

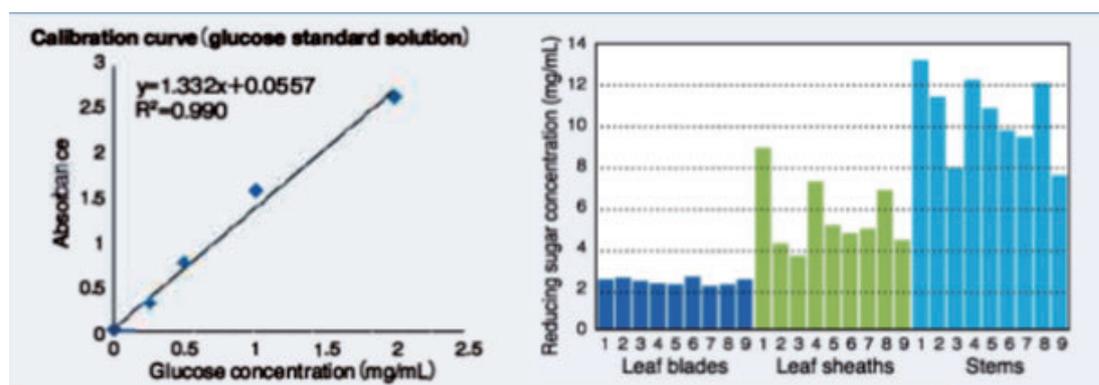


Quantification of glucose level (reducing sugar concentration) using PiCOEXPLORER

Overview The amount of glucose liberated from rice straw by enzymatic reaction is measured using absorbance meter PiCOEXPLORER.

- Procedure**
- Using a glucose standard solution (Wako Pure Chemical Industries 049-31165), adjusted concentration samples were prepared and a calibration curve was processed (0, 0.25, 0.5, 1, 2 mg/ml).
 - The rice straw used was collected from rice cultivated in pots from the field of Tohoku University's Faculty of Agriculture (cultivar: Nipponbare, collected at time of ear emergence).
 - The rice straw was divided into stems, leaf sheaths, and leaf blades, which were then crushed and sorted into samples of 0.015 g each.
 - 1000 μ l of an enzyme solution containing cellulase (Sigma C6105, C2730) was added to each sample and reaction was allowed to proceed for 48 hours.
 - After 5 minutes of centrifugation at 4°C, 20 μ l of supernatant was collected, to which was added 180 μ l of water and 600 μ l of DNS reagent (prepared using Wako Pure Chemical reagent*).
 - Afterwards, the samples were heated with boiling water (100°C) for five minutes and then cooled on ice.
 - 200 μ l was then drawn, inserted into PCR tubes (RIKAKEN, RS-PCR-1 F), and three measurements were taken for each sample using PiCOEXPLORER under the following conditions. (OD₆₀₀=0.01, 0.025, 0.05, 0.075, 0.1, 0.25, 0.5, 0.75, 1, 2.5, 5, 7.5, 10) 30 μ l, 50 μ l, and 100 μ l was then drawn, inserted into PCR tubes (RIKAKEN, RS-PCR-1 F), and measurements were taken with PiCOEXPLORER under the following conditions. Equipment conditions: Green color sensor (530 nm Maximum absorption wavelength, 455-630 nm Wavelength range), 10% LED output level
- * Sodium hydroxide (198-13765), potassium sodium tartrate tetrahydrate (191-03005), Phenol (160-17362), sodium hydrogen carbonate (191-01305), and 3,5-Dinitrosalicylic Acid (040-03642).

Calibration Curves



Result The amount of glucose (reducing sugar concentration) from each part of the rice straw was quantified based on the calibration curve, and it was confirmed that the stems exhibited the best saccharification.

SINCE 1889



Photo Absorbance Meter

PiCOEXPLORER

PAS-110-YU



Quantification of glucose level (reducing sugar concentration) using PiCOEXPLORER cont'd

Comments

Yukihiro Ito / Associate Professor, Environmental Biotechnology Lab, Environmental Bioscience Department, Tohoku University Graduate School of Agricultural Science

Our laboratory develops and studies rice that is suitable for biofuel production using cellulase genes. In aiming for the efficient production of biofuel, we have verified improved saccharification of cellulose contained in cell walls. An indispensable requirement for such verification is quantifying the amount of glucose liberated by enzyme reaction from harvested rice straw. I discovered PiCOEXPLORER at a conference, and thinking that it could be used for quantification of glucose levels, I introduced it to my students. While our lab is equipped with a droplet-type absorbance meter, we often use it for DNA and RNA measurement, so introduction of PiCOEXPLORER for quantification of glucose lets us increase the overall efficiency of laboratory work. I think it is very important to provide students with an environment in which they can devote greater attention to their research.

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When quantifying glucose, sometimes we will measure more than 100 samples at a time. Previously we had to do this using the droplet-type absorbance meter, so it was physically very challenging. Also, since the droplet-type absorbance meter was shared by everybody in the lab, users hesitated to monopolize it for two hours at a time. PiCOEXPLORER enables measurements that are comparable to the droplet-type absorbance meter, and the way in which it enables measurement in tubes rather than measurement of a drop at a time makes the work much easier. Measurements are simple to take, don't require a lot of time, and measurement data can be managed in Excel, all of which contributes to greater work efficiency.

Reference: Abe T, Ito K, Takahashi Y, Sato K, Matsuzaka A, Sonoki T, Ito Y (2016) Difference of saccharification yields between organs and growth stages in rice. Plant Biotechnol 33, 105-110.



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