

PRELIMINAR ASSAY TO ESTABLISH A MICROPROPAGATION PROTOCOL FOR MANGO (Mangifera indica, L.) CULTIVATED IN BRAZILIAN SAVANNAH

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INTRODUCTION

Mango (Mangifera indica, L.) is currently one of the most important cultivated fruits in Brazil. The most important region of its cultivation is the San Francisco Valley in the State of Pernambuco. In 2001, approximately 94.000 t of mangoes produced in this region were exported mainly to the United States and Europe resulting in \$50 million of earnings to the country (Manga, 2002).

The mango production in Brazil and its exportation can be increased by overcoming some problems, such as pathogen

diseases, soft-nose disorder and improving fruit quality (taste and flavour) which occur in one of the most important produced cultivars that is Tommy Atkins.

In order to introduce and adapt mango in the Brazilian Savannah region, a genetic improvement program was initiated by Embrapa Savannah Agriculture and Livestock Research Center during the 80's (Pinto, 2001). This program intended also the development of superior genotypes for commercial ends. Mango

tissue culture is a possible auxiliary tool to micropropagate the genetic improved cultivars and, few studies have been made regarding this issue (Litz et al, 1982; Litz, 1984; DeWald et al, 1989; Raghuvanshi & Srivastava, 1995; Thomas & Ravindra, 1997; Ara et al, 2000).

This work presents preliminary results of assepsy using microcuttings and young leaves obtained directly from the field as explants to establish a micropropagation mango protocol.

MATERIAL AND METHODS

PLANT MATERIAL

Mango genotypes (Alfa - Embrapa 141; Tommy Atkins and Hybrids selections Embrapa/CPAC 136-86 and Embrapa/CPAC 2-97) used in this work as a source of explants were collected in the Experimental Fruit Crop Area of Embrapa Savannah Agriculture and Livestock Research Center.

ASSEPSY PROTOCOL ON MICROCUTTINGS EXPLANTS

Nodal and internodal segments from mature trees (Alfa; Embrapa/CPAC 136-86 selection and Tommy Atkins) were collected from January to October during 2000 and 2001. The segments were cut into small shoots and eliminated the apical buds. Microcuttings were washed with soap in running tap water and then

disinfected with Sodium hypochlorite solution 2 % + 5 drops of Tween 80 for 15 minutes and Benomyl 200 mg.l⁻¹ for 30 minutes. The tips of the shoots were dabbed in a Benomyl paste and then put into sterile magenta vessels containing expanded vermiculite and then incubated in a growth chamber under controlled conditions (28 °C \pm 2°C and 12h photoperiod at an intensity 40-50 μ mol c s -1 2 .s -1).

ASSEPSY PROTOCOL ON LEAF EXPLANT

Leaves were collected from trees (Embrapa/CPAC 136-86 and Embrapa/CPAC 2-97) during February and September of 2001. They were washed with soap in running tap water and cut into small pieces in water, disinfested with Sodium hypochlorite solution in the following concentrations: 0.5%; 1%; 1.5% and 2% plus 12 drops

of Tween 80 for 5 minutes, transferred to Petri dishes containing SP medium (Barrueto Cid, 1999) supplemented with carbenicilin 200 mg.l-1; Benomyl 200 mg.l-1; polyvinilpirrolidone (PVP) 10 g.l-1; naphthalene acetic acid (ANA) 5μ M; benzilaminopurine (BAP) 7.5μ M and MS-B vitamins (Barrueto Cid, 1994) except for thiamin which was 10 mg.l-1, then incubated in a growth chamber under controlled conditions.

DATA ANALYSIS

Data from both experiments were analyzed in SAS program (version 8.0) as an experiment in a completely randomized design in factorial scheme. Analysis of variance data were transformed in $y = \arcsin(x+0.5)0.5$. Percentage means were compared by Tukey's test when p = 0.01 or p = 0.05, according to the results.

RESULTS AND DISCUSSION

DISINFESTATION AND BUDDING ON MICROCUTTINGS

Microcuttings presented 40% to 60% of contamination and, sprouting (small, middle and large-sized) in microcuttings, varied from 20% to 50% during all period analyzed. Statiscally significant large-sized buds were obtained only during January, April, August and September during year 2000.



Fig. 1- Small and large-sized buds in Alfa variety microcutting during September/2000. Arrow points to the buddings and Benomyl paste on the tip of the microcutting.

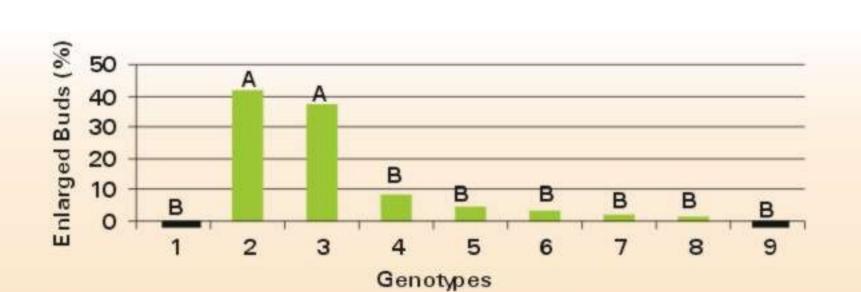


Fig. 2 - Percentage analysis of statiscally significant large-sized buds in one month observation during 2000. Treatments correspond to: 1 - Alfa in July/2000; 2 - Alfa in January/2000; 3 - Alfa in April/2000; 4 - Embrapa/CPAC 136-86 in January/2000; 5 - Embrapa/CPAC 136-86 in April/2000; 6 - Tommy Atkins in April/2000; 7 - Alfa in September/2000; 8 - Tommy Atkins in August/2000; 9 - Tommy Atkins in October/2000. Columns with different letters differ significantly from each other using Tukey's test (p = 0.01).

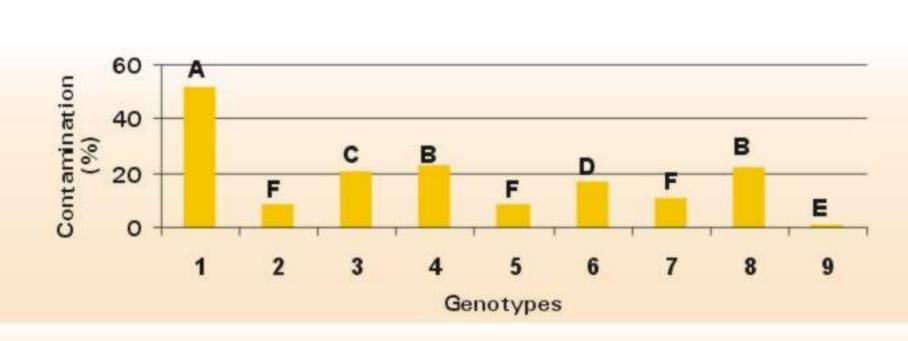


Fig. 3 - Contamination percentage of large-sized producing microcuttings corresponding to Fig.2. Columns with different letters differ significantly from each other using Tukey's test (p = 0.01).

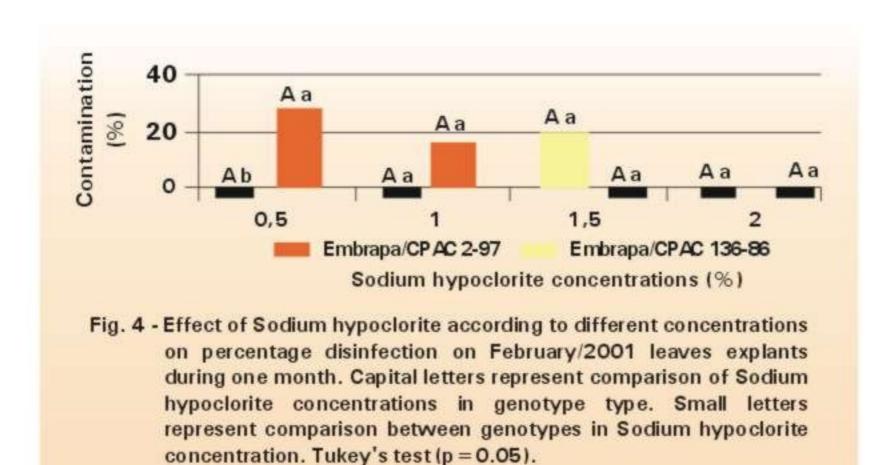


Table 1 - Climatic data at Embrapa Savannah Agriculture and Livestock
Research Center, Planaltina, DF, Brazil during 2000 and 2001
(means values 1)

Temperature
Relative
Humidity
Month
(°C)
(%)
Precipitation
Solar Irradiation
SunShine
(cal/cm²/dia)
(h)

Month	(°C)		Humidity (%)		(mm)		(cal/cm²/dia)		(h)	
	Jan.	22.2	22.2	80	80	2.9	2.9	510.67	510.67	7.0
Feb.	21.9	22.6	87	75	6.7	1.4	468.38	491.45	5.8	8.1
Mar.	21.8	21.6	88	81	7.2	6.6	436.76	391.91	6.0	4.7
Apr.	21.8	22.3	81	64	2.2	2.3	441.97	469.78	8.2	8.7
May.	20.6	21.4	70	64	0.0	0.4	454.25	464.50	9.5	7.9
June	19.6	20.5	64	55	0.0	0.0	431.25	412.76	9.1	8.0
July	19.6	20.7	63	49	0.1	0.0	428.06	417.55	9.2	8.9
Aug.	21.7	20.6	52	47	1.3	1.1	456.62	453.01	9.0	9.3
Sept.	22.0	23.0	67	51	1.8	1.7	454.87	457.68	7.0	6.1
Oct.	23.7	22.0	60	68	2.7	2.5	522.97	407.05	7.8	4.9
Nov.	21.1	21.7	89	80	8.8	7.8	395.20	408.96	4.1	4.5
Dec.	21.8	21.9	89	78	7.6	8.0	453.10	421.94	4.2	4.3

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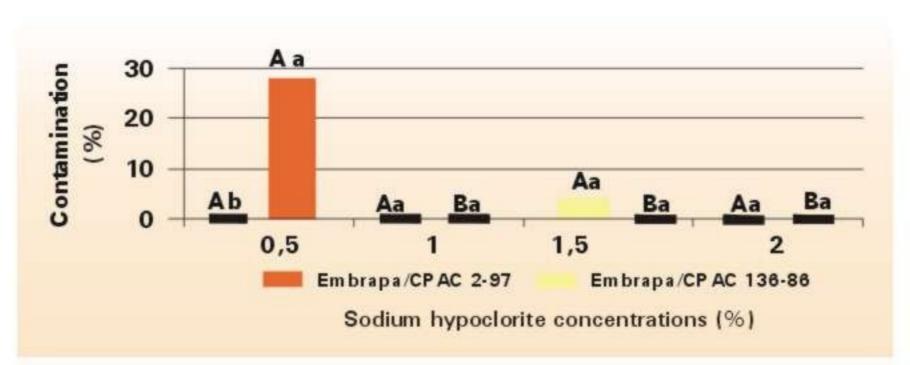


Fig. 5 - Effect of Sodium hypoclorite concentration on percentage disinfection on February/2001 leaves explants during the first week observation. Capital letters represent Sodium hypoclorite concentration comparison in genotype. Small letters represent comparison between genotypes in Sodium hypoclorite concentration. Tukey's test (p = 0.01).

DISINFESTATION AND BROWNING ON LEAVES EXPLANTS

Leaf contamination experiments showed that 0.5 % Sodium hypoclorite concentration had the highest percentual of contamination (28%) and the lowest percentual of browning production (28%) in Embrapa/CPAC 2-97 explants during February/2001.

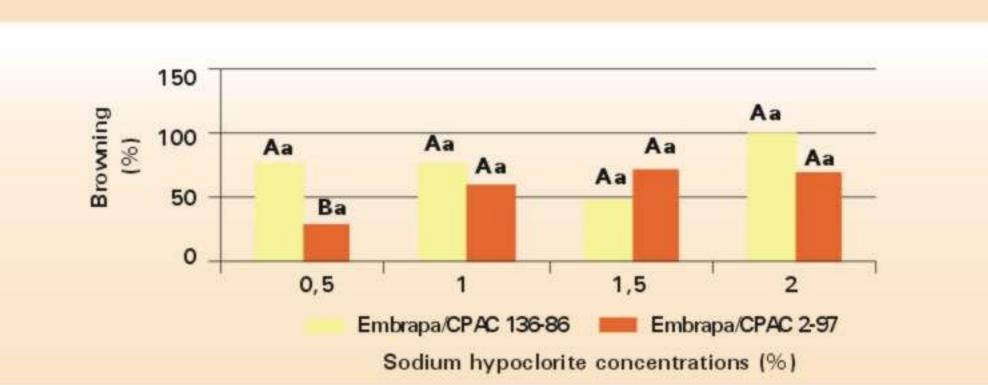


Fig. 6 - Effect of Sodium hypoclorite concentrations on percentage of browning on February/2001 leaves explants in the fourth week.of evaluation. Capital letters represent Sodium hypoclorite concentration comparison in genotype type. Small letters represent comparison between genotypes in Sodium hypoclorite concentration. Tukey's test (p = 0.01).

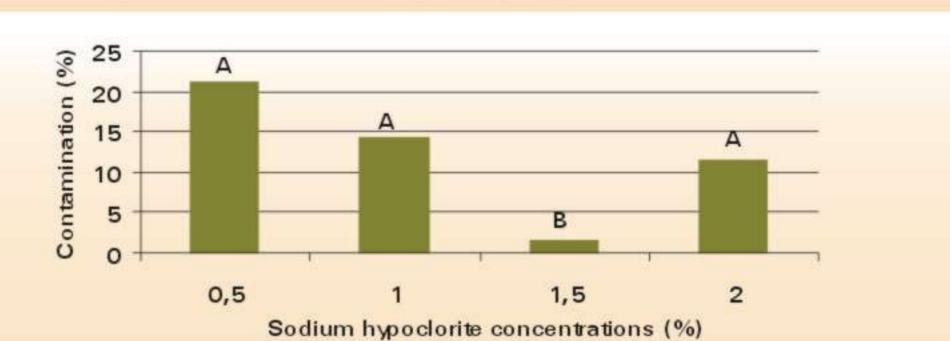


Fig. 7 - Comparison between February and September Embrapa/CPAC 2-97 leaves explants disinfestation experiments in the first week of observation. Capital letters represent Sodium hypoclorite concentration comparison. Tukey's test (p = 0.05).

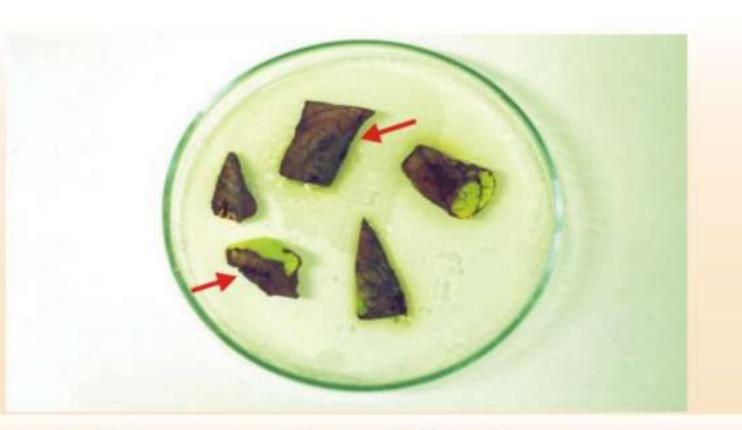


Fig. 8 - 21 days disinfested Embrapa/CPAC 2-97 leaves explants in September with Sodium hypoclorite 1.0% and incubated in SP medium. Arrows points to partial and total browning in explants.

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CONCLUSIONS

- 0.5% Sodium hypoclorite concentration was more efficient against explant browning but with higher percentage of contamination in Embrapa/CPAC 2-97 genotype during February/2001.
- Susceptibility to browning seems to be genotype dependent.
- Browning process in explants is the most important event to be controlled in order to establish a mango tissue culture protocol.
- Contamination is not a limiting factor to establish a mango tissue culture protocol.
- Large-sized buds in microcuttings was greater during year 2000.

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