2023 ARBC

Online Qualifier Paper Round

Paper Round Answer Key

Article 1:

https://www.science.org/doi/10.1126/science.aat9077

1. What is the purpose of photorespiration? (1pt)
   Possible answers include: Photorespiration is used to regenerate RuBP which is consumed during oxygenation by Rubisco, because the plant needs to be able to continue carbon fixation. (scientists speculate photorespiration may also produce useful compounds for defense though). It reduces overall efficiency because it results in the loss of a carbon atom (which is metabolically costly), and the loss of a nitrogen atom in the form of ammonium. It is also energetically costly to transport substrates across so many membranes within the cell. These effects are most pronounced at high temperatures and low CO2 concentrations.
2. In figure 1A, this paper schematically represents the alternative pathways (APs) that they introduced into the plants. What happens to the hydrogen peroxide produced from the conversion of glycolyate→glyoxylate in the native pathway? How is this different in AP2? (2pts)
   In the native pathway, hydrogen peroxide is broken down into H2O and O2 by catalase in the peroxisome. In AP2, H2O2 is broken down by catalase from E. Coli within the chloroplast.
   a. Would you expect adding the gene for Catalase (with an appropriate promoter) into the vector construct for AP3 to increase or decrease overall photosynthetic efficiency? Why? (2pts)
      It most likely would not increase overall photosynthetic activity, perhaps even decrease efficiency. This is because AP3 does not produce hydrogen peroxide, so catalase is not really needed to break it down (and expressing an unnecessary protein wastes resources).
3. What technique was used to produce the image in figure 1c? (1pt)
   Western blot
   a. Aside from the introduced proteins, why was Actin content also assessed in the blot? Why was PGL35 also measured? (1pt)
      Actin content was assessed in the blot to verify that their findings were normalized to total protein content (to rule out that differences in measured protein expression weren't due to there being simply a different concentration of total protein). PGL35 is a chloroplast marker, and was used to rule out cytoplasmic contamination.
   b. This blot analyzes samples from three distinct parts of the plant cell. In which sample would you expect to find the highest content of Photosystem II? What about

Cytochrome b6f? What about Rubisco? (2pts)

You would expect Photosystem II and Cytochrome b6f to be highest in the insoluble chloroplast membrane fraction, and Rubisco to be highest in the chloroplast sample.

c. Which of the introduced proteins for AP3 seems to be found in the lumen rather than the thylakoid membrane? (1pt)

Malate Synthase (MS)

4. What was the authors' purpose of analyzing the ratio of Fv'/Fm'? (1pt)

To assess the resistance to photoinhibition of the modified seedlings, and to preliminarily screen out which APs were the most promising (without needing to grow out all the seedlings).

Related Article:

https://journals.plos.org/plosbiology/article/file?id=10.1371/journal.pbio.2006352&type=printable

https://www.farmprogress.com/biotechnology/nitrogen-fixing-corn-farming-s-holy-grail-when

1. What types of macromolecules are nitrogen likely to be incorporated into? (1pt)

   Proteins and Nucleic acids

2. In what soil conditions might plants favor ammonium vs nitrate as a nitrogen source, and why might this be? (1pt)

   Plants that prefer acid soil conditions seem to prefer ammonium, while those adapted to higher pH soils prefer nitrate

3. Referencing figure 4 in the image below, by what factor is the amount of oxygen reduced? Why might this be beneficial for the bacteria and the plant? (1pt)

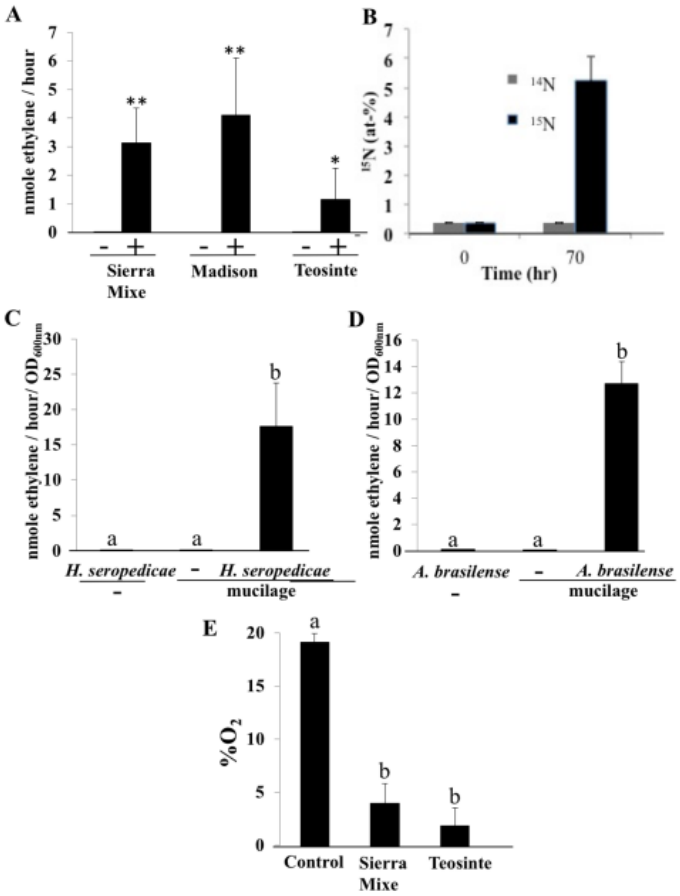

Fig 4. Nitrogenase and N_2 fixation activity in mucilage produced by Sierra Mixe maize. (A) Mucilage of various Sierra Mixe maize lines collected in Sierra Mixe or field-grown plants in Madison, USA, and of teosinte display strong acetylene reduction activity. (-, no acetylene; +, 10% acetylene). Asterisks indicate significant differences ($^{*}P < 0.05$; $^{**}P < 0.01$, Mann-Whitney test). (B) Nitrogen fixation in Sierra Mixe maize mucilage by $^{15}N_2$ assimilation. Mucilage collected from Sierra Mixe maize grown in Sierra Mixe was incubated in gas-tight vials filled with $^{15}N_2$ or $^{14}N_2$ gas for 70 hours at 37 °C. ^{15}N (atom % excess) was determined by IRMS. (C and D) *H. seropedicae* and *A. brasilense* display acetylene reduction activity when added to nonfixing mucilage, whereas the same mucilage supplemented with sterile medium (-) or the same bacteria without mucilage (-) do not. (E) Oxygen concentration at 8 mm inside of the mucilage. Means and standard errors are shown. Different letters indicate statistically supported groups (Kruskal-Wallis test). (Data at DOI: 10.6084/m9.figshare.6534545). IRMS, isotope-ratio mass spectrometry.

¼ or 25%

This is beneficial because O_2 is an inhibitor for dinitrogenase, so by reducing the amount of O_2 the bacteria is exposed to, the plant can increase the amount of available nitrogen

4. Often, fertilizers are added directly to the soil of the plant, and it is uptaken by the roots. However, excess fertilizer often has negative effects on the environment and the plant. Explain why scientists are interested in pursuing plants that culture nitrogen-fixing bacteria:
   Reduce the contamination of the environment with fertilizers (which often contain nitrogen) by having the plants produce nitrogen directly

5. Why are many scientists and experts skeptical about the potential of this corn strain to improve agriculture? What are the other viable alternatives?
   Using conventional breeding, scientists expect it would take 5-10 years for this adaptation to be in commercially useful corn strains (that can deal with higher latitudes, etc.)

   Other viable alternatives include genetically engineering microbes to produce bioavailable nitrogen more efficiently in the soil

Synthesis questions:

1. Both of these modifications/adaptations are attempts to combat the inhibitory effect of what substance/factor on overall productivity?
   O_2

2. Hypothetically, would you expect introducing the synthetic APs mentioned in the first article into the corn strain discovered in the second article to increase crop productivity similar to the results found with tobacco? Why or why not?
   No, because corn is already a C4 plant (it already has mechanisms for reducing photorespiration), so the APs would not have a similar effect

    a. In reality, what scientific barriers would probably make it difficult for scientists to introduce APs into this new strain of corn if they wanted to?
       Scientists lack sufficient genetic information about this new corn strain, making gene editing infeasible.