

# A Closed-Loop Biorefinery using Renewable Deep Eutectic Solvents with Engineered Biomass

## Background

- Down-regulation of cinnamyl alcohol dehydrogenase (*CAD*) genes results in the formation of an abnormal and potentially valuable aldehyde-rich lignin.
- Deep eutectic solvents (DESs) have gained increasing attention due to their application-friendly properties, including universal solvating capabilities and wide tunability.

## Approach

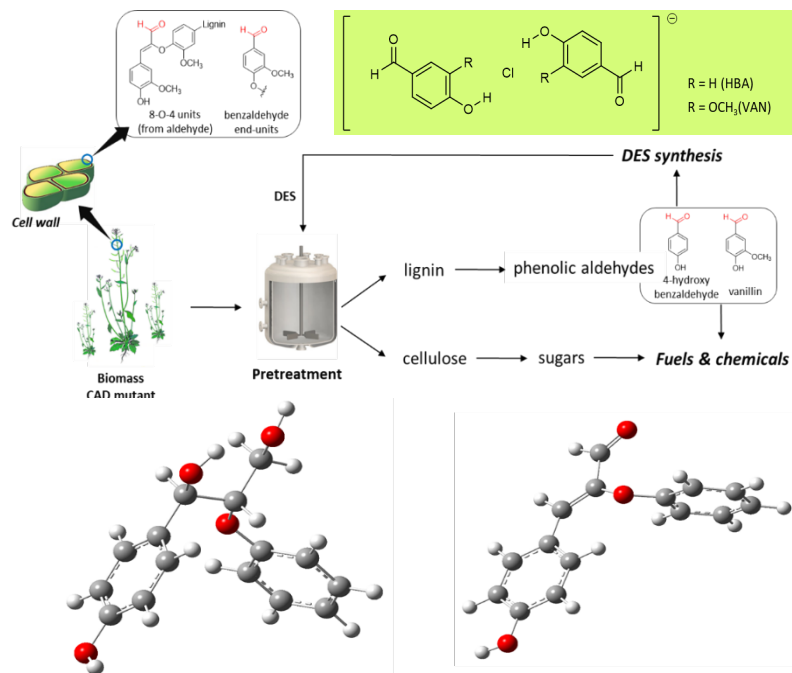
- *CAD*-down-regulated plants is pretreated with DESs derived from phenolic aldehydes.
- The recovered lignin after pretreatment can be further processed to extract phenolic aldehydes for the production of renewable biomass-based DESs.

## Outcome

- 8-O-4 dimer from *CAD* mutant has a higher electrophilicity index, indicating that this structure is chemically more reactive than the normal  $\beta$ -O-4 structure
- The transgenic *Arabidopsis thaliana CAD* mutant was pretreated with the DESs and showed a twofold increase in the yield of fermentable sugars compared with wild type (WT) upon enzymatic saccharification.

## Significance

- Integration of renewable lignin-derived DES with low-recalcitrant genetically engineered biomass is a promising strategy in developing a closed-loop process.



(a)  $\beta$ -O-4 dimer from WT

Electrophilicity index: 1.88 eV

(b) 8-O-4 dimer from CAD mutant

Electrophilicity index: 3.00 eV

