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Absorption and metabolism of formaldehyde in solutions by detached banana leaves

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Detached banana leaves are one of the by-products of banana production. In this study, the absorption and metabolism of formaldehyde (HCHO) in solutions by detached banana leaves was investigated under submergence conditions. The results showed that banana leaves could effectively absorb HCHO in the treatment solutions, and the relationship between HCHO absorption and treatment time appeared to fit a radical root function model. ^{13}C nuclear magnetic resonance analysis was used to investigate the ability of detached banana leaves to metabolise H^{13}CHO, and the results indicated that the H^{13}CHO absorbed from the treatment solutions was converted into non-toxic compounds. High amounts of [U-^{13}C]glucose, [U-^{13}C]fructose, [3-^{13}C]serine and [3-^{13}C]citrate were produced as a result of H^{13}CHO metabolism in banana leaves, and the production of a small amount of [2,4-^{13}C]citrate and [2,3-^{13}C]alanine was also observed. These results suggest that detached banana leaves can metabolise H^{13}CHO and convert it to non-toxic compounds. The metabolic pathways that produce these intermediates in detached banana leaves are postulated based on our ^{13}C nuclear magnetic resonance data.

[**Key words:** Detached banana leaves; Aqueous formaldehyde; Formaldehyde absorption; Formaldehyde metabolism; ^{13}C nuclear magnetic resonance analysis]

Formaldehyde (HCHO) is toxic to the majority of organisms due to its non-specific reactions with macromolecules such as proteins, nucleic acids and lipids to form crosslinks, thereby leading to the loss of their biological functions (1). Many studies have demonstrated that phytoremediation is an effective method to eliminate HCHO pollution (2–4). Although HCHO uptake by plants has been investigated extensively (5,6), a few studies have been engaged in the metabolism of HCHO in plants (7,8). To date, the HCHO metabolic mechanism in plants has mainly been investigated using H^{14}CHO labelling to identify the metabolic products (7–9). However, as this method cannot determine the exact location of the ^{14}C label in the metabolic products, it is difficult to summarise the detailed pathways of HCHO metabolism in plants. Conversely, the exact location of the ^{13}C label in metabolites can be determined using ^{13}C nuclear magnetic resonance (NMR) technology. The treatment of plant tissues with ^{13}C-labelled metabolic precursors will lead to the redistribution of the ^{13}C-labelled isotope in the metabolic network, selectively enhanced the ^{13}C NMR signals of the metabolites (10). Thus, ^{13}C NMR analysis can be directly used for the description of metabolic pathways and metabolic flux. Indeed, our recent study (9) indicated that ^{13}C NMR analysis is an effective method to investigate HCHO metabolic pathways in plants.

Banana (*Musa sapientum* L.) is a monocot herbaceous plant in the *Musaceae* family and is one of the main fruits in tropical and subtropical areas. Although banana leaves are one of the only by-products in banana production, the leaf yield is approximately the same as that of the banana fruits (11). In general, the banana trees need to replace after 1 or 2 years of planting. The vast majority of leaves are removed manually from the trees and piled in an open space or removed as garbage. In this study, the ability of detached banana leaves to absorb HCHO in solutions was investigated under submergence conditions. To understand the metabolic pathways of H^{13}CHO in banana leaves, detached banana leaves were treated with H^{13}CHO solutions, and the metabolites were analysed by ^{13}C NMR technology. Based on the ^{13}C NMR data obtained, metabolic pathways are postulated for H^{13}CHO metabolism in detached banana leaves.

MATERIALS AND METHODS

Measurement of HCHO uptake by detached leaves of banana and other plants Detached leaves (3 g fresh weight [FW]) of banana (*M. sapientum* L.), *Canna indica*, *Eichharnia crassipes*, *Chlorophytum comosum*, *Scindapsus aureus* and *Hedera helix* were surface-sterilised with $HgCl_2$, cut into small pieces (2 × 2 cm) and then divided into two groups: one was autoclaved at 121°C for 20 min to kill the leaf cells and was used to measure HCHO adsorption by the leaves; the other group consisted of fresh leaf pieces used to measure HCHO absorption. The autoclaved leaf (AL) pieces and fresh leaf (FL) pieces were submerged in 100 ml of HCHO solution containing 5 mM $KHCO_3$, 0.1% MES (W/V) and 2 (all plant leaves), 4 (only banana leaves) or 6 (only banana leaves) mM HCHO in a flask. The flask was shaken at 100 rpm at 23°C under continuous light (100 μmol m^{-2} s^{-1}). The concentration of the residual HCHO in the treatment solutions after 4, 8, 12, 24, 32 and 48 h was determined as described by Nash (12). The same treatment solutions without plant material under the same conditions were used as controls to monitor the volatilisation of HCHO. The value of HCHO elimination was calculated as 100% (the initial level) - residual HCHO% − volatilised HCHO%. HCHO elimination by AL was defined as the HCHO adsorption of leaves. The HCHO absorption of leaves was calculated as the HCHO elimination by FL − the HCHO elimination by AL.

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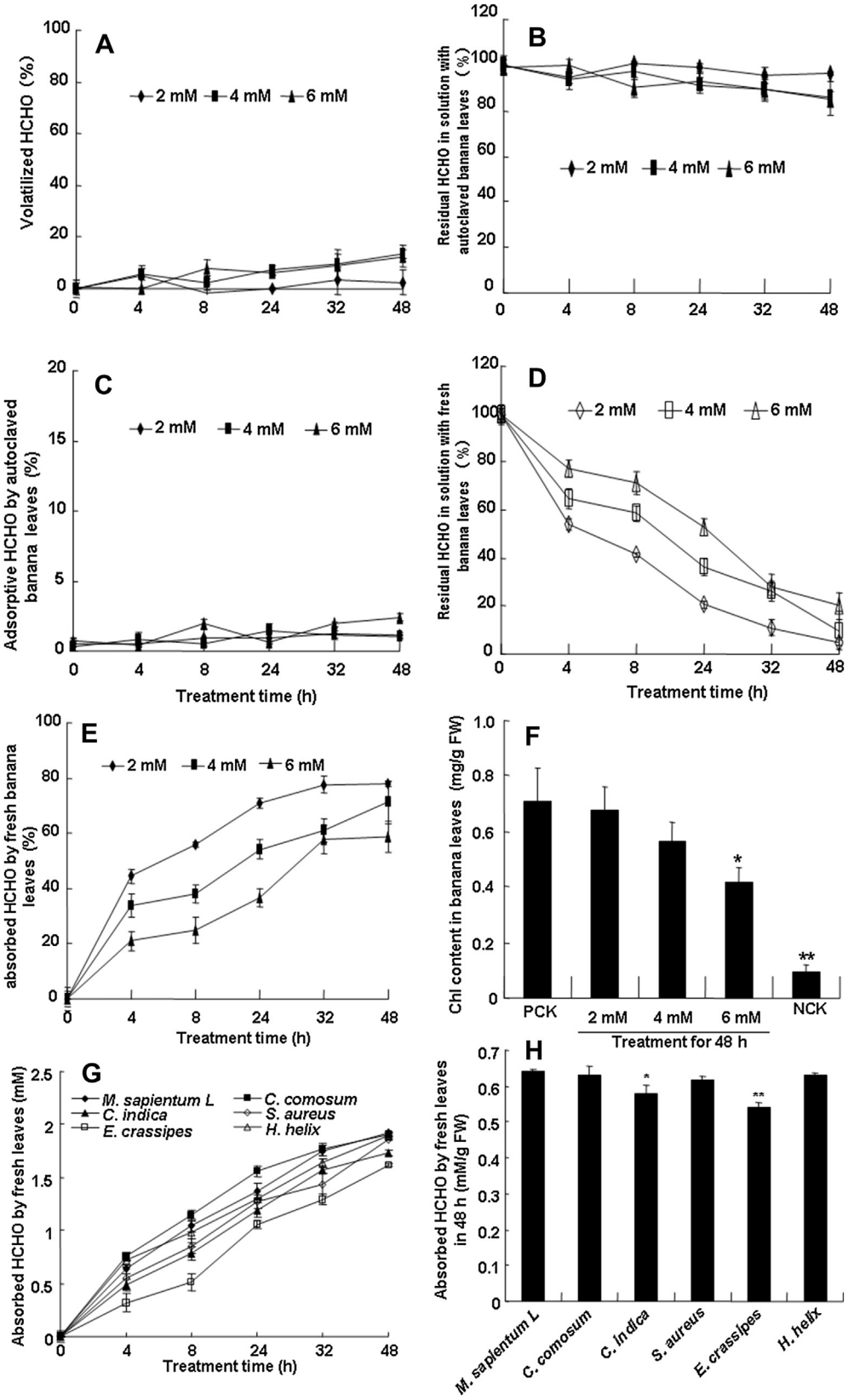

FIG. 1. Adsorption and absorption kinetics of HCHO by banana and other plant leaves treated in HCHO solutions. HCHO adsorption and absorption was measured as described in Materials and Methods. The values are the means ± SD of three independent replicates (A–H). (A) Amount of volatilised HCHO from 2, 4 and 6 mM solutions. (B) HCHO remaining in the treatment solutions with autoclaved banana leaves for 4, 8, 24, 32 and 48 h, respectively. (C) HCHO adsorption curves by autoclaved banana leaves processed according to the content of the residual HCHO in the treatment solutions and the HCHO volatilised from the treatment solutions with autoclaved leaves. (D) Remaining HCHO in the solutions treated with fresh banana leaves for 4, 8, 24, 32 and 48 h, respectively. (E) HCHO absorption curves by fresh banana leaves processed according to the content of the residual HCHO in the treatment solutions, the HCHO volatilised from the treatment solutions and HCHO adsorption by autoclaved banana leaves. (F) Chl content in fresh banana leaves (PCK), autoclaved banana leaves (NCK) and fresh banana leaves treated with 2, 4 and 6 mM HCHO solution for 48 h. (G) Comparison of HCHO absorption ability between fresh detached banana leaves and other plant leaves. (H) Comparison of the absorbed HCHO by fresh detached banana leaves and other plant leaves in a 48 h period.