Measurement of ethylene production during banana ripening.

Abstract — Introduction. This protocol aims at measuring fruit ethylene production during ripening. It can be used to compare ethylene production between different banana varieties or to compare ethylene production between fruit produced in different pedo-climatic conditions. The principle, key advantages, starting plant material, time required and expected results are presented. Materials and methods. This part describes the required laboratory materials and the three steps necessary for calculating the amount of ethylene produced during banana postharvest ripening. Possible troubleshooting is considered.

1. Introduction

Application

This protocol aims at measuring fruit ethylene production during ripening. The ripening process involves the production of ethylene, which, in turn, controls several physiological events such as loss of firmness, peel discoloration, sugar biosynthesis, etc. This method can be used to compare ethylene production between different banana varieties or to compare ethylene production between fruit produced in different pedo-climatic conditions.

Principle

Bananas are climacteric. This method is designed to initiate fruit ripening with an ethylene-like gas such as acetylene or propylene and to measure fruit ethylene production by gas chromatography [1].

Key advantages

This protocol makes it possible to obtain an accurate measurement of the ethylene production potential of banana fruits.

Starting material

This protocol uses green bananas harvested 2–3 days prior to testing.
Time required
Each measurement of ethylene production lasts 2 min.

Expected results
Around 2–50 µL ethylene·kg⁻¹·h⁻¹ should be released during the initial ripening phases and can be measured with the method proposed.

2. Materials and methods

Laboratory materials
The protocol requires a gas chromatograph (GC) with a flame ionisation detector (FID), a 2–3-L jar with a sampling septum, a climatic room at (25 ± 0.5) °C, acetylene or propylene, a 250-µL glass syringe, a 50-mL plastic syringe and a test tube.

Measurement of fruit ethylene production during ripening

- Step 1. Choose and cut the fruit:
  – choose a representative hand of the bunch for analysis (the middle hand is generally selected),
  – cut off a banana from the hand,
  – store the fruit for 2–3 days at 20 °C to get rid of ripening inhibitors [2],
  – weigh the banana and place it in the jar,
  – measure air volume around the banana in the container (fill the jar with water and measure the water volume).

- Step 2. Acetylene (or propylene) treatment of bananas:
  – to initiate ripening, inject approximately the equivalent of 10 000 µL·L⁻¹ of acetylene or propylene with a 50-mL plastic syringe through the septum. Note: lower concentrations of acetylene or propylene can also be used according to the fruit sensitivity threshold,
  – store the jars with bananas at 25 °C (optimal temperature for fruit ethylene synthesis).

- Step 3. Analysis of ethylene production:
  – turn the jar over several times to homogenise the internal atmosphere,
  – take 250 µL from the head-space of jar air and inject it into the GC column,
  – aerate the jars (leave them open for a few minutes),
  – reseal the jars and store them again at 25 °C for further measurement,
  – after each measurement, calculate the amount of ethylene produced.

Note: the time between two measurements should be set prior the experiment. This time depends on the objective of the experimentation: if the objective is the analysis of ethylene production during the first stages of fruit ripening, a short interval between two measurements has to be selected (1–2 h); if the objective is to measure total ethylene production during ripening, a long interval (24 h) can be chosen.

Caution: if a 24-h interval is chosen, the jar must be large enough so that the CO₂ released during fruit respiration does not affect ethylene synthesis (2–3% CO₂).

Troubleshooting
Two main problems can occur:
(a) No ethylene is detected: there is a leakage via the septum.
Solution: change the septum.
(b) The measurements indicate very low ethylene concentrations: there is too high a CO₂ concentration. This can occur when the ratio [(total volume of the container) / (volume of the fruit)] is too small.
Solution: choose a container better adapted to the volume of the fruit. As an example, for an 8-h interval between two measurements, a 3-L container is suitable for a 200-g banana fruit.

References