10.1071/FP10055_AC

Accessory Publication: Functional Plant Biology, 2010, 37(12), 1161–1174.

## Accessory Publication

**Table S1. Allocation of $^{14}C$ to various metabolite pools by N3.2 and Q117 cells after 4 h labelling period using exogenous [U-$^{14}C$] fructose in Day 1 and Day 4 cultures**

The total activity of each component is expressed as kBq $mg^{-1}$ protein. Each value is the mean ± s.d. of three separate samples. Values in parentheses indicate the percentage of total label allocated to the particular pool. Significant differences between cell lines, determined by *t* tests are marked: *, $P$ <0.05; **, $P$ < 0.01

| Components | $^{14}C$ distribution from a [U-$^{14}C$] fructose pulse | | | |
|---|---|---|---|---|
| | Day 1 culture | | Day 4 culture | |
| | Q117 | N3.2 | Q117 | N3.2 |
| Total uptake | 11.95 ± 1.69 (100%) | 16.88 ± 1.74* (100%) | 11.91 ± 1.34 (100%) | 18.31 ± 0.70** (100%) |
| $H_2O$-soluble | | | | |
| Sugars | 3.17 ± 0.61 (26.4%) | 6.08 ± 0.88** (36.0%)** | 3.26 ± 0.62 (27.3%) | 5.75 ± 1.07* (31.4%)* |
| Organic acids | 2.54 ± 0.18 (21.3%) | 3.14 ± 0.29* (18.6%)* | 2.18 ± 0.54 (18.3%) | 3.74 ± 0.49* (20.4%)* |
| Amino acids | 1.68 ± 0.19 (14.1%) | 1.99 ± 0.25 (11.8%) | 1.75 ± 0.56 (14.7%) | 1.83 ± 0.82 (10.0%) |
| $H_2O$-insoluble | 3.37 ± 0.42 (30.2%) | 3.61 ± 0.55 (21.4%) | 3.58 ± 0.21 (30.1%) | 4.56 ± 0.49* (24.9%)* |
| Lipid-soluble | 0.11 ± 0.04 (0.9%) | 0.15 ± 0.05 (0.9%) | 0.13 ± 0.07 (1.1%) | 0.15 ± 0.07 (0.8%) |
| $CO_2$ | 1.07 ± 0.09 (8.9%) | 1.79 ± 0.21* (10.6%)* | 1.07 ± 0.33 (9.0%) | 1.98 ± 0.26* (10.8%) |

The BioLC conditions for these sugar phosphate separations on a Dionex CarboPac PA10 column with PAD detection were: Eluent A: 1M NaOAc in 50 mM NaOH; Eluent B: 50 mM NaOH.

| Time (min) | Eluent A (vol%) | Eluent B (vol%) |
|---|---|---|
| 0 | 5 | 95 |
| 5 | 10 | 90 |
| 20 | 15 | 85 |
| 30 | 20 | 80 |
| 35 | 75 | 25 |
| 35.1 | 100 | 0 |
| 40.1 | 5 | 95 |
| 50 | 5 | 95 |

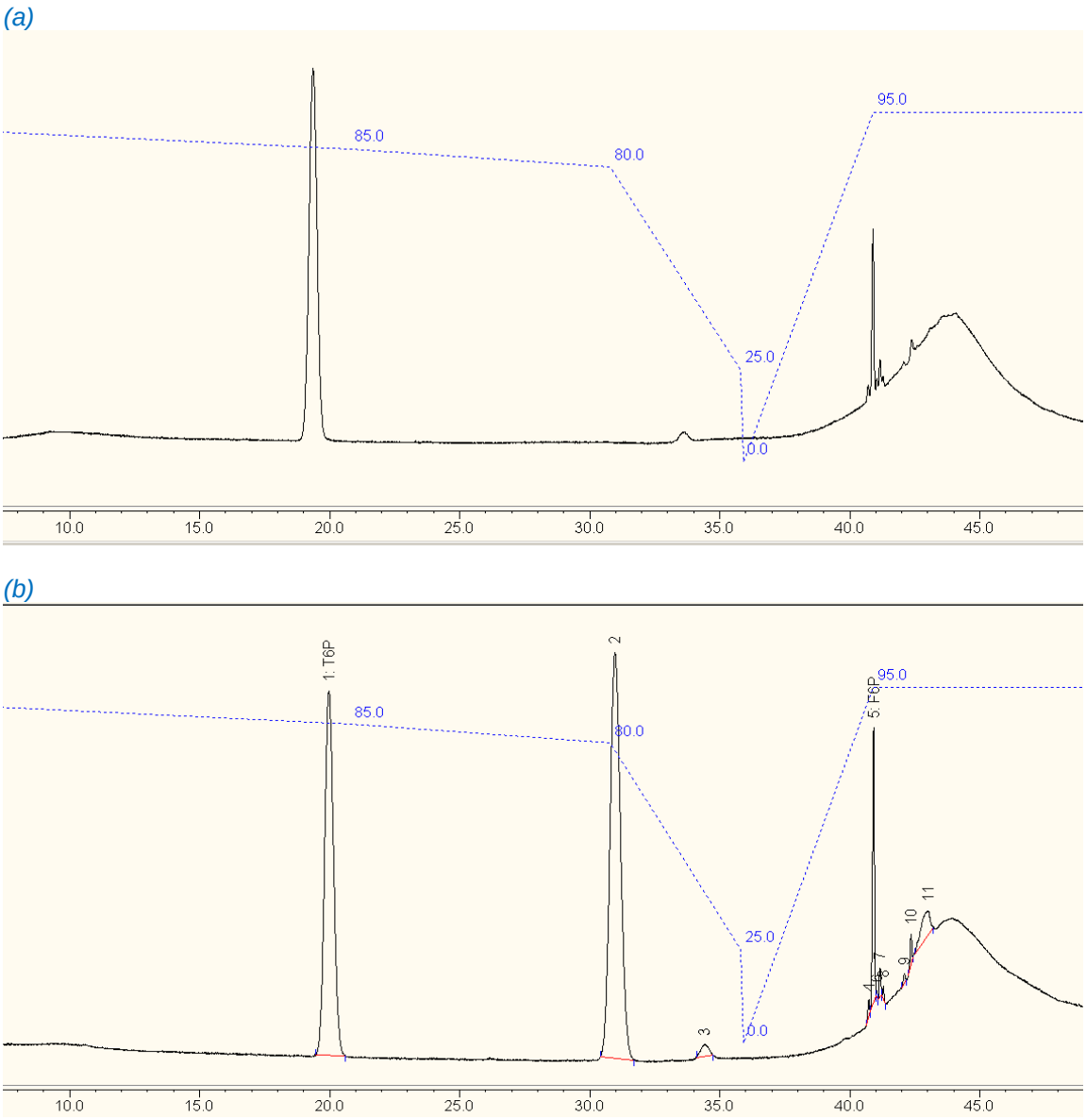


**Fig. S1.** Confirmation of T6P standard. (*a*) Chromatogram from HPAEC-PAD after 10 μL injection of 50 μM T6P standard (Sigma-Aldrich lot 1614848 specified as 100% pure by TLC assay). The major peak at 19.5 min was verified as T6P by mass spectrometry using an ABSciex 4000 QTRAP MS with TurboV electrospray source operated in negative-ion, multiple-reaction-monitoring mode. This yielded the expected major parent ion for T6P at $[M\text{-}H]^-$ 420.9 m/z and the phosphate ion fragment at 79.0 m/z. MS analysis in precursor ion scan mode indicated very high purity. (*b*) Aligned region of a chromatograph after injection of T6P and S6P standards, showing clear separation of these disaccharide monophosphates by approximately 11 min.

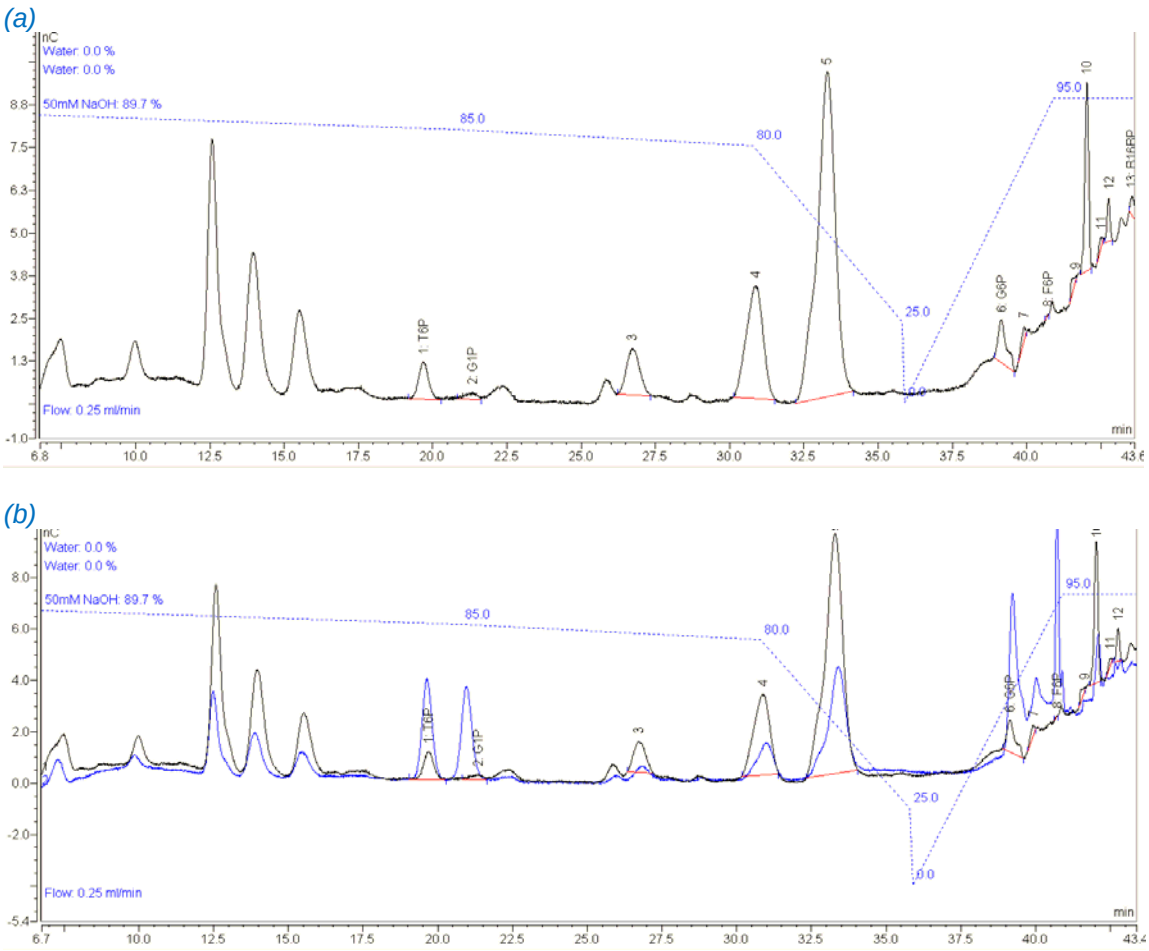


**Fig. S2.** Detection of T6P in sugarcane suspension cell extracts. (*a*) Chromatogram from HPAEC-PAD after 10 µL injection of material obtained from N3.2 transgenic sugarcane suspension cells through liquid extraction and SPE. (*b*) Overlaid chromatogram (blue) of the same sample spiked with 18.18 µM of each standard: T6P, G1P, G6P and F6P.

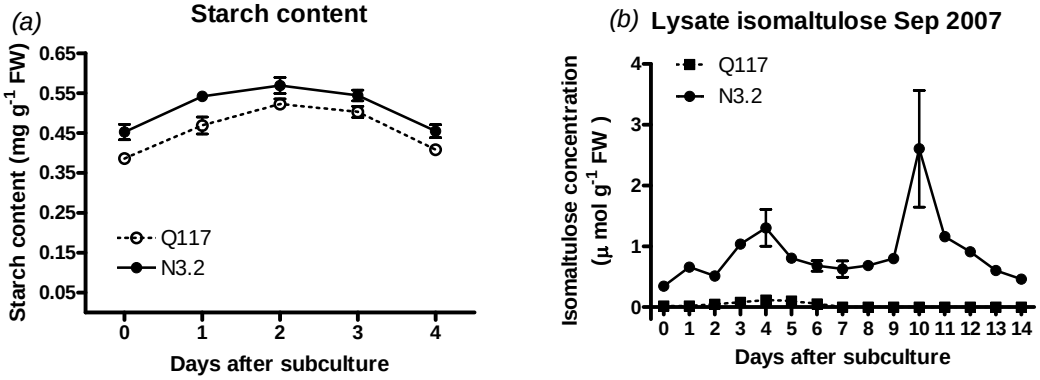


**Fig. S3.** (*a*) Starch contents and (*b*) isomaltulose concentrations in suspension cultured cells of sugarcane transgenic line N3.2 (closed symbols with solid line) and control Q117 (open symbols with dotted line) during a 4-day batch culture. Results are means of three replicates with standard error bars.

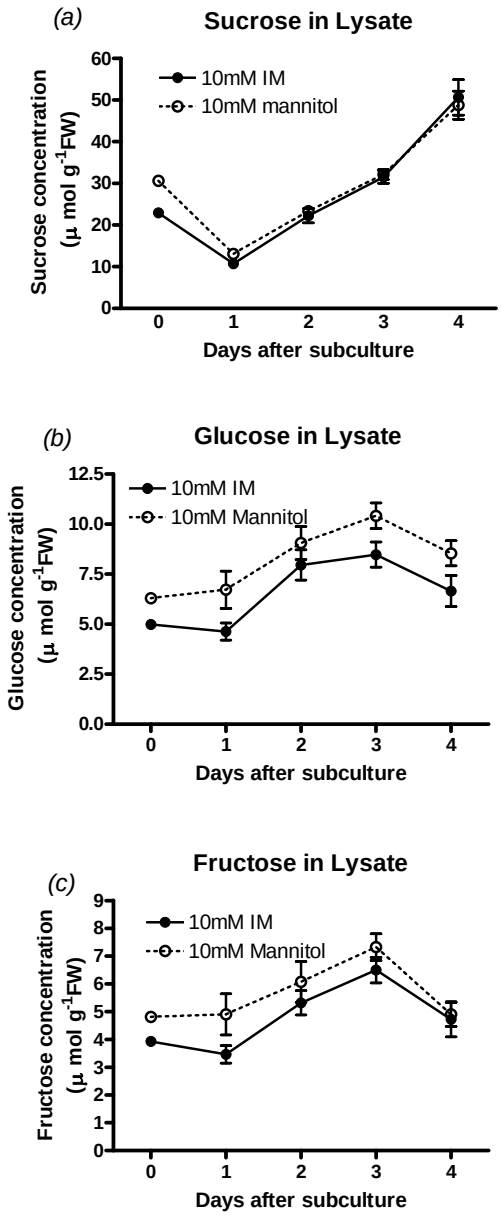


**Fig. S4.** Sugar concentrations in Q117 suspension cultured cells supplied with 10 mM isomaltulose (closed symbols with solid line) and control cultures supplied with 10 mM mannitol (open symbols with dotted line) during a 4-day batch culture. Results are means of four replicates (except Day 0 with only one replicate) with standard error bars. ANOVA with Bonferroni posttests showed no significant difference between isomaltulose-treated and control cells at any time point.