



## **Environmental and Genotypic Effects on the Growth Rate of in Vitro Cassava Plantlet**

**By**

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### **Abstract**

Two cassava genotypes were evaluated in vitro in the screen house and culture room using five different media treatments. Each treatment was replicated 12 times and observed for 5 weeks before sub-culturing. There were significant differences in the growth rate of plantlets in different media, which suggests an interaction between treatments and environments. The survival of TMS 188/00106 was significantly different from TMS 083/00125 in the culture room than in the screen house. The study confirms that tissue culture derived plantlets can be raised in the screen house but suggests that plantlets should be allowed to survive in the culture room before transferring to the screen house for further growth.

### **Introduction**

Tissue culture offers a unique opportunity to mass propagate plant materials especially, disease free plantlets. Vegetative propagation through tissue culture has played significantly in the mass production of vegetative propagating materials (Ajithukumar and Seemi 1998). It is faster and requires less space than that required for conventional methods of preparing cassava cuttings. The provision of electricity in the laboratory and the cost of maintaining a generating plant for regular power supply are very high in Nigeria and other developing countries. Even if the government can afford it, the private laboratories and seed companies may not be able to break even.

Cassava has just entered the international market in Nigeria. The need to rapidly produce disease-free planting materials to meet the growing local and international demand was a propelling factor for this investigation. Secondly, the space in many laboratories cannot accommodate the commercialization of major crops as demanded by big time

farmers, hence the need to look beyond the laboratory environment. In vitro culture in the screen house will not only reduce the cost of production, it will also enhance quick acclimatization. There is rapid growth rate due to high temperature in the screen house than when in the laboratory. This study aims therefore to assess the use of screen houses to maintain cultures and rapidly propagate important crops with less contamination at a reduced cost.

## Materials and Methods

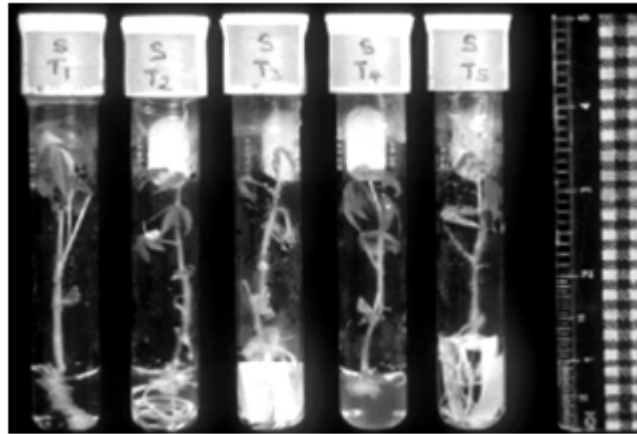
Two genotypes of cassava (TMS 188/00106 and TMS 083/00125) were obtained from the International Institute of Tropical Agriculture (IITA, Ibadan, Nigeria), while five different media were prepared using Murashige and Scoth (1962) with minor adjustments as follows:

- Treatment 1 (T1) - Liquid only
- Treatment 2 (T2) - Liquid with 50% normal agar (2 g L<sup>-1</sup>)
- Treatment 3 (T3) - Liquid media with filter paper embedded
- Treatment 4 (T4) - Media with normal Agar (4 g L<sup>-1</sup>)
- Treatment 5 (T5) - Liquid media with filter paper projecting out

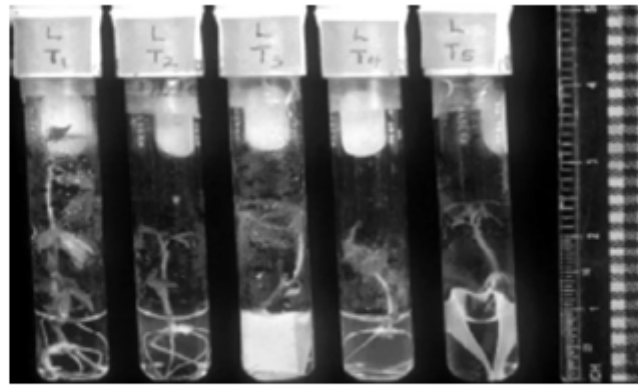
The pH was measured and dispensing was done at the rate of 3 ml before autoclaving. The sub-culturing was done the following day. A total of 120 test tubes were used for each cultivar with 12 test tubes per treatment. A complete set of 60 test tubes with 5 treatments of 12 replicates was placed in the laboratory while the second set was placed in the screen house in the first day for TMS 188/00106. The same procedure was adopted the following day for TMS 083/00125. Data were recorded weekly for 5 weeks before sub-culturing. The second generation was observed for only two weeks to ensure the repeatability of the data recorded during the first generation. The data records include explants survival, shoot development, root development, nodal formation, leaf growth and increase in height. Each of the records were scored on a 0–3 scale [0: dead, 1: alive, but not growing, 2: growing slowly, and 3 growing very well]. Survival rate was recorded for two weeks only while the other five traits were scored continuously for three weeks consecutively. Only the screen house explants were subculture after 5 weeks to ensure the repeatability of the findings. The subculture materials from the screen house explants were also placed in both screen house and culture room (laboratory). The same recording was undertaken for responses of the explants to the culture medium and environment as in the first generation explants.

## Results and Discussion

The two environments were not affecting significantly shoot, root, node, leaf and height development (Figs. 1–2). The laboratory plantlets grows better in liquid, and liquid with filter paper embedded media than when placed in the screen house, which might be due to high temperature recorded at the time of placement (32 -36oC viz. a viz. to 22-25oC in the laboratory).



**Fig. 1** – Screen house performance of the 5 treatments. T1: liquid only, T2: liquid with 50 % normal agar ( $2 \text{ g L}^{-1}$ ), T3: liquid media with filter paper embedded, T4: media with normal agar ( $4 \text{ g L}^{-1}$ ), and T5: liquid media with filter paper projecting out.



**Fig. 2** – Laboratory performance of the 5 treatments T1: liquid only, T2: liquid with 50 % normal agar ( $2 \text{ g L}^{-1}$ ), T3: liquid media with filter paper embedded, T4: media with normal agar ( $4 \text{ g L}^{-1}$ ), and T5: liquid media with filter paper projecting out.

There was a major difference in growth rate of plantlets (Table 1), which indicates an interaction between treatment and environment. This result suggests that for long storage, the laboratory may be ideal while for short storage, screen house can be adopted. TMS 188/00106 survived better in liquid media than TMS 083/00125 (Table 2). The two genotypes grew equally in the media except on survival in liquid media only (Figs. 3–7).

**Table 1. Environmental effect on *in vitro* cassava survival, shoot root node leaf height**

Survival		Shoot		Root		Node		Leaves		Height	
SH	LAB	SH	LAB	SH	LAB	SH	LAB	SH	LAB	SH	LAB
1.77	2.41	1.92	1.93	0.73	1.55	2.00	1.93	1.92	1.93	1.92	1.96
1.67	1.44	1.56	1.42	1.67	1.33	1.86	1.63	1.67	1.46	1.61	1.33
1.02	1.81	1.21	2.04	0.46	1.42	1.46	2.17	1.29	2.00	1.21	2.04
1.73	1.27	1.83	1.00	1.00	0.75	2.04	1.29	2.00	0.96	1.79	1.00
1.73	1.27	1.88	1.13	1.63	1.04	2.13	1.29	1.79	1.13	1.92	1.17
<i>CV</i>	42%	49%		89%		48%		51%		48%	
<i>Lsd</i>	0.38.4	0.4470		0.2987		0.4889		0.470		0.439	
<i>Std</i>	0.193	0.2266		0.5892		0.2479		0.2383		0.2228	

SH = Screen House LAB = Culture room in the Laboratory

The survival was significantly different according to the media used (Table 3), which suggests that before the explants can be transferred to the screen house, one needs to ensure their survival in the laboratory. It can therefore be concluded that when the need arises, *in vitro* plantlets of cassava can be raised adequately in the screen house and even be raised faster than in the laboratory.

**Table 2. Genotypic effect on *in vitro* growth rate of cassava survival, shoot Root node, leaf height**

	Survival		Shoot		Root		Node		Leaves		Height	
	G <sub>1</sub>	G <sub>2</sub>	G <sub>1</sub>	G <sub>2</sub>	G <sub>1</sub>	G <sub>2</sub>	G <sub>1</sub>	G <sub>2</sub>	G <sub>1</sub>	G <sub>2</sub>	G <sub>1</sub>	G <sub>2</sub>
I	2.37	1.51	2.25	1.34	1.29	0.77	2.29	1.39	2.29	1.30	2.21	1.46
II	1.67	1.37	1.46	1.53	1.79	1.05	1.67	1.84	1.54	1.60	1.46	1.47
III	1.13	1.71	1.21	2.04	0.71	1.17	1.25	2.38	1.13	1.17	1.29	1.96
IV	1.54	1.46	1.33	1.50	1.00	0.75	1.79	1.54	1.54	1.42	1.42	1.38
V	1.54	1.46	1.58	1.42	1.00	1.67	1.63	1.79	1.42	1.5	1.54	1.54
<i>CV</i>	43%		50%		92%		48%		52%		51%	
<i>Lsd</i>	0.3898		0.4494		0.5889		0.4791		0.4684		0.4600	
<i>Sed</i>	0.1976		0.2278		0.2985		0.2429		0.2375		0.2332	

G<sub>1</sub> = TMS 188/00106 G<sub>2</sub> = TMS 083/00125

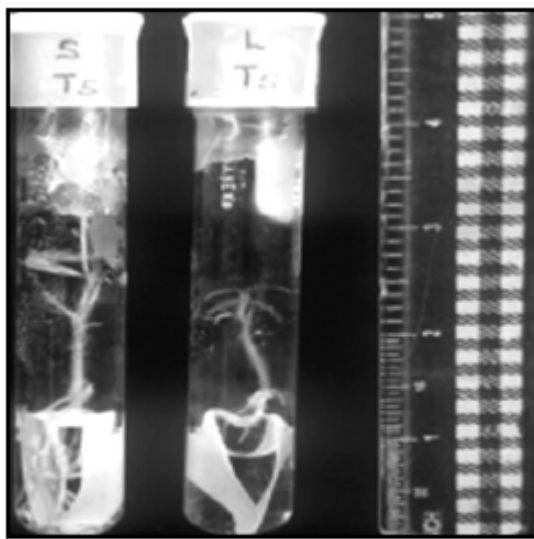


Fig. 3 – Comparison of treatment 1 [liquid media] in two environments [S = Screen house L = Laboratory]

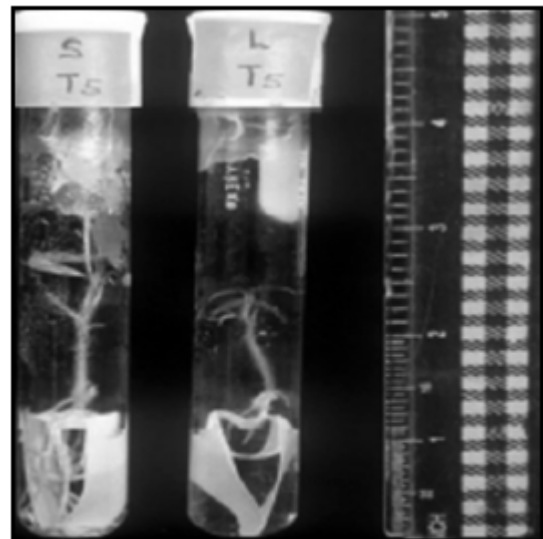


Fig. 4 – Comparison of treatment 2 [liquid with 50 % normal agar ( $2 \text{ g L}^{-1}$ )] in two environments [S = Screen house L = Laboratory]

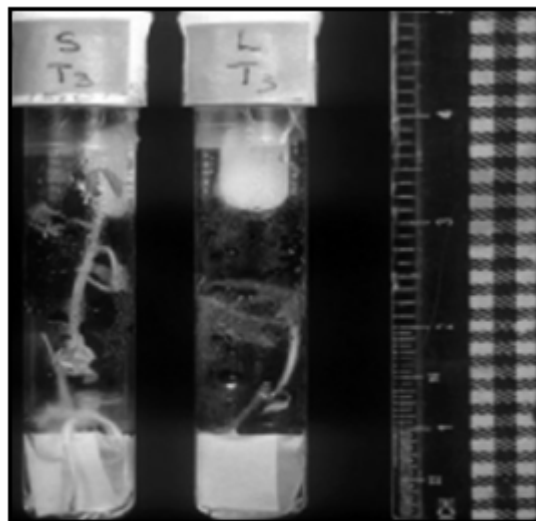


Fig. 5 – Comparison of treatment 3 [liquid media with filter paper embedded] in two environments [S = Screen house L = Laboratory]

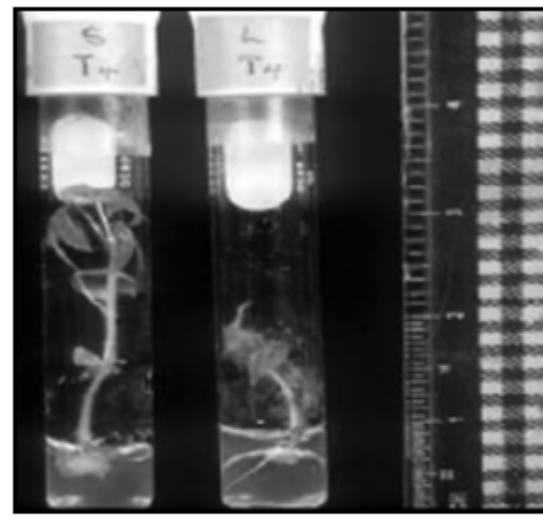


Fig. 6 – Comparison of treatment 4 [media with normal agar ( $4 \text{ g L}^{-1}$ )] in two environments [S = Screen house L = Laboratory]

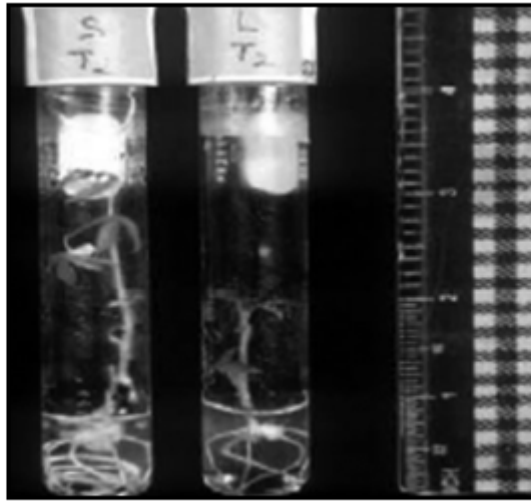


Fig. 7 – Comparison of treatment 5 [liquid media with filter paper projecting out] in two environments [S = Screen house L = Laboratory]

**Table 3. Analysis of variance of the effect on five treatment media on *in vitro* growth rate of cassava survival, shoot Root node, leaf height**

Parameters	DF	Sum of square	Mean square	Variation Ratio	F. Probability
Survival	4	14.1374	3.5343	7.87	<001
Shoot	4	7.7280	1.9320	3.12	0.008
Root	4	13.349	3.337	3.12	0.160
Node	4	2.6084	0.6521	0888	0.474
Leaves	4	75502	1.8875	2.77	0.029
Height	4	8.4901	2.1225	3.56	0.008

### Acknowledgment

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