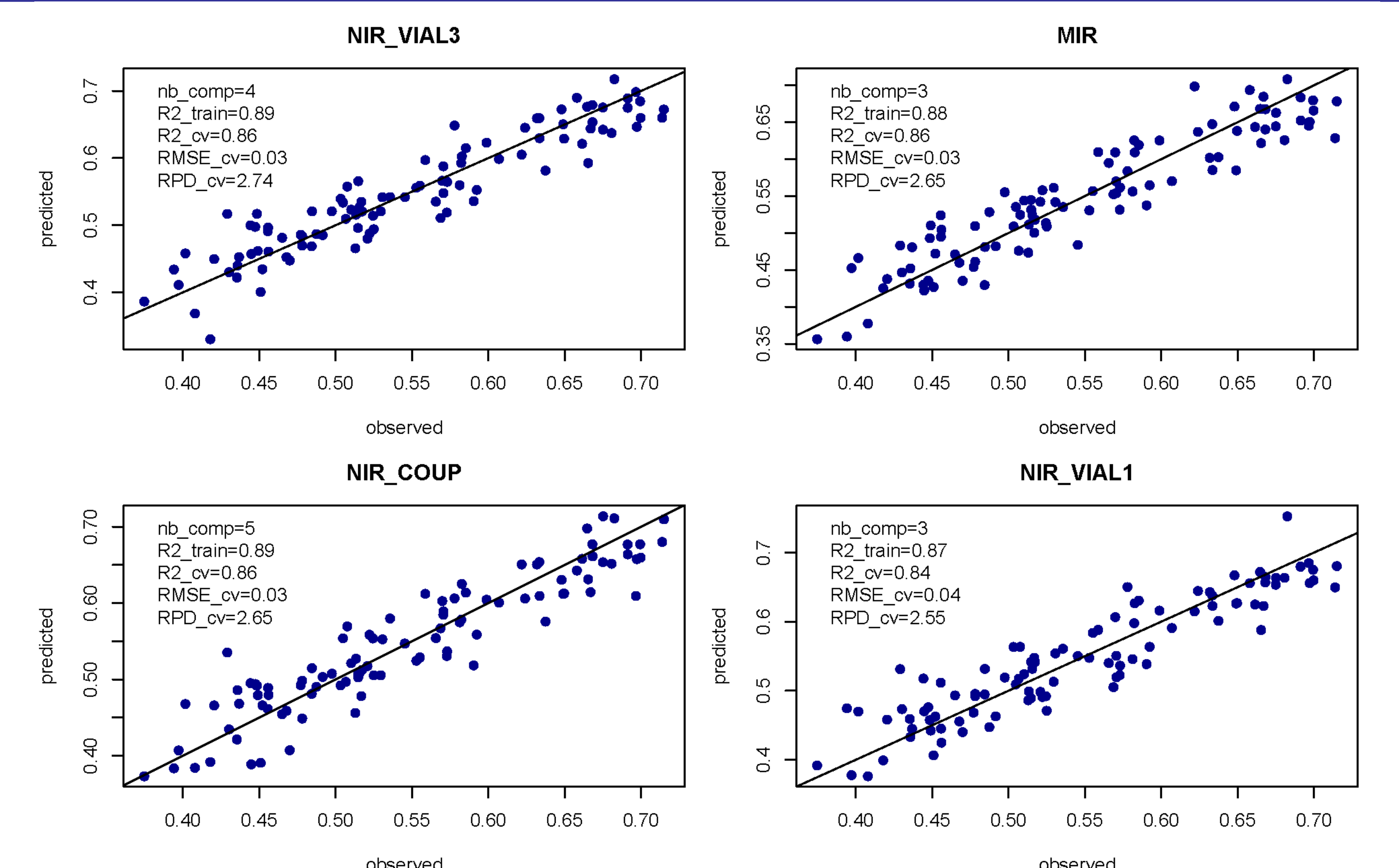
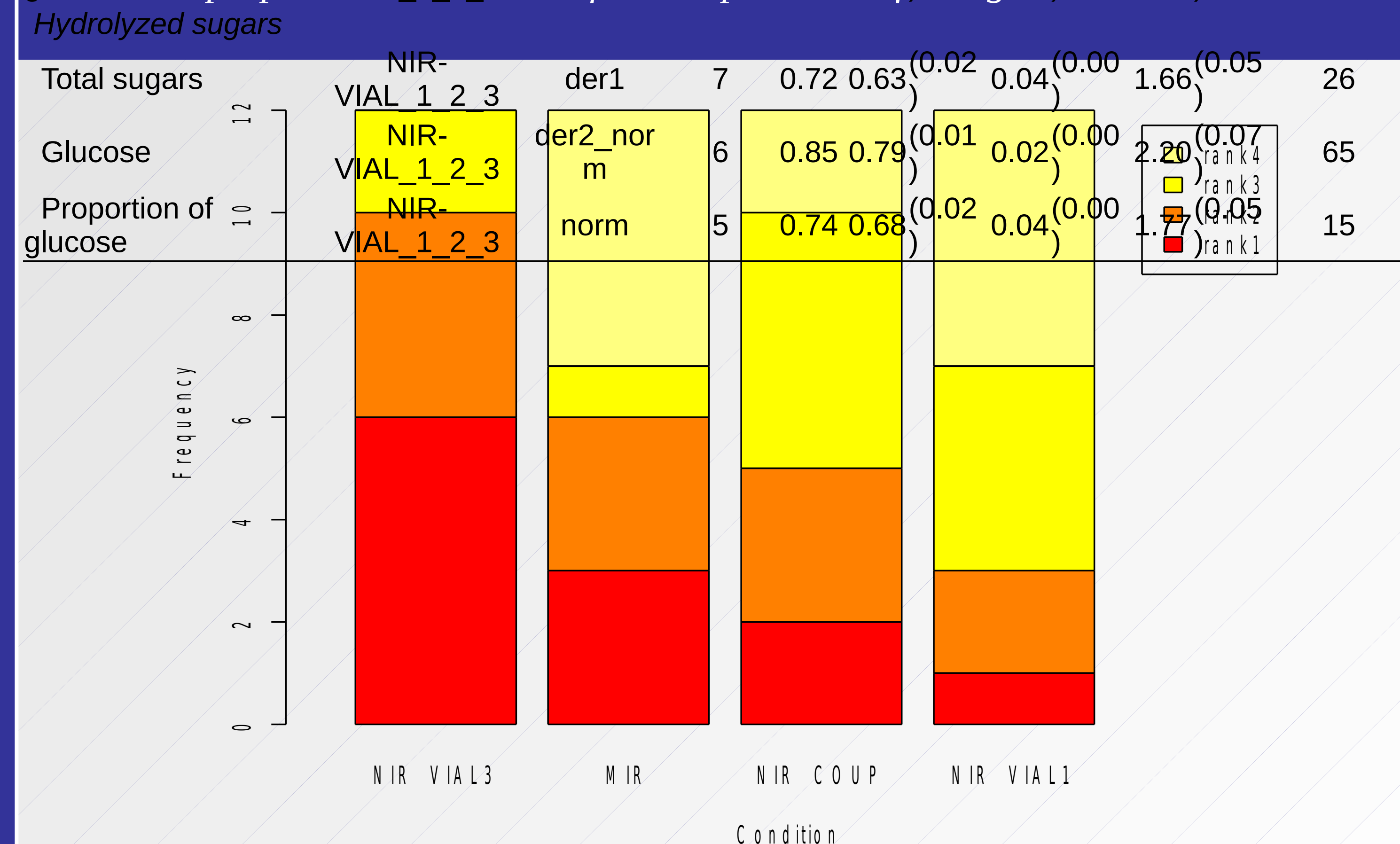


Table 1. Best calibration models for phenotypes related to the chemical properties and saccharification potential of *P. nigra* wood.

comp: number of latent variables; R²train: training R²; R²cv: cross-validation R²; RMSEcv root mean square error of cross validation; RPDcv: Ratio of standard error of performance in cross validation to standard deviation; # lambda: number of wave numbers selected by CARS.

Trait	Condition	Treatment	# Comp	R ² tra in	R ² CV	RMSECV	RPDCV	# Lambda
Chemical composition								
Extractives	NIR-COUP	der1_norm	7	0.85 0.79	(0.01)	1.25(0.04)	2.20(0.07)	37
Lignins	NIR-COUP	der2_norm	4	0.72 0.65	(0.02)	0.95(0.03)	1.69(0.05)	335
Holocellulose	NIR-VIAL_1	norm_der1	6	0.81 0.73	(0.02)	1.28(0.04)	1.95(0.06)	40
Saccharification potential								
<i>Soluble sugars</i>								
Total sugars	MIR	norm	5	0.95 0.93	(0.00)	0.02(0.00)	3.91(0.11)	50
Glucose	MIR	norm	6	0.94 0.92	(0.01)	0.00(0.00)	3.50(0.11)	98
Proportion of glucose	MIR	norm_der2	5	0.88 0.86	(0.01)	0.05(0.00)	2.65(0.08)	61
<i>Solubilized sugars</i>								
Total sugars	NIR-VIAL_1_2_3	norm	6	0.78 0.73	(0.02)	0.04(0.00)	1.94(0.05)	48

Figure 2. Graphical representation of the frequency of calibration model ranks depending on the spectra acquisition condition over 12 phenotypes related to the chemical properties and saccharification potential of *P. nigra* wood.



References

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