# **VETIVER PROPAGATION**

# **Nurseries and Large Scale Propagation**

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## 1. INTRODUCTION

The Vetiver Network promotes the use of sterile vetiver cultivar to avoid it becomes a weed in the new environment. DNA tests showed that the sterile vetiver cultivar used around the world is genetically similar to Sunshine cv in the US and Monto cv in Australia, which all originated from southern India. Because of its sterility, vetiver grass has to be propagated vegetatively.

In most major applications, a large number of plant are required and the quality of the planting material is one of the most important criteria for application the Vetiver System (VS). Therefore the establishment of nurseries capable of producing large quantity, high quality and low cost of planting materials are essential for application of VS in Kuwait.

#### 2. METHODS OF PROPAGATION

The two commonly used methods for large scale propagation of vetiver are:

- Separation or splitting of tillers from a mature vetiver clump.
- Tissue culture

## 2.1 Vegetative Propagation

The separation or splitting of tillers from a vetiver mature clump produces at least four kinds of planting materials:

- 1. Bare root slips
- 2. Culm slips
- 3. Polybags or tubestock
- 4. Planting strip

#### Bare root slips

A planting slip is defined as a stem, root, twig, etc. cut or broken off a plant and used for planting or grafting. The splitting from the mother clumps has to Be done carefully so that each slip must have at least 2-3 tillers and a part of the crown. After splitting the bare root slips are

dipped in rooting hormones and keep in shallow water pools until new roots start growing. The slips should be kept in good sun light for faster growth and wet until planting out.

## Culm slips

A culm is defined as a stem, stalk of various grasses. The culm of the vetiver grass is solid (not hollow), stiff, hard, having prominent nodes with lateral buds that can form roots and shoots upon exposure to moist condition. Laying or standing the cut pieces of culms on moist sand, or better under mist spray, results in the rapid formation of roots and shoots at each node.

## Polybags or tubestock

When the bare root or culm slips are ready, they are put in small pots or small plastic bags containing half soil and half potting mix. They are kept in these polybags for 3-4 weeks, depending on the temperature conditions. They are ready for planting out when at least 3 new tillers or shoots appear.

# Planting strip

Planting strips are a modified form of polybags, instead of individual bags, bare root slips or culm slips are planted at close spacing in specially prepared long furrow medium, which would facilitate transportation and planting. It is a labor saving practice at planting on difficult sites such as steep slopes, with high survival rate since the roots are not disturbed as in the case of using polybags.

## 2.1.2 Advantages and Disadvantages of Different Vegetative Propagation Methods

## Bare root and Culm slips

The advantages of this method of propagation are:

- Very efficient, low cost and fast to prepare the planting material
- Small volume for transport, ie lower delivery cost
- Very easy to plant out by hand
- Can be mechanically planted out for large areas

The disadvantages of this method of propagation are:

- Vulnerable to dryness and extreme temperature
- Limited storage time on site
- Need to be planted to moist soil
- Need more frequent irrigation in the first few weeks.
- Recommended for good seedbed sites with easy access to irrigation

**Polybags and Planting strips** 

The advantages of this method of propagation are:

- Very hardy and is not affected to exposure to high temperature an moisture stress
- Lower irrigation frequency after planted out
- Faster establishment and growth after planted out
- Longer period of storage time on site
- Recommended for harsh and hostile environments

The disadvantages of this method of propagation are:

- More costly to produce
- Longer period to prepare, 4 -5 weeks or more
- Large volume and heavy load for transport, ie lower delivery cost
- More maintenance cost at site after delivery, if nor planted out within a week

## 2.2 Tissue Culture

Tissue culture is another method of large scale propagation of vetiver planting materials. Instead of using a large part of the mother clumps, tillers and culms, tissue culture uses only some special tissues of the vetiver plant, such as shoot tips, young flower inflorescences or nodal buds. The procedure is fairly routine in the horticultural industry around the world.

The tissue culture methods varies with the individual laboratories, but basically it involves the use of a very small bit of tissue, growing it in a special agar medium under aseptic conditions, plant the very small plantlets out to appropriate medium until fully developed into a small plant.

# 2.2.1 Tissue Culture Methods

One method of tissue culture is described below by Dr.Malee Nanakorn, Botany Department, Kasetsart University, Bangkok, Thailand

# Mass Propagation of Vetiver Grass

Vetiver grass can be rapidly propagated through tissue culture by culturing sterile explants on sterile medium to induce a mass of plantlets (small plants) having the roots and shoots. Two basic methods can be achieved by using young inflorescence or node as follow:

I. Shoot induction from young inflorescence culture

II. Shoot induction from single node culture

# A. Shoot induction from young inflorescence culture

This method can be accomplished in 5 steps:

Step I: Surface sterilization

- Wiping young inflorescence that still enclosed in the flag leaf, 10-15 cm in length, with 75% alcohol.
- Spraying with (or dipping into) 75% alcohol and flame.

- Cutting sterilized inflorescence into 10-mm pieces.
- Step II Callus induction
  - Culturing sterilized explants on Murashige and Skoog medium supplemented with 15 \_mol/l 2,4-D for 30-45 days.
  - Transferring the callus to MS medium supplemented with 10 \_mol/l 2,4-D for 30-45 days to proliferate the callus.
- Step III Shoot induction
  - Selecting compact callus with creamy color and culturing on hormone free MS medium for 45-60 days. Plants or plantlets with roots are ready for transferring to greenhouse conditions.

Step IV Shoot proliferation (multiplication)

- Separating plantlets into single shoot.
- Transferring single shoot to MS medium supplemented with 10 \_mol/l BA to proliferate more shoots.
- Subculturing every 30 days.
- Step V Root induction
  - Separating plantlets into single shoot.
  - Transferring shoot to MS medium supplemented with 5 \_mol/l IBA for 15 days.

#### **B.** Shoot induction from single node culture

This method can be accomplished in 4 steps:

Step I Surface sterilization

- Immersion nodal segments, 3-4 cm length, in 10% solution of commercial bleach added with 2-3 drops of surfactant and shaking gently for 10 min.
- Washing the explants three times with sterilized distilled water.

## Step II Shoot induction

• Culturing sterilized nodal segments and transferring onto MS medium supplemented with 10 \_mol/l BA for 60 days.

Step III Shoot proliferation (multiplication)

- Separating plantlets into single shoot
- Transferring single shoot to MS medium supplemented with 10 mmol/l BA to proliferate more shoots.
- subculture every 30 days.

Step IV Root induction

- Separating plantlets into single shoot
- Transferring shoot to MS medium supplemented with 5 mmol/l IBA for 15 days

## C. Transplanting

• Keeping the culture bottles of rooted-shoots outside the culture room for a few days to acclimatize these plantlets.

• Transplanting the plantlets to well-drained growing medium in high relative humidity condition.

Gradually lowering the humidity within 1-2 weeks

## 2.2.2 Advantages and Disadvantages of Tissue Culture Methods

The advantages of this method of propagation are:

- A very large number of plant can be produced very quickly
- No need for a large scale nursery
- Smaller volume and weight for transportation
- Free from pest and pathogen in nursery

The disadvantages of this method of propagation are:

- The need to set up a small laboratory, which can be expensive for a small nursery
- The need for a well trained technician and other skilled staff
- The need for more manual labour to transfer the seedling to different size pots during its growing period.
- It takes longer to get the plantlets ready for planting
- More susceptible to pest and disease on site and adverse conditions.

# 2.3 Vetiver Nursery

A nursery is needed to provide stock materials for vegetative and tissue culture propagation of vetiver grass. The following are criteria needed to plan and establish a productive and easily managed vetiver nursery:

- **2.3.1** Soil type: For the ease of harvesting and minimizing root and crown damages, sandy loam to loam is the best. Clay loam is acceptable but heavy clay is not recommended.
- 2.3.2 **Topography:** A slightly sloping land is recommended to avoid water logging in case of over watering. Flat site is acceptable but watering must be monitored to avoid water logging, which will affect young seedling growth. Vetiver will grow well under water logging conditions when mature, but young slips growth will be affected by water logging after planting.
- 2.3.3 *Planting layout:* Planting should be done in long and neat rows across the slope for easy mechanical harvesting.
- 2.3.4 *Harvesting method:* Harvesting the mature plants can be done either mechanically or manually. For machine harvesting, the mature stock should be uprooted at 20-25cm below ground. Care must be taken to avoid damaging the crown of the plants, this can be best done by a single blade mouldboard plough or a disc plough with special adjustment.

- 2.3.5 *Irrigation method*: Overhead irrigation is recommended to evenly distributing water in the first few moths after planting. Flood irrigation can be used later on more mature plants.
- 2.3.6 *Training of operational staff*: Availability of well trained staff is essential to the success of the nursery
- 2.3.7 *Mechanical planter:* For large scale planting at the nursery and/or desert rehabilitation later, a specially designed vetiver planter (Australian manufactured) is recommended.
- 2.3.8 *Availability of farm machinery:* Some basic farm machineries are needed for seed bed preparation, weed control, cutting. harvesting etc.