INSTITUT DE RECHERCHES
POUR
LES HUILES ET OLEAGINEUX

GENERAL INSTRUCTIONS FOR HAND POLLINATION OF THE COCONUT (COCONUT I.G.F.)

Document No. 1654
Copy No. 9

1980

11, SQUARE PÉTRARQUE, 75016 PARIS

C O N T E N T S

I.G.F. 1

G E N E R A L

1. The coconut is a plant of allogamous tendency whose natural pollination is assured by the wind and by insects.

For plant breeding programmes, hand pollination only will be used. It is a delicate operation to carry out, requiring considerable precautions to ensure perfect isolation of the inflorescences and good final results.

2. The practical tasks involved in hand pollination are tiresome to perform, and the personnel concerned do not always fully grasp their importance. It is therefore indispensable to assure strict daily inspection.

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I.G.F. 2

A REMINDER OF SOME FEATURES OF THE COCONUT PALM'S FLORAL BIOLOGY

The coconut is monoecious and its inflorescences are hermaphrodite.

The inflorescence is a composite spadix constituted by a central rachis on which the spikelets are inserted.

The very numerous male flowers grow all along the spikelets, except at the base, where the female flowers are found (0 to 3 or more per spikelet).

The inflorescences appear roughly every month, the interval being shorter in the dry season (a little over 20 days) than in the rainy season (about 30 days).

As soon as the spathe opens flowering starts with the blooming of the male flowers at the tips of the spikelets, and spreads gradually all along the spadix.

Male flowering lasts about 20 days and is influenced by the seasons.

The average length of female flowering is from 3 to 5 days in Talls; it can be as much as 15 days in Dwarfs.

Each female flower is flanked by two accompanying male flowers, which open normally in the male phase and give viable pollen.

The female flower is receptive as soon as the stigmata open, at which time nectar is emitted; the stigmata necrose quickly (24 hours after the emission of nectar).

All the female flowers on an inflorescence do not give nuts. Usually anything up to 60 % of the flowers fall in the first six weeks. It has been proved that this is not due to lack of pollen.

At surrounding temperature the pollen keeps for several days; after 8 days it can be considered to be dead.

Coconuts are classified in four groups according to their mode of reproduction :

- Group I : short female phase with no overlapping with the male phase of the same inflorescence or with that of the following inflorescence : strict allogamy (W.A.T. 1st. population).

- Group II : Short female phase without overlapping by the male phase of the same inflorescence but with considerable or total overlapping with the male phase of the following inflorescence : indirect autogamy (W.A.T. 2nd. population, R.L.T., P.Y.T., M.L.T., N.H.T.).

- Group III : Long female phase completely overlapped by the male phase of the same inflorescence, with or without overlapping by the male phase of the following inflorescence : direct autogamy (M.Y.D., C.R.D., M.R.D., S.G.D.)

- Group IV : Short female phase partially overlapping with the male phase of the same inflorescence and with that of the following inflorescence : semi-direct autogamy (E.G.D., B.G.D., M.Y.D. x E.G.D., M.Y.D x W.A.T.).

I.G.F. 3

VISITING THE PARENT TREES

I. ORGANIZATION OF PERSONNEL

POLLEN HARVESTER

Each one is responsible for about 60 trees. He does all the harvesting, preparation and conditioning of the pollen.

BAGGER

Depending on the size of the trees, he looks after 80 trees (large) or 100 (small). It is he who does the emasculations, puts on the ♀ bag and takes it off.

POLLINISER

He looks after 160 - 180 trees (Talls) or 100 - 110 (Dwarfs). He makes up the pollen-talc mixture in the dusting flasks and does the hand pollinations.

All this personnel is supervised by overseers.

One person is responsible for recording and harvesting the nuts.

II. PLANNING

In the case of pollen harvesting, the trees are visited every other day.

A table showing the planning of hand pollinations is posted up (see model overleaf). The days of the month are shown horizontally, the parents, represented by their field number, vertically.

Every day, the different operations to be carried out on each tree are marked in the corresponding column with staples of different colours.

The work programme for the day is based on the results of previous visits.

The colours used on the planning sheet to indicate the different operations to be done are as follows :

- Red : Visit
- Grey : Male flowering
- White : Female flowers too young - expected date of bagging (the number of days' wait to be indicated in roman numerals).
- Light green : Bagging
- Yellow : Pollination
- Dark green : Removal of bag

Note : For the autogamous varieties of coconut, as the bag is put on as soon as the spathe is opened by hand, the grey and white staples are not used.

III. VISIT MANIFOLD

The observations made or the tasks carried out on each tree are entered in a manifold notebook as follows :

POLLEN HARVESTER

- B = Bagging
- PH = Pollen harvested and bag removed

BAGGER

- Spathe open, phase ♂ (8) = Visit and emasculation in 8 days.
- Female flower too young (5) = Bagging to be done in 5 days.
- B (2) = Bagging postponed 2 days.
- B = Bagging.
- RB (2) = Removal of bag postponed 2 days as too many flowers not yet necrosed.
- RB = Removal of bag.
- Flowers cut (1) = When the bag is removed, all the non-necrosed flowers are cut, in this case 1 flower.
- P (3) = Pollination postponed for 3 days.

POLLINISER

- F2 (45) = 2nd. pollination in the case of Dwarfs, done with pollen collected 45 days previously.

The Dwarfs are pollinated three times, the Talls once.

This organization enables the work to be done to be foreseen in detail and its execution to be checked each day.

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PLANNING I.G.F.3

Days / Field No ♀	1	2	3	4	5	6	7	8	9	10	11				28	29	30	31
S 20 - 12 - 15							III			B								
S 20 - 12 - 19	Visit																	
S 20 - 21 - 24			II		B													
S 20 - 32 - 16				Removal of bag														
S 22 - 15 - 17		Pollination									Removal of bag							
S 23 - 04 - 02			Visit															

Grey — Male flowering

White — III — ♀ too young, Bagging in 3 days

Light green — B = bagging

Yellow — Pollination

Dark green — Removal of bag

Red — Visit

I.G.F. 4

EQUIPMENT & PREMISES FOR HAND POLLINATION

I. EQUIPMENT

(a) Inflorescence Isolation Bags (see FIGURE 1)

The isolation bags for pollen collection or pollination are made of thick canvas, tightly woven to prevent insects or wind-borne pollen getting in, but allowing air to circulate. The canvas is green DICKSON CD 72 plastinic.

The bags are 80 cm high and 60 cm wide, and are of two types :

- ♀ bag for isolating inflorescences to be pollinated : it has three Rexon windows, H 12 cm x W 14 cm, two on one side and one on the other. The two windows on one side are centred widthwise, and the highest is 10 cm from the top of the bag, the second 5 cm below the first.

  The window on the other side is opposite the highest of the two.

- ♂ bag for pollen collection : this has two Rexon windows, H 12 cm x W 14 cm) 10 cm from the top on either side of the bag.

  There is a CD 72 canvas sleeve 2 cm below the edge of the window; it is 27 cm long and slopes towards the bottom of the bag. The diameter at the tip is 13 cm, where it can be fitted with a rigid plastic tube Ø 10 cm, L 7 cm, to which a transparent plastic bag is attached. The whole is held in place by a clamping band. The plastic bag receives the spikelets; it is 20/100 mm thick, measures 37 x 42 cm, and at one corner there is a sleeve 14 cm long, 11 cm in diameter which fits over the end of the rigid plastic tube of the canvas bag.

  On the right-hand side of the bag, 15 cm from the top, is another sleeve, L 26 cm, W 17 cm.

Both types of bag are made by DICKSON CONSTANT, 249, rue du Faubourg de Roubaix, 59000 - LILLE, France.

(b) Isolation Box (see FIGURE 2)

This box is used for all handling of pollen. It must be perfectly air-tight and sterilizable. Sterilization is by two 1000-watt infra-red tubes which raise the inside temperature to 150°C.

The box is in aluminium, 2mm thick, W 60 cm x L 60 cm x H 30 cm. It has a tempered glass window 24 x 44 cm on top, and a door in one side which has two openings Ø 17 cm through which the arms can be passed in order to work with the door shut in conditions of isolation. These openings are fitted with thick, tightly-woven canvas sleeves which cover the operator's arms, The door is only opened to clean the box after use.

In the Ivory Coast, these boxes are made by FERRIVOIRE, ABIDJAN.

(c) Box for sterilizing the Dusting Flasks (see FIGURE 3).

This, too, is in 2-mm-thick aluminium. There is a 100 watt lamp maintaining a temperature of 40 - 50°C within the chamber. The dusting flasks can be taken out by a lid in the top. Openings fitted with sleeves similar to those on the isolation boxes enable the flasks to be introduced and handled in the sterile chamber without fear of contamination.

This box is heat-sterilized every day by two 1000-watt infra-red tubes.

Makers : FERRIVOIRE, ABIDJAN, Ivory Coast.

(d) Ovens

Three types of oven are required :

- 1 ventilated oven adjustable with precision to 40°C for drying male flowers (Brands : MEMMERT or HORO).
- 1 ventilated oven adjustable to 105°C to test pollen humidity (same brands).
- 1 small oven, adjustable to 35°C, for germinating pollen (PROLABO).

(e) Equipment for conditioning Pollen

- Freeze-dryer, model EF4 "MODULYO", made by EDWARDS, Manor Royal, Crawley, West Sussex RH 102 LW, England. Agent in France ZIVY & CO., 31, rue de Naples, 75008 - PARIS.

The pollen can be conditioned either in tubes or in flasks, so that the freeze-dryer will be equipped with :

. a rack of 48 nipples for tubes,

and/or . trays enabling flasks to be vacuum-stoppered.

- Air-gas sealing torch, type 3A Blowpipe, brand NATIONAL, San Francisco, for the tubes.

- Compresser pump, model 80041, made by UNIVERSAL ELECTRIC CO., Owosso, Michigan, U.S.A., or type AEB 63 a-2, from BREY, Memmingen, for the tubes.

- Round-bottomed glass tubes, Ø $7^{8}/8^{3}$ mm, L 100 mm, thickness 6/7-10ths, from BERTHON ET REMEUR, 2, rue de la Madeleine, 77170 - Brie Comte Robert, France.

- Glass flasks (penicillin type), capacity 5 ml, with grooved stoppers and aluminium capsules (VERRERIES GENERALES, 29, Route de Bonneuil, 94370 - SUCY-EN-BRIE, France) and plastic pill-boxes, L 22 cm and Ø 1 cm, with pierced cork.

(f) Metal Ladders of various sizes

Suppliers : BOUET, ABIDJAN, Ivory Coast or FAVOME, 85, rue Eugène Caron, COURBEVOIE, France.

(g) Equipment for Checking of Pollen Quality

- Ovens (see (d)).

- Electric precision balance, maximum range 160-200 g, precision 1/10 mg, 1 covered pan, direct reading.

- Binocular microscope, WILD M5 HEERBRUNG.

- High-frequency vacuum checker. EDWARDS model T2, suppliers ZIVY & CO., 31, rue de Naples, 75008 - PARIS.

- 5 ml hypodermic syringe for flasks.

(h) Small Equipment

- Aluminium sieve, Ø 21 cm, stainless wire gauze, gauge 200. From :Ets. TRIPETTE et RENAUD, 39, rue J.J. Rousseau, 75001 - PARIS, France.

- Polythene dusting flasks, capacity 125 ml, supplied by PROLABO, B.P. 200, 75526 PARIS CEDEX 11, France.

- Metal labels : Ets. J. TULOUT VERNIER, 59680 - COLLERET, France.

- Shears, SANDVIK-PRADINE No. 3-23 for the baggers and No. 3-20 for the pollen harvesters. From Ets. PEYRISSAC, ABIDJAN, Ivory Coast.

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- Staplers
- Gloves : Ets. PEYRISSAC, ABIDJAN.
- Markers : Ets. J. TULOUT VERNIER, 59680 - COLLERET.
- Paper bags (21 x 31 cm).
- CD 72 plastinic canvas bags (22 x 32 cm) made on the Station.
- Roller for crushing spikelets.
- Slim taper file NICHOLSON HOLLAND for the tubes.
- Rubber thongs, cut from inner tubes.
- Pyrex Petri dishes, Ø 70 mm, supplied by PROLABO.
- Paint brush
- Pyrex weighing bottles, low, fitted cover, Ø 50 mm, H. 30 mm, capacity 30 ml. Supplier : PROLABO.
- 1-l polythene flasks fitted with a 100 ml dosimeter. C.N.T.A., 12, Avenue Georges V, 75008 - PARIS, France.
- Glass pencils
- Bunsen burner

(i) Products

- Surgical spirit 95°
- Teepol
- Formol
- Talc (or lycopodium)
- Kapok
- Cotton wool
- Sticking plaster
- Scotch tape
- P_2O_5
- Butane
- Prosevor
- Timor insecticide aerosol
- Morestan
- Raticide
- Agar-agar
- Sugar
- Distilled water
- Filter paper
- Oil for vacuum pump

Note : Each year the Stations will send Head Office their orders for materials not made on the spot, making sure they always have 6 months' stock in hand to allow for delivery dates.

II. DISTRIBUTION OF MATERIALS BY OPERATION

(a) Pollen Harvesting

Bagging : Male isolation bag, ladder, shears No. 3-20, Timor aerosol, Prosevor, surgical spirit, Scotch tape, kapok.

Collection of spikelets : Ladder, shears No.3-20, stapler, marker.

(b) Pollen Preparation

Stripping and drying of flowers : isolation box, surgical spirit, paper bag, canvas bag, roller, oven at 40°C.

Sieving and putting in tubes : isolation box, surgical spirit, sieve, glass tubes or glass flasks and pillboxes, labels, paper, cotton wool.

Conditioning of pollen : freeze dryer, blowpipe with compresser, freezer.

(c) Pollen-talc Mixture

Box for sterilizing dusting flasks, isolation box, talc with dosimeter, file, dusting flask, tube or flask of pollen.

(d) Pollination

Bagger : Female isolation bag, ladder, shears No.3-23, gloves, Timor aerosol, Prosevor, surgical spirit, sticking plaster, rubber thong, kapok.

Polliniser : ladder, Timor aerosol, surgical spirit, sticking plaster, dusting flask, metal label, Scotch tape.

(e) Removal of bag

Ladder, Morestan, raticide if necessary.

(f) Check of Pollen Quality

Vacuum : High-frequency vacuum checker or syringe.

Viability : Petri dish, agar-agar, sugar, dropping bottle with distilled water, bunsen burner, butane, filter paper, paint brush, oven at 35°C, microscope, glass pencil.

Humidity : Weighing bottle, precision balance, oven at 105°C.

III. PREMISES

The size of the premises depends on the annual pollination programme.

A pollination laboratory should have the following rooms :

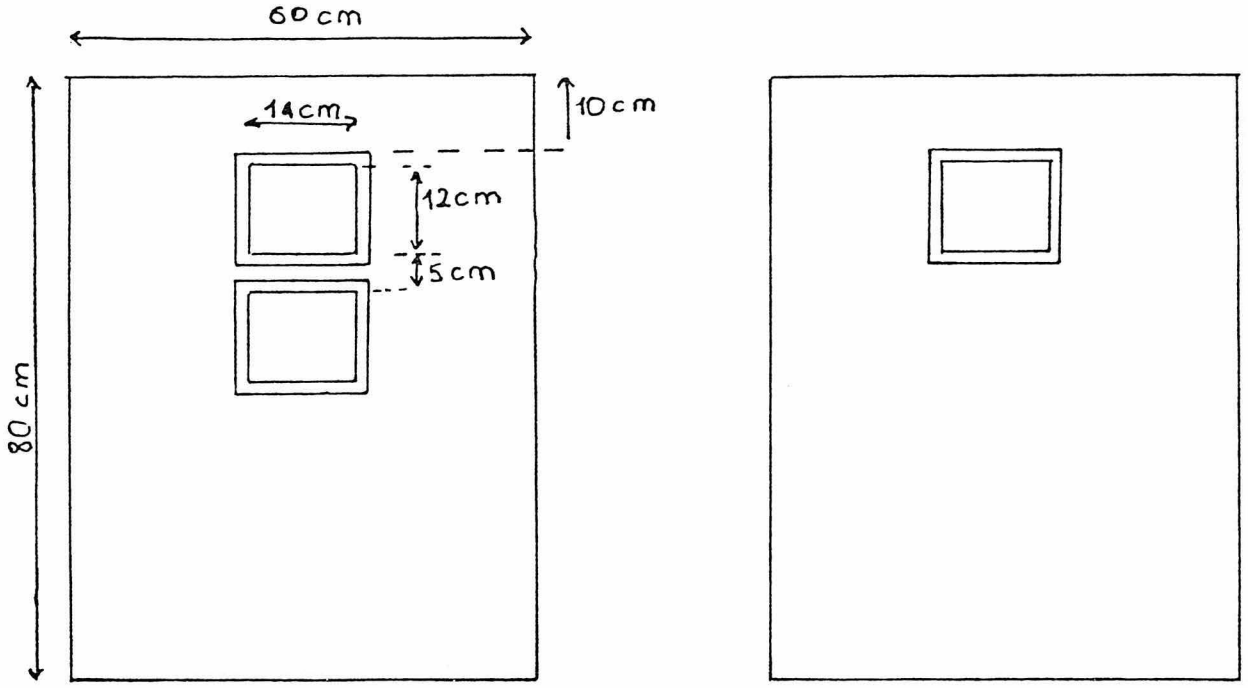
- an office for the person in charge;
- a large office for the baggers, pollinisers and pollen harvesters;
- 1 laboratory for the extraction and conditioning of pollen;
- 1 laboratory for the storage and quality control of pollen;
- 1 laboratory for filling the dusting flasks for the hand pollinations.

FIGURE 4 shows the lay-out of the installation, with the circulation between laboratory I (preparation-conditioning), laboratory II (storage-checking) and laboratory III (use).

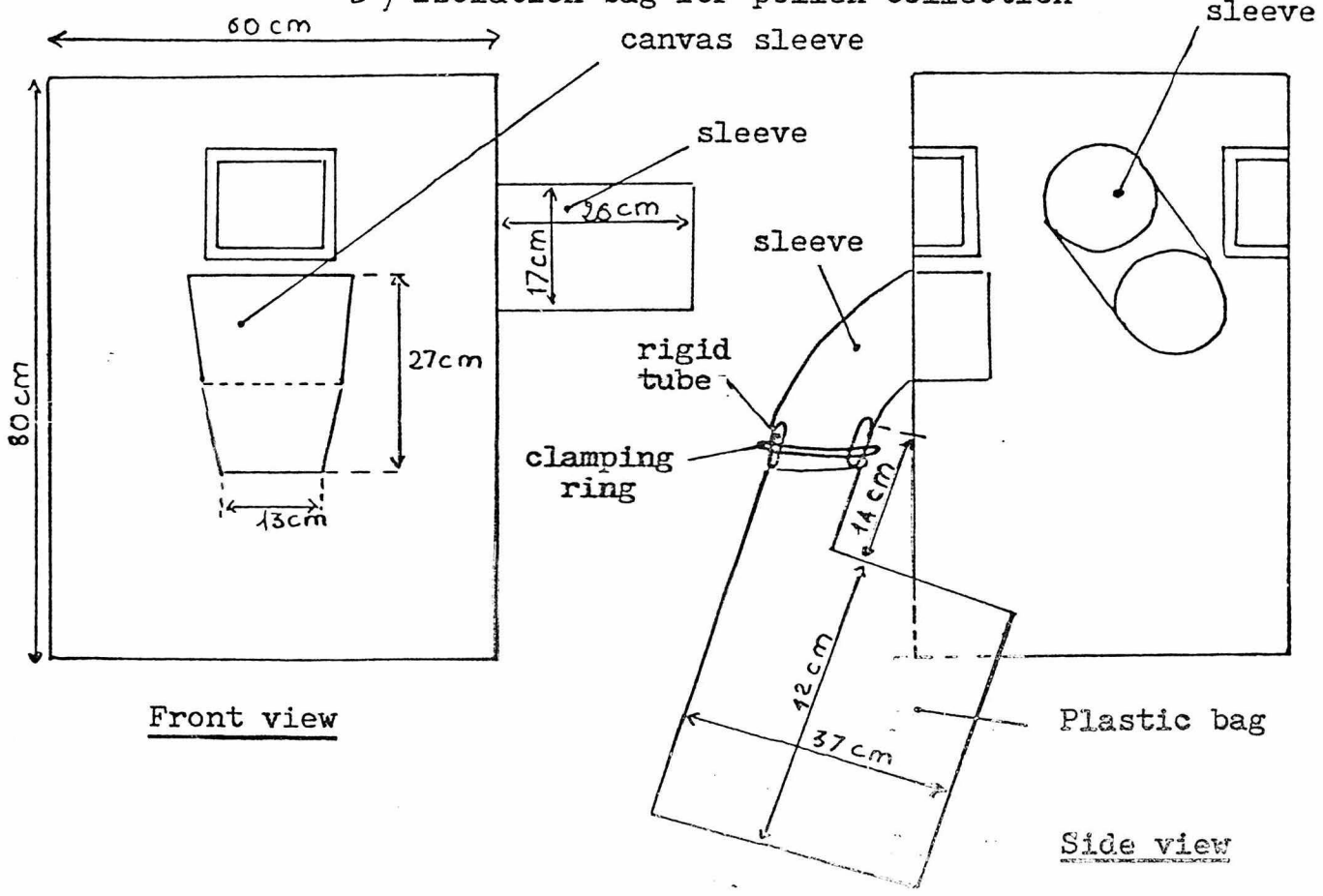
FIGURE 1

ISOLATION BAGS

1°) Female isolation bag



2°) Isolation bag for pollen collection



Front view

Side view

FIGURE 2

ISOLATION BOX

Materials : Aluminiun sheet, 2 mm thick.
Window in tempered glass.

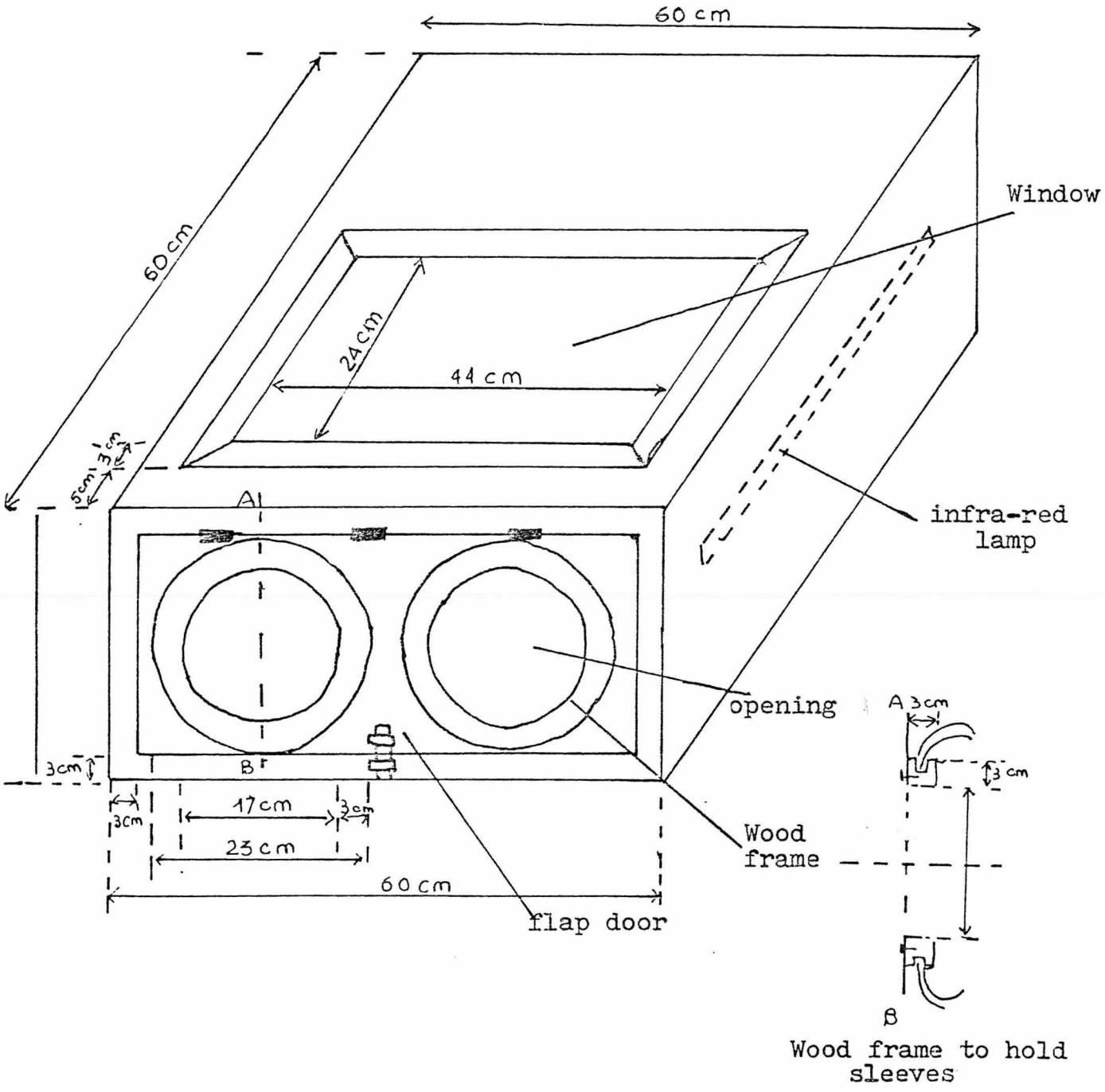
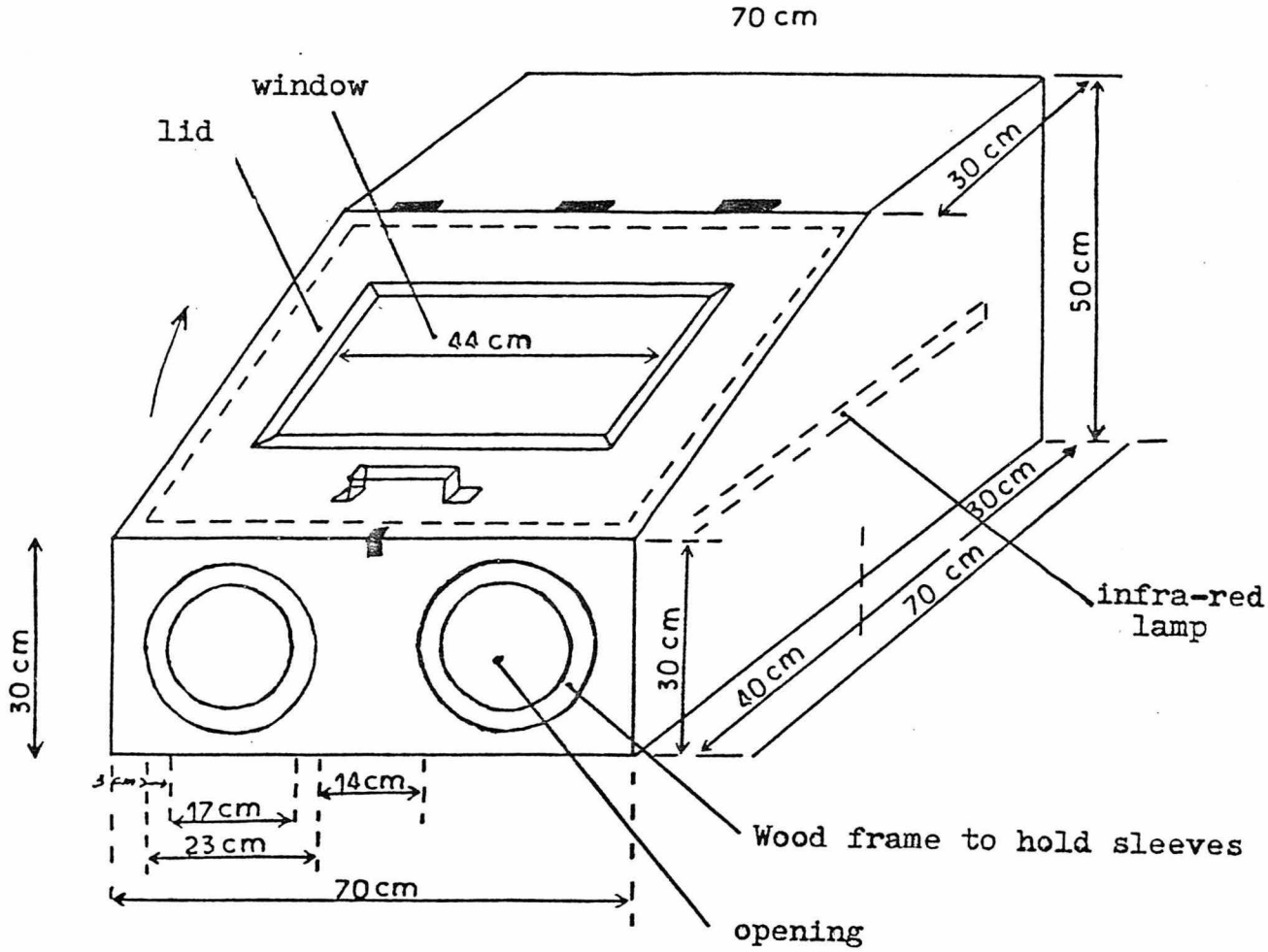


FIGURE 3

BOX FOR STERILIZING DUSTING FLASKS

Materials : Aluminium sheet 2 mm thick.
Window in tempered glass



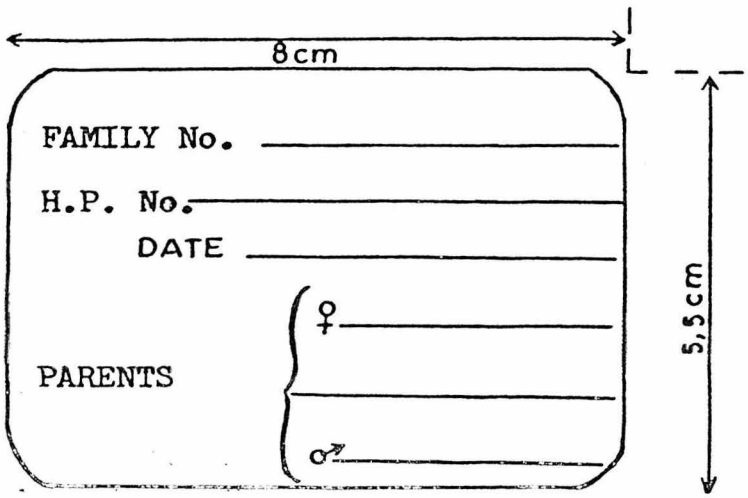


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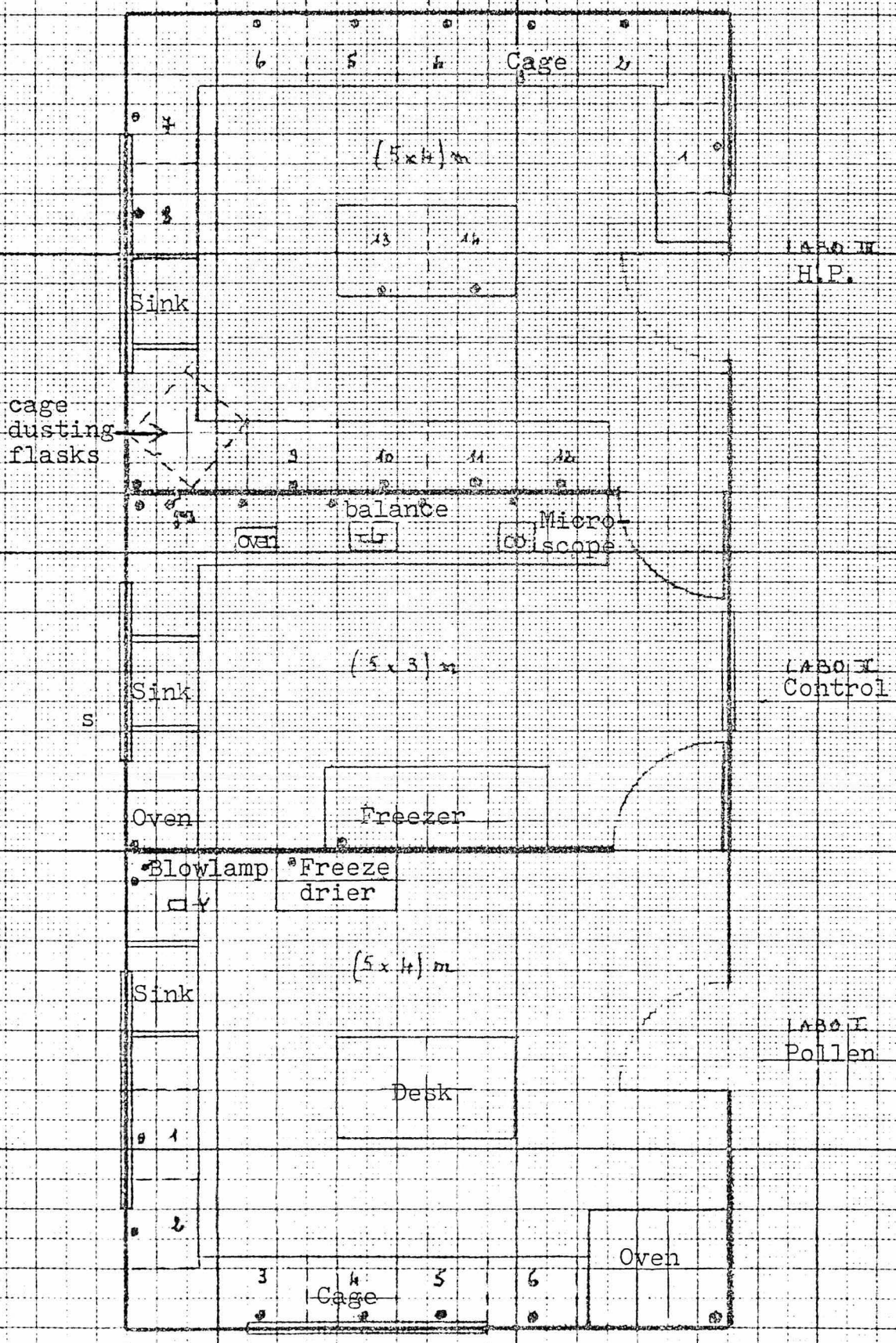
FIGURE 4

LABORATORIES

THREE AIR-CONDITIONED ROOMS



I.G.F. 5

DISINFECTION OF EQUIPMENT & PREMISES

I. GENERAL RULES AND MATERIAL IN COMMON USE

(a) The disinfection of the equipment and premises used for hand pollination must be carried out with the greatest care.

In order to be sure of the legitimacy of the nuts it is, in effect, very important that no viable pollen should be present when new pollen is introduced, be it in an isolation box, a sieve, a tube, a dusting flask or anywhere else.

Those responsible for hand pollination should wash their hands with surgical spirit at 95° each time they pass them into a bag or a handling box, or when doing the pollination itself; in fact, before any handling.

(b) The isolation bags will be washed in fresh water then soaked for 10 minutes in a 10 % formol solution. After that, they will be dried in the shade.

(c) The isolation box will be sterilized by infra-red heat at 105°C for 15 minutes. All the material needed for the operation in view and which can support this temperature should be put into the box and sterilized with it : sieve, paper and canvas bags, glass tubes or flasks, paper labels.

The boxes should be allowed to cool off for about half an hour. The number of boxes in use depends on the quantity of male inflorescences to be prepared in one day and the number of pollinations to be done.

A disk placed on each box indicates whether it can be used or not; it is divided into four sections inscribed : "To be disinfected", "Being disinfected", "Disinfected hot" and "Disinfected cold". A movable pointer is set to the corresponding position.

II. MATERIAL SPECIFIC TO POLLEN HARVESTING

(a) The shears used by the harvesters will be cleaned with surgical spirit immediately before being introduced into the bag.

(b) Any material which cannot stand high temperatures, such as the cotton, the grooved stoppers and the pillboxes (when the pollen is packed in flasks) will be put into

plastic bags marked with the date on which this is done and not used until a fortnight later, by which time any pollen which may be in lost its viability.

(c) These plastic bags and those containing the male spikelets are disinfected with surgical spirit at 95° just before being put into the cooled isolation box.

III. MATERIAL SPECIFIC TO HAND POLLINATION

(a) The dusting flasks will be washed with Teepol and rinsed with surgical spirit. Afterwards they will be stored open in a box previously sterilized at 105°C for 15 minutes. All the time they are stored the temperature in the box will be kept between 40 and 50°. Before the flasks are used they are closed and the tip of the tube is sealed with sticking plaster, and only then will they be removed from the isolation box.

(b) Just before the pollination is done, the dusting flask and above all the tube which is passed into the bag will be cleaned with surgical spirit.

I.G.F. 6

P E S T I C I D E S

INSECTICIDES

The products to be used are :

1. Prosevor 80 - 85 % Carbaryl (Sevin), made in the Ivory Coast by PROCIDA represented by SOFACO, is used to impregnate the packing of kapok or cotton placed in the neck of the bag round the peduncle of the inflorescence.

2. Timor, a commercial brand, in aerosol form, to kill the insects on the inflorescence at the moment of bagging.

ACARICIDE

Morestan 25 WP : a solution at 2 g/litre will be sprayed on the young nuts just after the bag is removed (control of Eriophyes).

RATICIDE

Raticide blocks (basis paraffin wax + coumafene) placed in the crown in case of attack.

I.G.F. 7

ISOLATION OF INFLORESCENCES FOR THE HARVESTING OF MALE FLOWERS

I. TIME OF BAGGING

The inflorescences are bagged on the day on which the spathe opens. In the case of autogamous coconuts (Dwarfs), in which the male phase starts as soon as the spathe opens naturally, the bag will be put on 72 to 48 hours before the presumed date of opening. The safety margin thus left (8 days) between bagging and harvesting of the pollen is sufficient to ensure that virtually all the pollen grains which were on the inflorescence when it was bagged will be dead. The few which might still be viable at the time would probably not survive the preparation and conditioning of the pollen.

II. BAGGING

The harvester cuts the spathe at the base with a shears, opening it first if it has not yet done so (case of autogamous coconuts). He surrounds the peduncle of the inflorescence with a packing of kapok or cotton impregnated with insecticide (base Sevin) and sprays the inflorescence with Timor insecticide from an aerosol.

Then, taking care not to knock off the male flowers, he fits on the bag, the mouth of which comes down over the peduncle. He pleats the open end of the bag and lashes it down over the kapok or cotton packing with a rubber thong to stop insects getting in. The sleeve is also tied up with a rubber thong, and the plastic bag is kept rolled up with sticking plaster.

Care must be taken to see that the bag does not fit too tightly over the upper part of the inflorescence, so that the latter has room to open out a little more.

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I.G.F. 8

ISOLATION OF FEMALE FLOWERS

I. TIME OF EMASCULATION AND BAGGING

The dates of emasculation and bagging vary according to whether the variety of coconut concerned is allogamous or autogamous. In practice, the inflorescence should be isolated for at least 8 days before the female flowers start becoming receptive. The coconuts are classed in four types having different intervals between the opening of the spathe and the receptivity of the first female flower (see I.G.F.2).

- Types I and II : Interval : 22 - 24 days.
  Emasculation 5 days after spathe opening.
  Bagging 3 days later.

- Type III : Interval : 0
  Emasculation and bagging 48 hours before natural opening.

- Type IV : Emasculation the day of natural opening.
  Bagging 3 days later.

Note : On Types I and II one can therefore collect the pollen from an inflorescence and then bag it again for hand pollination of its own female flowers. This is impossible for Types III and IV, where the inflorescence serves for either pollen harvesting or for pollination.

It is important that no female flower should be receptive at the time of bagging. In certain autogamous Dwarfs, the female flowers closest to receptivity (usually the lowest down) are removed when the bag is placed.

The safety margin (8 days' isolation) is considered sufficient for it to be certain that any pollens on the inflorescence at the time of bagging will be destroyed, as the life of pollen emitted naturally generally does not exceed 6 - 7 days.

As the presence of the bag has a marked depressive effect on fruit set, the inflorescences should not be bagged too early.

I.R.H.O.

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II. ELIMINATION OF MALE FLOWERS : EMASCULATION

After opening the spathe if it has not opened by itself (autogamous coconuts), the bagger cuts it at the base with a shears. He clears the spadix of male flowers, not forgetting the axiliary ones on either side of the female flowers. The branches of the spadix should not be cut as they serve to support the bag and stop the female flowers touching the walls.

Note : When a spathe has been opened by hand, care must be taken not to damage the female flowers or break the spikelets, as the tissues are tender.

III. BAGGING

After spraying Timor insecticide, the bagger fits on the bag, the mouth of which comes down round the peduncle of the inflorescence. He pleats the bottom of the bag and secures it over the packing of cotton or kapok impregnated with insecticide (Prosevor) round the peduncle, so that insects cannot get in. He sees that the windows are not too much exposed to sunlight.

IV. REMOVAL OF THE BAG

The bag is removed 10 days after the last pollination. If, at that time, there are any female flowers which have not necrosed, they are cut.

I.G.F. 9

HARVESTING OF MALE FLOWERS AND OBTAINMENT OF POLLEN

I. HARVESTING OF SPIKELETS

The operator checks the development of flowering through the Rexon windows and decides on the exact date of harvesting; the average is 6 - 8 days after bagging.

After disinfecting his hands, arms and the shears with surgical spirit, he opens the sleeve on the bag and passes in his arm holding the shears. He cuts the spikelets with at least 20 % male flowers open and puts them into the plastic bag (unrolled beforehand) fixed under the window. If the spikelets are too long he cuts them in two before putting them in the plastic bag.

The latter is then closed hermetically (staples or sealing) then separated from the isolation bag and sent to the laboratory. The isolation bag is removed.

Note : The number of the parent and the field number of the tree from which the spikelets are being removed are inscribed on the plastic bag at the time of bagging.

II. STRIPPING OF MALE FLOWERS

By one of the sleeves, the preparer introduces into the "disinfected cold" isolation box the plastic bag, disinfected with surgical spirit (see I.G.F. 5) and containing the spikelets. If the bag will not go into the box completely, he inserts part of it and empties the spikelets into the box.

He strips the male flowers off and puts them in a paper bag on which the identity of the pollen and the date of harvesting are written, seals it hermetically and puts it into a canvas bag which will protect it during the following operation. The whole is then removed from the box.

III. CRUSHING

The male flowers are simply crushed with a roller to open them and favour drying.

IV. DRYING

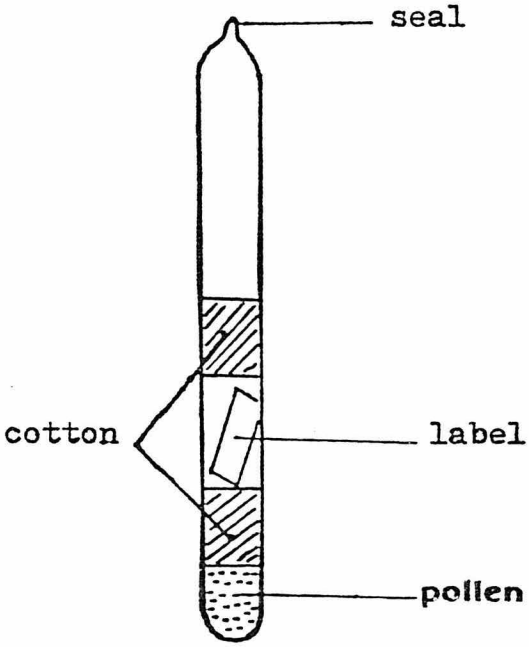
The canvas bags are put in the ventilated oven, set to 40°C, and left for about 20 hours.

V. OBTAINMENT OF POLLEN - SIEVING AND PACKING IN TUBES

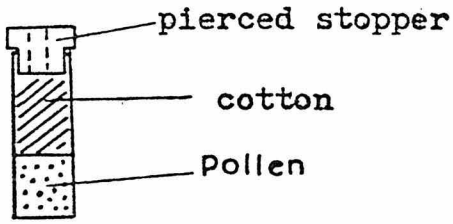
The canvas bag is taken out of the oven, and the paper bag is transferred via one of the sleeves into the isolation box containing the sieve, the glass tubes or pill boxes + flasks, the paper labels and the cotton. The operator empties the crushed flowers into the sieve, which he closes; he shakes it and collects the pollen, which is put into tubes or pill boxes at the rate of about 0.25 g each, which is the amount required for one pollination. In each tube, a plug of sterilized cotton is put on top of the pollen, then a paper label showing the identity of the pollen and the date of harvesting, then another plug of sterile cotton. In the case of pill boxes, the pollen is put in, then some cotton, and the pill box is closed and placed with the label in a flask, which is corked (see sketch).

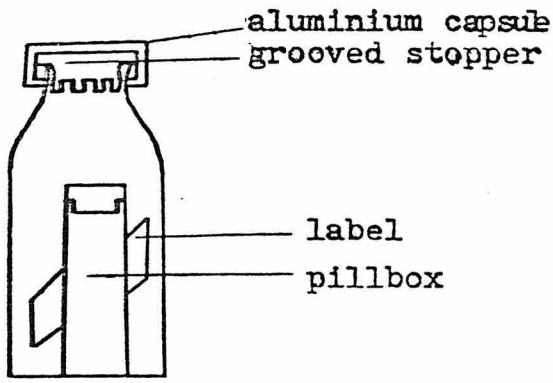
CONDITIONING OF POLLEN

TUBE



PILLBOX AND FLASK





I.G.F. 10

STORAGE OF POLLEN

I. VACUUM PACKING

The glass tubes or flasks are taken out of the isolation box.

(a) Tubes

The filled glass tubes are pre-drawn with a blow-pipe. They are then placed on the teats of the rack of the freeze drier Five minutes after the vacuum has reached 6 10-2 Tor, the tubes are sealed at the neck.

(b) Flasks

The stoppers are loosened slightly so that a vacuum can be formed in the flasks and pill boxes (pierced stopper). The flasks are stood on the freeze drier trays, and five minutes after the vacuum is formed, they are re-corked under vacuum by drawing the trays together by means of a screw.

Method

- Check that the P_2O_5 of the water trap is still usable.
- Place the tubes on the teats or the flasks on the trays.
- Switch on the apparatus.
- Shut the air intake.
- Start the vacuum pump.
- Check that the vacuum has reached 6 10-2 Tor.
- After 5 minutes of vacuum, seal the tubes with a torch or stopper the flasks.

II. FREEZING

The tubes or flasks thus prepared are regrouped by parent and by programme in plastic boxes and kept in the freezer at - 20°C. The maximum storage time is 6 months, but in principle the pollens are used within 3 months.

I.G.F. 11

CHECKING POLLEN QUALITY

One pollen unit (in tube or flask) is sampled for each pollen harvested or for each parent, after preparation and conditioning. If the results are not good a second tube or flask in the lot concerned is tested, and if the first check is confirmed, the whole lot is eliminated.

I. VACUUM QUALITY

In the tubes the vacuum is checked with a high frequency apparatus, in the flasks with a syringe, measuring the retraction of the piston.

II. POLLEN VITALITY

The vitality of the pollen is measured by culturing on a gelose-saccharose-distilled water medium.

1. Preparation of the medium

Put 1.2 g gelose flakes in an Erlenmeyer flask.

Add 100 cm^3 distilled water with a dropping bottle.

Heat on a bunsen burner over a low flame until all the gelose is dissolved, stirring continuously. Once the gelose is dissolved, allow to stand for 1 or 2 minutes, then add 11 g sugar (saccharose) to the Erlenmeyer. Stir to dissolve. Make up to 110 cm^3. Pour into petri dishes and cover.

When the gelose has set, open the dishes and wipe the surface of the medium with slightly moist filter paper to remove droplets of water due to condensation. Wipe the covers of the dishes.

Note : The petri dishes are washed and kept in an oven at 105°C before use.

2. Seeding

Place the tubes or flasks containing the pollen in a humidor at surrounding temperature for 1 hour. Take a little pollen on a paint-brush, shake it so as to form a cloud of pollen grains, and pass a petri dish through the cloud once.

See if sufficient grains have been deposited on the gelose, otherwise pass the dish through the cloud again (if necessary check under the microscope that there are 10 - 20 grains in the field).

Close the dish. Mark the number of the pollen on it with a glass pencil and place in an oven at 35°C for 2 hours.

3. Counting

On removal from the oven, use a microscope to count the grains.

Count all the grains seen in one field and note :

- the normal grains which have germinated,
- the normal grains which have not germinated,
- the abnormal grains.

Repeat the counts several times until about 100 grains have been counted (if there is an average of 20 grains on one field, 5 fields should be observed).

Choose different fields at random - move the petri dish around without looking in the microscope.

Example :

		Normal grains germinated	Normal grains ungerminated	Abnormal grains
1st. position of dish	Field 1	10	8	3
2nd. position	Field 2	11	7	1
3rd. position	Field 3	13	5	1
4th. position	Field 4	8	10	4
5th. position	Field 5	9	14	1
		51	44	10

Abnormal grains : $\frac{10}{105}$ = 9.5 %

Vitality : $\frac{51}{105}$ = 48.6 %

III. HUMIDITY

1. Method

Before putting the tubes or flasks in the humidor, part of the pollen is transferred to a weighing bottle. The checker determines the fresh weight, then the dry weight after 24 hours in the oven at 105°C. The weighing bottle is open in the oven and closed for weighing.

2. Calculation

W 1 = Weight of weighing bottle

W 2 = Weight of weighing bottle + fresh pollen

W 3 = Weight of weighing bottle + dry pollen

$$\% \text{ humidity} = \frac{W3 - W1}{W2 - W1} \times 100$$

IV. QUALITY NORMS

% germination	$\geq$ 30
% humidity	= between 4 and 8
Vacuum in flasks	= 5

I.F.G. 12

DESPATCH OF POLLEN

I. PACKING

The pollen tubes or flasks are placed in a polystyrene box specially designed to receive them.

II. DESPATCH

Pollen is always sent from one country to another by air freight.

A quarantine certificate is enclosed with the parcel if necessary.

III. DESPATCH ADVICE NOTE

A despatch advice note (model overleaf) is enclosed with each parcel. It specifies :

- the variety,
- the field number of the tree and its parent number,
- the date of harvesting,
- the number of tubes or flasks,
- the percentage of germination and humidity after preparation of the pollen.

IV. RECEPTION

The consignee checks that the contents of the parcel conform to the advice note.

I.R.H.O.

COCONUT I.G.F. 12.2

April 1980

I.R.H.O.

MARC DELORME STATION A.F. No. .....

DESPATCH ADVICE NOTE FOR POLLEN No. ..... of .....

Unit of ...th. consignment of pollen to :

Consignor :

Addressee :

Pollen sent :

Unit :

Parent No.	Variety	Date of harvest	Number (1) of units (2)	% germination	% humidity

(1) Unit of

(2) Unit of

Plant Breeding Service Director

I.G.F. 13

RATE OF POLLEN

Normally, one unit of pollen (0.25 gram) is mixed with 4.75 grams of talc (or lycopodium) before the pollination.

I.G.F. 14

CARRYING OUT THE POLLINATION

I. TIME OF POLLINATION

The operation is done when the majority of the flowers have their stigmata open and are secreting nectar. There are two cases, according to the length of the female phase :

- flowering lasts 3 or 4 days and the female flowers are receptive for 2. One pollination suffices - this is the case of Talls.

- flowering lasts about 14 days and receptivity of the female flowers no more than 2 : three pollinations are carried out at 3-day intervals, generally the 4th., 7th., and 10th. days after the female phase starts.

The polliniser follows the evolution of flowering by watching the inflorescence through the windows in the bag.

II. PREPARATION OF POLLEN FOR POLLINATION

The pollen to be used is determined by the crossing plan (see I.G.F. 15). One tube of pollen from a single pollinator palm is used for each pollination.

(a) Filling the dusting flasks

The isolation boxes containing talc, a measuring test tube and a file are disinfected the day before.

A disinfected box serves for one preparation only; if there is another manipulation in view, it must be disinfected again.

If several pollinations are to be done with the same pollen (same parent), the tubes or flasks of this identical pollen can be handled in the same box at the same time.

The technique is as follows :

- Switch off the lamp in the dusting flask isolation box. Wash the hands in surgical spirit and introduce via the sleeves. Close the dusting flasks and stop the tip of the tube with sticking plaster. Open the box and take the flasks out.

- Wash hands, tube(s) or flask(s) containing pollen and dusting flask(s) with surgical spirit, and put them through the sleeves into the isolation box "disinfected cold".

Inside the box :

- With the file, saw through the pollen tube between the two plugs of cotton, i.e. at the level of the label, or open the flask. Put the talc, the pollen and the paper label in the dusting flask, close it and shake to mix well. Take the dusting flask(s) out of the box and turn the disk to the "To be disinfected" position.

(b) Labelling - Identification

Two labels per H.P. have been prepared before handling and placed in the box where mixing of the corresponding pollen is done. When taken out of the box, the labels are attached to the dusting flask, each of which receives :

- a paper label stuck on it and showing the field number of the female parent and the parent number of the male;

- a metal I.G.F. 14 label giving the number of the family, the H.P. number, the date of pollination, the field number and parent number of the female parent, the male parent number and, on the back, the reference programme. This information is written on with a black marker, except for the H.P. number, which is stamped. This label is strung on a piece of wire.

III. THE POLLINATION ITSELF

The polliniser checks that the field number and parent number inscribed on the tree, the flask and the label are exactly the same. He climbs the tree with the help of a ladder, taking care not to damage bunches or fronds.

The operations are as follows :

- Attach the metal label to the base of the bag at the level of the rubber thing.

- Spray insecticide around the bag to keep off insects.

- Wash hands and dusting flask with surgical spirit, especially the tip of the tube which is going to enter the bag.

- Remove the sticking plaster closing the flask and that over the hole in the Rexon window, and insert the tube of the flask quickly through the hole.

- Squeeze the flask several times to blow the talc-pollen mixture over the inflorescence in different directions.

- When all the mixture has been sprayed, withdraw the flask tube and close the hole in the window with sticking plaster again. Shake the bag gently to spread the mixture properly.

- Open the flask, take out the paper label and, after checking the exactitude of the pollen identity against the metal label, stock the former to the back of the latter with Scotch tape.

The polliniser then climbs down the tree and records the pollination in his manifold notebook.

When the bag is removed, the metal label is firmly fixed to one of the spikelets. If, at this moment, there are still some flowers which have not necrosed, the spikelets bearing them are cut off.

I.G.F. 15

RECORDING THE DIFFERENT OPERATIONS

The I.G.F. 3 visit manifold book and the I.G.F. 11 pollen quality check manifold are used for keeping the following records :

1. Pollination Register

A model with very strong board covers is chosen, so that it will last many years. Format : 33 x 50 cm.

The pollinations are numbered in series with figures and one letter. This letter changes on 1st. January each year and the figures start again at 1.

In the register, the pollinations are filed by number. For each one, the following information, occupying two pages, is entered :

♀ parent : Number
Field No.
Variety

♂ parent : Number
Field No.
Variety

No. of family

Programme

Date of emasculation

Date of bagging

Done by

Pollination : 1st. date
2nd. date
3rd.

Date pollen harvested

Age of pollen

H.P. done by

Bag removed : Date
No. ♀ cut

Fruit set : No. ♀ set

Harvest : Date
Total no. of nuts
No. good nuts

2. Crossing Plans

These are established on squared cards, format 21 x 29.7 cm (single) or 42 x 29.7 cm (double).

At the top are inscribed : the programme, the family number, the type of cross and the number of HP to be done per mother-tree.

The field numbers and parent numbers of the mother-trees are entered in the first column.

Opposite each mother tree, the line is divided into spaces in which are written : in the first, the field number of the ♂ parent, and in the others (one per HP) :

- date of pollination,
- number of male parent,
- pollination number,
- number of nuts set.

3. Pollen Harvest Register

This should be in strong board covers, format 38 x 38 cm.

The pollens are numbered in sequence from 1 on 1st. January to 'x' on 31st. December, followed by the last two figures of the year; e.g. 1105/80.

For each pollen, the information covers two pages, and is as follows :

Person responsible :

♂ parent : Variety
Field number
Number

Inflorescence order No. : Date of spathe opening
Date of bagging
Harvest :
. date
. hour
. Number of spikelets

Programme No. :

Oven : Entry :
. date
. hour
. number of bags

Withdrawal :
. date
. hour
. number of bags

Sieving : number of tubes

Conditioning : date
length in mins.
number of tubes successful

Quality control : % germination 1
2

% humidity 1
2

vacuum

Observations :

Passed by :

Note : for the % of germination and humidity, a second check is made if the norms are not respected, and the result will appear in column 2.

4. Drying Oven Book

The format will be 18 x 23 cm, and the following will be entered :

Date :
Heating temperature :

Entry : Parent No.
Number of bags
Hours

Withdrawal : Parent No.
Number of bags
Hours

5. Pollen Use Book

Format : 18 x 23 cm. There are usually two pages recto-verso per male parent; at the top are written the field number and the parent number, with, at the head of each column :

Date of harvest
Date of conditioning
Tube : as many lines as there are tubes prepared
Programme

When a pollen is used, a tube is crossed out and the programme for which it used marked in.

For exported pollens, mark 'Exp.' and the name of the country under 'Programme'.

An identical book is kept to follow up the use of imported pollens.

6. Pollen Receipt Book

Format : 18 x 23 cm. It gives the following information about imported pollens, inscribed on two pages :

Country of Origin
Date of harvest
Date of receipt
Varieties
Number of tubes : broken
sound
Programme
Quality control : % germination 1
2
% humidity 1
2
Vacuum

Observations
Passed by :

7. Despatch of Pollen

See despatch advice, I.G.F. 12.

1°/ - POLLINATION REGISTER

PAGE I

Pollination N°	♀ Parent			♂ Parent			Family N°	Programme	Date emasculation	Date of bagging	Done by
	N°	Field N°	Variety	N°	Field N°	Variety					

PAGE II

Pollination			Date harvest pollen	Age pollen	HP done by	Bag removed		N° ♀ + set	Harvest			Observations
Date 1	Date 2	Date 3				Date	N° ♀ + cut		Date	Total N° of nuts	N° good nuts	

2°/ CROSSING PLAN

PROGRAMME : ..... FAMILY N° : .....

TYPE OF CROSS : .....

N° HP per ♀ parent : .....

	Field n° ♀ Parent n°	♂ Parent Field No.	Date HP, ♂ Parent n°, HP n°, N° nuts set					
1				do.	do.	do.	do.	do.
2	"	"	do.	do..				
3	"	"	do.	do.				
4								

3°/ POLLEN HARVEST REGISTER

Page 1

Person Responsable	♂ Parent Variety	♂ Parent Field n°	♂ Parent N°	Pollen order n°	Inflorescences Date open	Inflorescences Date of bagging	Inflorescences Harvest Date	Inflorescences Harvest Hour	Inflorescences Harvest N° of spikelets	Programme n°	Oven Entries Date	Oven Entries Hour	Oven Entries N° of bags	Oven Withdrawals Date	Oven Withdrawals Hour	Oven Withdrawals N° of bags

Page 2

Sieving N° tubes	Conditioning Date	Conditioning Length min.	Conditioning N° of tubes successful	Quality Control % Germination 1	Quality Control % Germination 2	Quality Control % Humidity 1	Quality Control % Humidity 2	Vacuum	Observations	Passed by

4°/ - DRYING OVEN BOOK

Date :

Temperature :

Movement

Entry :

Withdrawal :

Parent n°	N° bags	Hour	Parent n°	N° bags	Hour

5°/ - POLLEN USE BOOK

♂ PARENT

Field n° :

N° :

Date harvest	Date Cond.	Tube	Programme	Date harvest	Date Cond.	Tube	Programme	Date harvest	Date Cond.	Tube	Programme
2/1/80	3/1/80	1	GC 26								
		1	"								
		1	Exp.								
		1	GC 26								
		1									
		1									
10/2/80	11/2/80	1									
		1									
		1									
		1									
		1									
		1									
		1									

6°/ - POLLEN RECEIPT BOOK

Origin	Date Harvest	Date Receipt	Variety	N° of tubes		Programme	Quality control					Observations	Passed by
				Broken	Sound		% Germination		% Humidity		Vacuum		
							1	2	1	2			

I.R.H.O.

COCONUT I.G.F. 16.1

April 1980

I.G.F. 16

MONTHLY REPORT

The object of this report is to follow up the evolution of the various hand pollination programmes each month.

There are four tables to be filled in :

I : Pollinations this month

II : Fruit set on pollinations in the month considered.

III : Harvest this month

IV : Pollen movements

Models of the tables follow.

I.R.H.O.
COCONUT I.G.F. 16.2

MARC DELORME
STATION

Month : ..... 19..

MONTHLY REPORT - PLANT BREEDING

TABLE I

POLLINATIONS

Trial	Type of cross	Family No.	No. of H.P. done this month	No. of H.P. since start of programme (cumulative)	Estimated no. of H.P. included in programme

COCONUT I.G.F. 16.3

MARC DELORME STATION

Month : ..... 19..

MONTHLY REPORT - PLANT BREEDING

TABLE II

Fruit set on pollinations for month of : ..... 19..

Trial	Type of cross	Family n°	This month			Programme realisation			
			N° of HP	Flowers set	Flowers set by HP	N° of HP	Flowers set	Flowers set per HP	Seed nuts required

Blank Pollination

N° of blank pollinations :
N° of blank pollinations with nuts :
Polliniser reponsible for HP with nuts :
Trials :

I.R.H.O.

COCONUT I.G.F. 16.4

Month : ..... 19..

MARC DELORME
STATION

MONTHLY REPORT - PLANT BREEDING

TABLE III

HARVEST - PLANT BREEDING

Trial	Type of cross	Family N°	This Month		Implementation of programme				
			N° of bunches	N° of nuts	N° of bunches (cumulative)	N° of nuts (cumulative)	Nuts per bunch	Set flowers/bunch in	Nuts required

I.R.H.O.

MARC DELORME STATION

COCONUT I.G.F. 16.5

Month : ..... 19..

MONTHLY REPORT - PLANT BREEDING

TABLE IV

MOVEMENTS OF POLLEN FOR H.P.

Trial	Type of pollinator	N° of inflorescences harvested	N° of flasks					
			Obtained	Imported	Exported	Lost	Used	In stock

I.G.F. 17

C H E C K S

All the operations involved in the obtainment of pollen and in hand pollination are checked daily by the overseers and heads of service.

These checks are indispensable to ensure perfect legitimacy of the seed obtained.

Any errors observed during pollen harvesting and preparation or during the hand pollinations lead automatically to the elimination of the pollen or pollination concerned.

1. Checking Isolation

Each month, for one bagger, 2 female inflorescences normally bagged will be pollinated with talc without pollen. The Head of the Plant Breeding Service will indicate which ones are to be done to the overseer in charge of pollinations, just before the operation is due to take place. On no account must the bagger know when carrying out his task which inflorescences are not going to be pollinated.

The bag is removed after the normal length of time and the various operations involved in this bagging recorded in a special "Blank pollination" book.

Ten to twelve weeks later, the number of nuts set is counted and recorded in this book.

If there are any nuts set, the Head of the Plant Breeding Service will find out why as quickly as possible and take steps to remedy the situation.

2. Various Checks

The overseers will make daily checks of the baggers and pollinisers : bagging, pollination, removal of bag.

Pollen harvesting, preparation and conditioning are checked by the senior staff member in charge of pollens.

3. Checking Work Quality

Control of Fruit Set

Three months after pollination, the number of set nuts is counted. This enables the quality of the pollination work to be checked and the progress of the programme concerned followed.

Check of Pollen Quality

See I.G.F. 11

I.G.F. 18

HARVESTING - SEED-BED - NURSERY IDENTIFICATION

HARVESTING

Seven to eight months after pollination, the H.P.number inscribed on the metal label fixed to the bunch is marked on each nut with a marker. In this way the nuts can be identified at harvest even if they have dropped to ground by the time the harvesters pass.

Harvesting takes place every month; the nuts from the same H.P. are tied together and to the metal label.

SEED-BED AND NURSERY

On arrival in the seed-bed, an aluminium ribbon giving the number of the H.P. as inscribed on the metal label is fixed to the husk of the nut. The ribbon is marked with a Dymo punch. After checking that the number on the nuts corresponds to that on the H.P. tag, the nuts are detached and regrouped in the seed-bed by programme and family number.

In the nursery, when the plants are 2 or 3 months old, an aluminium ribbon bearing the same H.P. number as that attached to the nut is placed in the axil of a leaf. Regrouping in the nursery is also by programme and order number attributed to the corresponding family and programme.

Example : 2A = Family No. 2 of programme A.

I.G.F. 19

DESPATCH OF H.P. NUTS

I. IDENTIFICATION

An aluminium ribbon giving the H.P. number as shown on the metal label is fixed in the husk of the nut.

II. TREATMENT

The standard treatment is to remove the calyx from the nut and soak it for 3 minutes in the following solution :

- fungicide : Organyl 66 - 500 g commercial product/hl;
- insecticide - mite killer : Monocrotophos - Nivacron @ 33 g/hl.

III. DESPATCH

III.1 - Packing

- by boat : gunny bags
- by air : plastic bags.

III.2 - Despatch Advice Note

A despatch advice note (model overleaf) is sent for all shipments. The numbering of the bags allows the different varieties and hybrids to be separated. With this advice is enclosed a separate list for each variety and cross, giving for each bag the H.P. number of the nuts it contains. In this way, if the bags are opened or the nuts mixed up, the H.P. number marked on the nut can be checked against this list and its origin found out.

A quarantine certificate is also drawn up for each shipment.

III.3 - Mode of Shipment

Shipment is made by air freight or boat according to the client's wishes.

IV. CONTROL OF CONSIGNMENTS

Wherever possible (number of nuts sufficient) a sample of the lot shipped is kept after treatment as a control; it is placed in the seed-bed, and serves to check germination and legitimacy. It should be representative of the different varieties and hybrids.

I.R.H.O.

COCONUT I.G.F. 19.2

MARC DELORME
STATION

DESPATCH ADVICE

Advice n° :

A.F. N° :

Shipment on : ..... 19..

Client :

Mark on bags :

Family N° (if necessary)	Cross	N° of nuts	N° of bags	Bag N°

Port Bouet ..... 19..

Plant Breeding Service

DIRECTOR