

SI - nonstructural carbon reserves

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Calculating nonstructural carbon reserves for the 2017 experiment at Harvard Forest

To estimate changes in soluble sugars (here we refer to only glucose and) and starch (as glucose equivalent) for various tissue, we scaled the concentrations derived from each tissue using colorimetric assays as described in the methods. Below we describe the scaling assumptions for nonstructural reserves in a stem section, as well as estimates at the organ-level for the stem, roots and leaves. The differences of total values between the four sampling points in turn provide estimates of net gains or losses of soluble sugars and starch for each stem section, organ and tree. To scale concentrations that were derived from measurements, we estimated the dry matter for each stem section, organ and tree and multiplied it by the nonstructural carbon concentration measured for that tissue similar to Furze *et al.* (2019). Below we provide examples for each tissue.

For stem sections

We measured nonstructural carbon concentrations for the first centimeter of the stem core (from the cambium towards the pith) as detailed in the methods section. While nonstructural carbon concentrations follow a generally decreasing gradient from the bark towards the pith [Hoch, Richter and Körner (2003);furze_whole-tree_2018], we only consider nonstructural carbon reserves in the first centimeter underlying the bark for each stem section. We limit our estimation to the first centimeter, because deeper reserves are more difficult, and thus less likely, to be accessed on timescales considered here.

Assuming that the bulk of easily accessible nonstructural carbon can be found in the first centimeter and that our measurements are an average of these reserves, we derive the total amount of nonstructural carbon in sugars and starch ($M_{sugar,i}$ and $M_{starch,i}$, respectively) by multiplying the mass of the hollow cylinder ($M_{s,i}$) of each stem section i with the section's measured concentrations of sugar and starch ($c_{sugar,i}$ and $c_{starch,i}$, respectively).

$$M_{sugar,i} = c_{sugar,i} \times M_{s,i} = V_{s,i} \times \rho_w \times c_{sugar,i}$$

The mass of a hollow cylinder representing the outermost centimeter of material is derived from its volume ($V_{s,i}$) and an average density of the wood of ... (ρ_w). In turn the volume calculated using a height (h) of 10 cm and the section's circumference ($cbh_{s,i}$; in *cm*), which was measured in the field using a tape measure. We use $289.9 \pm 40.4 \text{ kg m}^{-3}$ as density, which is the mean \pm standard deviation of wood density of 16 cores from nine trees in the control group as determined using x-ray film.

$$M_{s,i} = V_{s,i} \times \rho_w = \pi \times h \times \rho_w \left(\left(\frac{cbh_{s,i}}{2\pi} \right)^2 - \left(\frac{cbh_{s,i}}{2\pi} - 0.01 \right)^2 \right)$$

```
# Get the tree measurements of circumference from the googlesheet
key <- '1K8Vk9VHRdwNjIa28frc-Zt4uyk07G6CZe1EkJunU1l0'
suppressMessages (predictors <- gs_read (gs_key (key), ws = 'predictors', col_types = cols ()))

# Declare the height of the stem section (h, m)
h <- 0.1

# Declare mean stem wood density (rho_s, kg m-3)
```

```
rho_w <- 289.9
```

```
# Determine the volume of each stem section (m3) at various height (0.5m, 1m, 1.5m, 2m, 2.5m)  
# cbh is in cm, therefore divide by 100 and by 2.0 and pi to get the sections radius in m
```

```
predictors <- add_column (predictors,  
  V50 = ((predictors[['cbh50']]/(100.0*2.0*pi))^2.0*pi*h) -  
        ((predictors[['cbh50']]/(100.0*2.0*pi)-0.01)^2.0*pi*h),  
  V100 = (predictors[['cbh100']]/(100.0*2.0*pi))^2.0*pi*h -  
         ((predictors[['cbh100']]/(100.0*2.0*pi)-0.01)^2.0*pi*h),  
  V150 = (predictors[['cbh150']]/(100.0*2.0*pi))^2.0*pi*h -  
         ((predictors[['cbh150']]/(100.0*2.0*pi)-0.01)^2.0*pi*h),  
  V200 = (predictors[['cbh200']]/(100.0*2.0*pi))^2.0*pi*h -  
         ((predictors[['cbh200']]/(100.0*2.0*pi)-0.01)^2.0*pi*h),  
  V250 = (predictors[['cbh250']]/(100.0*2.0*pi))^2.0*pi*h -  
         ((predictors[['cbh250']]/(100.0*2.0*pi)-0.01)^2.0*pi*h))
```

```
# Determine the mass of each stem section (Msection, kg)
```

```
predictors <- add_column (predictors,  
  M50 = predictors [['V50']] * rho_w,  
  M100 = predictors [['V100']] * rho_w,  
  M150 = predictors [['V150']] * rho_w,  
  M200 = predictors [['V200']] * rho_w,  
  M250 = predictors [['V250']] * rho_w)
```

```
# Read the sugar and starch concentration (means for needles, branches and roots and first centimeter of  
suppressMessages (source ('processNSCDataForExp2017.R'))
```

```
## Warning in processNSCs(rawData = rawData3, cvLimitSample = 0.25,  
## cvLimitTube = 0.25, : Warning: There is no LCS Oak for batch 9 analysed on  
## the 2019-03-21.  
  
## Warning in processNSCs(rawData = rawData3, cvLimitSample = 0.25,  
## cvLimitTube = 0.25, : Warning: There is no LCS Oak for batch 10 analysed on  
## the 2019-03-21.  
  
## Warning in processNSCs(rawData = rawData3, cvLimitSample = 0.25,  
## cvLimitTube = 0.25, : Warning: There is no LCS Oak for batch 11 analysed on  
## the 2019-03-21.  
  
## Warning in processNSCs(rawData = rawData3, cvLimitSample = 0.25,  
## cvLimitTube = 0.25, : Warning: There is no LCS Oak for batch 12 analysed on  
## the 2019-03-21.  
  
## Warning in processNSCs(rawData = rawData4, cvLimitSample = 0.25,  
## cvLimitTube = 0.25, : Warning: 2 samples are excluded because their sample  
## mass is below the threshold of 5 mg.
```

```
# Create responseVariables tibble
```

```
responseVariables <- tibble (year = 2017,  
  month = c (rep (7, 41), rep (8, 41),  
             rep (10, 41), rep (11, 41)),  
  tree = rep (1:41, 4),  
  sugarW250 = NA, starchW250 = NA, sugarW200 = NA,  
  starchW200 = NA, sugarW150 = NA, starchW150 = NA,  
  sugarW100 = NA, starchW100 = NA, sugarW50 = NA,  
  starchW50 = NA)
```

```

# Wrangle data to get sugar and starch concentrations (g kg-1) for each section and
# multiply the concentration by the dry mass of the section (kg) to obtain total sugar and
# starch reserves (kg)
for (r in 1:dim (responseVariables) [1]) {
  treeID <- responseVariables [['tree']] [r]
  iMonth <- responseVariables [['month']] [r]
  if (unique (stemData [['treatment']] [stemData [['treeID']] == treeID]) == 1) {
    responseVariables [['sugarW150']] [r] <-
      predictors [['M150']] [predictors [['tree']] == treeID *
        (stemData [['sugar']] [stemData [['treeID']] == treeID &
          month (stemData [['date']] == iMonth) / 100) * 1000
  } else if (unique (stemData [['treatment']] [stemData [['treeID']] == responseVariables [['tree']] [r]) == 1) {
    responseVariables [['sugarW100']] [r] <-
      predictors [['M100']] [predictors [['tree']] == treeID *
        (stemData [['sugar']] [stemData [['treeID']] == treeID &
          stemData [['sampleHeight']] == 1 &
          month (stemData [['date']] == iMonth) / 100) * 1000
    responseVariables [['sugarW200']] [r] <-
      predictors [['M200']] [predictors [['tree']] == treeID *
        (stemData [['sugar']] [stemData [['treeID']] == treeID &
          stemData [['sampleHeight']] == 2 &
          month (stemData [['date']] == iMonth) / 100) * 1000
  } else if (unique (stemData [['treatment']] [stemData [['treeID']] == responseVariables [['tree']] [r]) == 2) {
    responseVariables [['sugarW50']] [r] <-
      predictors [['M50']] [predictors [['tree']] == treeID *
        (stemData [['sugar']] [stemData [['treeID']] == treeID &
          stemData [['sampleHeight']] == 0.5 &
          month (stemData [['date']] == iMonth) / 100) * 1000
    responseVariables [['sugarW150']] [r] <-
      predictors [['M150']] [predictors [['tree']] == treeID *
        (stemData [['sugar']] [stemData [['treeID']] == treeID &
          stemData [['sampleHeight']] == 1.5 &
          month (stemData [['date']] == iMonth) / 100) * 1000
    responseVariables [['sugarW250']] [r] <-
      predictors [['M250']] [predictors [['tree']] == treeID *
        (stemData [['sugar']] [stemData [['treeID']] == treeID &
          stemData [['sampleHeight']] == 2.5 &
          month (stemData [['date']] == iMonth) / 100) * 1000
  }
}
}

```

Total above ground biomass

According to Jenkins *et al.* (2004), we calculated total aboveground biomass (B ; in kg) from the diameter at breast height of tree t (d_t ; in m) as follows:

$$B = \exp(\beta_0 + \beta_1 \ln d_t)$$

```

biomass04 <- exp (2.4800 + 2.4835 * log (predictors [['cbh150']] / pi))
#biomass08 <- exp (-2.6177 + 2.4638 * log (predictors [['cbh150']] / pi)) # with specific gravity < 0.4
#biomass08 <- exp (-3.0506 + 2.6465 * log (predictors [['cbh150']] / pi)) # with specific gravity >= 0.4
#biomassxx <- 2.2874 * exp (0.1968 * (predictors [['cbh150']] / pi)) # From peichl

```

with generic pine species (*Pinus* spp.) parameters of $\beta_0 = -2.4800$ and $\beta_1 = 2.4835$. This total biomass includes foliage, stemwood, bark and branches. To get tissue specific values for stemwood, bark and foliage we estimated the fraction of the total biomass in each tissue using $\alpha_0 = \sim 0.3737$ and $\alpha_1 = \sim 1.8055$ from Table 2 in Jenkins *et al.* (2004) and the following equation:

$$f = \exp(\alpha_0 + \frac{\alpha_1}{d_t})$$

```
rootFraction <- exp (-1.5619 + ( 0.6614 / (predictors [['cbh150']] / pi)))
stemFraction <- exp (-0.3737 + (-1.8055 / (predictors [['cbh150']] / pi)))
barkFraction <- exp (-2.0980 + (-1.1432 / (predictors [['cbh150']] / pi)))
foliFraction <- exp (-2.9584 + ( 4.4766 / (predictors [['cbh150']] / pi)))
branFraction <- 1.0 - foliFraction - stemFraction - barkFraction
```

Branch biomass was calculated as the residual of total aboveground biomass and the three other tissues estimated above. Here, the mean stem, bark, branch and foliage fractions were 62.5%, 11.5%, 19.4%, and 6.6%, respectively.

$$f_{branches} = 1.0 - f_{foliage} - f_{stemwood} - f_{bark}$$

Root biomass was also estimated as a fraction of total aboveground biomass with an average of 21.7%.

For each organ and the entire tree we multiplied the respective biomass with the average sugar and starch concentration derived from colourimetric assays. The total nonstructural carbon estimates are the sum of the mass of sugar and starch.

```
foliageMass <- biomass04 * foliFraction
```

References

- Furze, M. E. *et al.* (2019) ‘Whole-tree nonstructural carbohydrate storage and seasonal dynamics in five temperate species’, *New Phytologist*, 221(3), pp. 1466–1477. doi: 10.1111/nph.15462.
- Hoch, G., Richter, A. and Körner, C. (2003) ‘Non-structural carbon compounds in temperate forest trees’, *Plant, Cell & Environment*, 26(7), pp. 1067–1081. doi: 10.1046/j.0016-8025.2003.01032.x.
- Jenkins, J. C. *et al.* (2004) *Comprehensive database of diameter-based biomass regressions for North American tree species*. NE-GTR-319. Newtown Square, PA: U.S. Department of Agriculture, Forest Service, Northeastern Research Station. doi: 10.2737/NE-GTR-319.