TISSUE CULTURE FOR FARMERS: PARTICIPATORY ADAPTATION OF LOW-INPUT CASSAVA PROPAGATION IN COLOMBIA

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(Accepted 12 July 2005)

SUMMARY

The lack of good quality planting material of farmers' cassava varieties, produced locally and at low cost, is a major constraint limiting the expansion of cassava production in Colombia. This article describes the adaptation of conventional cassava propagation to a low-input scheme for rural tissue-culture multiplication, developed and run by small, resource-poor farmers (referred in this article as an informal-farmers' seed production system). Developed through a two-phase participatory process by a group of women farmers, a non-governmental organization and International Center for Tropical Agriculture scientists in a farmers' community in the hillsides of southern Colombia, the project resulted in alternative, economical and readily available sources of tissue-culture material and equipment. Rates of multiplication achieved with the system were as high as with conventional tissue-culture procedures.

INTRODUCTION

Cassava (*Manihot esculenta*) is a crop of tropical American origin that today sustains more than 500 million low-income people. A staple in the small farmer's 'market basket', due to its highly efficient production of carbohydrates per unit of cropping area, cassava's low requirements for inputs, water, and technology also give it a margin of profitability for the small farmer.

Cassava has become increasingly important to industry because of the variety of its starches, its potential as a substitute for maize in feed concentrates and for wheat in bakery goods, and its role in the production of glue, paper and biodegradable plastics. This potential has helped change cassava's standing from being a 'poor man's crop' to being seen as a profitable cash crop.

Average world production of cassava in the last 10 years was 162 million t, with Africa, Asia, and South America being the principal producers. Colombia is the

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third largest producer in South America, after Brazil and Paraguay, with an average production in the last 10 years of 1.8 million t (FAOSTAT, 2001).

Cassava is generally cultivated on soils that are too degraded and marginal for normal agriculture. In Colombia, there are two types of cassava-producing areas, based on the crop's use:

- 1. For *human consumption*, it is grown in areas near centres of consumption, in all types of soils and climates, such as in the coffee zone, Santander, North Santander, Piedmont Eastern Plains and Valle del Cauca.
- 2. For *industrial use and animal consumption*, it is grown where competition with other crops is unlikely, i.e. in soils that are of low fertility and usually marginal, such as in the Atlantic Coast, Middle Magdalena, Eastern Plains and the Cauca Department in southern Colombia. Cassava is a high-priority crop in the Cauca region, where there are about 200 local artisanal systems for extracting starch. Known as *rallanderías*, they employ most of the region's labour and supply almost 80 % of the national demand for sour starch.

Despite the crop's importance, the average yield of fresh cassava roots is $10-12 \text{ tha}^{-1}$, whereas, with improved varieties and adequate agronomic management, yields could be as high as 36 tha^{-1} .

Cassava is conventionally propagated by planting 'stakes', pieces of the plant's woody stem. Although this system is practical, it generates a series of challenges such as short shelf life, low propagation rates, and inconvenient weight and bulk of material, with high handling and transport costs. Sanitary health is particularly difficult to maintain, and stakes often become the means for disseminating pests and diseases. This problem is so serious that regulations have been put in place to control the movement of germplasm between production areas, countries and continents.

In addition, the lack of alternative markets, unstable and uncertain prices, social unrest in production areas, lack of credit and lack of incentives to grow the crop have resulted in a failure to realize cassava's potential as a profitable crop for small farmers in the region.

Among all these constraints, one of the most important is the lack of good quality planting material for varieties of interest to farmers and processors. During the 1980s, the International Center for Tropical Agriculture (CIAT) attempted to address this problem by developing a tissue-culture system to eliminate pathogens and to propagate and conserve cassava genetic resources (Roca, 1984). Shortly after that, in 1995–1997, there was a decrease in maize imports in Colombia, which led to cassava being considered a profitable alternative. The Colombian Government also set up a programme to substitute illicit crops with other crops, including cassava. This caused a sudden increase in demand that led to a major bottleneck because of the lack of local, low-cost supplies of healthy planting material for local varieties.

This paper presents our group's experience in establishing a participatory scheme for strengthening the informal-farmer system of cassava seed production through the use of low-cost *in vitro* techniques that can be carried out by resource-poor farmer groups. The Women's Group in the community of Santa Ana, CIAT and the Foundation for

Agricultural Research and Development (FIDAR), a Colombian non-governmental organization (NGO), joined their efforts to solve what seemed, at first, a fairly complicated technical agenda. However, having mutually desired agendas, and close collaboration and exchange of insights throughout proved key, from the first stages of idea development, through to the testing of prototypes, and eventually to the community's taking over the management of successful propagation models (Ashby and Sperling, 1995). This paper provides details of the technical specifics, as well as the social, participatory processes, which led to the results achieved.

MATERIALS AND METHODS

Plant material

The local variety 'Algodona', which is widely accepted by small farmers, was used in this study. This is a variety high in dry matter (28 %), of which 83 % is starch with 22 % amylose content (T. Sánchez, personal communication, 1999). Algodona's highly marketable fresh roots are long and white, and the root peel is a dark coffee colour. The variety tolerates thrips and *Phoma*, but is highly susceptible to frog skin disease (FSD) (a very detrimental virus), and to cassava bacterial blight (CBB).

Most farmers classify 'Algodona' as 'the variety that gets ill the most'. Even so, it covers 80 % of the area planted to cassava on the hillsides, and despite being considered as late maturing (14–18 months for harvest), it has a fresh-root yield of 7–11 t ha⁻¹, with an average harvest index of 0.36, which is considered to be fairly poor by farmer standards.

The site

Our project was developed on the hillsides of the Santa Ana Village District in the Department of Cauca, Colombia. Soils are mostly shallow to moderately deep, red, with limited organic matter, and acid, with high interchangeable aluminium content and low levels of phosphorus with high fixation rates. Rainfall is heavy. Planting on slopes is carried out without soil management or conservation practices, leading to serious erosion problems (G. Jaramillo, personal communication, 1999).

The principal constraints to cassava production in the area are a high incidence of FSD, whitefly (*Bemisia tuberculata*) and burrower bug (*Cyrtomenus bergi*), together with the fungus *Phoma* and leaf-cutting ants (*Atta* spp.). Frog skin disease is considered to be the greatest constraint to crop production, causing losses as high as 90 % or more in fresh root yield (Calvert *et al.*, 2001). The disease is spread mainly through contaminated planting stakes.

In Cauca, cassava is produced by about 5000 small farmers and their families (approx. 24 500 people) of indigenous, mestizo and African origins. Poverty in the area is high and is associated with marginal agriculture. In spite of the widespread lack of schooling, training and technical assistance, these people generate over 58 000 t of fresh cassava roots on farms averaging 2–4 ha. They sell 80 % of the production to the sour starch *rallanderías* for income generation. The remainder is consumed or sold in the region's markets.

The economic benefits of cassava production to small farmers have been limited by problems in the supply of planting material of sufficient quantity and of suitable genetic and sanitary quality.

Project participants and their functions

This project began with support from the Cassava Biotechnology Network (CBN) in 1998, with the purpose of establishing a local, low-cost production system for clean planting material for small farmers. Other participants included FIDAR, which has the mission to promote production models in communities with limited economic resources in both urban and rural sectors. FIDAR works on agricultural and environmental issues, including cropping systems, water and soil conservation, reforestation, management of local diversity and integrated pest management. Its function within this project was to serve as a bridge between farmers and researchers during the experimental phase (including participatory and gender components) and, in the diffusion phase, to help other groups in the area.

CIAT, located in Palmira, Colombia, is a non-profit entity belonging to the Consultative Group on International Agricultural Research (CGIAR). Its mission is to reduce hunger and poverty in the tropics through collaboration in the use of agricultural research aimed to improve productivity while maintaining good management of natural resources. The centre has broad experience in such areas as management of genetic resources, biotechnology, breeding, entomology and pathology in crops such as cassava, beans, pastures, forages and rice. CIAT's function in the project was to provide know-how on tissue culture and to develop, in collaboration with the farmers, low-cost conditions appropriate for establishing a rural tissue-culture laboratory.

In addition, there were contributions from the CGIAR's Program on Participatory Research and Gender Analysis for Technology Development and Institutional Innovation (PRGA) and the CBN. PRGA contributed strategies for managing gender and following-up participatory developments. CBN contributed with feedback on the needs of small cassava farmers, updating and articulating the agenda among them and the researchers.

The target farmer group was identified through surveys and meetings held either in the Cauca area or CIAT headquarters. Project objectives and mechanisms were discussed with 20 farmer groups, including farmer associations from El Turco, Morales, Buenos Aires and Santander, as well as governmental and non-governmental organizations within the municipalities of Caldono, Santander and Caloto. Following the farmers' suggestions, CIAT and FIDAR identified a group of mostly women farmers from the Santa Ana Village, Santander de Quilichao Municipality. This group had a background of community work, for example, road and house improvement, household vegetable gardening, school construction and a parents' board for action, all of which indicated that the group was organized and committed to its objectives.

The group was made up of nine women, three of whom were household heads, all more than 38 years old and owners of the land on which they lived. In total, they had 35 children, 21 (60 %) of whom were minors attending a nearby rural primary school; the older children had formed new family groups, but most of them still resided in the

maternal house (Ospina, personal communication, 2001). Of the women, four had not completed primary school, three had a primary education, and two were high school graduates. As participants in the project, they contributed their knowledge of cassava production, management and constraints, and assisted in establishing and leading a joint working scheme with other project's participants.

All group members were experienced with cassava and identified the need 'to improve the quality of its seeds' because the 'native varieties are becoming exhausted and the scourge of pests and diseases to the crops of the region have affected the community's economy'.

Selecting a farmer-facilitator in the community

A normal day for the women who participated in this project includes activities such as attending to the home and family (i.e. caring for the children, preparing food and doing housework) and maintaining the household's vegetable garden. On Wednesdays and Fridays, they go to market to buy and sell produce. They also plant and maintain different crops (cassava, coffee, pineapple, plantain) and attend to other duties to complement their spouses' activities. Sometimes they take up casual labour in other plots.

Their involvement in the project was defined as 'extra activity'. Some women agreed to programme these activities for the afternoons (after 14.00 hours.), after having done their daily tasks, and others, after watching their mid-day 'TV soap show'. The women have a crucial role in household activities and, with only the afternoons available for them to go to CIAT (which is about one hour's drive by car from Santa Ana), training was difficult. To confront this challenge, a farmer-facilitator was selected by the project to work with the women's group in their own village.

This facilitator was trained at CIAT headquarters three to four days per week (for a period of six months), and then visited the women's group in the village for one to two days per week, to help the development and adaptation of the *in vitro* technology. Several criteria were used to select the farmer-facilitator. These included: (i) the person had to have a high interest and motivation to be involved in the project; (ii) he/she had to be farming in the area and had to have experience with cassava; (iii) the person had to be able to project confidence and credibility; (iv) the person had to possess good communication skills; and (v) it had to be someone with enough time to fulfil the needs of the project. The person's gender was not seen as a critical selection criterion, as having time to travel to CIAT for training activities, was the really key point. Women consider that *'it's not easy for us with house-activities and children to neglect our duties'*. Using these criteria, a male-facilitator was selected who had experience with and a role in other participatory research projects, as well as some experience with agricultural techniques.

Establishing the pilot site

The farmers and the facilitator made several visits to CIAT's headquarters to learn conventional *in vitro* multiplication of cassava. They also helped set up a workplace at CIAT that would reflect the actual day-to-day conditions of the field. The tissue-culture pilot site was located in a rudimentary storeroom of CIAT's gardening section.

Constructing a common language

Constructing a common language between the women's group, the facilitator and the CIAT researcher took place primarily during monitoring and feedback sessions. These sessions were carried out in two ways: 'as the farmer does it and as the laboratory does it', as the farmers would say. Although this took some time, a common language was seen as crucial to effective communication and the success of the project overall.

In vitro conditions

Roca (1984) established the conditions for cassava pathogen elimination, propagation and conservation using conventional tissue-culture techniques. Medium 4E, used for propagation, included MS salts (Murashigue and Skoog, 1962), $0.04 \text{ mg} \text{l}^{-1}$ BAP, $0.05 \text{ mg} \text{l}^{-1}$ GA₃, $0.02 \text{ mg} \text{l}^{-1}$ NAA, $1 \text{ mg} \text{l}^{-1}$ thiamine, $100 \text{ mg} \text{l}^{-1}$ m-inositol and 2 % sucrose, at pH = 5.7–5.8. Medium 17N, used for rooting, included MS/3, $25 \text{ mg} \text{l}^{-1}$ of the fertilizer Plantex[®], $0.1 \text{ mg} \text{l}^{-1}$ NAA, $0.1 \text{ mg} \text{l}^{-1}$ GA₃, $1 \text{ mg} \text{l}^{-1}$ thiamine, $100 \text{ mg} \text{l}^{-1}$ m-inositol and 2 % sucrose. These media continue to be used in the laboratories of CIAT's Biotechnology and Genetic Resources Units, and have been adopted by many of the region's laboratories.

In conventional research, results and quality of processes are ensured by the use of high-purity reagents, including mineral salts, hormonal regulators, vitamins and agar. These are costly inputs, to which importation tariffs and other duties must be added. In addition, some of these chemicals are regarded as dangerous to human health, making it necessary to follow the restrictions and regulations that govern international transport of chemical products, which subsequently generates a considerable increase in cost.

Although tissue-culture propagation is a simple technique, it requires a basic set of equipment, such as a flow chamber to ensure 'freedom from contaminants' during operations, a de-ionizer to control the total ionic concentration of water, an autoclave to sterilize culture media, an analytical scale to weigh small amounts of solids, a pH meter, a stove to heat solutions and a refrigerator to store media components, among other things.

Special working facilities are also needed to prepare culture media, to sterilize and wash equipment and materials, and to store cultures. Conditions in the tissue growth area – temperature (28 ± 2 °C), illumination (1000 lux), photoperiod (12 h of illumination), and relative humidity – must be controlled to achieve optimal cassava growth and development during *in vitro* propagation.

RESULTS

Having established both the social basis for collaboration (a defined challenge, an interested farmer group, a communication bridge with farmers and a common farmer-researcher language) and the technical basics (prototype equipment and tools, work spaces and inputs for culture media), the group moved forward to investigate solutions. Our agreed upon objective was to develop alternative economical and readily available sources of materials and equipment that would make the adaptation

and implementation of the tissue-culture methodology feasible for any farmer group. Below we discuss the processes and product modifications made to arrive at something useful and usable. This includes specific discussion of: the tools, laboratory equipment, modification of both the CIAT and rural laboratory sites, weighing of different culture media and food for plants, and altering of transplant methods.

Tools for a rural laboratory

Test tubes and special plastic-ware were replaced by recyclable jars of any type, provided they had wide mouths and could be autoclaved, e.g. baby food, mayonnaise, and preserve jars or soft drink, juice and local liquor bottles.

Because few women used electric stoves and because of frequent electricity blackouts, the stove and sterilizer were converted to gas, which turned out to be a cheaper alternative. Nevertheless, the gas stove represented some difficulties for the group, in that some members were more familiar with using wood stoves for domestic tasks. The gas stove was also regarded as dangerous, and some people were afraid to use it.

To replace the Petri dishes (used for excising plant parts), glass plates (3 to 4 mm thick and 15 to 20 cm wide), bathroom tiles, Kraft paper, printing paper, or newspapers, dinner plates, or metal tops of preserve jars were used. Newspaper also served to wrap implements for sterilization.

Tools for cutting plant tissues included tweezers, scalpel handles and No. 10 blades. Sharpening with a knife sharpener and wet/dry sandpaper grit 400 prolonged the blades' useful life. Periodically, they were washed with a little soap and a brush, and care was taken not to place them in contact with strong cleaners or hypochlorite. In cases where not enough funds were available to purchase these implements, substitutes were made with razor blades, glue and wooden sticks.

To disinfect work areas, hands and tissues, 70 % alcohol (an antiseptic that is freely sold) and 96 % alcohol (for burners) were used.

Adapting and constructing laboratory equipment for rural conditions

In a conventional laboratory, precision weighing and volume determination is done with analytical balances, pipettes and micropipettes. These tools were replaced with 1, 5, 10 or 60 ml capacity syringes to measure volumes and knives or spoons of different sizes to measure weights (for example, dessert spoons and soup spoons; with four level soup spoonfuls equalling approximately 20 g of sucrose). Although some precision was lost, an economical and practical system was generated.

To prepare culture media, glassware such as beakers and Erlenmeyer flasks were replaced by containers obtained by adapting plastic soft-drink bottles or buckets, or enamelled pots for use on the stove. A common laboratory equipment agitator-heater was replaced by a gas stove and manual agitation with the aid of a wooden spoon. Cold boiled water for preparing media was stored in clean liquor or soft-drink bottles.

The pH of the culture media can be determined with either indicator paper or using a pH meter. Both methods have advantages and disadvantages. Indicator paper is highly economical and is available in different scales, but provides only approximate values. The pH meter is more precise but requires regular calibration and access to

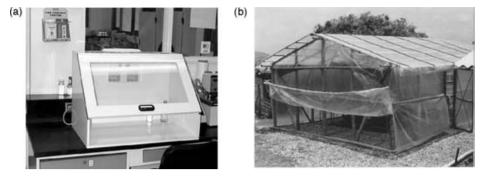


Figure 1. (a) A low-cost wooden flow cabinet for a rural community tissue-culture laboratory. (b) A plastic-covered growing area on a patio, where micro-propagated cassava plantlets were grown. Note that the plastic walls can be lowered or raised to control temperature.

special calibrating solutions. It is also more delicate and expensive. Based on cost and practicability, it was decided to use indicator paper with a unitary scale for adjusting the pH to 6.

In tissue culture, close attention must be paid to possible sources of contamination, such as plant tissues, the worker and the environment, in order to avoid incalculable losses in work time, inputs and other resources. Some equipment, such as the laminar flow chamber, helps ensure asepsis and freedom from contaminants during tissue-culture processes, but is expensive (US\$4000). The Santa Ana Women's Group was provided with a flow cabinet, but when the project was transferred to another group, such a donation was not possible, so a substitute had to be devised.

One of the options considered for replacing flow cabinets was to work near alcohol burners. However, the high temperatures would dehydrate the tissues and the possibility of workers being burnt led to this option being ruled out. Another option was the use of plastic cabins, which was first tried out in Vietnam in 1980, in an experiment on potato propagation by farmers. A 20-watt UV lamp was placed inside the plastic cabin 30 to 40 minutes before starting tissue propagation (N. Van Uyen, personal communication, 1999). The cost of this equipment in 1998 was US\$1000, so other options were considered, including aquariums, a glove chamber and acrylic boxes. The option finally used was a homemade wooden cabinet at a total cost of US\$350. It minimized the risk of burns and used a filter (Holmes Inc., reference HAP 240C) that was easy to remove (Figure 1a).

Adapting CIAT's pilot work site

Three working areas were established at the CIAT pilot site: a kitchen, a workroom and a patio. Equipped with a bench, washing area and stove, the kitchen was where the containers were washed, culture media prepared, and heating and autoclaving carried out.

The workroom, where plant tissue propagation was carried out, was well lit and ventilated. The only rules for entering this room were to wear clean clothes and not to have visited any fields on the day of entry. The flow cabinet was located in this room, opposite the door, on the side where there was the lowest airflow. By preventing strong air currents and cleaning the room daily with a wet rag environmental contamination was controlled.

The patio, containing the growth room, had a transparent roof and plastic 'walls' to filter the light. The 'walls' were secured with cord so that the plastic could be raised or lowered, like blinds, thus regulating temperatures (Figure 1b).

The floor was covered with gravel to control weeds and prevent puddles during the rainy season. When temperatures within the growth room climbed to 40 °C watering the floor cooled the area; however, in the rural community in Cauca, water pressure was insufficient for this, and during the dry season, water was scarce and its use was confined to household consumption.

Adapting the pilot site to the rural community site

With the experience gained at the CIAT pilot site, a rural laboratory prototype was built at the Santa Ana field site.

First, a structure was built using bamboo (*Bambusa guadua*) canes set in concrete foundations to prevent rotting and to prolong useful life. Group members and FIDAR, with suggestions from researchers and the facilitator, agreed to locate the community laboratory at a member's house, as it was close to the community hall and easy to reach. A propagation room (2.5 m^2), a kitchen (1 m^2), and a patio with a growth room (10 m^2) were constructed. The women's group participated in the construction, painting, installing lights and clean-up of the laboratory.

The patio of the pilot laboratory at CIAT had a fence of different plants, including a balsa tree (*Ochroma pyramidale*), which provided shade and helped to maintain moderate temperatures. The roofing sloped gently at an angle that favoured the accumulation of branches, leaves and seed silk that fell from the tree. In contrast, in Santa Ana, the patio was located on a high flat area near the house, where there was little vegetation, making it difficult to regulate temperatures. However, the roofing had two sloping sides (Figure 1b).

The optimal temperature for *in vitro* cassava propagation is 28-30 °C (Roca, 1984). Different types of mesh, located within and outside the patio, were evaluated for their capacity to regulate temperature within the patio. The best results were obtained with a white mesh made with empty 50 kg fertilizer bags and installed as over-roofing, and a sanitary PVC tube placed 1.2 m above the ground to act as a chimney. The bags were washed, opened along the middle, and sewn to each other to cover the roof area. The resulting covering was then attached to the roof by its corners, leaving the rest free to maintain a layer of cool air circulating between the roof and mesh. The mesh also reflected part of the solar radiation during the day. On average days, this reduced the temperature from 38 to 30 °C and, on cool days, increased humidity from 50 to 80 %.

Culture media for the rural community laboratory

A conventional culture medium usually contains the nutrients that plants need for adequate development, including (a) a basal solution (which provides major and minor

mineral elements), (b) a source of carbon (i.e. sucrose), (c) some vitamins (especially B vitamins), (d) growth regulators, (e) a gelling agent for solid media, and (f) other supplements.

The principal growth regulators include auxins, abscisic acid, ethylene, cytokinins and gibberellins. The auxins, cytokinins and their interaction are generally considered of great importance in regulating growth and organized development. These ingredients are usually acquired abroad, with the agar and growth regulators being the most costly. The selection of alternative products to replace imported culture media was key to the laboratory's success because '*the food for plants*', as the farmers called the culture media, was prepared from these alternatives.

Some herbicides found on the market-contained compounds known to have an auxin-like effect, e.g. 2,4-D, picloram, dicamba and TDZ. Some commercial root promoters contained IBA or NAA as an active ingredient, and compounds that helped maintain flowering, break seed latency, and retard or accelerate fruit maturation contained gibberellins.

The Mexican *Diccionario de Especialidades Agroquímicas* (Rosenstein, 2000), published annually, lists agricultural inputs distributed without restriction in Colombia, including the active ingredients, correct use, care and dosage. The products $ProGibb^{(R)}$ (10 g having at least 10 % GA₃) and Hormonagro^(R) liquid (250 ml containing 17.2 g l⁻¹ of NAA) were considered. Based on the price in the Sigma catalogue for 2002–2003, 1 g of imported GA₃ cost US\$32.50 (not including shipping and import costs) compared to the local product at US\$1.40. Likewise, 25 g of imported NAA costs US\$14 versus US\$5.20 for the non-reactive local product. Such price differences represented savings of 96 % and 54 %, respectively, on costs alone, without taking into account savings in time and effort with import transactions.

Table sugar was used as a source of carbon. Thiamine and m-inositol were substituted with a complex of B vitamins, easily acquired from the drug store.

As gelling agents, cassava starch, unflavoured gelatine and a cheap agar distributed for school laboratories were all tried. One kilo of imported agar costs US\$450, whereas school agar costs US\$35, a savings of 92%. School agar has a clear matrix that facilitates the determination of contaminants. Although cassava starch was economical, it was not the best initial choice because contaminants were hard for farmers to detect. Moreover, the quality of gel depended on the type of starch – whether it was sour or fresh, as Nene *et al.* (1996) have suggested – and it disintegrated over time, becoming liquid. With time and experience, cassava starch probably could have been used, or it could have been mixed with agar to reduce final costs and improve quality. The unflavoured gelatine gave a very rigid, plastic matrix that expelled tissues and prevented contact with the medium. Moreover, it re-melted when placed near heat, and in terms of price, it was more expensive than the school agar.

Based on catalogue prices, the cost of a litre of medium 4E is US0.6, with the agar representing 43 % of the cost and the NAA and GA₃, 0.2 %. If local inputs were used with this medium, without taking the MS salts into account, costs dropped more than 50 %.

The initial transparency of the culture medium was taken as an indicator of *'cleanness'*, which facilitated the implementation of a practical system to detect contaminants by associating the *'appearance of coloured spots'* (e.g. green, orange, yellow, black, red) with contamination. The number of *'coloured spots'* was directly related to the level of contamination.

To replace the basal solution, several fertilizers were tested. The conventional ones (N:P:K 10:30:10 and 15:15:15) presented problems with the sediments they produced in the medium, which affected the plant tissues. Some, such as Cosmocel[®] (20:30:10) prevented stabilization of the pH at 6 in the culture medium. Generally, the concentrations tested in these products were toxic to the tissues: in most cases, chlorosis appeared within three to five days; at other times, the whole tissue appeared burned. However, Walker (1999) has reported success with fertilizers when attempting to propagate the carnivorous plants *Pinguicula* and *Drosera*.

Medium preparation had to be simple, and similar to a kitchen recipe. During the project's socialization phase, an example was used of the care taken with the amount of salt used in cooking, so that the food was neither unsalted nor too salty. This helped the group understand the need to be careful with the quantities used for each ingredient of *'the food for plants'*.

In all, more than 130 possible combinations of fertilizers, substitutes for growth regulators, organic supplements, and gelatines were tested to find a substitute for medium 4E used by Roca (1984).

Preparing 'working solutions' (WS) was an easy way to make the medium. For Hormonagro[®], 1 ml of commercial product was added to 9 ml of water; 1 ml of this working solution could then be diluted in 99 ml of water. For Progibb[®], the working solution required 200 mg of the product dissolved in 200 ml of water.

Food for plants and propagation rates

Of the combinations tried, the best results were no. 8 (BM + $0.02 \text{ mg} \text{l}^{-1} \text{ NAA} + 0.05 \text{ mg} \text{l}^{-1} \text{ GA}_3$), no. 9 (BM + $1.2 \text{ ml} \text{l}^{-1} \text{ Hormonagro}^{\$} WS + 0.5 \text{ ml} \text{l}^{-1} \text{ ProGibb}^{\$} WS$) and no. 10 (BM + $1.2 \text{ ml} \text{l}^{-1} \text{ Hormonagro}^{\$} WS + 5 \text{ ml} \text{l}^{-1} \text{ ProGibb}^{\$} WS$), where BM consists of the MS salts + 3 g l⁻¹ sucrose + $1 \text{ mg} \text{l}^{-1}$ thiamine + $100 \text{ mg} \text{l}^{-1}$ inositol + 0.9 % agar; pH = 6.

The propagation rates (3:1) under farmers' conditions were no different from those normally obtained using medium 4E under conventional tissue-culture conditions. The rate is obtained by comparing the number of planting '*heads or small trunks*' (small plant shoots or buds) recovered in relation to the initial tissue set, after 45 days of culture.

Tissues recovered from media no. 9 and no. 10 were more vigorous and of a more intense green colour than tissues from medium no. 8. The media selected displayed homogeneity across several trials.

When supplementing these media with banana extract (1-10%), coconut milk (5-10%), or tomato or pineapple juices (0.5-2%), we observed that despite there being no increase in propagation rates, the appearance (stronger, greener and more vigorous)



Figure 2. (a) A woman farmer works in a rural community laboratory that she helped build for *in vitro* propagation of cassava. (b) A woman farmer planting a cassava plantlet propagated in a rural community laboratory in Cauca Department, Colombia.

of tissues was significantly improved, especially when tomato or pineapple juice was used.

Once the women's group had adjusted to the tissue-culture propagation process (Figure 2a), the percentage of initial contamination dropped from 40–60 % to 5 %. The women's observations during the process also enabled them to exercise quality control: when the agar was exhausted in the flasks and a new lot was added, they referred to the associated changes in tissue behaviour as the 'fish smell of the new agar'.

Transplanting from flasks to soil

For this phase, the methods known for cassava as proposed by CIAT (Roca, 1984) and the International Institute for Tropical Agriculture (IITA) (Ng *et al.*, 1994) were tested. In the CIAT method, each plantlet is covered with a perforated polystyrene cup that acts as a humidity chamber. In IITA's system, plantlets are placed inside plastic bags, which are then sealed and hung in the shade. Although both methods lead to good results, they need care in watering. They also tend to incur extra costs, and, at the end of the process, the cups and transparent bags have to be stored until the new cycle begins.

Preliminary trials showed that, in the IITA system, contamination foci developed in the bags over repeated cycles. Moreover, at the end of the process, each bag contained not only a 10–15 cm plant, but also 1 kg of soil, requiring the use of a truck for transporting the plants to their field site. Because of this, Escobar (1991) adapted another simple and economical system to prevent contamination of plantlets and reduce transplanting trauma: one week before transplanting the plantlets to soil, the sealing tape was removed from the jars containing the plantlets. This permitted a gradual reduction of humidity within the jars, ensuring that the plantlets would begin to adjust to more field-like conditions. At the end of the week, the plantlets were removed from the jars and the agar gently washed off their roots. The jars were also washed and enough tap water was added to cover only the roots. The plantlets were placed in the water and left for one week with the water changed daily to prevent rotting. The plantlets were then planted in plastic bags containing 500 g of steam-sterilized soil made up of coarse sand and soil at a ratio of 3:1. The soil was sterilized with equipment constructed from two metal drums joined with a plastic hose; steam was generated using firewood collected locally. The plantlets were watered and given fertilizer (one small spoonful of complete fertilizer per 5 l of water, applied every 30 days to favour plant development) for two months until they were 10 to 15 cm high, when they were transplanted to the field. To be successful, transplanting should be done on a cloudy day or during the late afternoon, during the rainy season or with irrigation. The day before transplanting to the field, the plants were not watered.

Farmers carried the bags with the plants to the field site and placed them, still in the bags, on the ground (Figure 2b). Plants were then removed from the bags and placed within a hole as large as the bag, and up to the lowest two nodes of the shoot. The soil around the plant was pressed and care was taken to maintain high soil humidity after transplanting. This procedure not only reduced the handling of fragile plantlets, but also reduced contamination and subsequent losses in transplanting from agar to soil. Currently, losses due to this method of transplanting under farmers' conditions are about 10 %, versus 40 % when the conventional CIAT system is used.

DISCUSSION

Use of tissue-culture techniques allows for the propagation of large amounts of planting material, economically and in a short time. Such techniques can contribute to the rapid adoption of new varieties as well as the sustained use of local ones (as farmers can maintain and multiply them widely). While 1 ha of maize can produce sufficient seed for more than 100 ha of crop, 1 ha of cassava produces stem cuttings for only 7–10 ha (Ceballos, 2000). The participatory and scientifically rigorous methodology developed in this study can support more effective, informal (farmer-based) cassava production with propagation rates of three from each *in vitro* plant every 45–60 days. Theoretically, this system could produce in a year between 729 to over 6561 new plants from one initial *in vitro* plant.

A conventional cassava stake costs US0.012. Recently, private enterprises in Colombia have marketed cassava-planting material derived from *in vitro* plants for US0.295 per plant. With a planting density of 10 000 stakes ha⁻¹, this is equivalent to an investment as high as US2950 for one hectare – an investment that is well out of the reach of a small farmer.

Our study shows that a well-organized group can adapt a low-cost, small-scale multiplication technique and establish an informal-farmer decentralized system to produce disease-free planting material of local varieties, in sufficient quantities to generate a micro-business. However, cassava farmers usually do not buy seed; they use better-quality seed only if it is given to them. This poses the additional challenge of integrating other crops, whose seed can be sold, into the scheme developed for cassava so that investment and effort could be recouped. CIAT is following the group's suggestions and projections to add other crops to the scheme and thus establish a platform from which to increase the range of crops and strengthen the idea that clean planting material can be produced in a rural laboratory.

Some reflections on the project process

While the results of the project have been considered useful for all, we want to emphasize that the process was not always an easy one. For instance, in the beginning, the various groups had different perceptions of this project. Some groups, like that at Santander of Quilichao, were made up only of men, and others, like that of Santa Ana, only of women. In general, men had different expectations and reactions to the project – they were interested in the planting material, or seed; they saw the work itself as very delicate or feminine. In contrast, the women were more receptive to discovering how to produce this particular seed, seeing it as a process. However, a deeper reason was, according to one man's perception, that '*women had more time*', by which he meant that their time was not remunerated, whereas the men's own work activities were. The men were not willing to commit themselves to the project and put their already precarious economic situation even more at risk.

In spite of the men's acceptance of their wives' participation in the project, they did nothing to help with the domestic work while the women were out. In addition, they accepted the women's participation only so long as the women's domestic responsibilities were not neglected.

Below we reflect further on some of the social insights and collaborative features which have made this project a useful one. This is not meant to be a comprehensive 'guide' to participatory research, but rather to highlight the salient process elements, which were important to our case.

Farmer involvement from the very beginning

Farmers do not always easily adopt new varieties, even for crops with which they are familiar. Sperling *et al.* (1993) pointed out that at the end of an improvement scheme, involving selection, trials and multiplication, it is the farmers who decide whether to adopt the new material or not. Including farmers at the beginning of any improvement programme is the key to anticipating possible problems in the technology design. Effective programmes ensure that farmers have a strong voice throughout the range of activities. In particular, farmers and their communities should help to define (and redefine) goals, weigh the different options to be tested, evaluate options actively and have their feedback taken seriously. Farmers themselves also have to make steps to learn the concepts and incorporate appropriate insights of other partners – in this case, for instance, knowing and familiarizing themselves with *in vitro* material to the extent of regarding it as if it were part of their daily lives.

Respect for farmers' time and multiple duties

As suggested by Weltzien *et al.* (2001), respect for farmers' time is critical. The organization and time that farmers are required to devote to a project should not

conflict with bottleneck periods, such as harvesting or planting, or other occasions when the farmer's time may be especially limited. Even in our case, we had to respect the 'TV soap show hour' at mid-day, as some members would not go to appointments before 14:00 hours. Likewise, to encourage member participation, the social worker and the farmer-facilitator generated a space in which group, artistic and recreational activities (sometimes integrated with adults) were developed for the group's children. Group members could then devote themselves to their activities, assured of their children's safety.

Programming for a farmer-facilitator

It was not possible to establish a programme of frequent CIAT visits because of problems of social unrest in the rural area. The confidence and empathy that the group developed toward its farmer-facilitator thus became particularly important (Braun *et al.*, 2000). The farmer-facilitator helped correct some technical difficulties and his easing of two-way farmer-researcher communication helped the group consider and understand the opinions, proposals and objectives of the different parties involved in the innovation scheme. Because this person could also maintain greater contact than the farmers with the facilities of a conventional research laboratory, the facilitator could learn more about certain terms and managing certain equipment, and so teach the group. By the end of the experiment, as a mechanism for *expanding their knowledge*, some women also familiarized themselves with many of the relevant terms.

Being forthright about the risks and benefits: for individuals as well as the community

The group was easily discouraged when their work became contaminated, or the material did not grow, or they could not control the temperature, because all their hopes, expectations and efforts to improve their conditions became centred on these small jars. This is unlike the situation of researchers, who have no such personal stake and who might see such failure as normal – or as the point from which to begin new research or to adjust the process. Generally, group members expected their efforts to be rewarded with quick benefits and effective responses over the short term, preferably generating income. This means that, in developing this type of activity, project leaders must, from the very beginning, explain the risks involved in research and continually discuss successes and setbacks in the project's progress.

Even where the women perceived the possible benefits to the community and the region, they could not find a clear personal benefit, especially an economic one. To increase a little the individual participant's own income, the project also supported other initiatives, such as raising chickens and establishing community plots for cassava production (from which produce could be sold).

Negotiating a common language

The building of a common language helped encourage mutual learning – and allowed members to be mutually understood. Aided by the farmer-facilitator, such language building was based on comparing the conventional laboratory and the farmers' systems. '*How to do it*' allowed the farmers to construct a concept of the process. They could then establish equivalences or similarities among the systems. Together, the group built a simple definition of '*propagation in little jars* (in vitro) *consists in planting heads or small trunks* (small plant shoots or buds) *to give them food* (culture medium) *so that they may grow and form new plants again*', as farmer Hilda M. Gómez, 50 years old, put it at the end of the experiment's first phase.

Building farmer self confidence – and providing constant motivation

At first the group saw the researcher as being that '*person who qualified*' their work as being either good or bad, increasing their nervousness before new experiences. At the beginning of the project, they were also insecure when dealing with the new ways, confronting as they did, years of inherited tradition. Even among themselves, they were nervous, saying they '*had to learn to overcome* [the idea] *that others would see us*'. So building confidence was a constant process and the project leaders gave the members the personal challenge of trying to solve a community problem and showing their children that '*while their father works in the field, their mother is doing something important and is getting trained*'.

Lack of schooling itself was not usually a problem for participation. Use of the farmer's simple everyday language and '*learning by doing*' helped personalize and favour the adaptation and final use of the jointly developed solutions. Even so, phrases such as '*I am too old for this*' or '*I'll never learn*' were frequent and the self-esteem of group members had to be strengthened and promoted so that they would believe in both their own and the group's possibilities, offering mutual support. While some believed that their time for learning was past, the oldest woman in the group (Nohemi Larrahondo, 64 years old) maintained her interest and readiness to develop activities in accordance with her capacities. Her visual limitations did not permit her to continue with laboratory activities, particularly measuring with the syringes, but she agreed to handle transplanting to soil and field. Because of the differences in age and schooling, the way each group member became qualified to do the laboratory work was customized to her specific abilities.

The need to encourage and motivate farmers through every step of the propagation process, however minor, was particularly illustrated when the planting materials were taken to the field. The farmers saw the plantlets as being very weak, compared with stakes, and probably unable to survive under field conditions. Only after two to three months in the field, when farmers could find no differences between the *in vitro* and traditional materials, did they understand that tissue culture did not create differences in the appearance of the crop and that it did indeed produce benefits such as propagating desirable materials in less time, in greater quantities, and in less space than the traditional methods. They also saw that the crop was disease-free and remained so during the crop cycle. When they harvested the plants derived from *in vitro* propagation, some participants commented, *'What good cassava from those little jars, very soft!'* On seeing the success with 'Algodona', the women added five more varieties.

Sometimes, unexpected factors arose, such as the association of social status with different aspects of the propagation process. For example, some group members who

managed the low-cost flow chamber suggested changing to a commercial one as the former had already given them a beginning. In contrast, they would not accept changing the Petri dishes for glass plates, explaining that the dishes were '*famous*' and the women '*would lose status by the change*'.

Concluding remark – from farmers themselves

In the end, this tissue culture project resulted in not only a useful, usable and potentially lucrative propagation system, but in significant, positive, changes among the participating women of Santa Ana. As the women themselves concluded, '*At first we were afraid we wouldn't be able to carry out the laboratory work. The equipment seemed sophisticated.* But now it comes as naturally to us as sowing seed.'

Acknowledgements. The authors express their thanks: especially to the Women's Group of Santa Ana (Ana Gloria Pérez, Doris Castillo, Hilda M. Gómez, Luz Dary Vivas, Luz Mila Larrahondo, María Genis Banguero, Mariela Larrahondo Nohelia Palacios y Nohemi Larrahondo), Department of Cauca, Colombia, for their valuable assistance, leadership and collaboration in the effective development of this project; to the Program on Participatory Research and Gender Analysis for Technology Development and Institutional Innovation for funding the development of the project; and to Louise Sperling for her support and suggestions. Finally, the authors acknowledge the useful comments of anonymous reviewers.

DEDICATION

We dedicate this work to Chusa Ginés and Verónica Mera, Coordinator and Social Worker, respectively, of the CBN, who lost their lives while carrying out their duties in January 2001.

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