

# EFFECT OF MODE OF RIPENING ON ETHYLENE BIOSYNTHESIS DURING RIPENING OF ONE DIPLOID BANANA FRUIT

HUBERT O., CHILLET M., JULIANNUS P., FILS-LYCAON B., MBEGUIE-A-MBEGUIE\* D.

*\* CIRAD/UMR 94 QUALITROP, Neufchâteau, Capesterre-Belle-Eau,  
Guadeloupe, French West Indies, F-97130 France  
[didier.mbeguie-a-mbeguie@cirad.fr](mailto:didier.mbeguie-a-mbeguie@cirad.fr)*



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# INTRODUCTION



- For fruit, quality traits including nutritional and organoleptic aspects can be considered as a source of biodiversity and increase the added value of the product and consequently the incomes for the producers
- Fruit Quality is a complex criteria (external and internal aspects as color, flavour, texture, aroma etc.) that is highly affected by both biotic and abiotic cues, some of them being antagonistic
- Elaboration of fruit quality traits involved number of complex physiological processes resulting from the coercitive action of different genes

# INTRODUCTION



- As other fruits, the main quality traits of banana are set up during an important phase of its development namely ripening
- Banana fruit undergoes a ripening climacteric process characterized by a peak of respiration and a burst of an autocatalytic production of ethylene
- Concomitantly, there are molecular and biochemical changes leading to fruit ripening (peel degreening, aroma volatiles, sugar accumulation, fruit softening). Some of these changes are ethylene-dependant, ethylene-independent or both.

# OBJECTIVES



- Characterize IDN 110, a dessert (AA) and diploid variety, in term of ethylene biosynthesis. IDN110 is a member of the core collection of banana varieties used as parents in breeding program
- Get more insights into the molecular mechanism that govern the ethylene biosynthesis process during ripening of IDN110, in order to identify the major (candidate) genes involved and related molecular marker
- The availability of characterized varieties parents of banana and, related marker of fruit quality traits are the key prerequisites for the implementation of the strategy (breeding and marker assisted selection) developed at CIRAD to improve of banana fruit quality.

# MATERIAL

## Growing – Harvesting



- MATERIAL - GROWING CONDITIONS :

IDN 110 (dessert cultivar, *Musa acuminata*, AA)

Guadeloupe ; altitude: 250 m; andosol; rainfall: 3500 mm / year



- HARVESTING *in-planta*:

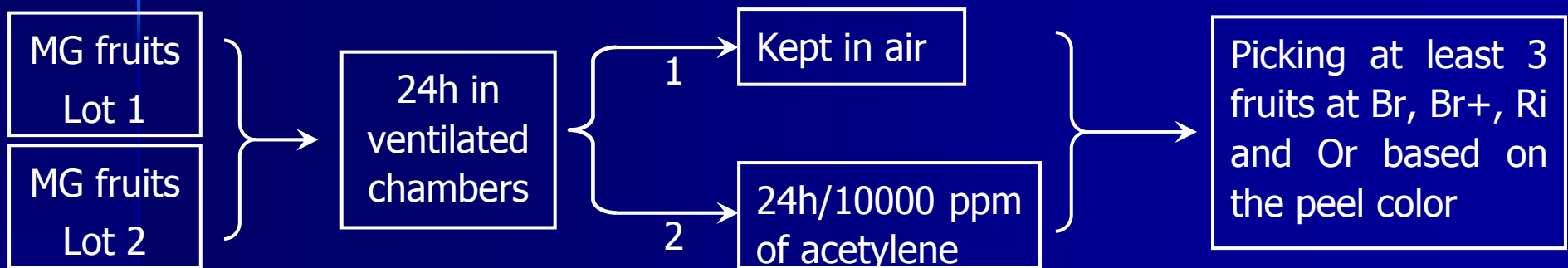
Fruits were harvested at 6 developmental stages for all ripening conditions including IMG (immature green) and MG (mature Green), Br (breaker), Br+ (more breaker), Ri (ripe) and Or (over ripe). IMG and MG stages were harvested based on the heat units concept (Ganry and Meyer, 1975). While Br, Br+, Ri and Or stages were harvested based on the peel color

# METHODOLOGY

## *Postharvest ripening - Physicochemical and genes expression analyses*



### Postharvest ripening at 20°C (*ex-planta*):



### Physico-chemical analyses:

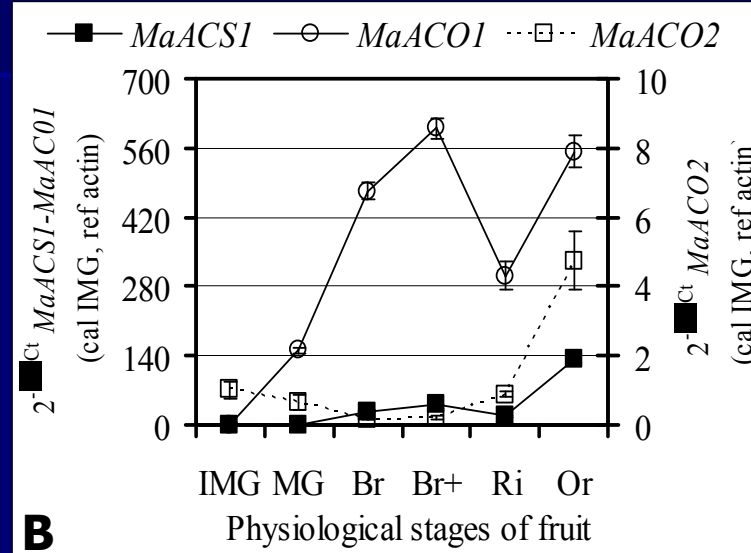
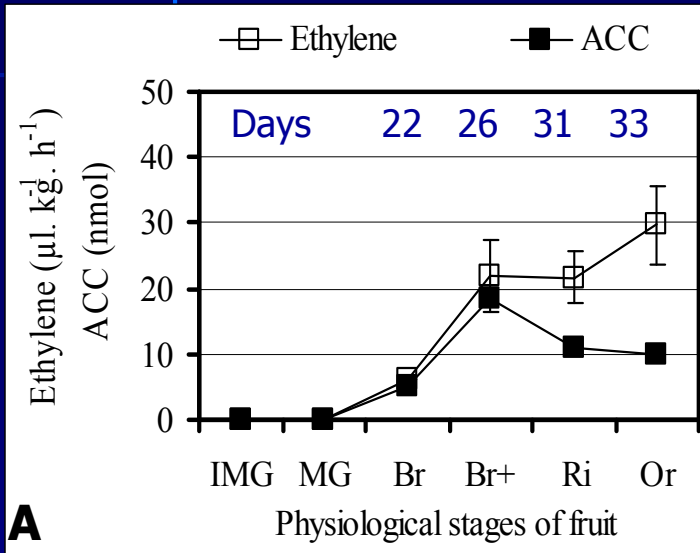
Measurement of ethylene production of the whole fruit and ACC content of the pulp tissue

### Gene expression analyses throughout Real-time Quantitative PCR:

- Two ACC oxidase genes, MA-ACO1 and MA-ACO2 (Inaba et al., 2008)
- One ripening and inducible ACC synthase gene, MA-ACS1 (Liu et al., 1999)
- mRNA level quantified using Double-Delta formula (Livak and Schmittgen 2001) with actin gene as reference and IMG stage as calibrator

# RESULTS

## Ethylene biosynthesis *in-planta*



Each data point is the mean of values obtained from at least three fruits originated from three replicate bunches. Vertical bars indicate standard deviation (S.D.). When no bar is shown, S.D. was smaller than the symbol. The numbers indicated within "ethylene-ACC" graphs represent the days taken by fruit, since IMG stage, to reach the corresponding physiological stage indicate at the abscise axe.

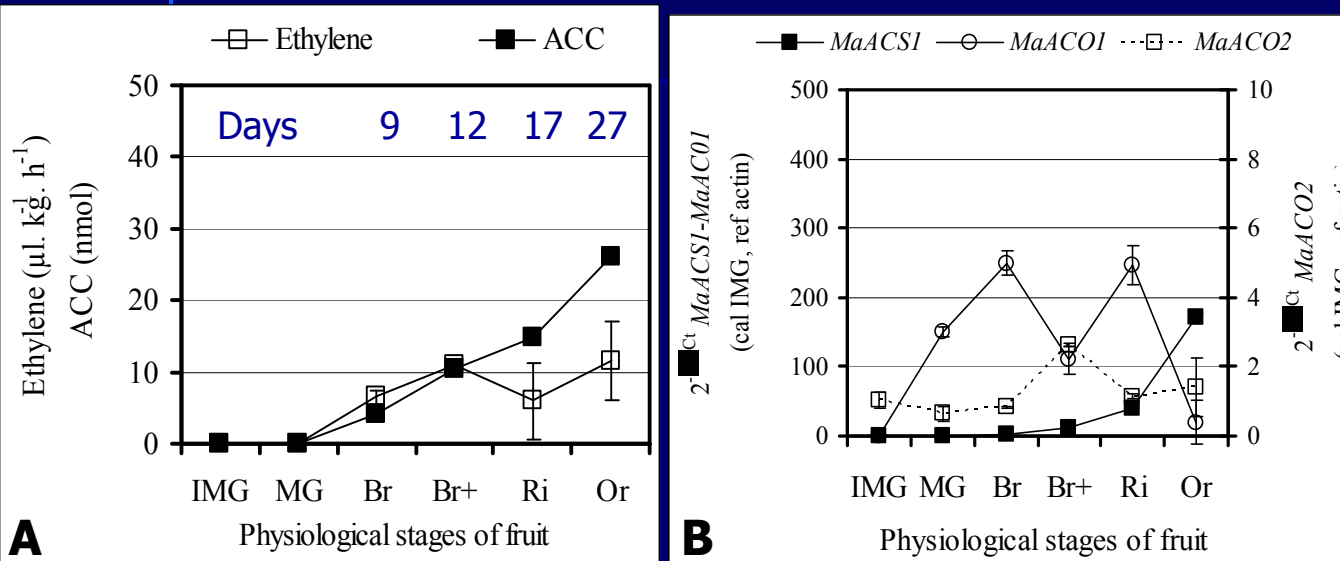
Ethylene production of the whole fruit and ACC content (A), ACC synthase and ACC oxidase genes expression (B) in pulp tissue of IDN 110 fruit ripening *in-planta*.

IMG =immature green; IM=mature green; Br=Breaker; Br+=more breaker; Ri=ripe.

- Increase of C<sub>2</sub>H<sub>4</sub> production throughout ripening with no fall in the rate as it was observed in triploid Cavendish banana.
- No direct correlation between ACC content and the level of MaACS1 mRNA in pulp
- A direct correlation between ethylene production, MaACS1 and mainly MaACO2 mRNA accumulation in pulp

# RESULTS

## Ethylene biosynthesis *ex-planta* in air



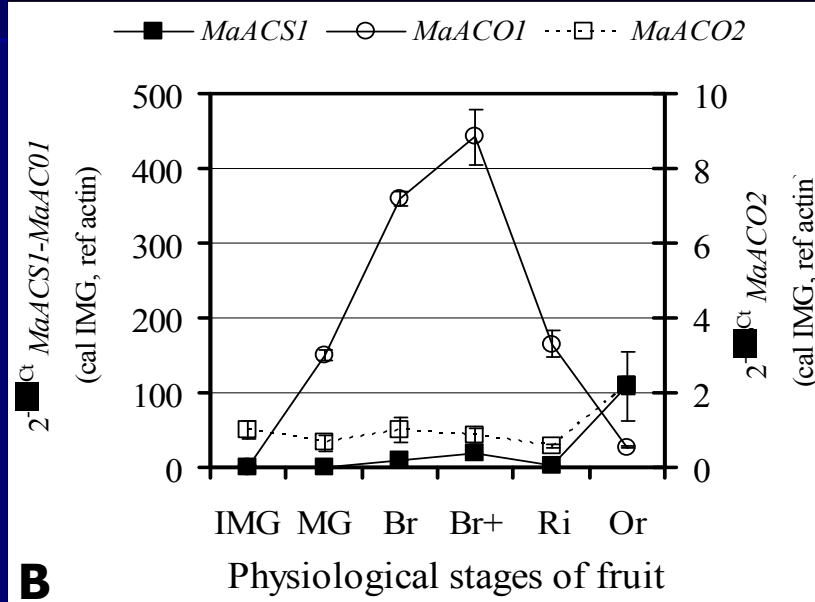
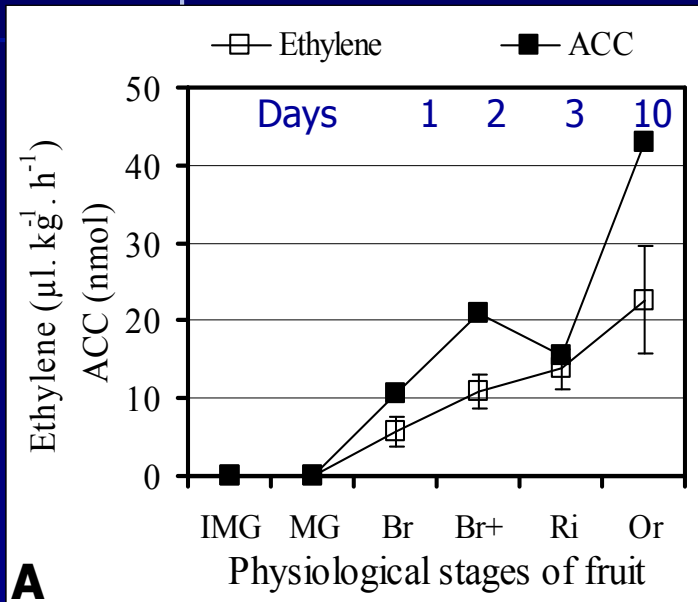
Each data point is the mean of values obtained from at least three fruits originated from three replicate bunches. Vertical bars indicate standard deviation (S.D.). When no bar is shown, S.D. was smaller than the symbol. The numbers indicated within "ethylene-ACC" graphs represent the days taken by fruit, since IMG stage, to reach the corresponding physiological stage indicate at the abscise axe.

Ethylene production of the whole fruit and ACC content (A), ACC synthase and ACC oxidase genes expression (B) in pulp tissue of IDN 110 fruit ripening *ex-planta* in air. IMG =immature green; IM=mature green; Br=Breaker; Br+=more breaker; Ri=ripe.

- Increase of  $\text{C}_2\text{H}_4$  production throughout ripening with no fall in the rate as it was observed in triploid Cavendish banana.
- Direct correlation between ACC content and, MaACS1 mRNA and the level of ethylene production
- A direct correlation between ethylene production and only MaACS1 mRNA accumulation in pulp

# RESULTS

## Ethylene biosynthesis ex-planta in air and after acetylene treatment



Each data point is the mean of values obtained from at least three fruits originated from three replicate bunches. Vertical bars indicate standard deviation (S.D.). When no bar is shown, S.D. was smaller than the symbol. The numbers indicated within "ethylene-ACC" graphs represent the days taken by fruit, since IMG stage, to reach the corresponding physiological stage indicate at the abscise axe.

Ethylene production of the whole fruit and ACC content (A), ACC synthase and ACC oxidase genes expression (B) in pulp tissue of IDN 110 fruit ripening *ex-planta* in air and after acetylene treatment. IMG =immature green; IM=mature green; Br=Breaker; Br+=more breaker; Ri=ripe.

- Increase of ethylene production throughout ripening with fall in the rate as it was observed in triploid Cavendish banana.
- Direct correlation between ethylene production of the whole fruit and ACC content, *MaACS1* mRNA and *MaACO2* mRNA accumulation in pulp

# CONCLUSIONS



- IDN 110 fruit ripen slowly *in-planta* than *ex-planta*, and *ex-planta* in air than after acetylene treatment.
- No correlation was observed between the level ethylene production of the whole fruit and its ripening speed
- Whatever the ripening conditions, IDN110 displays a pattern of ethylene production different to that of triploid Cavendish banana, with no fall in the rate during the late ripening stage

# CONCLUSIONS



- Difference between the time course of ethylene production of the whole fruit and, the pattern of ACC content and mRNA accumulations of ethylene biosynthesis genes

	<i>C2H4</i>	<i>ACC content</i>	<i>MaACS1</i>	<i>MaACO1</i> (highly expressed)	<i>MaACO2</i>
<i>In-Planta</i>	↗	↘	↗	↗↘↗	↗
<i>Air</i>	↗	↗	↗	↗↘↗↘	↗↘↗
<i>Acetylene</i>	↗	↗	↗	↗↘	↗

# PROSPECTS



- Characterize a large number of banana varieties in order to get more information the banana diversity in term of ethylene biosynthesis
- Examine, within these varieties, the relationship between the structural diversity of ethylene biosynthesis genes and ethylene biosynthesis in order to identify the putative marker usable in breeding to improve banana quality traits
- Investigate the mechanism of ethylene responsiveness process related with banana fruit ripening



# TEAM



## **INRA/CIRAD UMR 94 QUALITROP**

C. GALAS

P. JULIANNUS

B. FILS-LYCAON

D. MBEGUIE-A-MBEGUIE

D. RINALDO

## **CIRAD UMR 95 QUALISUD**

O. HUBERT

M. CHILLET