VETIVER ROOTS

THE VETIVER SYSTEM TECHNOLOGY HIDDEN HALF

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CONTENT

TRIBUTES

In Commemoration of
HIS MAJESTY KING BHUMIBOL ADULYADEJ THE GREAT

Mr R Grimshaw OBE

Founder, The Vetiver Network International, USA

John Greenfield's handbook "Vetiver Grass - A Hedge Against Erosion"' was brought to His Majesty's attention by Dr. Sumet Tantivejkhul - both saw the potential for its application to mitigate Thailand's erosion problems. I recall visiting His Majesty to present an award from the World Bank for his work with vetiver. After the formalities he pulled out of his pocket a thick pack of photographs (ones that he had taken himself) showing with great enthusiasm what was being done with vetiver - we spent a delightful hour discussing our favorite plant and its potential applications. His Majesty was passionate about what the Vetiver Grass Technology could do for Thailand and her people. He was a practical man who wanted to understand how and what Vetiver could do, and he closely followed its development. Over the next 30 years the Royal Development Projects Board, the Land Development Department, and the Universities put a huge effort into vetiver research and development that not only benefited Thailand but the world at large.

Additionally he supported, through the Chaipattana Foundation, all the International Vetiver Conferences, as well as a number of training programs - especially those associated with vetiver handicrafts. This support was vital for the growth of the technology and the spread of its applications - support that we are forever grateful. His Majesty was revered by his people - in return he was a leader who cared greatly for their wellbeing. The Thai people are fortunate to have such a King, and we in the world of vetiver are fortunate to have had such a "friend".

Dr Paul Truong

Technical Director, The Vetiver Network International, Australia

Due to its unique and phenomenal attributes, Vetiver Grass is now known as a Holy plant, Magic plant and many other names in local communities worldwide. This status could not be achieved without the early recognition, support and promotion by **His Majesty King Bhumibol Adulyadej** almost 40 years ago.

His Majesty's vision and dedication to numerous applications of the Vetiver System Technology ranging from Environmental Protection, poverty Alleviation and Climate Change, inspired me to compile this book.

INTRODUCTION

What inspired the author to prepare this book were these publications - ***Plant Roots: The Hidden Half*** by **Eshel and Beeckman (2013)**, which provided an up to date and exhaustive review of the role and development of the plant root system in the soil.

As root constitutes a very important and significant proportion of the Vetiver plant, whose behavior under various environmental conditions must be highly influenced by its root system. Therefore a better understanding of plant roots in general and of vetiver roots in particular is vital for increasing its effectiveness in various applications.

Extensive information was also drawn from – ***Plant Roots: Growth, Activity and Interaction with Soils*** by **Gregory (2006)** which pointed out the importance of the root system and its interactions with surrounding environment.

Complete Guide for Growing Plants Hydroponically by **Benton Jones (2014)** - gave a comprehensive update of plant root growth and development under hydroponic conditions.

In addition to these, information was also drawn from numerous scientific publications and reports by vetiver users, particularly the Australian, Chinese, Indian, Indonesian,Thai and Vietnamese engineers and scientists, including this author's own research, experience and application of the Vetiver System over 35 years.

This book has five chapters:

1. Introduction to Vetiver grass
2. Basic information and understanding of the root system as reported by the above references
3. Information and understanding of the vetiver root system under various conditions and how it compares with the roots of other plants in general.
4. How to apply the unique and superior attributes of the vetiver roots to maximise its effectiveness in various applications; and
5. Other important applications: Greenhouse gas and Carbon sequestration, Biofuel, animal fodder, thatching, handicraft Socio-economics in Africa, Asia and Latin America

The special aspect of this book is that in addition to scientific data, it also includes an extensive collection of photographic examples of various Vetiver System applications that the author has collected around the world. Full Photograph Credit (PC) is given to photographs from other sources.

CHAPTER ONE

INTRODUCTION TO VETIVER GRASS

VETIVER SPECIES

There are three well known vetiver species: Indian vetiver species (*Chrysopogon zizanioides),* African vetiver species (*Chrysopogon nigritana*) and South East Asian vetiver species (*Chrysopogon nemoralis).* In addition there are five less known species: *Chrysopogon lawsonii* and *Chrysopogon gryllus* in northern India and three Australian native vetiver species: *Chrysopogon filipes*, *Chrysopogon elongata* and *Chrysopogon rigidus.*

The Indian vetiver species (*C. zizanioides)* is further distinguished in to South Indian and North Indian cultivars.

C. zizanioides (S. India)

C. nigritanus

C. nemoralis

C. gryllus

C. filipes

C. ***elongata***

C. rigidus.

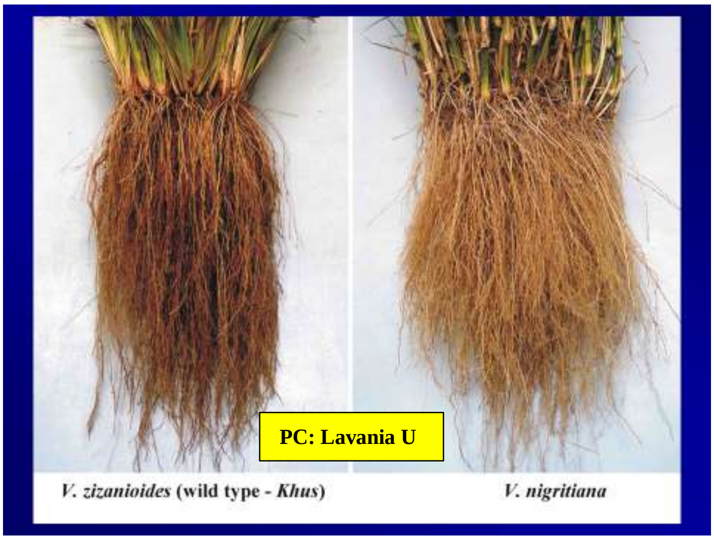
As shown above the shoot morphology of these species varies markedly, their root morphology also differs greatly.

C. filipes *C. nigritanus* C. *elongata* *C. rigidus.*



Two year old root mass of *C. zizanioides (South Indian)* and *C. nemoralis*

Roots of North Indian Vetiver and African Vetiver



Indian Vetiver Species

Among these, south Indian vetiver *C. zizanioides* is the best known and globally most widespread species due to its essential oil production and use in thatching. Presently the South Indian cultivar is cultivated for various applications in tropical and subtropical Africa, Asia, Americas, Oceania and Mediterranean Europe.

The North Indian cultivars is not commonly cultivated, it only exist in swamps in the northern states of India.

Lavania (U. C., 2008) of the Central Institute of Medicinal and Aromatic Plants in Lucknow India stated that *Vetiver is native to India, where it is known to be used both for its fragrant oil and as traditional medicine since antiquity, and its hedges have been applied for contour protection in India since centuries. The name "vetiver" is derived from the Tamil word "vettiver".*

Vetiver is found occurring in India in wild state throughout tropical and sub-tropical plains, particularly along the riverbanks and over marshy lands. It has wide range of ecological distribution ranging from sandy seacoasts and swamps to plains and foothills, and also on the hilltops up to elevations of 800m in the Kumaun hills of Uttar Pradesh. Two distinct morphological complexes of vetiver are found to inhabit spatially separated geographic regions in India

Based on geographical distribution patterns and detailed chromosomal evolutionary parameters it is suggested that south Indian peninsula is the area of its primary center of origin from where it has diverged in two directions:

- *one in the north along the Indo-gangetic plains and adjoining areas mainly in the states of Rajasthan, Madhya Pradesh, Uttar Pradesh and Bihar*
- *the other in the south along the east and west coasts of Indian peninsula in the states of Andhra Pradesh, Karnataka, Tamil Nadu and Kerala.*

The two cultivars are distinctly different. The north Indian wild types are profuse flowering high seed-setting having narrow leaves producing superior quality of laevorotatory root oil (ruh-khus or khus oil) and south Indian cultivated types are low/late flowering, low/non seed-setting with wider leaves producing lower quality of dextrorotatory root oil (vetiver oil).

In addition to Indian subcontinent, vetiver grows wild in subtropical and tropical areas of world. However, a primitive type of clone that had only male florets compared to the normal occurrence of hermaphrodite flowers has been reported from wild population in south India. Occurrence of tremendous diversity especially for reproductive features ranging from non-flowering to late-flowering and range of pollen sterility point to the possibility of India being the center of origin. It is one of the most widely distributed vetiver grass species in Southeast Asia and Oceania.

The South Indian C. zizanioides vetiver grass sports deep penetrating tufted root system and prolific clump of tillers above ground reaching the height of up to 2.5 meters, and the roots growing indeterminately reaching up to 3 meters in one year. The plant can be propagated vegetatively through planting of tillers. As stated above two diverse morphotypes varying for reproductive features are encountered in India. The north Indian types sport profuse flowering, open pollination and high seed formation. Whereas, heterogenous plant populations originating through seed propagation serve as a valuable genetic resource to tap genetic diversity to isolate desirable genotypes, but seed forming types are not preferred in ecological plantations on account of possibility of becoming weedy. Further, to utilize vetiver as an essential oil crop it is desirable to identify plant type where there is high root biomass containing higher concentration of essential oil of desirable quality. On the other hand for utilization of vetiver for ecological plantation the roots with least essential oil are preferred to avoid uprooting of plantations by commercial interests.

The root complex of vetiver is comprised of a tuft of fibrous roots that grow vertically and penetrate deep into the soil. However, this penetrating root system may have diverse architectural pattern, ranging from somewhat spread to vertically penetrating, smooth or with lateral branching along-with different grades of thickness lysigenous cavities, cortical sclerenchyma and essential oil *secretory cells. The primary fibrous roots are main source of essential oil, and there is little oil in the lateral hairy roots, but latter does help in formation of root-web facilitating strong soil binding.*

North Indian vetiver and its root types growing wild in the swamp 50 km from Lucknow

Lavania, (S., 2003) from Department of Botany, Lucknow University, India, in the paper *Vetiver Root System: Search for the Ideotype* which she presented at ICV3 in China pointed out that the Vetiver Root System has diverse applications:

- Source of essential oil
- Water and soil reclamation
- Detoxification and pollution mitigation
- Land/Slope stabilization
- Bio-Engineering; and
- Environment specific cultivation

Each application envisages specific a type of root system, hence search for **Root Ideotype** i.e. Ideal Root Type for specific applications. The following lists the more important features:

Botanical Organization of Vetiver Root System

- *Tufted vertically growing root system*
- *Primary roots are supported with secondary fibrous roots*
- *Juvenile primary / secondary roots are solid with persistent cortex and little oil*
- *Mature thick roots are spongy with schizogenous cortex and have well developed phloem*
- *Phloem is the site of Essential Oil synthesis and storage*

- *Solid vascular cylinder provides tensile strength to roots*
- *Schizogenous cortex facilitates root aeration suitable for submerged conditions*

Diversity in the Vetiver Root System

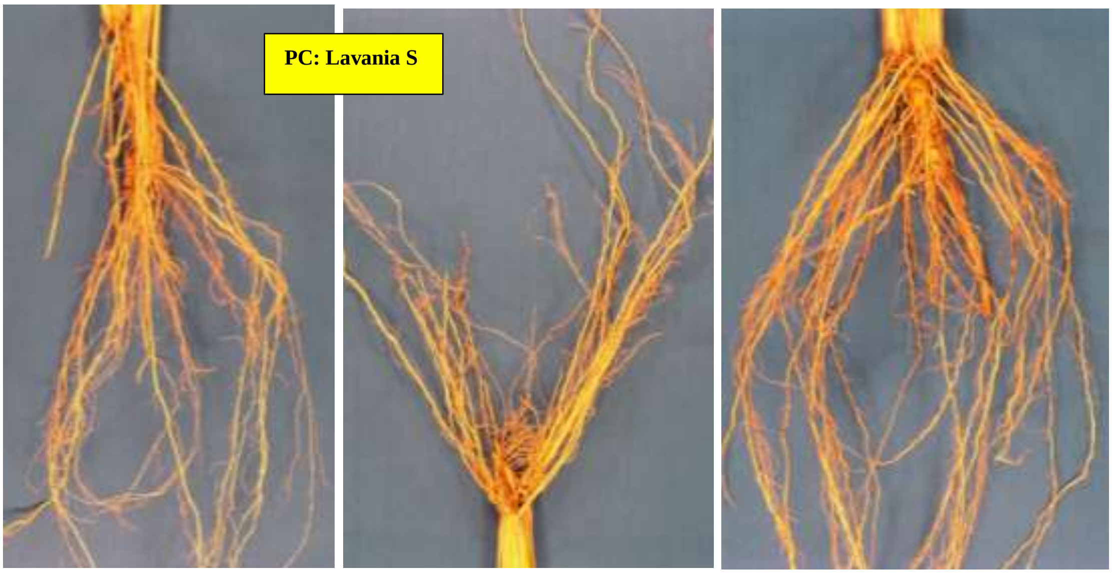
Organizational

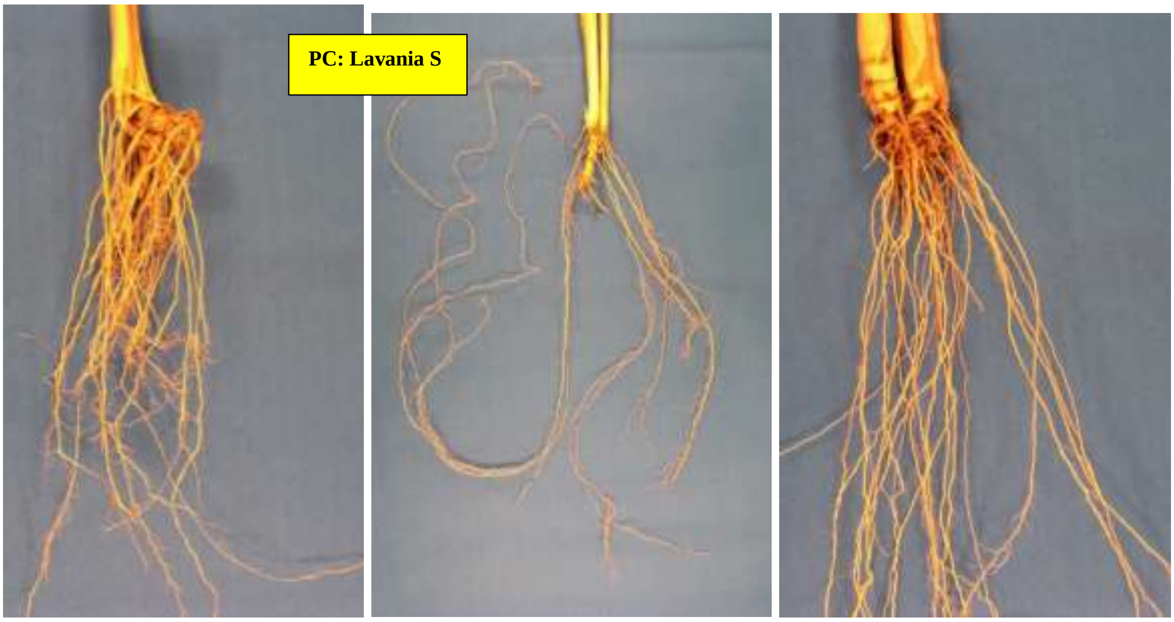
- *Tremendous diversity in structural organization*
- *Growth pattern – vertical deep growth to a more spreading type*
- *Diversity in root-thickness and branching pattern*
- *Smooth to fibrous-anastomosising root mass*

Physiological

- *Physiochemical absorption potential of agricultural and toxic residues*
- *Tolerance to a wide range of soil climates*

The following photos show the diversity of the Vetiver Root System





Vetiver Root Ideotypes

Essential Oil

Smooth and thick vertically growing roots with minimum branching, with well developed phloem

Land / Slope Stabilization / Bioengineering

Profusely branching, spreading type with the amouint least essential oil

Water and Soil Reclamation

High absorption potential for soluble N, P and pesticidal residues

Detoxification and Pollution Mitigation

High absorption potential and tolerance to heavy metals

Management of Waterlogged areas

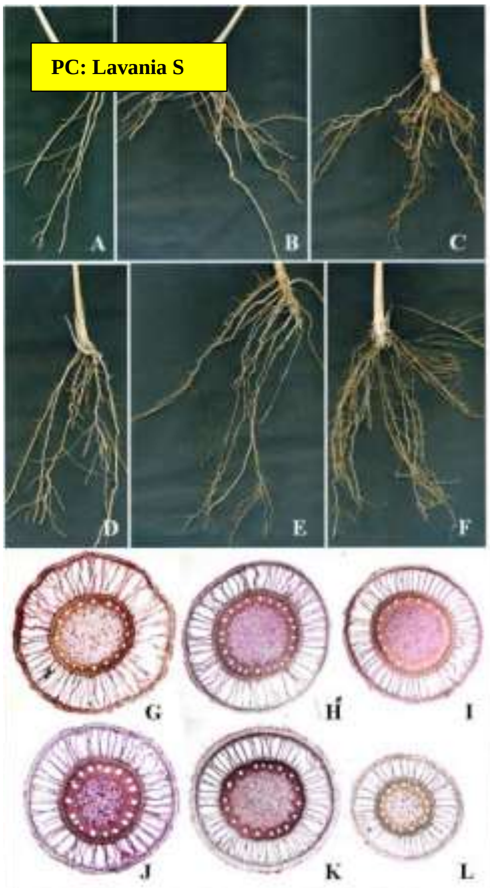
Spongy roots with schizogenous cortex

In a paper entitled *Vetiver grass model and phenomics of root system architecture*, Lavania, (S., 2019) stated that *with rising global interest in the vetiver root system, there is a need to explore the diversity in its root architecture for its multifarious applications to make the vetiver plantations ecosystem sustainable and globally acceptable. Two different models have been identified:*

- Industrial applications**:** *It is desirable to identify plant types that sport thicker roots with least lateral roots, lysigenous cavities containing higher number of essential oil secretary cells, shorter crop cycle and lower distillation period*
- Environmental applications**:** *The key requirements for environmental applications are non-seeding habit and low essential oil in the roots to realize non-invasive feature and deterrence to uproot the plantations for its otherwise industrial utility. In addition, the root architecture for ecological plantations should be such that the roots are able to penetrate deep into the subsoil horizon to realize Carbon sequestration; impart high soil binding potential achievable through fast growing web forming tufted roots; realize high absorbance of toxic chemicals and metalloids for pollution mitigation; should have capacity to sustain high lysis in the cortical region for enhanced aeration to sustain its survival under submerged conditions. An illustrated account of phenomic diversity in root architecture vis-a-vis its prospective industrial and environmental applications is shown below.*

Tufted roots of different vetiver genotypes showing variation in growth, length and density at the same age

Representative architectural diversity in primary roots depicting smooth to hairy with secondary and tertiary roots (A-F); Transverse section of primary roots showing diversity in root thickness, variation in cortex / stele ratio and cortical aerenchyma (G-L)



Genetic and taxonomic features

Vetiver grass (*Chrysopogon zizanioides* L.) belongs to the same grass family as maize, sorghum, sugarcane and lemon grass.

Scientific classification	
Kingdom	Plantae
Order	Poales
Family	Graminae (Poaceae)
Subfamily	Panicoideae; Tribe-Andropogoneae; Subtribe-Sorghinae
Genus	*Chrysopogon*
Species	*zizanioides*
Common name	Vetiver grass

Screening of numerous accessions of *Vetiveria zizanioides* (L. Nash) and other vetiver species such as *Chrysopogon fulvus* (Spreng.), *C. gryllus*, *Sorghum bicolor* (L.) and *S. halepense* (L.), revealed that *Vetiveria* and *Chrysopogon* are not separable by their Random Amplified Polymorphic DNAs (RAPDs). This led to the merging of the genus *Vetiveria* with the genus *Chrysopogon,* hence *Vetiveria zizanioides* (L. Nash) is now known as *Chrysopogon zizanioides* (L Roberty), with cytology of chromosome base number, x = 5 and 10, 2n = 20 and 40. (Adams et al, 2003)

Although vetiver is a typical C4 tropical grass, it can survive and thrive under subtropical conditions and in some very cold settings with temperate climate conditions. Vetiver is a non-invasive plant; it flowers but sets no seed, it produces neither above nor underground runners (stolons) and it has to be established vegetatively by root (crown) splitting or other vegetative propagation means. Hence it is non-invasive and can easily be controlled by uprooting the plant at the crown and drying out the exposed roots or by using a herbicide such as Glyphosate.

Vetiver grass has a deep and massive root system, which is vertical in nature descending 2-3 meters in the first year, ultimately reaching five meters under tropical conditions. (Truong 2002)

Designer Vetiver genotypes

In addition to the above naturally existing species and genotypes Lavania U *et al* (2020) have identified/developed a number of new vetiver genotypes specially designed for environmental and industrial applications.

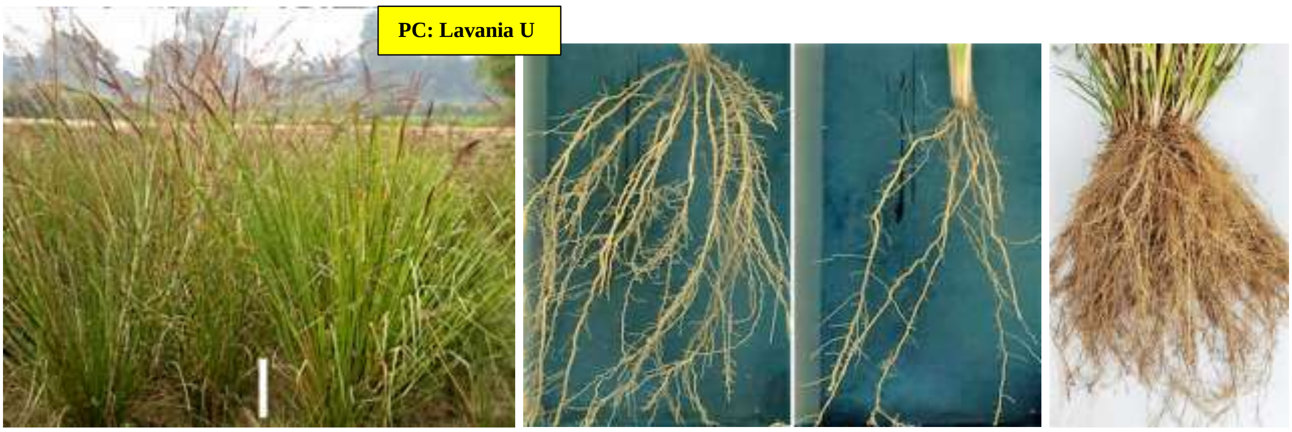
They have done this because traditionally vetiver is valued in the fragrance industry for its essential oil extracted from roots, but it is now extensively used as a green technology for its multifarious environmental applications. With a rising global interest in Vetiver System Technology (VST), it becomes desirable to identify/develop designer genotypes of vetiver for specific applications to increase its implementation efficiency. The three distinct applications are:

- Efficient soil binder and soil ameliorator combined with fodder qualities,
- Root based sequestration of atmospheric carbon deep into subsoil horizon likened to trees; and
- Short duration crop for high essential oil productivity.

Keeping in mind the concept of 'root ideotype' and 'root phenomics' suggested for vetiver grass for specific applications, extensive efforts were made to isolate genotypes from the range of diversity prevalent across the length and breadth of India. Further selection pressure and genomic manipulation were applied to identify genotypes that meet the 'best fit' required by environmentalists in a global perspective, including non-invasiveness. The genotypes were tested for their efficiency as a soil ameliorator by growing them in iron mine spoil dumps, nutritional and palatability qualities of leaves desirable as a fodder, growth efficiency and deep penetrability of roots for carbon sequestration potential, and concentration and quality of essential oil in the roots harvested after six months.

The following three designer genotypes were developed for environment and industry specific applications:

- *Genotype 'BL'* is a non-seeding ideal plant type that has profuse rooting with high secondary roots (with the least oil) for enhanced soil binding properties, coupled with profuse tiller and shoots rich in high fiber content and nutritional qualities suitable for fodder. This genotype is suitable for ecological plantations in degraded soils (iron mine spoil dump.



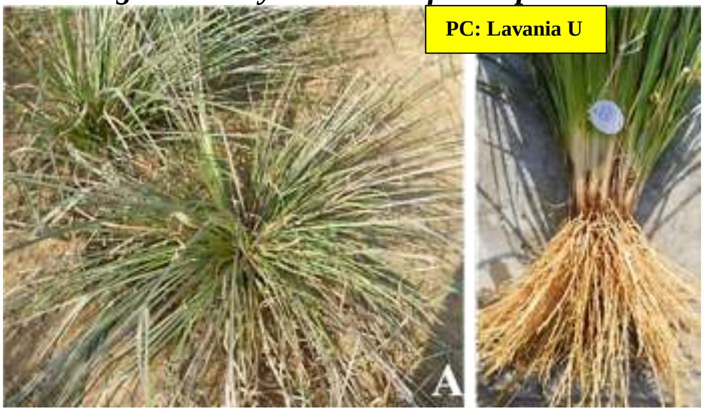
CIM Vriddhi   Genotype BL   Genotype BL (Lab.) CIM Vriddhi (Lab) Genotype BL (Field)

• *Genotype CIMAP-KH40* is an infertile plant type that has thick deep penetrating roots with least secondary roots and high biomass. This plant type is ideal for sequestration of atmospheric carbon deep into subsoil region likened to trees as well as suitable for improving soil fertility through enriching the soil carbon pool.

Profuse and deep root relative to shoot

• *Genotype CIMAP- KHUSHINOLIKA* is a short duration clone that could yield high amount of essential oil of desirable quality from its roots harvested just after six months of plantation, otherwise obtainable after 18 months in the standard cultivars.

Strong secondary roots ideal for oil production

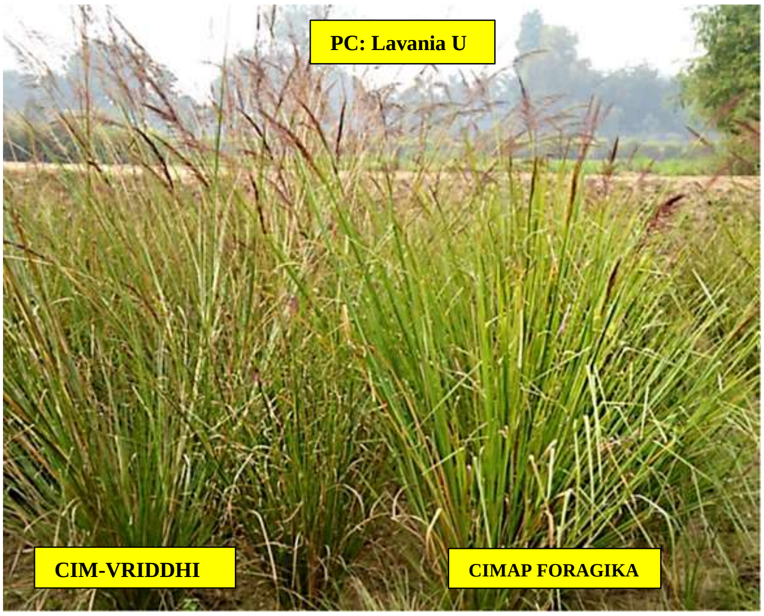


• *Genotype CIMAP- FORAGIKA* was identified by Lavania et al (2021) as an endemic new cultivar near Lucknow, which is sterile, having softer leaves, very high biomass with 1.5 times the number of tillers (this would equate roughly with total leaf mass of the existing cultivars), extensive root system (2.5 times the root mass) and high forage value.

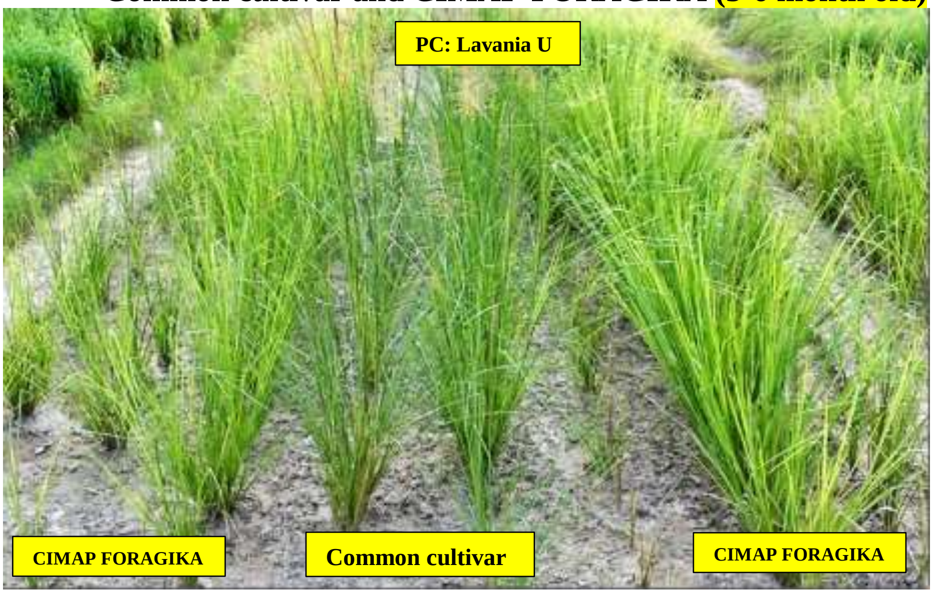
The cultivar is morphologically quite different from other clones currently used and has very low oil content so would not be of interest to the vetiver oil producers. With the added forage value it could be attractive to farmers. It is currently available for public use and could be obtained from Central Institute of Medicinal and Aromatic Plants (CSIR), Lucknow, India.

CIMAP- FORAGIKA

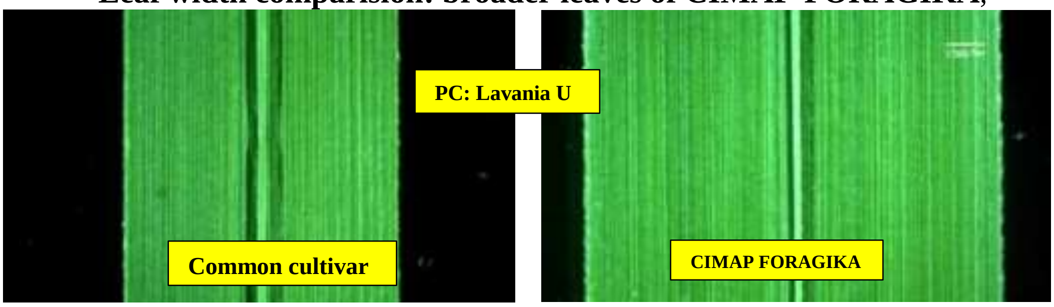
Fully mature plant of Clone CIM-VRIDDHI and CIMAP-FORAGIKA. Note erect tillers, broad and drooping leaves of CIMAP-FORAGIKA



Common cultivar and CIMAP-FORAGIKA (3-6 month old)



Leaf width comparision: broader leaves of CIMAP-FORAGIKA,



Root pattern of CIMAP-FORAGIKA.
Left – under field conditions, Right – under experimental conditions grown in microcosm. Note profuse primary, secondary and tertiary roots.

Profuse secondary roots in CIMAP-FORAGIKA as compared with CIM-VRIDDHI

CHAPTER TWO

BASIC INFORMATION AND UNDERSTANDING OF PLANT ROOT GROWTH AND DEVELOPMENT IN SOIL MEDIUM

PLANT ROOT GROWTH AND DEVELOPMENT IN GENERAL

This review extracts information in various chapters of the book ***Plant Roots: The Hidden Half by Eshel and Beeckman (2013)*** that are relevant and supporting the recorded/observed characteristics and behaviour of the vetiver root system.

Role of Plant Roots

Plant roots have to serve two important functions: They anchor the plant to the soil and allow the uptake of important nutrients and water from the soil. To be able to fulfill these tasks, plant roots have to be able to grow into the soil to reach the nutrients and water, and to be able to do this, they need the energy provided by cellular respiration. For this they need a sufficient supply of oxygen that they must either get from the air present in variable amounts in between the soil particles or more commonly from a special tissue called ***aerenchyma****, which supply oxygen from plant shoots.*

Basic Features of Root Anatomy

Plant root structure is a lot simpler than that of the shoot. This is mainly due to the fact that the shoot has a complicated branching pattern and bears leaves, flowers, and fruits, and often some additional structures such as thorns or hairs. Disregarding whether the plant is a dicotyledonous or monocot species, its root has a radially symmetric structure with the vascular tissues inside the root central cylinder. The outermost part of this cylinder is formed by a layer of cells from which the lateral roots will eventually emerge. Inside this cylinder are the actual vascular tissues, xylem, and phloem, organized in a radial symmetric fashion: In monocotyledonous plants, there are normally several bundles of xylem and phloem; hence, the root is called polyarch, while in dicots and gymnosperms, the number of bundles is less than eight leading to oligarch organization of the central cylinder.

The tissue outside the central cylinder is called the root cortex. The innermost layer of this is the endodermis, which has a profound effect on the water and nutrient uptake by the root. The rest of the cortex is formed mainly of parenchymal tissue with small or larger air-filled intercellular spaces. It is in this cortical tissue that aerenchyma is formed in plant roots.

Aerenchyma Formation and Functioning in Plant Root

In wetland plants, cortical aerenchyma is formed constitutively, but it is even more pronounced in roots growing under oxygen-deprived conditions. Dryland plants do also develop root cortical aerenchyma in the case of a sudden flood or other circumstances where oxygen diffusion to the roots is hindered.

Aerenchyma can be formed by three very different processes:

- *It can develop when the intercellular spaces between cells enlarge through the breakdown of peptic substances in the middle lamella.*
- *It can form from programmed cell death, PCD. The air spaces formed where cells have disintegrated during PCD*
- *It can develop from honeycomb aerenchyma, which is characterized by intercellular spaces through cell division and expansion.*

All these events are developmentally controlled. Even though the air spaces in roots, rhizomes, and stems take up a large part of the volume of the organs, the risk of water filling these *spaces in the case of a wound does not seem to be great. There are two main factors preventing the water flowing in: the air cavities do not form continuous space through the different plant organs, but there are thin septa separating the root cavities from the shoot, and in the shoot, there are septa in each internode. Another fact affecting the influx of water is the presence of hydrophobic compounds lining the walls of the air cavities.*

PLANT ROOT GROWTH, ACTIVITY AND INTERACTION WITH SOILS

This review extracts information in various chapters of the book ***Plant Roots: Growth, Activity and Interaction with Soils, Gregory (2006)***, which pointed out the importance of the root system and its interactions with surrounding environment**.**

Plants, Roots and the Soil

It has long been appreciated that plants influence the properties of soils and that soil type can, in turn, influence the type of plant that grows. This knowledge of plant/soil interactions has been put to use by humans in their agriculture and horticulture. Although the focus of much plant and soil science has been on the return of leaves to the soil both as a stock of C in the soil and as a substrate for soil organisms, root returns to soil are larger than shoot returns in several regions and several grasses produced more organic matter below ground than above ground.

Functional Interdependence of Root and Shoot

The different morphologies, anatomies, physiologies and functions of roots and shoots have frequently led to their being considered as two separate systems within the entire plant. Nevertheless, while each system grows and functions as a discrete site for the capture of specific resources (carbon dioxide, light, water and nutrients), the two systems are coupled together and their functions have to form an integrated system. The total above-ground growth of plants is strongly dependent on the developmental stage of the root. Only when the root can fully develop will the above-ground plant reach its full potential. Therefore the size of both systems might be inter-related and the simpler notion that big shoots were associated with big root systems.

In experiments designed to investigate the equilibrium between root and shoot growth, it was found that characteristic equilibria were attained depending on the conditions prevailing. The experiments demonstrated the following:

- *When root growth is limited by a factor to be absorbed by the root system, then root growth is relatively favoured; conversely, when the limiting factor has to be absorbed by the shoot, its growth is relatively favoured.*
- *Disturbance of the ratio of root:shoot brought about by either root removal or defoliation leads to changes in the pattern of growth so that the original ratio is rapidly restored.*
- *Transfer of plants from one environment to another causes changes in the pattern of assimilate distribution so that a new characteristic root:shoot ratio is established over a period.*

The Root–Soil Interface

The interface between the root and the soil is complex and frequently an ill-defined boundary. Products are released from roots into the soil which change its chemical and physical properties, and stimulate the growth of various microorganisms. Concurrently, the root tissues and associated root products also provide physical shelter for many microorganisms. This complex environment where root and soil meet is known as the rhizosphere. It is now more widely used to describe the portion of the soil that forms the complex habitat of plant roots, the composition of which is altered by root activity.

Roots and soil particles are frequently in intimate contact, with root hairs, mucilage and microbes forming a zone of multiple interactions between the plant and the soil. Mucilage of both bacterial and plant origins is able to bind soil particles on drying, and to retain the particles on subsequent rewetting. This biologically active zone of soil means that root–root, root–microbe and root–faunal communications are likely to be continuous occurrences.

Architecture of Root System

Roots are complex structures that exist in diverse forms and exhibit a wide range of interactions with the media in which they live. They also exhibit a very wide range of associations with other living organisms with which they have co-evolved. Laboratory and field studies have revealed a great deal about this complexity, especially during the last 20 years or so when there have been several national programmes of research around the world focusing on below-ground processes.

Root architecture, the spatial configuration of a root system in the soil, is used to describe distinct aspects of the shape of root systems. From the architecture both the topology (a description of how individual roots are connected through branching) and the distribution (the presence of roots in a spatial framework) can be derived. Root architecture is quite complex and varies between and within plant species. Drawings of excavated root systems of crops and other species show the differences in shape between monocotyledons and dicotyledons and allow some broad generalizations to be made about the depth of rooting and the relative distribution of roots. Generally with the exception of the tap root which grows almost vertically throughout, most other root axes grow initially at some angle relative to the vertical but gradually become more vertically orientated. Gravitropic responses combined with responses to light, water and touch together, perhaps, with the predominance of vertical cracks in deeper soil layers, produce these patterns

The following section describes the essential anatomical and morphological features of some selected types of root that are relevant to vetiver as a background to understanding the diverse forms of its root systems and their functioning.

Important Components of the Root System

<u>*Root hairs*</u>

Behind the zone of elongation is a zone of maturation in which root hairs are produced as specialized projections from modified epidermal cells. In many plant species (nearly all dicots, some monocots, and most ferns), all epidermal cells of the root seem capable of producing a hair, whereas in others there are cells that have the potential to become root hairs

The development of root hairs is also greatly influenced by the surrounding environment such as Ca and P concentrations For example, when Arabidopsis was grown in a P-deficient soil, root surface area was increased sevenfold compared with plants grown under P-sufficient conditions, and root hairs constituted 91% of the total root surface area.

Root hairs can vary in length and frequency along a root but are typically 0.1–1.5 mm long, 5–20 μm in diameter, and vary from 2 per mm2 on roots of some trees to 50–100 per mm of root length in some grasses. Usually, the size of the root hair zone on roots is short because root hairs have a short life of a few days or weeks. Nuclear staining with acridine orange suggested that the average life of the root hairs was 1–3 weeks. Lifespan will, though, be affected by several environmental factors including soil water status and nutrition.

Root hairs play an important role in root/soil contact through the formation of rhizosheaths and in the acquisition of water and nutrients such as calcium, potassium, nitrate, ammonium, manganese, zinc, chloride and phosphate.

Root cap

The root cap responds to the soil environment to: (1) control the direction of movement; (2) facilitate penetration into soil; and (3) determine the microbial environment around the root. The root cap is a multifunctional, molecular relay station that not only detects, integrates and transmits information about the environment to appropriate plant organs, but also functions to specifically modulate properties of the soil habitat in advance of the growing root. The cap maintains its own independent developmental patterns in response to the environment while simultaneously directing movement generated by the root meristem and region of elongation'.

Mucilage

The gel-like mucilage secreted by the cells of the root cap contributes to many interactions between the plant and the soil including root penetration, soil aggregate formation, microbial dynamics and nutrient cycling. As the root extends through the soil, so mucilage and associated root cap cells are left behind along the root–soil interface. Mucilage mixed with border cells penetrates between soil particles and into aggregates close to roots, and plays a major role in maintaining root–soil contact. In many grasses and some dicots, a coherent sheath (a rhizosheath) of soil permeated by mucilage and root hairs develops around the root and remains intact on root axes until the large xylem vessels mature. While the mucilage per se has almost no capacity to store water in the rhizosphere, the chemical and physical properties of mucilage influence the supply of water to the root

Many functions have been ascribed to mucilage, but one of the most common is that it acts as a lubricant to ease the passage of the root through the soil. The resistance to root penetration in a soil is the sum of the frictional resistance to root penetration plus the pressure required to form a cavity. Friction can be 80% of the penetration resistance experienced by roots as they move through soil, so reducing this resistance would be advantageous to plants in their exploration of soil resources.

Development and Growth of Root System

Soils are optically opaque so that continuous visual observation of growth is impossible, while disturbance of soil to expose roots substantially changes their environment which may, in turn, lead to modifications to growth and function. For example, measurements of root mass at particular times are frequently obtained by washing roots from soil, whereas measurements of root longevity and turnover are commonly obtained by direct observation of roots growing against a transparent glass or plastic surface (a mini-rhizotron).

One of the commonest sets of measurements required by ecologists and agriculturists is that of the size of the root system, how it is distributed with depth, and how it changes with time. In addition to the failure to recover roots from the soil, root mass can also be lost during washing and storage. Root respiration may result in 5–10% loss of weight in the 24 hours after sampling unless samples are kept cool (4°C). The downward velocity of the root was linearly related to thermal time (the summation over time of temperature above a specified base) and were substantial differences between species.

Size and Distribution of the Root System

Mass and length

In croplands and temperate grasslands, most of the roots are fine roots (≤2 mm diameter), but in many biomes coarse roots make up by far the majority of the root biomass. It is the fine roots that are of particular interest because they constitute the primary pathway for water and nutrient uptake.

Depth of rooting

Genetic and environmental factors both influence the depth of rooting. The depth to which roots are able to grow has many implications for the hydrological balance and biogeochemical cycling of ecosystems. In 290 observations of maximum rooting depth of 253 woody and herbaceous species from the major terrestrial biomes, it was found that maximum rooting depth varied from 0.3 m for some tundra species to 68 m for Boscia albitrunca in the central Kalahari. Twenty-two species had roots that extended to 10 m or more but 194 species had roots that were at least 2 m deep. For example, grouping the species across biomes, the average maximum rooting depth were:

- *Cropland 2.1m ± 0.2;*
- *Temperate grassland 2.6m ± 0.2;*
- *Sclerophyllous shrubs and trees 5.2m ± 0.8;*
- *Desert 9.5m ± 2.4; and*
- *Tropical grassland/savanna 15.0m ± 5.4.*

In practice, environmental conditions may also play an important role in determining rooting depth either because of limited soil depth or hostile soil conditions. The maximum depth of rooting on deep soils is genetically determined and differs between species grown under identical conditions

Root and Shoot allocation of dry matter

The proportion of total plant dry matter allocated to roots differs substantially between different groups of plants and ecosystems. In annual plants, the allocation of dry matter to roots changes during their life cycle and with growing conditions. Typically, relatively more assimilates are channeled to roots during early growth but, as development proceeds, the growing reproductive structures come to dominate and the amount of assimilate translocated to roots decreases In general, a shortage of resources in the root environment causes a shift of as similates in favor of the root system relative to the shoot and vice versa. This is clearly seen, for example, in the response of cereals to applications of N fertilizer where there was a sharp decline in root proportion to total plant mass immediately following N fertilizer application. In drier conditions, too, fertilizer applications can have similar effects whereby shoot growth is increased substantially but root growth is less affected so that root:total mass ratio is decreased.

Root longevity and turnover

The median lifespan of roots is highly variable, ranging from a few weeks in some plants (annual crops like sorghum and groundnut) to many months (sugar maple). In grasses, thicker roots and high tissue density have also been associated with increased longevity. For example, a study of four grass species in The Netherlands found that the species from N-rich habitats (L. perenne) had significantly finer roots and shorter longevity than species from N-poor habitats. In nutrient-poor environments thicker roots with a longer lifespan may increase the residence time of nutrients in the plant, and provide an important means of nutrient conservation.

Longevity also varies within a species depending on growing conditions and the interaction with fungal symbionts and pathogens. Application of ammonium sulphate increased the production of fine, white roots relative to the control but these had high mortality (60% compared with 30% in the control), whereas application of nitrogen-free fertilizer decreased production and mortality (8%). Mycorrhizal colonization can decrease rates of root mortality through diverse effects such as improved nutrition, enhanced tolerance to drying soil, and reduced deleterious effects of pathogens and herbivory.

Root growth and root mortality occur throughout the year but the net balance is highly seasonal in perennial plants, with a burst of production in the spring and considerable mortality in the autumn.

The Functioning of the Root System

The root system has to serve several functions simultaneously. It has to provide a stable platform for the shoot so that the photosynthetic organs can intercept sunlight, and also has to provide a network that can exploit the water and nutrient resources of the soil. The availability and movement in the soil of resources varies depending on the particular resource being considered, so that in contrast to the shoot which is essentially harvesting only two resources, light and carbon dioxide, the roots and root system have evolved to cope with a more challenging environment

Root anchorage

The force required to pull a root from a soil is dependent on the area of contact between the root and the soil, and the shear strength of the soil. The resistance to being pulled out is, then, greatest for long, thick roots in strong soil. Roots, though, may not be able to withstand this force (their breaking strength is proportional to their cross-sectional area) and may break before lower parts of the root are stretched.

The uprooting tests lead to two important conclusions: (1) a plant cannot improve its anchorage just by increasing its root length or strengthening the bond between root and soil; and (2) anchorage will be improved by strengthening the base of the root (e.g. by lignification and/or secondary thickening.

The anchorage systems of self-supporting plants must, then, be able to transmit rotational torque to the soil rather than transmit simple upward forces. This means that the fibrous root systems that are so good at preventing uprooting are much less good at preventing overturning because each root will simply bend at its base. Resistance to rotation requires at least one rigid element at the base of the stem to act as a lever; this can be provided by a tap root, or plate root systems, or by leaves growing at the base of the stem in a rosette, or by having several stems which grow horizontally along the ground before growing upwards

Soil properties influence the exact mode of failure. If the soil is wet, then it will compress easily allowing rotation deep underground and the permanent leaning over of the plant, whereas if it is dry, the tap root or stem may fail

Water Uptake by Plant Root System

Water is essential to the life of terrestrial plants and for the biota that live in the soil. It carries nutrients in the soil to the roots, is the solvent for, and medium of, most biochemical reactions within plants, and its loss from plants is an inevitable consequence of CO2 exchange with the atmosphere. For most plants, the soil is the major source of their water, so that the acquisition of water from soils by roots has been, and continues to be, a major topic of soil/plant research.

There still remain a great number of uncertainties in our understanding of how water uptake by root systems occurs, and no single approach has yet found universal acceptance. Nevertheless, some broad generalizations have been found useful in describing the activity of root systems in taking up water. In wheat it was found that water loss from the soil profile over the growing season was better correlated with maximum rooting depth than with total root length. Inflow (uptake per unit root length) was highest in soil layers that were moist from recent rain or in which root length was expanding most rapidly.

The soil–plant–atmosphere continuum

Central to the understanding of how water moves from soil to atmosphere via plants is the cohesion-tension theory, which can be summarized as follows:

- *Water forms a continuous hydraulic system from soil, via plant, to the atmosphere.*

- *Evaporation from leaves reduces their water potential causing water to move from the xylem to the evaporating surfaces; this, in turn, lowers the water potential of the xylem.*
- *Gradients of water potential within the plant result in water inflow from the soil into the roots and thence to the leaves.*
- *Water has high cohesion and can be subjected to tensions (negative pressures) up to several hundred MPa before the column will break; and*
- *Walls of vessels are the weakest part of the system and can contain air and/or water vapor.*

Water movement from root xylem to stomata

Water flow in the shoot is largely controlled by stomata which actively regulate the loss of water to the atmosphere in response to the water status of the shoot and signals, especially abscisic acid (ABA), from the root. The cuticle of leaves can be regarded as almost watertight, especially in young leaves, so the stomata are the main sites through which water vapor is lost to the atmosphere. Atmospheric factors determine the potential rate at which water can be lost to the atmosphere but plants control the size of the stomatal opening so as to match the rate of loss to the supply available from the soil. Many environmental factors such as CO2 concentration, irradiance, humidity and water potential of the CO2 leaf affect stomatal opening

Nutrient Uptake by Plant Root System

Nutrient movement in soil solution

Roots are in direct contact with only a very small part of the nutrients in the soil solution, so that nutrients must move from the bulk soil to the root surface. This movement occurs through the processes of mass flow and diffusion. Mass flow (convection) occurs as a result of transpiration; dissolved ions are carried to the root surface in the hydraulic continuum formed by the soil–plant–atmosphere mentioned before.

Nutrient uptake and movement across the root

Many of the considerations pertaining to water transport across the root to the xylem, such as the role of the cell walls, membranes, exodermis and endodermis, also apply to the uptake and movement of nutrients, although there are some important differences.

Uptake into the plant almost always involves the passage across a membrane at some point in the transfer between soil and xylem, and this requires the expenditure of energy provided via the process of respiration. Uptake of nitrate is among the most costly in terms of energy expenditure, and its uptake across the plasma membrane is energy-dependent over almost the whole range of nitrate concentrations found in soils.

Some Responses Root System to Components of the Environment

Temperature

As with the shoot, temperature affects both the expansion of the root system through effects on development and growth. Growth can be specified either in terms of root length or root mass.

The response of a root system to temperature is similar to that of the shoot system with a minimum temperature below which no growth occurs, an optimum at which growth is maximal, and a maximum at which no growth again occurs. The minimum (base) and optimal temperatures depend upon the plant species and are typically in the ranges of 0–12°C and 25–35°C, respectively, while the maximum is almost always around 40–45°C. The optimum
is often broad rather than a sharp peak, and there is frequently a broad range of temperatures at

which root growth rates are =50% of their maximum. Both total root mass and total root length show similar overall responses to temperature although the size of the response to a particular temperature may differ between the two measures.

Plants exposed to temperatures below the minimum (ice and frost in temperate regions) and above the maximum (surface soil temperatures in tropical regions may exceed 45°C), have evolved mechanisms to cope with these extremes that are beyond the scope of this book.

Many studies have shown that the extension rate of roots is faster as temperature increases up to the optimum and decreases thereafter. However, there is no unique relation between extension rate of a particular root axis and temperature because elongation rates depend on the size and development stage of the shoot, canopy light interception and the age of the root axis itself.

The total length of the root system is also influenced by temperature and root mass depends on assimilate supply as well as temperature, but there appears to be some difference of opinion in the literature about the relative importance of these two factors in determining the size of the system.

Root elongation and mechanical impedance

Root growth occurs as the cells behind the root tip elongate longitudinally and radially to push the root tip forward. Almost all roots growing through soil experience some degree of mechanical impedance, and if continuous pores of appropriate size do not already exist then the root tip region must exert sufficient force to deform the soil. There is not much information about the minimum size of pore into which a root can grow without having to enlarge it, but few plants have roots smaller than 10 µm in diameter and most roots are much larger For example, in perennial ryegrass roots in pots of perlite containing sheets of steel mesh and found that roots were able to penetrate rigid pores as small as one-third of their nominal thickness by reducing their diameter. The degree of constriction was limited by the size of the root cap and that of the stele, which remained unchanged even in severely constricted regions.

The response of the root to hard objects and the importance of the root tip in sensing them were observed in maize. For roots with an intact root cap, the rate of elongation decreased from about 15 µm min–1 in the 20 minutes before contact to about 3.6 µm min–1 in the first 5 minutes after contact, before gradually increasing to the initial rate of elongation after 15 minutes. When the root cap was removed, the same initial rate of elongation was measured but no significant change in elongation rate occurred.

Root responses to mechanical impedance

There are two very obvious responses of plant roots to mechanical impedance – slowing of the rate of extension and an increase in root diameter immediately behind the root tip. It was found that the relative rate of elongation in response to soil strength differs between plant species but that at commonly encountered values of mechanical impedance >2 MPa, the root elongation rate of all species was reduced by at least 50%. The strength of most soils increases as they dry, so that shortage of soil water and hard soils are commonly interlinked.

The ability of roots to penetrate compact soils is not dependent on root thickness, but on the ability of the thickened root both to reduce the impedance ahead of the growing tip and to resist buckling or deflection when encountering strong layers. Lateral root proliferation appears to be a common response to compaction but the precise effects are variable

Soil water

Water affects root growth in many ways. A survey of water limited environments, showed that the rooting depth of vegetation was more strongly related to mean annual precipitation than to potential evapotranspiration.

In general that root systems become deeper as the environment becomes drier. For a given canopy size, herbaceous plants have deeper maximum rooting depths in drier environments, but because canopy size increases along the rainfall gradient, mean rooting depth also increases.

For a given location, numerous publications have confirmed that relatively dry soil conditions can induce plants to develop a more extensive root system and roots grown at low water potential were also thinner. Increasing soil strength (which is normally concurrent with soil drying) tends to result in thicker roots.

Soil aeration

It is well known that for most plants to grow in soil, part of the pore space must be gas-filled. This space allows the supply of O2 to roots to maintain respiration, and the removal of CO2 from the root, and if these processes do not occur fast enough then root growth is restricted. While there is some evidence that CO2 at high concentrations (although this is often accompanied by other toxic products) may be deleterious to root growth, the main effect of poor aeration is the lack of O2.

It was also found that roots whose elongation was reduced by oxygen shortage were thicker and elongation of individual roots was slower in waterlogged than freely drained soil.

Waterlogging and aerenchyma

Waterlogging has multiple detrimental effects on plants because of disruption to gas flow below ground. It is a common constraint to the production of agricultural crops, affecting about 10% of the global land area. Aquatic and marsh plants have adapted to cope with such conditions but many plants are very vulnerable.

The severity of the effects of waterlogging on roots depends on the developmental stage at which it occurs, the duration of the event, and other environmental factors such as temperature which affect the demand for oxygen by roots and microbes, and the production of chemicals (some of which are toxic to plants) in reduced soils

Aerenchyma, plant tissue containing enlarged gas spaces exceeding those found as intracellular spaces, occurs in many plants and is formed either as part of the normal developmental process, or in response to stress, particularly hypoxia as a consequence of waterlogging. It provides the plant with an alternative way for its root tissues to obtain O2 because the O2 interconnected lacunae provide an internal aeration system that transfers O2 from the atmosphere O2 to the root rather than relying on the soil atmosphere.

Two types of aerenchyma have been identified:

- *schizogenous aerenchyma forms as a result of differential cell growth and subsequent cell separation and is common in wetland species; and*
- *lysigenous aerenchyma forms after cells in the root cortex die and disappear to leave gas-filled spaces. Lysigenous aerenchyma formation is important in many crops plants including maize, wheat, barley and rice. The end result of aerenchyma formation is to leave a root where the gas spaces are separated by lines of cells bridging the space between the stele and epidermis, like spokes of a wheel. Aerenchyma formation increases porosity above that resulting from the usual intercellular spaces to form typically some 15–50% of the root volume.*

The network of spaces is continuous between roots and shoots allowing long-distance transport of gases. Until relatively recently most attention has been directed at the transport of oxygen to the roots but it is now appreciated that the system also transports CO2, ethylene and methane from waterlogged soils to the shoot and thence the atmosphere.Oxygen transported to the roots in aerenchyma is either consumed by adjacent cells, or diffuses towards the root apex, or diffuses out of the root into the rhizosphere.

The Soil Chemical Environment

Plant nutrients.Fertilizer applications to many soils produce substantial increases in shoot growth and yield while responses below ground are often less marked. For example applications of 50 and 100 kg N ha^{-1} to barley increased shoot dry weight by 13% and 52%, respectively, relative to the control at maturity while root dry weight was increased by only 8% and 16%; there was no further significant response to 150 kg N ha^{-1}

Low pH and Aluminium. Acid soils occupy almost 50% of all non-irrigated, arable lands and are very common in the tropics where high rainfall for prolonged periods has leached soluble bases. Acid soils can lead to negative effects on plant growth because of toxicity caused by H+, aluminium or manganese, or through deficiency of calcium or molybdenum. Plants can be separately adapted to H+ or Al3+ toxicity with different effects on root growth and anatomy.

Aluminium is now widely regarded as the most common limitation to growth on many acid soils because as pH falls to less than about 5.0–5.5, Al-containing minerals become soluble causing phytotoxicity. Even micromolar concentrations of Al3+ can inhibit growth in many plant species. Aluminium has similar effects on a wide range of plants with symptoms of toxicity evident first in the roots which appear shortened and swollen, before symptoms appear in the shoot.

Plants differ markedly in their response to aluminium toxicity both between and within species, with most of the proposed mechanisms involving either external avoidance or internal tolerance. External avoidance is brought about in several plants through the release of organic acids, especially citrate, oxalate and malate, which chelate aluminium in the rhizosphere Internal tolerance is also associated with processes resulting in the formation of Al–organic acid complexes. For example, in buckwheat (Fagopyrum esculentum) which accumulates Al in its leaves. In contrast, in Brachiaria species (tropical forage grasses), Al is accumulated at high concentrations in the roots with 70–85% of the Al taken

Salinity. Salinity affects about 7% of the world's land area with the area increasing as a consequence of clearing of native, perennial vegetation and the introduction of irrigation schemes without proper drainage. Irrigated plants in arid regions are especially susceptible to salinization, alkalization and waterlogging because waters often contain dissolved Na^+, Ca^{2+} and Mg^{2+} which are excluded by plant roots during water uptake and therefore accumulate in soils. Soil salinity inhibits growth through effects of both Na^+ and Cl^-. If salinity is high and the plant's ability to exclude NaCl is poor, then Na^+ or Cl^- (or both) accumulate in transpiring leaves and eventually exceed the ability of cells to compartmentalize these ions in the vacuole. The ions then build up in either the cytoplasm inhibiting enzyme activity or the cell walls causing them to dehydrate and shrink.

Root growth is often much less affected by salinity than leaf growth, in common with the effect of dry soil, suggesting that the effect is probably due to factors associated with water stress than a salt-specific effect. Generally, root cells of non-woody plants have Na+ and Cl– concentrations lower than those in the external solution (this rarely occurs in leaves), and do not accumulate Na+ at concentrations likely to become toxic

Roots and the Biological Environment

The rhizosphere is a zone that is densely populated with soil organisms, including bacteria, yeasts, fungi, protozoa and insects, feeding on a wide range of substrates. Much research has now demonstrated that compounds released from roots may act as messengers that communicate and initiate interactions between roots and a wide range of soil-dwelling organisms.

Interactions with Mycorrhizas. As with rhizobial symbioses, many plant species cannot complete their life cycle in their usual habitats without forming mycorrhizas, while others form mycorrhizas but can complete their life cycle without them. Arbuscular mycorrhizas (AM) – are the most common mycorrhizal class occurring in about 80% of terrestrial plants including trees, shrubs, forbs and grasses.

The nutritional benefits to host plants arising from mycorrhiza have received considerable attention and this aspect of the symbiosis tends to dominate the literature. Many experimental studies have shown that colonization by AM fungi increases P uptake and plant growth compared with non-mycorrhizal controls. For example, in leek seedlings both shoot growth and tissue P concentrations were increased by mycorrhiza.

AM infection decreased the critical level of soil P for maximum yield from 150 to 50 mg P kg–1 soil. The followings are two more important of the four possible explanations:

- *The main theory is that fungal hyphae outside the root are able to access supplies of P at some distance from the root that would otherwise take considerable time to diffuse to the root surface. The fungus takes up the ions external to the plant, translocates them to the internal mycelium, and transfers them to the host, thereby bypassing the slow diffusion process in the soil. This mechanism is similar to that of root hairs, but the hyphae can extend many centimetres from the root surface.*
- *The hyphae may be more effective than roots in competing with other soil microbes for P and thus circumvent the immobilization and/or adsorption of P in soils. There is some evidence that AM fungi can intercept inorganic P released during mineralization and thereby prevent immobilization in the microbial biomass or adsorption by minerals.*

The benefits of mycorrhiza for the acquisition of soil nutrients depend greatly on the fungal hyphae that extend well beyond the usual rhizosphere of the root to form a large 'mycorhizosphere'. AM hyphae can spread more than 250 mm from a root and ECM hyphae can grow several metres, thereby exploiting soil nutrients substantial distances from plant roots. Coupled with this is evidence that hyphae can link the roots of more than one plant to form an extensive underground network, thereby possibly allowing the transfer of C and nutrients from one plant to another, or at least from fungal mycelium in one plant to mycelium in another

Interactions with Nematodes. *The majority of nematodes in the rhizosphere feed on bacteria, but most plant-parasitic nematodes are soil-dwelling and infect plants via the roots. They cause major damage to many plant types but it is their impact on economically important crops that has received most attention. Most of the damage is caused by a small number of the many nematode genera that infect crops, especially the sedentary root-knot (Meloidogyne spp.) and cyst (Globodera and Heterodera spp.) nematodes, and several migratory nematodes including Pratylenchus and Radopholus spp.*

Genotypic Differences in Root Systems

There is widespread evidence for genotypic diversity in the root characteristics of many crop species, and increasing interest in using this diversity to exploit soils more effectively.

Size and architecture. *The majority of studies on root genotypic variation have focused on establishing differences in the depth of rooting, size of the root system, or rate of increase of the size of the root system, because these are measures that are conceptually easy to relate to the exploitation of soil resources*

In environments subjected to drought, roots have an obvious direct role in moderating the supply of water through the depth of rooting and the quantity of roots in a particular layer. Deeper rooting and subsequent extraction of water can be an important contributor to drought tolerance because drought is effectively avoided.

Thickness of roots and the ability of roots to penetrate compact layers have repeatedly been suggested as root traits linked to the ability of rice to avoid drought.

Root System as Management Tools

Optimal root systems and competition for resources. *The issue of the size of root system necessary to take up resources has been around in the crop production literature for a long time. However, as shown in section 1.2.1 it is impossible to give a single answer because it depends on the size,* *architecture and activity of the roots as well as the behaviour of the particular resource under consideration in the soil. In general, a large, more intensely branched.*

Biological drilling. In some parts of the world, soils may be deep but their subsoils dense resulting in a physical impediment to the downward growth of root systems of annual crops. Such soils are often associated with long periods of weathering leading to clay subsoils; in Australia they are called duplex soils and pose a range of problems for farmers. Mechanical disruption of the subsoil by deep ripping can reduce the physical constraints imposed by such soils but the process is often expensive and the benefits may be short-lived.

An option that has been proposed is to grow plants with roots that can penetrate the subsoil to create stable pores which, following decay of the roots, leave open biopores that can be exploited.

Utilization of subsoil water. The downward growth of roots into the soil profile is a major means of securing an uninterrupted supply of water should rainfall cease during crop growth and, indeed, most crops rely on water stored in the soil for at least part of their growing season. Attempts to increase the depth of rooting through breeding and varietal selection have been detailed in Chapter 8, but agronomic means have also been attempted with moderate success. Deeper roots will only be beneficial if the depth of wetting is greater than the usual depth of rooting so that in northern Syria, for example, where wetting is generally limited to the upper 0.8–1.0m, deeper roots in cereals such as wheat and barley would offer no advantage. Simulation analysis in the wheat belt of Western Australia showed that deeper roots gave the greatest benefit on sandy soils in high-rainfall zones where water and nitrogen would otherwise pass below the root zone.

Phytoremediation. Phytoremediation is an emerging technology that uses plants for the remediation of contaminated land. Phytoremediation of some organic compounds is possible, but most attention has been focused on inorganic metals and metalloids where the use of phytostabilization to reduce the flow of contaminants in the environment and phytoextraction to remove the pollutant from the environment are used. Efficient phytoextraction is determined by two factors: the biomass produced and the element concentration in the biomass relative to that in the soil (the bioconcentration factor). Roots cannot be harvested easily, so it is the size of the shoot system and the ability of of a plant to take up and transport metals to the shoots that determine the phytoextraction potential.

BASIC INFORMATION AND UNDERSTANDING OF PLANT ROOT GROWTH AND DEVELOPMENT IN HYDROPONIC MEDIUM

This review extracts information in various chapters of the book. ***- Complete Guide for Growing Plants Hydroponically by Jones (2014),*** which gave a comprehensive update of plant root growth and development under hydroponic conditions, which is considered as a technique for growing plants by placing the roots in liquid nutrient solutions rather than in soils.

In soil, any root restriction can have a significant impact on plant growth and development due to the reduction in soil–root contact. Root pruning, whether done purposely (to bonsai plants) or as the result of natural phenomena (due to the presence of plow or clay pans), will also affect plant growth and development in soil. In most hydroponic growing systems, roots may extend into a much greater volume of growing area or medium than would occur in soil.

Root size, measured in terms of length and extent of branching as well as color, is a characteristic that is affected by the nature of the rooting environment. Normally, vigorous plant growth is associated with long, white, and highly branched roots. It is uncertain whether vigorous top growth is a result of vigorous root growth or vice versa.

Root growth is dependent on the supply of carbohydrates from the tops and, in turn, the top is dependent on the root for water and the required essential elements. The loss or restriction of roots can *significantly affect top growth. Therefore, it is believed that the goal should be to provide and maintain those conditions that promote good, healthy root development, neither excessive nor restrictive.*

The physical characteristics of the root itself play a major role in elemental uptake. The rooting medium and the elements in the medium will determine to a considerable degree root appearance. For example, root hairs will be almost absent on roots exposed to a high concentration (100 mg/L, ppm) of NO_3^-*. High P in the rooting medium will also reduce root hair development, whereas changing concentrations of the major cations,* K^+*,* Ca^{2+}*, and* Mg^{2+}*, will have little effect on root hair development. Root hairs markedly increase the surface available for ion absorption and also increase the surface contact between roots and the water film around particles in a soilless medium; therefore, their presence can have a marked effect on water and ion uptake. Normally, hydroponic plant roots do not have root hairs.*

The question that arises is, "What constitutes healthy functioning roots for the hydroponic growing system?" The size and extent of root development are not as critical as in soil. It has been demonstrated that one functioning root is sufficient to provide all the essential elements required by the plant, with size and extensiveness of the roots being primarily important for water uptake. Therefore, in most hydroponic systems, root growth and extension are probably far greater than needed, which may actually have a detrimental effect on plant growth and performance.

It should be remembered that root growth and function require a continuous supply of carbohydrates, which are generated by photosynthesis. Therefore, an ever expanding and actively functioning root system will take carbohydrates away from vegetative expansion and fruit growth. Therefore, some degree of root growth control may be essential for extensive plant growth and high fruit yields.

A large and extensive root system may not be the best for most hydroponic growing systems. Rather than the large root mass, active, efficiently functioning roots are needed, since the nutrient solution continuously bathes most of the root system, thereby requiring less surface area for absorption to take place.

It should also be remembered that roots require a continuous supply of O_2 *to remain healthy and functioning. Roots will not survive in anaerobic conditions. Hydroponically speaking, a large, ever expanding root system probably does not necessarily translate into greater top growth and yield and, in fact, may actually have some detrimental effect.*

Temperature is another important factor that influences root growth, as well as the absorption of water and essential element ions. Water absorption by plant roots declines with decreasing temperature, decreases with increasing ion content of the water surrounding the root, and decreases with decreasing O_2 *content of the surrounding root mass environment The optimum root temperature will vary somewhat with plant species, but in general, root temperatures below 68°F (20°C) begin to bring about changes in root growth and behavior. Below the optimum temperature, there are reduced growth and branching, leading to coarser looking root systems. The maximum root temperature that can be tolerated before significant reduction in root activity occurs is not clearly known. Roots seem to be able to tolerate short periods of high temperature. Roots are fully functional at 86°F (30°C) and probably can withstand temperatures up to 95°F (35°C). However, the current literature is not clear as to the exact limits of the optimum temperature range for best plant growth. In order to avoid the hazards of either low or high temperatures, the roots and rooting medium should be kept at a temperature between 68°F and 86°F (between 20°C and 30°C). Reduced growth and other symptoms of poor nutrition will appear if root temperatures are kept at levels below or above this recommended temperature range.*

Aeration is another important factor that influences root and plant growth. Oxygen is essential for cell growth and function. If not available in the rooting medium, severe plant injury or death will occur. The energy required for root growth and ion absorption is derived by the process called "respiration," which requires O_2*. Without adequate* O_2 *to sup- port respiration, water and ion absorption cease and roots die. Oxygen levels and pore space distribution in the rooting medium will* *also affect the development of root hairs. Aerobic conditions, with equal distributions of water- and air-occupied pore spaces, promote root growth, including root hair development.*

All essential mineral element ions in plant root cells are at a higher concentration than that present in the surrounding environment. Therefore, how the mineral element ions are able to move against this concentration gradient? It was shown that the absorption of ions by the root is by both a passive and an active process. Passive root absorption means that an ion is carried into the root by the passage of water; it is carried along in the water taken into the plant. It is believed that the passive mode of transport explains the high concentrations of some ions, such as K^+, NO_3^-, *and* Cl^-, *found in the stems and leaves of some plants. The controlling factors in passive absorption are the amount of water moving into the plant (which varies with atmospheric demand), the concentration of these ions in the water surrounding the plant roots*

Currently the Hoagland/Arnon nutrient solutions are considered to provide adequate/optimal nutrient level for plant growth.

Element content of Hoagland/Arnon nutrient solutions (ppm)

Elements	***Hoagland #1***	***Hoagland # 2***
Nitrogen (NO_3)	*242*	*220*
Nitrogen (NH_4)	—	*12.6*
Phosphorus (P)	*31*	*24*
Potassium (K)	*232*	*230*
Calcium (Ca)	*224*	*179*
Magnesium (Mg)	*49*	*49*
Sulfur (S)	*113*	*113*
Boron (B)	*0.45*	*0.45*
Copper (Cu)	*0.02*	*0.02*
Manganese (Mn)	*0.50*	*0.05*
Molybdenum (Mo)	*0.0106*	*0.0106*
Zinc (Zn)	*0.48*	*0.48*

CHAPTER THREE

VETIVER ROOT GROWTH AND DEVELOPMENT

GROWTH, DISTRIBUTION AND SOME PHYSICAL ATTRIBUTES OF VETIVER ROOTS

In Chapter One, *Introduction to Vetiver Grass,* it compared the different root architectures of various vetiver genotypes. This chapter concentrates on the root system of the South Indian species. *C. zizanioides* commonly used for various applications of the ***Vetiver System Technology (VST).***

John Greenfield was posted to Dick Grimshaw's World Bank agricultural team in New Delhi in the 1980s at a time when Watershed Development Projects were in vogue. He recommended (based on his Fiji experiences) that vetiver hedgerows, planted on the contour, should be introduced in India as a more effective and less costly approach than hard engineered contour bunds for soil and water conservation. Small-scale pilot programs and some excellent research were then implemented under the watershed projects under Greenfield's review. Subsequently in 1987 when Grimshaw took responsibility for the World Bank's Asia Agriculture Technical Division he extended VGT first to other Asian countries including Thailand, Philippines, Malaysia and China, then globally to Africa and the Americas. In 1994 Grimshaw retired from the World Bank and established the Vetiver Network as a knowledge-based international non profit organization that supported training, publications, small grants and research (including the author's work in Queensland, Australia) to further extend and develop the technology. In 2000 all applications of VGT were brought together under the collective term "Vetiver System". (Grimshaw, 1995, 1997, 2008, 2009, 2014).

In his book entitled *Vetiver Grass: An Essential Grass for Conservation of Planet Earth"* Greenfield (2002) recounted his experience in Fiji in the 1950s, where he and his colleagues of the Colonial Sugar refinery extended the use of vetiver grass as an effective and low cost soil conservation measure on a large scale. Whilst in India Greenfield wrote the farmer handbook *'Vetiver Grass - A Method of Vegetative Soil and Moisture Conservation"* (1985, 1987) that was subsequently updated several times and published as, *Vetiver Grass - A Hedge Against Erosion*. These handbooks have been translated into more than 20 languages and more than 100,000 have been distributed globally. For this work Greenfield is known as "The Father of the Vetiver System"

Further recognition of VST was promoted by the Board of Science and Technology for International Development of the National Research Council and the National Academy Press, Washington DC, publishes the book entitled *Vetiver Grass, A Thin Line Against Erosion* in 1993, (Vietmeyer and Dafford, 1993)

In a review on the importance of vetiver roots in the essential oil industry and various environmental protection applications, Lavania S. (2019), Department of Botany, Lucknow University, Lucknow, India**,** pointed out that most grasses have fibrous roots, which spread out from the underground part of the crown and hold the soil in a horizontal pattern. The vetiver roots, however, penetrate vertically into the soil, whether it is the main root, secondary root or their fibrous ramification. In vetiver, the roots are biologically the most important and economically the most useful part of the plant. In addition to absorbing water and stabilizing soil moisture, vetiver roots help facilitate the absorption of toxic substances, excess chemical fertilizers, pesticide residues and heavy metal ions, improve physical elements and decomposition of organic matters. As such, their plantating are useful in soil and water conservation and the maintenance of good soil health. They serve as an important resource for the extraction of aromatic essential oil, which is used in the fragrance and medicinal applications. Dry roots are used as the raw material for preparation of various household products and handicrafts.

Structural Dynamics of Vetiver Root

Vetiver roots are comprised of the tufted mass originating from the crown from which shoots also arise. In general, growth and behaviour of roots is coupled closely to the growth and behaviour of shoots. Mature vetiver roots, about 12 to 18 months old, evince well-developed vascular cylinder and persistent cortex. Whereas the bast region (extra axillary secondary phloem) in root is the source of essential oil, the vascular cylinder (secondary xylem) provides physical and tensile strength to the vertically growing penetrating roots.

Vetiver Root Ideotype

Semi-technically 'ideotype' means 'ideal type' and with respect to plant this means 'ideal plant type'. Depending upon the requirement, the definition of a particular ideotype may suit specific needs. Since, the vetiver root system has numerous applications, a root ideotype has to make best use of root characteristics both in terms of growth potential and qualitative features. It is, therefore, necessary to define the characteristic features of the root suite to specific applications. In a broad sense utilization of the vetiver root system could be categorized into three major categories:

- Vetiver roots for environmental applications;
- Vetiver roots for essential oil; and
- Dry roots for handicrafts, and other household purposes.

This Review concentrates on the first application.

Vetiver roots for environmental applications

The various environmental applications of vetiver root include planting of vetiver hedgerows for soil and water conservation, detoxification of soil and water, and bioengineering uses. For soil binding in degraded soils, it is desirable that the soil surface is least disturbed/displaced. Accordingly a root form that binds to the root surface and covers more area with less plant population is desirable. *Therefore, a root ideotype should have more fibrous horizontally spreading type forming anastomosis of root mass.*

Presence of secondary and tertiary fibrous roots would add value to such roots by increasing their soil binding potential, making them unattractive to root diggers as these roots have the least oil content. For water conservation and detoxification of soil and water, high absorbing potential of roots is desirable. More spongy and fibrous roots provide large surface area, high tolerance to toxicity, deep growing roots are ideal for the purpose.

Vetiver root biomass of different genotypes

Xia et al (1999) of the South China Institute of Botany in Guangzhou, China investigated 12 genotypes of vetiver grass. The first 10 were obtained from the USA by Mark Dafforn and Robert Adams and the last two were collected locally.

- **USA**: Capitol, Huffman and Sunshine (1,2,3)
- **Malawi**: Lilongwe and Zomba (4,5)
- **Sri Lanka**: Kandy (6)
- **India:** Karnataka (7)
- **Malaysia**: Malaysia, Parit Buntar and Sabak Bernam (8,9,10)
- **China**: Domesticated Guangdong* and Wild Vetiver (Wuchan) (11,12)
  (* Karnataka, introduced by Dick Grimshaw from Bangalore into China in November 1988).

They reported that the biomass of roots for different ecotypes varies greatly. The least root biomass was in the local Wild Vetiver - Wuchan, which was introduced to China last century for essential oil production, and the most biomass was in the Zomba ecotype, in spite of the fact that

Zomba had the poorest ability to produce new tillers.

Root performance of 12 different vetiver genotypes

Item	**Ecotypes**											
	1	2	3	4	5	6	7	8	9	10	11	12
A	0.87	0.82	.88	1.25	1.44	0.77	0.42	0.51	0.97	0.58	0.57	0.39
B	50	60	60	20	30	40	80	70	60	70	40	30

Item A is the root weight in mg/tiller, and Item B is percentage of the root with diameter <1mm to the total root amount (%)

Root age and biomass

In a separate study Xia et al (1999) also noted that both vetiver root length and biomass of all commonly used ecotypes peaked at 6 months except the local Wild Vetiver. This suggests for commonly used ecotypes pruning at approximately 6 month after planting is advisable.

Cultivars	**Treatments**	**Root length (cm)**	**Root biomass (g/pot)**
Karnataka (tissue culture)	3 month - old	198	79
	6 month old	214	102
	2 year old	212	78
Karnataka (slip)	3 month old	238	60
	6 month old	240	364
	2 year old	229	306
Sunshine	3 month old	230	92
	6 month old	**227**	**173**
	2 year old	193	28
Wild Vetiver	3 month old	116	9
	6 month old	123	25
	2 year old	133	32

Effect of pruning on root development and biomass

An experiment was conducted to study the effects of pruning on growth of vetiver roots of different cultivars. Xia (2007) selected three cultivars: Sunshine, Karnataka with tissue culture, Karnataka with crown division (slip), and the wild native in Wuchuan from Guangdong.

They were divided evenly into 2 groups, one group was pruned once every three months to 30cm from the ground, and the other was kept intact (non-pruned). One year later, vetiver roots were dug up. It was found that the roots of the Wuchuan were only 110-130 cm long, significantly shorter than those of the other two cultivars, which were all between 193-238cm**.** *It seemed that cutting had no significant influence on root growth***.** Therefore the local wild cultivar is not suitable for erosion control and slope stabilization, while Sunshine and Karnataka (either tissue-cultured or slip produced) were quite ideal for the purpose. Moderate cutting is necessary in order to keep good landscape, furthermore cutting does not produce adverse influence on the growth of vetiver roots.

Root boxes

Effects of pruning on root length of various cultivars

Unpruned Karnataka slip and the wild native Wuchuan (L)

Pruned Karnataka tissue culture and the wild native Wuchuan (L)

Pruned Karnataka slip and Sunshine (L)

Unpruned Karnataka tissue culture and the wild native Wuchuan (L)

Pruned Karnataka tissue culture and Sunshine (L)

To determine the development of vetiver root under the tropical climate in Brazil, Dr. Aloísio Rodrigues Pereira - Director of Deflor Bioengenheria (deflor@deflor.com.br) (pers com.) conducted an experiment with a series of eight PVC tubes (0.5, 1.0, 1.5, 2.0, 2.5, 3.0 3.5, and 4.0m length) as shown below to evaluate Vetiver root development over a period from three months to at least two years. Watering was carried out daily from the top.

Due to the exposure to very hot tropical sunlight, the tubes were very hot, particularly the long tubes resulting in the death of roots deeper than 1.5m, after nine month of growth. Although the experiment could not achieve its original plan of two years, it has shown that the roots reached 1.5 m in length after nine months, indicating that a nine - month old vetiver plant would be able to improve slope stability in most applications. The experiment will be repeated using 40cm X 40cm wooden boxes in a future date.

Root 0.60m long 3 months old ***Six months old 1.10m*** ***Nine months old 1.50m***

Water use by Vetiver

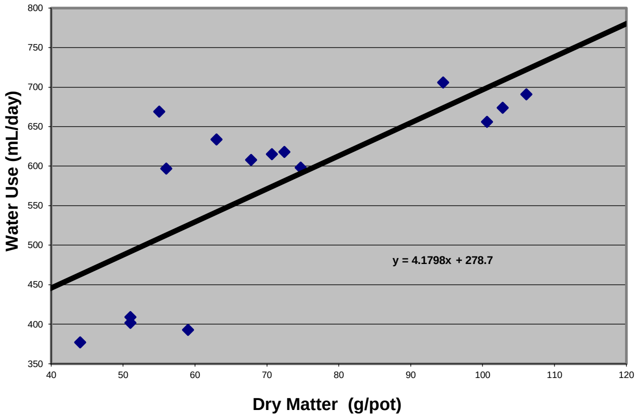
Water use or transpiration rate of Vetiver varies with:

- Growth stage: young (young plant or regrowth after cutting, mature and old).
- Climatic conditions: Temperature, rainfall, humidity, wind etc.
- Soil types; and
- Water availability

Under wetland conditions, vetiver had the highest water use rate as compared with other wetland plants such as *Iris pseudacorus, Typha spp*, *Schoenoplectus validus*, and *Phragmites australis*. At the average consumption rate of 600ml/day/pot over a period of 60 days, vetiver used 7.5 times more water than Typha spp (Cull *et al.* 2000).

To quantify the water use rate of vetiver, a glasshouse trial showed a good correlation between water use (soil moisture at field capacity) and dry matter yield. *From this correlation it was estimated that for 1kg of dry shoot biomass, vetiver would use 6.86L/day*. If the dry matter yield of a 12-week-old vetiver were 40.7t/ha, at the peak of its growth cycle, a hectare of vetiver would potentially use 279KL/ha/day.

Relationship between water use and dry matter (r = 0.7286)



Vetiver Root Research and Development under Field Conditions

Vetiver possesses a lacework root system that is abundant, complex, and extensive. The root system can reach 3-4 meters in the first year of planting and acquires up to a total length of 7 meters after 36 monthsin certain soil type. This feature support its survival under extreme drought conditions as it can utilize deep subsoil moisture. The root system prevents vetiver from dislodgement under high velocity water flows. However, its roots may not penetrate all the way into the groundwater table.

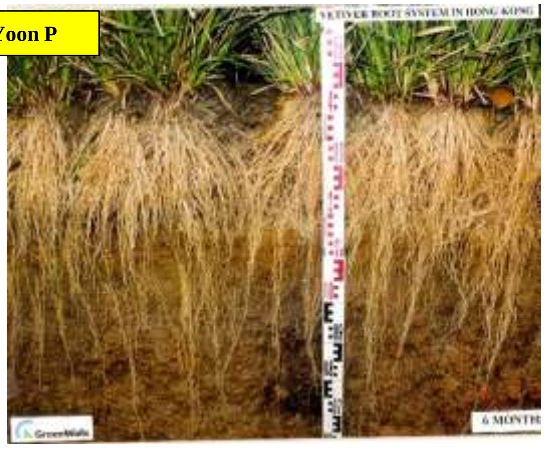
Massive, penetrating and deep root systems (3.3m in 12 months) (PC: TVNI Archive)**.**

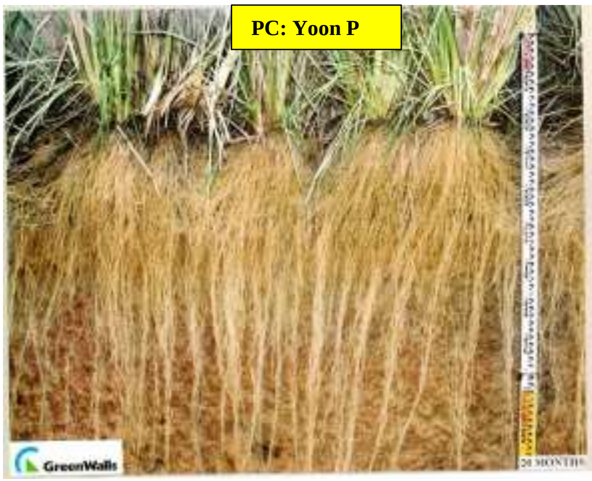
Generally at locations with high groundwater level, its root system may not be as long as in drier soil. Wang (2000) found that most roots of vetiver are very fine with an average average diameter of 0.66 mm (range from 0.2-1.7 mm). The vertical growth rate of vetiver root reaches a plateau of approximately 3cm per day at the soil temperature of 25°C. At the higher soil temperature, the root extension rate is higher but not statistically significant. At the lower soil temperature, 13-15°C, the underground root growth was still detected indicating that the vetiver is not dormant at this temperature. The horizontal spreading of lateral roots was in the range of 0.15 – 0.29 m with an average of 0.23 m. After 8 months after planting, vetiver produced 0.48 kg of dry roots per plant. The peculiarity of vetiver root system ensures high contact surfaces with soil particles and contaminants resulting in efficient phytoremediation of contaminated soils and wastewater.

Yoon (1989, 1992, 1994a, 1994b and 1995) of the Malaysian Rubber Research Institute a well-known and respected pioneer in the use of vetiver for bioengineering application in slope stabilization (cut and fill batters) described the vetiver root system as very deep and vigorous. With the use of quality planting materials and appropriate fertilizers rapid growth was obtained and a very immense network of roots formed in the soil. These were clearly demonstrated by plant excavations in Malaysia, Thailand, Vietnam and Spain. In Malaysia, 3-month old plants had massive root development in field planting. In the Mediterranean climate of Spain, the roots reached down to 2.1m, after 9 months of growth. After I4 months in the field, the roots reached 2.6m depth and this was despite having to go through a winter period when sub-zero temperatures killed the tops. In Vietnam, under very difficult soil conditions and six months of dry weather, one-year old plants produced roots of 1m depth. In Malaysia one year old plants had a massive root system of nearly 2m in one clump. This phenomenal root system even impressed the Prime Minister of Malaysia at the time.

PC: Yoon P

In Hong Kong, vetiver roots were between 1.2 and 1.5m deep after 6 months and over 3m after 20 months.





The root system of a 20-month old clump indMalaysia showed that the roots interlocked to form the immense underground web of vetiver roots. In the Royal Project in Thailand the massive root systems of many excavated plants were recorded. A root system of about 3m was harvested from a one year old plant.

Thus, the vetiver root system is unique in the depth, it can penetrate and the intensity and network it can form. The structural strength could be inferred from such observations. All those features will help the plant to anchor itself so firmly in the ground that it cannot be dislodged by the strongest floods.

Effects of fertilizer on Development of Vetiver Root System in the establishment phase

Lu Xiaoliang (2003) reported that in a soil pot experiment application of fertilizer had a distinct effect on the growth of vetiver root systems in the infertile soils during establishment period, but the development of the vetiver root system varied with different fertilizers. Chicken manure not only had a positive effect on the establishment of vetiver but also supplied the diversity of nutrients needed in shallow soil, resulting in vetiver growing a large number of new roots and branch roots in the shallow soil, and a subsequent increase of the surface area of the root system. So in this case applying chicken manure is better than applying other fertilizers to vetiver, and produces a better effect on water and soil conservation.

Four types of fertilizer were used:

1. Organic fertilizer: chicken manure (T2)

2. Compound fertilizer (N 25.7 P7.8 K20) (T3)
3. Organic and inorganic compound fertilizer (N10 P2.5 K6) (T4)
4. NaCl containing iodic salt
5. Five treatments were chosen in the experiment using the four fertilizer and Control (T1) with three replicates.

Root Growth of Vetiver in Different Treatments during Establishment

The experiment results show that fertilization during establishment not only increased the number of new roots and facilitated the anchoring of new roots but also helped to enhance the amount of root biomass and increase the transplant survival rate. There were not significant differences in new root biomass among different treatments. The total number of new vetiver new roots treated by chicken manure was highest in all treatments. On the other hand, KangBao fertilizer use resulted in the longest root growth.

Root growth of vetiver in different treatments

No.	Treatment	Root depth (cm)	No. of new root	Root biomass (g)
T1	CK	50.00c	10.0c	0.82b
T2	Chicken manure	66.67b	26.3a	1.76a
T3	BB fertilizer	75.33ab	14.3b	1.33a
T4	Kangbao fertilizer	81.33a	15.0b	1.70a
T5	Chicken manure+BB+NaCl	70.00ab	18.0b	1.19a

Each datum is the mean of three replicates. Values with the same letter in each column are not significantly different at $P<0.05$.

Shoot Growth of Vetiver in Different Treatments during Establishment

The results of the experiment indicated that fertilization had a distinct effect on the shoot growth of vetiver and the effects varied with different fertilizers. The results of variance analysis showed there were significant differences within the traits, such as the tiller height and shoot biomass, in different treatments. The effect of chicken manure was better than others. For plant height the effects of T2 and T5 were better than T1 and T4. Vetiver tillers and shoot biomass with chicken manure were much higher than others after two months.

Shoot growth of vetiver in different treatments

No.	Treatment	Plant height (m)	Tillers	Shoot biomass (g)
T1	CK	0.79c	3.1c	4.45c
T2	Chicken manure	1.55a	6.7a	13.76a
T3	BB fertilizer	1.34ab	4.3b	10.75ab
T4	Kangbao fertilizer	1.17b	4.7b	7.55b
T5	Chicken manure+ BB + NaCl	1.27ab	5.3b	9.98ab

Each datum is the mean of three replicates. Values with the same letter in each column are not significantly different at $P<0.05$.

Some root morphology traits of vetiver in different treatments during establishment

According to some root morphological traits of vetiver, fertilization had a distinct effect on the growth. All fertilizers showed positive effects on the root morphological traits of vetiver, such as new root total length, total surface area, average diameter and total volume. The number of new roots significantly increased in the chicken manure treatment in contrast to others.

It can be seen from four root morphology traits that new root total surface area is greater in T2 treatment than in T5, whereas new root average diameter of T2 treatment is greater than T3. It is obvious that the amount and diameter of new root increased by fertilizing with chicken manure, but there aren't any significant differences in the other two traits.

Some root morphological traits of vetiver in different treatments

No.	Treatment	Total length (cm)	Total surface area (cm^2)	Average diameter (mm)	Total volume (cm^3)
T1	CK	3138.42b	333.72c	14. 69c	7.23b
T2	Chicken manure	4889.16a	1152.18a	2.41a	22.78a
T3	BB fertilizer	4006.43a	999.86ab	14. 77b	21.22a
T4	Kangbao fertilizer	4882.55a	990.12ab	14. 91ab	16.36a
T5	Chicken manure + BB + NaCl	4048.67a	846.99b	14. 95ab	14.50a

Each datum is the mean of three replicates. Values with the same letter in each column are not significantly different at $P<0.05$.

It was concluded that chicken manure contains various organic nutrients with appropriate proportions and continually provides nutrition during establishment. The chicken manure mixed with the soil improves soil structure and enriches the poor soil. When vetiver was fertilized with chicken manure the growth of root and root biomasses had better development than with BB fertilizer.

According to experimental data the number of roots treated by chicken manure was highest among all treatments with the roots tending to be widely distribute in shallow soil with the root diameter appearing remarkably big and robust.

Shoot:Root Biomass Ratio of Vetiver.

The Shoot:Root Biomass ratio of vetiver generally varies between 1:1 and 1.2:1.0 in mature and intact plants. Therefore because external N availability generally favours root over shoot growth (Gregory 2006), adequate N supply is vital at planting to encourage establishment and earlier growth.

Transfer of plants from one environment to another environment causes changes in the pattern of assimilate distribution so that a new characteristic Root:Shoot ratio was established over a period of time. Yoon （1994) conducted a meticulous study on the *Effect of Shade on Vetiver Growth* over the period of 2 years, under both shade house and natural light conditions in a rubber plantation. Treatments included:

- Different shade intensity;
- Leaf area;
- Leaf weight;
- Tiller number;
- Plant weight;
- Shoot weight; and
- Root weight.

After 3-4 months growth, it was concluded that the root/shoot percentage stayed rather constant, indicating shading affects vetiver physiology and morphology profoundly.

Shoot and root growth under shade (80%) (L) and in open after 4 - month growth (R)

Root system architecture of Vetiver.

Another extraordinary attributes of vetiver is its root system mass in relation to its overall biomass, particularly the proportion of extremely fine roots (averaging 0.2mm). As stated above N supply strongly modulates root branching by directly regulating lateral root development and growth, adequate N supply is vital at planting to encourage establishment and earlier growth

Lifespan of Vetiver Roots.

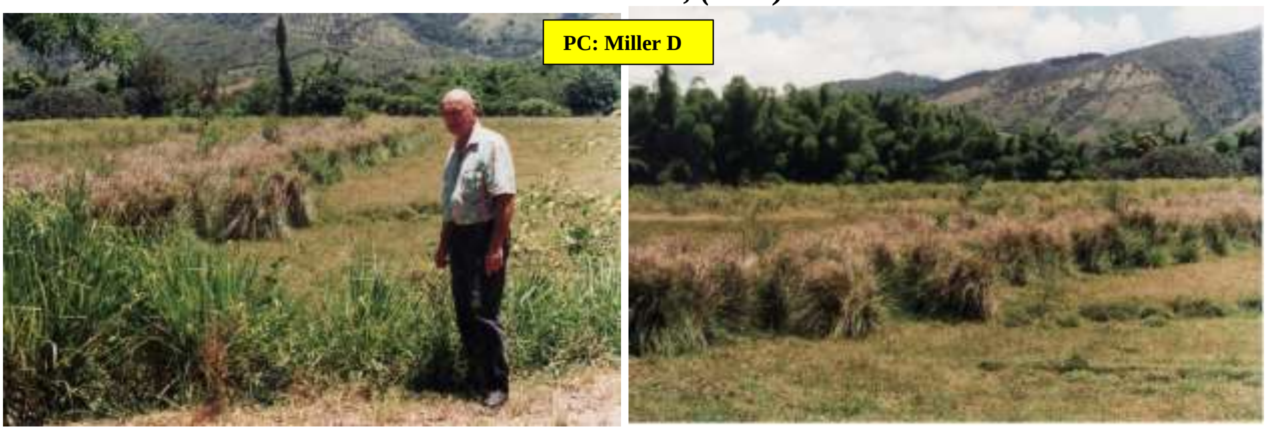
According to Eshel and Beekman **(**2013) in general, root lifespan varied widely. *The median lifespan of the finest roots can range from about 20 days at some times of the year in fast-growing trees and deciduous fruit crops to longer than a year in slow-growing forest trees. In a data set containing 190 studies in non-agricultural ecosystems, average root lifespans ranged from about 290 days in tropical ecosystems to about 3 years in high-latitude ecosystems, with considerable variation within each ecosystem. Studies based on tracer approaches have indicated that fine roots less than 2 mm in diameter may live considerably longer—averaging 4–8 years in some temperate forests. The estimate of lifespan, undoubtedly is affected by differences in environmental conditions and plant species*.

To date there has only been anecdotal information on the lifespan of vetiver roots, but it is generally accepted that the vetiver root system persists as long as its shoot growth is maintained. As life span and size of its root system play a very important role in both major applications of VST such as bioengineering and phytoremediation, measures that encourage root growth and persistence, even at the expense of its shoot growth (except for biomass production) should be encouraged. This includes the selection of late-flowering genotypes, whose resource would be diverted to root development instead of culms and flower heads.

In a review on the existence of Vetiver in the South West Pacific Islands, Don Miller (pers.com) discovered some long lived Vetiver plants in the region. Don is a New Zealander, who has lived and worked in the region in the last 30 years as a consultant to the New Zealand government on erosion and sediment control, reforestation and coral reef rehabilitation. During this time, he introduced vetiver to some islands and also discovered some old vetiver plants which had been introduced earlier, that could be more than 100 year old.

- ***New Caledonia*** was a French penal settlement and after the prisoners were released, but not allowed to return to mainland France, they were allocated plots of land to survive on and given thatching to roof their wattle and daub huts. Vetiver, possibly imported from Reunion Island, was grown in nurseries to supply the thatch and one of these nurseries still exists. *It is estimated to be over 100 years old*. Don Miller visited New Caledonia several times in 2000 and 2001 after seeing the extensive soil erosion caused there by nickel prospecting and mining. He met Georges Donskoff of the Department of Agriculture, their coffee growing expert, and was shown around by him to see points of interest. He was well versed in the value of Vetiver having researched its history in that country.

Georges Donskoff beside a Vetiver nursery once used for supplying thatch, in Grande Terre, New Caledonia, (2001).



- ***In Fiji*** Vetiver trials were done on sloping cane fields in the 1950s by John Greenfield and were proven to be very effective. In 1990 many of the Vetiver terraces were still evident three decades later, on a site established by Jonathon Subramanium, a former soil conservationist with the Colonial Sugar Refinery. He had planted vetiver hedges on the contour 25 years earlier and was pleased to show the height of sediment it had retained over that period of time. Many other examples of old vetiver erosion control hedges existed in the sugar growing areas of Fiji at that time and some were located again twenty years later, still growing in apparently the same locations. As these were already old hedges when photographed in 1990, judging by the height of retained soil, they could be between 40 and 60 years old.

Vetiver hedges on sloping cane land in 1990 (L) and 2009 (R)

50-60 year old Vetiver hedges on sloping cane land in 1990

- ***On Vanuatu's*** Aneityum Island, a project aimed to control the sediment discharge from many large eroding gullies that were severely affecting the fringing coral reefs of the island. On an initial assessment visit in 1995 a senior New Zealand forestry consultant recommended to start a vetiver nursery immediately, using plants already on the island. The plants were located along the walking track around the island and had apparently been established in about 1912 by a local chief, using his political prisoners. Remnants of those very early planting were also found at a number of locations around island coastal paths.

It seems highly likely that the Vetiver on Aneityum came from New Caledonia in the later 1800s or early 1900s. A dugout canoe with a simple crab claw rig would be able to sail directly across the prevailing wind to reach there and so could also return relatively easily.

The source of the plants used to establish the initial nursery in 1995 (L) and the track to a bluff stabilized with vetiver (R) in 1992.

Aerenchyma Formation and Functioning in Vetiver Root and Shoot

Anatomical research showed that vetiver has a very well developed aerenchyma network from the tip of its roots to the leaves.

The role of lysigenous intercellular spaces and aeration system was explained by Thammathaworn and Khnema (2011). They showed that leaves of all 11 cultivars of two Vetiver species (*C. zizanioides* and *C. nemoralis*) have large lysigenous intercellular spaces similar to water plants such as Typha spp. The lysigenous intercellular spaces appeared even in developing leaves but in smaller size and were larger in mature leaves. In principle the role of large lysigenous intercellular spaces within leaves of water plants is to eliminate hypoxia/anoxia effect in roots by promoting oxygen flow from leaves to roots. Aerenchyma at the cortex layer of roots is further evidence for sustaining oxygen to roots, which was also found in vetiver roots. In addition the parenchyma cells at the cortex layer on vetiver roots were lysed to aerenchyma and parenchyma cells in the inner vascular cylinder and intercellular spaces. Large lysigenous intercellular spaces in leaves and aerenchyma in roots could be evidence to confirm a great aeration system of vetiver as well as a potential for deep root penetration. These authors concluded that aerenchyma at the root cortex and air cavity at pith was strong evidences of aeration system from leaves to roots in vetiver. To avoid hypoxia/anoxia, large lysigenous intercellular spaces at laminas were a character of aquatic plants or long term flooding-tolerant plants by transporting O_2 from leaves to roots. Moreover, atmospheric O_2 could pass into vetiver via a pith cavity at culms, described by the theories called “Humidity-induced convection” and “Venturi-induced convection”.

Changes in anatomical features of Vetiver roots grown under dryland and wetland conditions.

In a study to select suitable species for a constructed wetland Liao et al (2002) showed that the cortex of *C. zizanioides* roots grown in a wetland, accumulated ergastic substance and had large air chambers than those grown on dryland. The thickness of aerenchyma tissue of roots grown in a dryland was significantly reduced ($P<0.01$) from 33.42µm to 24.57µm as compared to those grown in a wetland.

Changes in anatomical features of Vetiver leaves grown under dryland and wetland conditions

Liao et al (2003) showed that the leaf thickness of Vetiver was significantly reduced from 189.39µm in dry land situations to 147.31µm in wetland situations ($P<0.01$). Vetiver with thinner leaves is a method of adaptation to a wetland situation. The density of air chamber in wetland was higher than that in a dry land ($P<0.05$). They were 21.21/ cm^2 and 18.07/cm^2 respectively. This trait benefited the gas exchange of Vetiver under the wetland circumstance. He also found that the upper cortex of Vetiver leaves had fewer stomata than the lower side. The density of stomata on the surface of Vetiver leaves grown in wetland was larger than that in a dry land ($P<0.05$) and the area of the stomatic activity in wetland was larger than that in a dry land ($P<0.01$). ***This result agrees with previous research indicating that the area of stomatic activity tends to be smaller when the water supply reduced***.

Internal structure of vetiver leaf. **PC: Liao X**

Changes in anatomical features of Vetiver stems grown under dryland and wetland conditions

Liao et al (2003) found that the number of air chambers in the above ground tissue was closely related to the habitat. In the wetland circumstances, well-developed aerenchyma tissue was found and the number and the size of the air chamber were significantly increased ($P<0.01$) from dry land to wetland conditions to assure the adequate supply of air for the plant roots in a wetland. The author concluded that Vetiver a great potential to adapt itself to wetland conditions through aerenchyma and photosynthetic organs. In wetland, the thinner leaf, well-developed aerenchyma and increased stoma density was clearly shown in Vetiver. The anatomical features showed that Vetiver could adapt to the environmental change from dry land to wetland, and ***the transitional period is approximately three weeks***, by developing special aerenchyma in its roots to supply oxygen to the roots growing in the deep anaerobic soil or mud.

In addition, they found that the density of stomata and the area of the stomatic activity in a dryland was much lower than in wetland. ***This indicates that vetiver can lower its transpiration rate when the water supply is reduced.*** This finding explains how vetiver can tolerate a large volume of water under wetland conditions and yet can survive extreme drought.

Internal structure of vetiver leaf (L) and root (R)

Adventitious Root Formation (Eshel and Beekman, 2013)

Adventitious roots are usually formed on stems or leaves in response to external stimuli, e.g., mechanical wounding or abiotic stress. It has been shown that they are formed in cereals under flooding stress and can improve the flooding tolerance of the plants probably since they grow near the soil surface where some oxygen is present. To date there has been no information on the formation of adventitious root in Vetiver grass.

Effects of Soil Environment on Morphology and Physiology of Vetiver Roots.

Hamidifar et al (2018) investigated the morphological properties of the Vetiver grass root system including root area ratio (RAR), root diameter ratio (RDR), root diameter and density ratio (RDDR) and the root length density (RLD) in a clayey soil. The effects of morphological characteristics of the vetiver grass root system on the soil shear strength parameters including soil cohesion (C) and soil internal friction factor (φ) were studied. The results showed that RAR, RDDR and RLD decrease as the soil depth increases. Also, RDR was found to be correlated to the soil depth. The maximum RAR value was found to be 7.99% which is much higher than those reported by previous researchers for other plants used for soil protection (and conservation). The maximum RDR, RDDI and RLD values were 72.7, 4.4 and 0.1%, respectively. The results show that among the four root morphological traits studied, RAR and RLD are better correlated to C and φ, respectively. Furthermore, it is found that the plant density is not a significant parameter in the soil reinforcement in the range of densities studied here. Moreover, Vetiver grass roots can increase the soil cohesion and soil internal friction factor up to 119.6% and 81.96%, respectively. Based on regression analysis, some empirical equation were presented for calculation of the soil shear strength parameters as functions of the morphological characteristics of Vetiver grass root, which can be used for better management of natural waterways by means of a low-cost environmentally friendly technique.

Effects of Soil Environment on Morphology and Physiology of Plant Root in General as Compared with Vetiver Roots.

Effects of soil pH**:** (Gregory, 2006). *It has been known that the pH values in and around roots are not necessarily constant, and changes in pH are frequently observed. It is important to distinguish the changes in pH that occur during normal growth and development, from those that arise under adverse growth conditions. The growth-related pH changes that occur in the rhizosphere can have a profound effect on nutrient availability. Thus, the acidification of the rhizosphere and root apoplast that accompanies growth on ammonium, or to a lesser extent nitrogen fixation, favours the uptake of a range of nutrients, including both macronutrients, such as phosphate and minor nutrients, such as boron, iron, and manganese.*

Root pH values also respond to several environmental stresses, including flooding, drought, salt stress, and unfavourable soil pH values. The effect of oxygen deprivation on flooding-intolerant plants is particularly severe, since it causes an acidification of the cytoplasm that can lead to cell death.

pH tolerance of Vetiver: One of the most extraordinary attributes of vetiver is its tolerance to pH between 3.5 and 11.5 and in one case of a very sodic saline soil in India to pH 14.0. Very few food and industrial crop plants have such a wide range of pH tolerance. As mentioned above the pH changes that occur in the rhizosphere can have a profound effect on nutrient availability including both macronutrients, such as phosphate and ammonium, and micro nutrients, such as boron, iron, and manganese. To overcome these effects, fertilising is a must when vetiver is planted on poor and highly contaminated water and land.

Effects of nitrogen supply on root system architecture.

Both the size and the architecture of the root system are strongly dependent on external N availability and on internal N status of the plant. Root system architecture has been reported to be particularly affected by the NO_3 level in the growing medium, which has a limited impact on growth of the main root of species with a taproot system, but strongly modulates root branching by directly regulating lateral root development and growth. The size and architecture of the root system determine the total volume of soil explored by the plant and the size and location of the exchange surface between the soil and the plant.

Root system architecture of Vetiver.

Another extraordinary attribute of vetiver is its root system biomass in relation to its overall biomass, particularly the proportion of extremely fine roots (averaging 0.2mm). As stated above nitrate supply strongly modulates root branching by directly regulating lateral root development and growth, adequate N supply is vital at planting to encourage establishment and earlier growth.

Vetiver root crown or corm. The junction between vetiver plant shoot and root is commonly known as the CROWN but in botanical terms it is known as the CORM. One of the peculiar and special characteristics of vetiver is the formation of its unusual and prominent corm, a very dense mass of plant tissue containing the growing buds of both roots and shoots. On living plants the corm is obscured by the thick cover of shoots, it is exposed only after burning or severe trimming, showing dead old shoots. The following photos show a two year old corm growing in Australia.

Top of an old Vetiver corm

Side of Vetiver corm

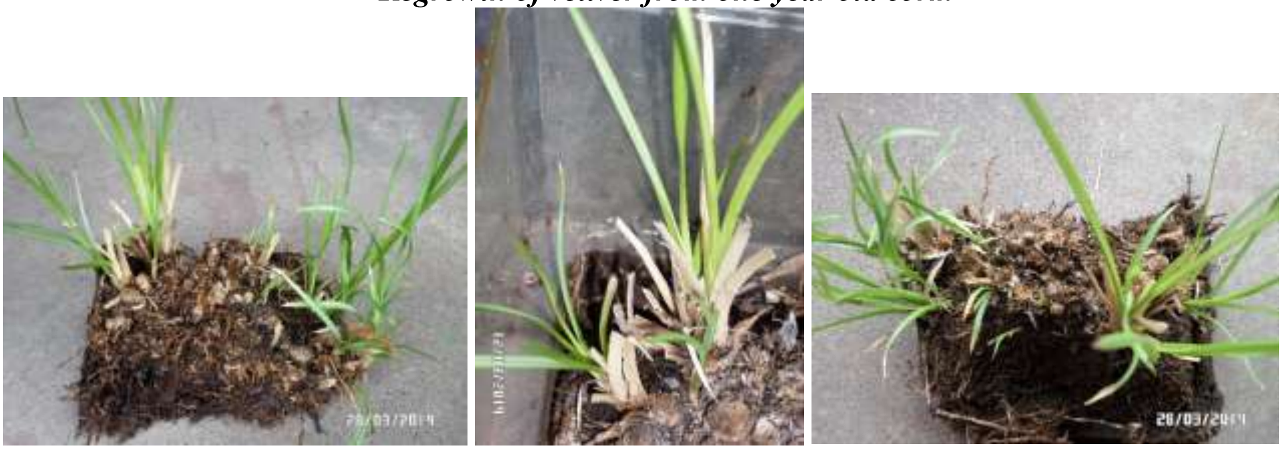
Bottom of Vetiver corm

Side of Vetiver corm with roots

Root development in Vetiver corm

In an old corm, the underground component is a very *compacted and solid root mass*. This 40cm deep corm was from a 4 year old plant.

Regrowth of vetiver from one year old corm



Effects of Nitrogen supply on the Shoot:Root Biomass ratio.

A decrease in the external N availability generally favours root growth over shoot growth, resulting in a decrease of the shoot:root biomass ratio, that can be quite dramatic in N-starved compared to well-fed plants. This does not necessarily mean that root biomass is increased in absolute terms by N deprivation. Root growth is often actually decreased by N-limitation, but to a much lesser extent than shoot growth. This very common adaptive response (seen in many plant species and also for other nutrients than N) aims at favouring the growth of the organ (the roots in this case) in charge of the acquisition of the element that is most limiting for growth (N in this case). However, little is known concerning the mechanisms responsible for it (Gregory, 2006).

***Shoot:Root Biomass ratio of Vetiver*.** The Shoot:Root Biomass ratio of vetiver generally varies between 1:1 and 1.2:1.0 in mature and intact plants. Therefore if external N availability generally favours root growth over shoot growth, adequate N supply is vital at planting to encourage establishment and earlier growth.

Soil Temperature Variation.

Temperature is one of the major external inputs plants are exposed to from germination to senescence. Temperature variation has a strong influence on the establishment and function of plant root systems; furthermore, temperature can affect overall plant productivity. Most variations in soil temperature result from temperature variations at the surface. The main components of the balance between incoming and outgoing energy at the soil surface are daytime heating from the sun and night-time cooling. Thus, heat is usually stored in the soil during daytime and released at night. The same mechanisms cause heat storage during spring and summer and release during autumn and winter. Soil temperature follows surface temperature with a time lag and progressive damping with depth.

Decrease of temperature with depth is typically governed by a damping depth, at depth of 5cm, the temperature variations are reduced to less than 1% of the fluctuation at the surface, thus being almost damped out. Diurnal fluctuations reach far less deeply into the soil than annual variations; due to the dependence on oscillation period, the damping depth for diurnal fluctuations is typically more than one order of magnitude smaller than for annual variations. (Gregory, 2006).

Root Responses to Low Temperature.

If the temperature of the soil differs significantly from the species-specific optimum temperature then the structure and function of the root system can be altered. This might even result in a total loss of function or at least severe damage of the root system. The negative effects of root zone chilling can appear in a wide range of temperatures, from just above 0°C to higher temperatures of up to 20°C, depending on species-specific adaptation.

The most obvious morphological response to temperatures lower than the species-specific optimum is that the plants produce smaller root systems and roots of thinner diameters. For example, maize roots showed a 40% reduction of growth rate at root zone temperatures of 16°C compared to 25°C. The biomass was reduced especially in the basal parts of the root systems as well as in the lateral roots. Furthermore, root systems grown at low temperatures are less branched, leading to more compacted root systems and therefore to a reduction in the soil volume explored by the root system.

The most significant physiological response of plants subjected to chilling events is that the uptake of water and nutrients is reduced. This results from a decreased root hydraulic conductivity and a malfunction of the stomata in the leaves and appears to be the most important difference between chilling-sensitive and chilling-tolerant plant species.

Finally, chilling events also affect plant nutrition. For example, root zone chilling decreased the $NH4^+$ *and* NO_3*– absorption by plant roots, affecting NO3– more strongly than NH4+ uptake. Hence, low soil temperatures cause a preference for* $NH4^+$ *over* NO_3*– nutrition. However, the root zone temperature is not the only factor governing* NO_3*– uptake during chilling. Furthermore, the uptake of other nutrients, such as boron, can also be suppressed by root chilling and thus in a second step result in severe deficiency effects*. (Gregory, 2006).

Vetiver Root Responses to Low Temperature

Although Vetiver is a tropical species, it is extremely tolerant to low temperature. In Australia, Vetiver growth was not affected by severe frost at –14°C ground temperature, it survived for a short period at –22°C (-8°F) in northern China. In Georgia (USA), Vetiver survived in soil temperature of -10°C but not at –15°C.

Adams et al (2003) reported that vetiver is tolerant to temperatures from -15^0 C (5^{0}F) to 55^0 C (131^0 F), depending on growing region. Under frosty conditions, shoots become dormant and purple, or even die, but the underground growing points in the corm survive and can regrow quickly when the conditions improve. The plant, however, cannot reliably withstand freezing conditions. Vetiver growth is definitely limited by cold temperatures.

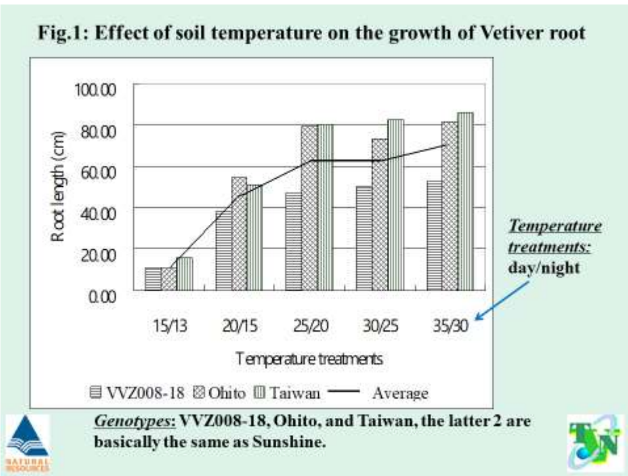
Australia

Jordan

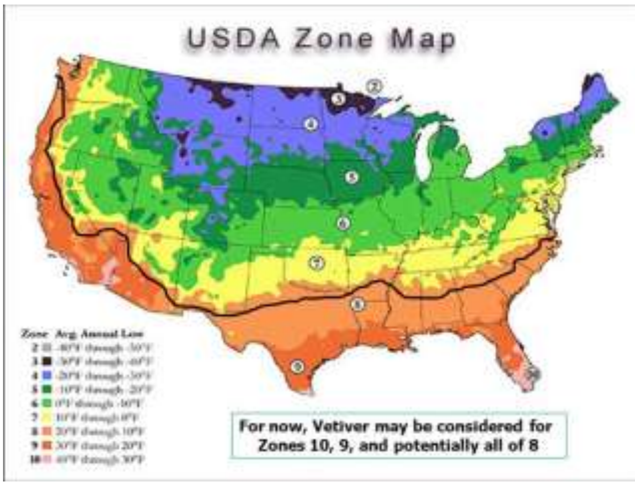
Chile

The growth characteristics of vetiver were studied in a phytotron by Wang (2000) of the National Taiwan University. On average, at temperatures above 25°C, daily root growth of 3 cm was recorded. Although very little shoot growth occurred at the soil temperature range of 15°C (day) and at 13°C (night), root growth continued at the rate of 12.6cm/day, indicating that Vetiver grass was not dormant at this temperature. Extrapolation suggested that root dormancy occurred at about 5°C. As its growing buds are on the corm, *Vetiver dies when the corm or the ground is frozen.*As soil is a very good insulator, its underground temperature is often above surface or air temperature when the surface freezes, vetiver root will continue to grow under snow or frosty conditions.

Effect of day and night temperature on vetiver root



Potential climatic zone for vetiver in the US



Survival of vetiver under cold temperature in Australia

Survival of vetiver under cold temperature in USA

In February 2021, an unexpected extremely cold weather period hit the USA, and surprisingly a very severe cold front occurred in Texas, including the subtropical region in the south near the Gulf of Mexico. The following table shows air temperature ranges of a number of phytoremediation sites operated by Leachate Specialist Co. in Texas and other Gulf States (E. Wiediger per. com)

Texas 2021 Winter Freeze - Air Temperature (°C)													
Day		Wed	Thur	Fri	Sat	Sun	Mon	Tues	Wed	Thur	Fri	Sat	Sun
February Date		10	11	12	13	14	15	16	17	18	19	20	21
College Station TX	Max	8.9	2.8	0.0	0.6	-0.6	-6.7	-2.8	1.1	1.1	5.0	15.6	22.2
	Ave	5.3	0.9	-0.6	-1.2	-4.3	-9.9	-7.0	-0.4	-1.6	-0.7	6.7	14.8
	Min	3.3	0.0	-1.1	-2.2	-8.3	-12.2	-14.4	-2.8	-3.3	-6.1	-1.7	6.7
	SNOW					10 cm							
Fort Bend TX	Max	17.8	6.7	2.8	2.8	1.7	-5.0	1.1	2.8	3.3	9.4	16.7	21.7
	Ave	12.8	3.9	1.7	1.7	0.0	-7.2	-3.9	1.7	1.1	2.8	8.9	15.6
	Min	6.7	2.8	1.1	0.6	-5.0	-10.0	-10.0	0.0	-1.1	2.2	-1.1	6.7
	SNOW					0.25 cm							
Sulphur LA	Max	25.6	21.1	5.6	7.8	4.4	-0.6	2.2	3.3	5.0	10.6	14.4	20.0
	Ave	20.0	11.7	4.0	3.8	2.3	-2.7	-3.8	1.6	2.2	3.5	9.1	12.9
	Min	15.6	5.0	2.2	1.1	-0.6	-6.7	-8.9	0.0	1.1	-1.1	3.3	3.3
Biloxi MS	Max	18.9	20.0	9.4	7.2	8.3	5.6	4.4	10.6	8.9	13.3	13.9	17.8
	Ave	17.4	17.8	7.6	5.5	5.2	1.1	-1.9	4.7	6.2	4.9	5.3	11.2
	Min	15.6	12.2	5.6	3.9	2.8	-4.4	-5.6	-3.3	2.8	0.6	-2.2	1.1

Although the minimum temperature was very low (- 14.4^{0}C), it was over a very short time, so the soil was not frozen, hence vetiver survival was not affected as shown below.

Snow cover on lakes in the area

Root Responses to High Temperature

Global warming will affect soil temperature and consequently also plant growth. *Generally, an increase in soil temperature is accompanied by an increase in root growth, but when root systems are exposed to temperatures that are higher than the optimum, plant roots generally show decreased elongation rates as a first response. Generally, the size of the root system is reduced by temperatures above the optimum. This size reduction is often due to a decreased number and length of lateral roots. The duration of exposure to high temperatures also appears to have an impact on the growth and development of root systems.* (Gregory, 2006).

Vetiver Root Responses to High Temperature.

An anecdotal report indicates that vetiver could survive at air temperatures up to 55^0 C (131^0 F) on a tailings dam. These powdery black tailings are the product of a very deep gold mine near Johannesburg in South Africa. Vetiver was planted there to control dust from the powdery tailings.

Vetiver is also very tolerant to fire, both wildfire or control burns used to control pest (insect and rodent) when planted for erosion control. The following photos show the effect of fire on vetiver and its regrowth after a control burn and wild fire in Australia.

On individual plant clump in Australia

On hedgerows in Australia

Although soil is a very good insulator, the extreme heat did not affect vetiver roots, but it was hot enough to burn all ground vegetation and even young trees.

Root Elongation in the Soil Matrix.

The bulk of water and nutrient uptake along root axes occurs in mature root tissue (mainly via the subtending lateral roots) some distance from the root; however, the placement of this mature tissue depends on the prior growth of the root tip through this region of the soil. Furthermore, uptake of water and nutrients diminishes with root age, so an effective root system depends on the continued generation of new root tissue. The elongation of roots depends on the balance between cell proliferation and differentiation and, in expanding cells, on the balance between cell wall loosening and wall stiffening. The key processes that determine the production of cells are the rate of cell division and the size of the meristem. A complex interplay of factors controls these processes, all occurring within a brief frame of time and space. When cell elongation ceases, the tissue is firmly fixed at a point in the soil matrix: the flux of water and nutrients and associations with rhizosphere microflora all occur at this local spot until the root dies. (Gregory, 2006).

Vetiver Root Growth.

Under phytotron conditions, on average, at temperature above the 25°C, daily root growth of 3 cm was recorded and at the soil temperature range of 15°C (day) and at 13°C (night), root growth continued at the rate of 1.26cm/day, indicating that Vetiver grass was not dormant at this temperature. (Wang, 2005)

RESPONSES OF PLANT ROOTS AND VETIVER ROOTS TO STRESS

Effects of Flooding on Plant Roots (Gregory, 2006).

The adaptation mechanisms of plants to the waterlogged environment include changes in the root morphology and anatomy as well as in the primary metabolism of root cells. During their evolution, some plant species have adapted well to soil flooding and this adaptation has been achieved by two different means: either by increasing the air spaces inside the roots thus allowing diffusion of gases or by changing their respiratory activity to reduce the consumption of oxygen while maintaining the supply of ATP for vital cellular maintenance. This review will concentrate on the development of air spaces, i.e., aerenchyma in roots and in particular, whether the aerenchyma formation is induced or constitutive.

In nature oxygen deprivation of the root system occurs during flooding season causing plant death or significant loss of biomass and yield in agriculturally important plants. The physiological response of the root, an organ that is most affected by flooding, has a major impact on plant survival under oxygen deprivation.

Uptake of water and Minerals under Flooding

Water is essential for virtually every metabolic reaction in living organisms. In higher plants, the root system effectively performs uptake of water and nutrients from the environment, redistributes it within the root (radial transport), and transports it to the shoot via xylem vessels (axial transport). The efficiency of water absorption by roots depends also on root anatomy and morphology of root system. A number of environmental factors such as oxygen deprivation, drought, nutrient deficiency, chilling, and high concentration of toxic ions can reduce the water absorption of roots

Mineral Uptake and Ion transport under Oxygen Shortage.

Roots and the rhizosphere in the soil have a complex interaction: The soil provides the plant roots with vital nutrients and water, while the roots affect the rhizosphere with the diffusion of oxygen and carbon dioxide and with the many organic compounds leaking from the roots. Flooding of the soil disturbs this interaction in many ways and often leads to root death especially in flooding-intolerant plants.

Waterlogging leads to multiple metabolic changes that affect nutrient uptake in a negative way through restricted energy supply for root growth and for transmembrane transport activities and through decreased transpiration and inhibited water transport. Nutrient uptake rate in roots is regulated by many factors: by physical and chemical processes in the root and in the surrounding soil, by soil microorganisms, by the root system architecture, and by root metabolism. Changes in the rhizosphere imposed by waterlogging also have an impact on nutrient uptake.

Cessation of root growth and decline in nutrient uptake are immediate responses to the energy saving strategy of the flooded plant. In waterlogged soils, inhibition of root growth may be also caused by secondary microbial metabolites—monocarboxylic and phenolic acids. (Gregory, 2006).

Physiological adaptations to flooding (low O_2).

Even though anatomical adaptations in roots and rhizomes play an important role in the survival of plants and tissues during low-oxygen conditions in both wetland and dryland plant species, there are instances such as total submergence, where anatomical adaptations are not enough. Still, we may find differences in tolerance between plant species. Such differences in sensitivity to flooding or anoxia are due to biochemical, not anatomical, features.

The three main metabolic processes that related to tolerance or intolerance of low-oxygen conditions:

- *the consumption of carbohydrates: Carbohydrate metabolism is strongly affected by low O_2*
- *the action of plant mitochondria and cellular energetics: Plant root cell mitochondria play a central role in the energy metabolism of root cell.*
- *the differences in accumulation of anoxia-specific metabolites. In waterlogged soils, roots also encounter an environment with low redox potential where reduced substances such as Mn^{2+}, Fe^{2+}, H2S, and nitrite can build up* (Gregory, 2006).

Physiological adaptations of Vetiver under flooding (low O_2).

Xia et al (2003) conducted a series of submergence tests under different conditons. They pointed out that at that time, there were still no ideal bio-engineering or economical conventional hard measures available to protect or stabilize the inner slopes of rivers, reservoirs, and lakes, as plants established on the inner slopes are almost all drowned by the elevated water level in the rainy season. Therefore to find ideal plants, the key is to screen for strongly tolerant plant species to submergence in order to effectively stabilize and vegetate the “wet” slopes.

A comparative study was conducted on the tolerance of eight grasses to submergence, up to 100 days. The eight selected grasses were vetiver grass (*Chrysopogon zizanioides* Nash), Bahia grass (*Paspalum notatum* Flugge), Aciculate Chrysopogon (*Chrysopogon aciculatus* Trin.), Bermuda grass (*Cynodon dactylon* Pars.), common Centipedegrass (*Eremochloa ophiuroides* Hack.), St. Augustine (*Stenotaphrum secundatum* Kuntze.), Carpet Grass (*Axonopus compressus* Beauv.), and Sour Paspalum (*Paspalum conjugatum* Bergius). They were all excellent plant species for soil and water conservation in southern China. The tested plants were raised in pots first and then put into a cement tank filled with water to determine their tolerance to total submergence. After three years of observation, it was found that vetiver and Bermuda grass could tolerate the longest time of submergence, at least up to 100 days and it was probably longer than that. Bahia grass ranked second, up to 60–70 days, followed by Carpet Grass, up to 32–40 days and then Aciculate Chrysopogon and Sour Paspalum, up to 25–32 days; St. Augustine was marginal, up to 18–32 days and the poorest species with very low resistance to submergence was Centipede Grass, only 7–10 days. Algae seemed to be an environmental factor influencing the tolerance of plants to submergence, inferring that muddy or polluted water kills plants more easily than clear water. The tests were conducted in clean and clear rain water, so the plants were exposed to sunlight during day- time hours.

Testing tank

In the first test, when Vetiver and six flood tolerant grasses were completely submerged for 70 days, only two out of three pots of Vetiver and Bermuda grass (*Cynodon dactylon*) survived. All other plants died.

In the second test, when Vetiver and Bermuda grass were submerged for 80 days, all three vetiver pots survived. Although they looked dead initially, after nearly one month of rejuvenation, they become normal again.

Plants before 80 day test

Looking dead initially after 80 days

A third test was conducted for 100 days of submergence. After the testing period, the plants look better than those after the 80 day test. It was affirmed that they all survived.

Plants before 100 day test

Plants after 100 day test

As vetiver fully recovered after 100 days of total submergence, the authors concluded that *vetiver roots could survive at least for 100 days in clean and clear water under normal daylight conditions*. They also noted that Vetiver was more tolerant than Bermuda grass.

The experiment will continue with the fifteenth submergence begin on 14 May 2003 and will finish on 14 September, lasting 120 days. The result will determine if vetiver or Bermuda would survive the 120-day submergence. Longer submergence trial will continue until all tested plants are killed to thoroughly ascertain the tolerance of vetiver and Bermuda grass to complete sub mergence. It is very important to point out that the survival duration mentioned above was under clear water with access to sunlight during day time.

These results are supported by field experience in South Africa and Venezuela, vetiver survived and flourished under prolonged inundation by excessive rain and floods. Rafael Luque (pers.com) also observed that in Venezuela as long as some shoots remain above water, vetiver can survive indefinitely under a semi submerged state. This indicates that its root system does not only survive but continues to grow under an extremely low oxygen supply delivered through the aerenchyma network mentioned before.

Vetiver growth was not affected after 7 months of flooding in Maracay region of Venezuela

In Portugal, Vetiver survived and regrew after being completely submerged by the lake for almost one month. (Schipper pers.com)

In China, Vetiver grew well when not completely submerged on a river bank in Guangdong province.

However in deep water and/or under muddy conditions, where light penetration is poor the survival of vetiver was severely affected. In a project to stabilize a section of the bank of the Mekong river 30km north of Phnom-Penh, Cambodia, vetiver died after 3 months under 10-15m of muddy water. This can be attributed to the lack of sunlight at that depth under muddy conditions.

The following photos show how vetiver was affected under deep and muddy water.

Erosion on the Mekong bank

Site preparation

Vetiver planting for erosion control

Two months after planting

Eight months after planting

Before flooding

10-15m deep flooding

Three months after flooding showing dead vetiver

Dead vetiver on the on the deepest part of the bank

On the section above the dead vetiver plants, where it was inundated but some part of the shoot remained above the muddy water, vetiver was badly affected but survived and resumed good growth later.

Vetiver recovered after partial flooding ***Two months after flooding*** ***Eight months after flooding***

In a report entitled Comparison of Current Status of protective planting of species in the draw down zone of the Danjiang reservoir area, Henan Province, China, Xia Hanping of the South China Institute of Botany, reported that for water storage of the middle reaches of the South-to-North Water Transfer Project, the area of the Danjiang reservoir, Hechuan County of Henan Province, has for many years had a low water level of between 150 and 157 meters. The Government had invested in protecting the reservoir area by growing trees and grasses in the draw down area (fluctuation zone). Since September 2017, the water level began to rise and in March 2018 it reached 167 meters - the highest water level in history.

The level then retreated to 160 meters by the end of April 2018. The water level fell by 7 meters in these six months. *Most of the trees and grasses planted in the draw down area were submerged and died, only the vetiver grass survived*. Jiang Jianggong, chief of the Yangtze River Committee, after seeing the scene said *"I have worked in the Yangtze River Valley for 30 years. I haven't seen a plant in the upper and lower reaches that can survive more than 3 months flooding, this vetiver grass has survived for six months under flooding water"*.

Vetiver before flooding at Danjiang reservoir, Henan Province, China

Vetiver after 6 month flooding at Danjiang reservoir, Henan province, China

Trees and grasses drowned around the reservoir area after six month of flooding

Effects of Drought on Plant Roots (Gregory, 2006)

The rhizosphere environment is neither static nor homogeneous; root growth, therefore, must respond in dynamic ways to local challenges, such as lack of soil moisture. Root growth is determined by the balance between cell division and differentiation and between cell wall loosening and wall tightening.

When there is little rainfall and upper soil layers are depleted of moisture, plants rely on the ability of root systems to proliferate throughout the soil profile to extract water. The patterns of root growth and soil drying are spatially heterogeneous, such that regions of the soil are dried unevenly, leaving patches or layers of moisture not yet explored by roots. In some circumstances, to reach moisture, roots must pass through soil that is already dry because other roots—perhaps of neighboring plants—have previously extracted the soil water. In other circumstances, seed sown in dry soil germinates when surface layers are wetted, but seedling roots must penetrate through dry soil to find moisture in deeper layers. In such situations, roots must be able to continue growing through a soil matrix that is often at water potentials that may be inhibitory to growth. For example, under drought conditions, the nodal root axes of maize (which are produced from the stem nodes) are challenged to grow into and through surface soil that may have become very dry. In these circumstances, the water for continued root growth can be supplied to the root tip via the phloem. Similarly, the primary root of several crop species is able to continue growing at low soil moisture that completely inhibits shoot growth. These findings indicate some form of internal regulation within the root growth zone that allows the maintenance of cell elongation under these conditions.

The mechanisms underlying primary root growth maintenance at low soil moisture have been studied extensively, but large gaps remain in our understanding of how the growth of roots in response to low soil moisture is controlled and how the molecular physiology that regulates root growth and development at high soil moisture is modulated to enable continuation of growth when *water is limiting. In this review, some brief references are made to maize (Zea mays L.), which is closely related o Vetiver, on its important physiological features involved in root growth maintenance at low soil moisture. The growth and function of the entire root system also depend on the initiation and growth of lateral roots and root hairs.*

Perception of Low Soil Moisture and Intracellular Signaling

Another important element to consider in root growth in dry soil is how low soil moisture is sensed by the root and how this signal is transduced throughout the tissue. In this process, the plasma membrane and Ca^{2+} feature as key players. A decrease in soil moisture induces changes in root cell membrane potential that are maintained under steady-state conditions, long after the initial stimulus disappears. In addition to Ca^{2+}, it is clear that cell wall pH and its regulation play an important role in wall extensibility through direct effects on acidification of the wall matrix and on wall-loosening (Gregory, 2006).

Root Growth in Drying Soil in the Field

In comparison with the amount of information on seedling root growth in laboratory experiments, there is considerably less information on how root growth responds to low soil moisture in the field. Due to the complexities of the field environment, experimentation is challenging, and extrapolation of laboratory results to the field must be done with caution. For instance, many soils also become hard as they dry; therefore, roots encounter both water stress and penetration resistance. There is some evidence to suggest that root behavior observed at the seedling stage under controlled conditions can indicate growth of mature root systems in the field (Gregory, 2006).

Vetiver Root Development and Depth

As mentioned earlier Vetiver has an extensive and deep root system as a result of selection over centuries for essential oil production in India.

Massive root grown in a bag container for essential oil production in Thailand

Massive, penetrating and deep root systems (3.3m in 12 months)

Thailand ***Central China*** ***Southern China*** ***Alabama, USA***

Under artificial inducement it can be in excess of 11m as in Guatemala. At that depth, oxygen level in the soil is extremely low, to sustain this underground growth, vetiver has to rely on the oxygen supply from the shoot through its phenomenal network of aerenchyma mentioned above.

Vietnam ***DR Congo*** ***China*** ***Guatemala***

Physiological adaptations of Vetiver under drought

Under drought conditions, vetiver extends its roots until they reach the moist subsoil and beyond to the water table. Significant physiological adaptations of its root must occur during this water stress period. The main inducement for its roots to reach the depths shown above is to limit water supply from soil surface and increase soil moisture from the bottom of the soil container (pipe) used to grow these demonstration plants.

Effects of Salinity on Root Growth (Gregory, 2006)

Salinity inhibits growth and development of most plants. Growth, morphology, anatomy, and physiology of roots are affected by salinity. Since root growth is usually less sensitive to salt stress *than shoot growth, an increased root/shoot ratio is often observed when plants are subjected to saline conditions.*

The restriction of root growth by salinity, which reduces the soil volume that can be explored by the root and hence the availability and uptake of water and essential minerals, diminishes the supply of nutrients to the shoot which may contribute to growth reduction. The increase in root/shoot ratio reduces the demand for element supply to the shoot and thereby has a potential to increase the ability of the root to supply those elements and present an adaptive advantage. A potentially negative effect of such a change is the decreased capacity of the shoot to supply assimilates to the root and the growing tissues, which is likely to affect plant development and survival particularly under long-term salinization.

The underlying mechanisms involved in the inhibition processes of root growth under salinity are not clearly established. Saline solutions impose on roots two types of stresses:

- ***_Osmotic stress** resulting from lowered water potential in the root-growing medium and ionic stress induced by changes in concentrations of specific ions in the root medium and inside the growing tissue.*
- ***_A secondary induced oxidative stress**_affects the root cells._The information available, concerning root responses to salinity and the mechanisms involved is limited compared to our current knowledge on shoot responses.*

More specifically salt stress imposes inhibition of root elongation in most plant species. The mechanism of root stress response and tolerance is complex and integrates numerous physiological aspects of regulation, metabolism, and biophysics. Salinity is conventionally considered to reduce root growth by osmotic effects, specific ion effects, and/or oxidative stress. Therefore, investigations have centered on regulation of Na^+ uptake and compartmentation, interaction between Na and acquisition of major mineral nutrients such as K^+ and Ca^{2+} and turgor regulation. It is yet unknown whether Na^+, Cl^-, or other ions are the predominant growth-limiting factors for root growth, and the mechanism for sensing the change in Na^+ concentration is also unknown.

Root Growth under Salt Stress

As roots are in direct contact with the soil solution, they are first to encounter the saline medium and are potentially the first site of damage or line of defense under salt stress. The shape and size of the root system are determined by extension growth of individual root tips as well as by the rate and location of lateral root development. Salinity affects these root developmental processes differently. While root elongation of many plants is severely inhibited by high concentrations of NaCl in the medium, lateral root formation is less affected or might even be stimulated by the stress.

The stress-induced inhibition of root elongation may result from effects on the rate of cell division, rate of cell expansion, duration of cell growth, or orientation of cell expansion. In principle, any of these processes could underlie the root's growth response to salinity. Information about salt stress effect on root-cell expansion is available from three types of indirect studies and from growth kinematic analysis:

- *Salinity inhibits the elongation rate of root segments in the elongation*
- *Swift changes in root growth rate following salinization or removal of salt. Such changes are due to cell expansion and not cell division because newly dividing cells do not contribute to organ elongation before several hours or even days have passed*
- *Cell length measurements suggested that cell growth is inhibited. Mature root-cell size was reduced under salt stress, and salinization resulted in shorter roots.*

Similar to cell expansion, only little evidence of salt effect on cell division rate has been reported. Salt stress inhibited cell number increase in roots and apparent root-cell production rate.

In addition to the effect on cellular expansion rate, salinity might also change the growth orientation (growth anisotropy) of cells. Salinization was shown to cause thicker but shorter roots.

Salinity-induced changes in root morphology may be involved in the root response to salinity and extent of root and plant salt tolerance. For example, some halophyte have a thickened primary root that acts as a sink for Na^+ sequestration, thereby preventing its accumulation in the lateral roots and young leaves, and glycophytes can adjust root growth and root system architecture in response to salinity to avoid locally high salt concentration .

Salinity Sensing and Signaling

In response to exposure of the root to salinity, the root needs to adjust to maintain its function under the altered conditions, as well as to signal to the shoot the occurrence of stress. Plants responds rapidly and specifically to increase in Na+ salinity; however, the mechanism for sensing the change in Na+ concentration is unknown.

Following salinization of the growth medium, a rapid reduction of leaf elongation rate occurs. The mechanisms involved in this swift growth restriction are not clearly known, but are likely to be regulated by long-distance signaling from the root to the shoot by hormones or their precursors, since the reduction of leaf growth rate under salinity is not limited by carbohydrate availability or water status.

Possible Factors affecting root Growth under Salt Stress

It is generally accepted that salt stress may reduce root growth by osmotic effects, by specific ion effects (toxicity, deficiency, or ion imbalance), and/or by induction of oxidative stress. The degree at which these factors affect root growth depends on the inherent plant sensitivity, the duration of exposure to the stress, the concentration and type of salts involved, and environmental variables (such as physical and chemical properties of the growing medium, air humidity and composition, soil and air temperature, and light intensity). The followings are important factors:

- *Osmotic Effects. High salt concentrations in the root media result in low soil water potentials at the root zone and eventually may lead to a water deficit. Roots that are exposed to a sudden event of salinity or water stress lose their turgor and respond in an immediate cessation of elongation. Recovery of elongation can occur without full recovery of the turgor in the growing cells of the root, as long as the turgor exceeds the wall "yield threshold".*
- *Specific Ion Effects. Generally, macronutrient concentration in roots is reduced under salinity, whereas some micronutrient concentration is increased. Salinity-induced toxicity or deficiency of one or more ions may cause growth reduction under stress. The term specific means that the ion under consideration causes an additional depression of growth beyond what could be expected from its osmotic effect.*
- *Toxicities. Although much research focused on ion-specific effects under salinization, it is yet unknown whether Na^+, Cl^-, or other ions are the predominant growth-limiting factors for root growth reduction. Salinization increases Na^+ content throughout the elongation region of cotton roots, but rates of sodium deposition are enhanced only in the region least affected by salinity. This suggests that in cotton roots, similar to leaves, sodium accumulation in the growing cells is not the main cause of root growth reduction under NaCl stress.*

Na^+ and Cl^- uptakes play very important roles in the level of tolerance. Growth inhibition, the most common effect of salinity, is often correlated with high Na^+ concentration in the plant. For some plants, especially woody perennials (avocado, grapevine, and citrus), Na^+ is accumulated in the roots or stems, and Cl^- is transported to the shoot where it causes tissue damage. However, for most plants, Na^+ is considered to be the primary cause of ion-specific damage and processes which control uptake of Na^+ into the root, and its delivery and entry to the xylem are central for salt tolerance.

Cl^- influx into plant roots is thought to require energy in most situations and to be catalyzed by a $Cl^-/2H^+$ symporter. The nature of the mechanisms of Cl^- exclusion by roots, which characterizes tolerance in some species (e.g., Citrus), is not understood.

***Deficiencies and Ion Imbalance*.**

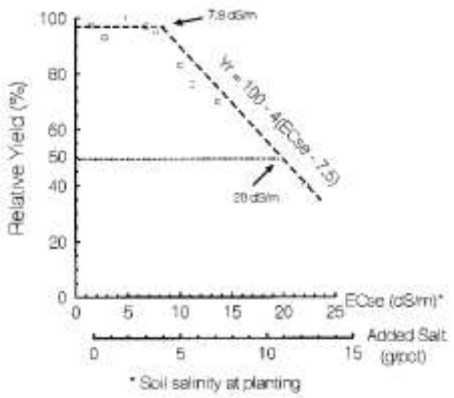
Salinity-induced root growth restriction reduces the volume of soil that can be explored by the root system and therefore the quantities of nutrients available to the plant. Additionally, high concentrations of Na^+ and Cl^- near the root surface can affect membrane uptake processes and thereby reduce nutrient uptake.

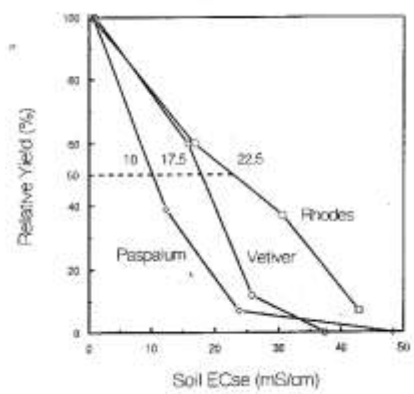
Antagonistic effects for influx into roots were reported for Na^+ and K^+, Ca^{2+}, Cl^- and NO^{3-}, and salinity can also reduce phosphorous uptake. Reduced Ca^{2+} and K^+ supply to the plant is often discussed in relation to salinity-induced growth inhibition. The rate of water and nutrient transport from root to the shoots is reduced under salinity. Therefore, in addition to effects on mineral uptake, salinity may also affect nutrient transport and distribution in the plant.

- *Reduced Ca^{2+} Supply: Calcium is an essential nutrient that is required for structural roles in the cell wall and membranes, as a counteraction for inorganic and organic anions in the vacuole and as an intracellular messenger in the cytosol. It plays a unique role in the response of plants to saline conditions. High concentration of Na^+ can displace Ca^{2+} involved in pectin-associated cross-linking and Ca^{2+} present at the binding sites of the root cell membranes, thereby affecting cell-wall and membrane stability and cellular functions. The reduction in cell growth when Ca^{2+} of the cell membrane is replaced by Na^+ was attributed to potassium leakage from the cells.*
- *Reduced K^+ Supply: Appropriate concentration of K^+ in plant cells is essential for growth and metabolic processes. However, K^+ and Na^+ ions compete for entry into plant cells; thereby, under saline conditions, plants may suffer potassium deficiency. Increasing concentration of NaCl significantly inhibits K+ influx into roots and increases the loss of intracellular K^+ Consequently, K^+ concentration in the plant is often reduced under salinity. Furthermore, most plants use K^+, rather than sodium, as a component for osmotic adjustment; therefore, reduced K^+ root intake under salinity may increase synthesis of organic osmolytes and have a negative effect on the energy balance in the plant. The selectivity of K^+ over Na^+ for root uptake is of major importance for plant function under saline conditions. The net root selectivity of K^+ over Na^+ differs between species and particularly between mono- and dicotyledonous halophytes, with a mechanism which is not yet clear* (Gregory, 2006).

Salt Tolerance of Vetiver grass

As shown in the graphs below, the saline threshold of Monto vetiver is EC_{se} = 8 dSm^{-1} (left) and soil EC_{se} values of 10 and 20 dSm^{-1} would reduce yield by 10% and 50% respectively (right). These results indicate vetiver grass compares favorably with some of the most salt tolerant crop and pasture species grown in Australia as shown in the table below. (Truong *et al*., 2003)

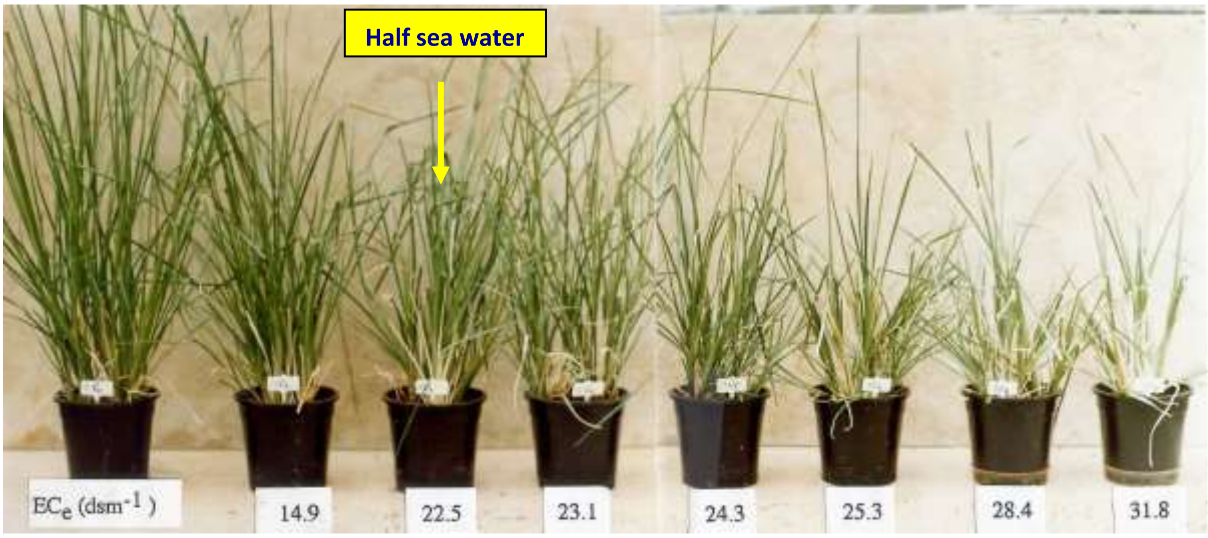




Salt tolerance level of vetiver grass as compared with some crop and pasture species grown in Australia

Species	Soil EC_{se} (dSm^{-1}) Saline Threshold	50%Yield Reduction
Bermuda Grass (*Cynodon dactylon)*	6.9	14.7
Rhodes Grass (C.V. Pioneer) (*Chloris guyana*)	7.0	22.5
Tall Wheat Grass (*Thynopyron elongatum*)	7.5	19.4
Cotton (*Gossypium hirsutum*)	7.7	17.3
Barley (*Hordeum vulgare*)	8.0	18.0
Vetiver (*Chrysopogon zizanioides*)	**8.0**	**20.0**

Response to varying levels of soil salinity



In Australia a trial was set up to select the most suitable species for the rehabilitation of a 23ha coal tailings dam. The substrate was saline, highly sodic and extremely low in nitrogen and phosphorus. The substrate contained high levels of soluble sulfur, magnesium and calcium. Plant available copper, zinc, magnesium and iron were also high. Five salt tolerant species were used: vetiver, marine couch (*Sporobolus virginicus*), common reed grass (*Phragmites australis*), cumbungi (*Typha domingensis*) and *Sarcocornia spp.* Complete mortality was recorded after 210 days for all species except vetiver and marine couch. (Radloff *et al*, *1995*; Truong, 1999b).

Complete mortality for all species except vetiver and marine couch

Under dryland salinity conditions, with drip irrigation, vetiver can survived under extremely saline surface soil

Dryland salinity trial at Kalgoorlie, Australia, note the salt crust on the surface near the vetiver

Saline toxicity symptom under field condition (L) and glasshouse (R)

A trial compared the performance of Vetiver and three other aromatic grasses, [Citronella (*Citronela java),* Lemon grass (*Cymbopogon citrates)* and Palmarosa (*Cymbopogon martini)*] for their suitability to saline water irrigation in a field experiment conducted on a calcareous sandy-loam soil at Bir Reserved Forest, Hisar, India. After the establishment of the four grasses Tomar and Minhas (2004) irrigated with fresh canal water or saline water at ECse 8.5dS/m. Results show that the vetiver biomass yield was 90.0 tonnes/ha as compared with Palmarosa at 29.1 t/ha and Lemon grass at 16.1 t/ha and Citronella did not survive, vetiver being the least affected.

Vetiver Root Development under Salt Stress

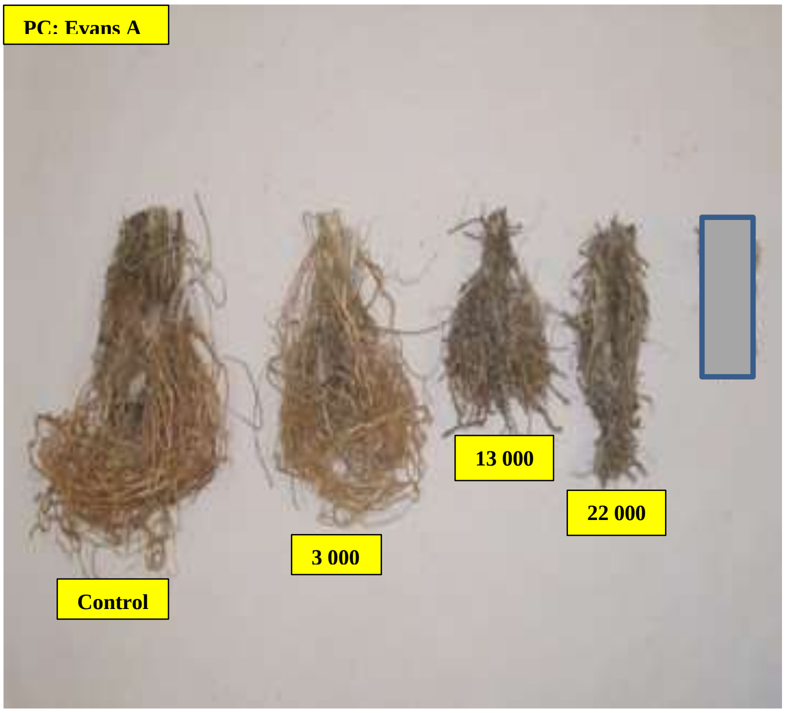
Extensive research conducted by the author on dryland salinity in Australia, where soil salinity was determined by the Electrical Conductivity of the soil extract, - $EC_{se,}$ instead of normal EC. The EC_{se} measure indicates the salinity level of the soil at field capacity, so it is a more accurate measurement. Results show that the salinity threshold of Vetiver is between 15.79 and 25.82mS/cm, which very high compared with corn and sorghum crops of approximately 8mS/cm

Salinity tolerance of vetiver ***Vetiver roots at different soil salinity levels***

Comparison of Vetiver root growth at different soil salinity levels

Evans et al (2008) from Louisiana State University Agricultural Center reported that after 11 weeks of growth and under hydroponics conditions both shoot and root biomass decreased when salinity increased from 0 (control) to 3,000, 13,000 and 22,000ppm of ocean salt. Shoot growth reductions from the control were 68%, 18% and 7% for 3,000, 13,000 and 22,000ppm treatments respectively. Root weights recorded for control, 3,000, 13,000 and 22,000 ppm treatments were 25g, 17g, 14g and 16g respectively. These results show that in the 22,000 ppm treatments, shoot growth was only 7% of control, while root growth was 64% (16g/25g) of control. *As mentioned in literature root growth is usually less sensitive to salt stress than shoot growth, this confirms that under salinity stress, vetiver root growth was less affected than shoot growth.*

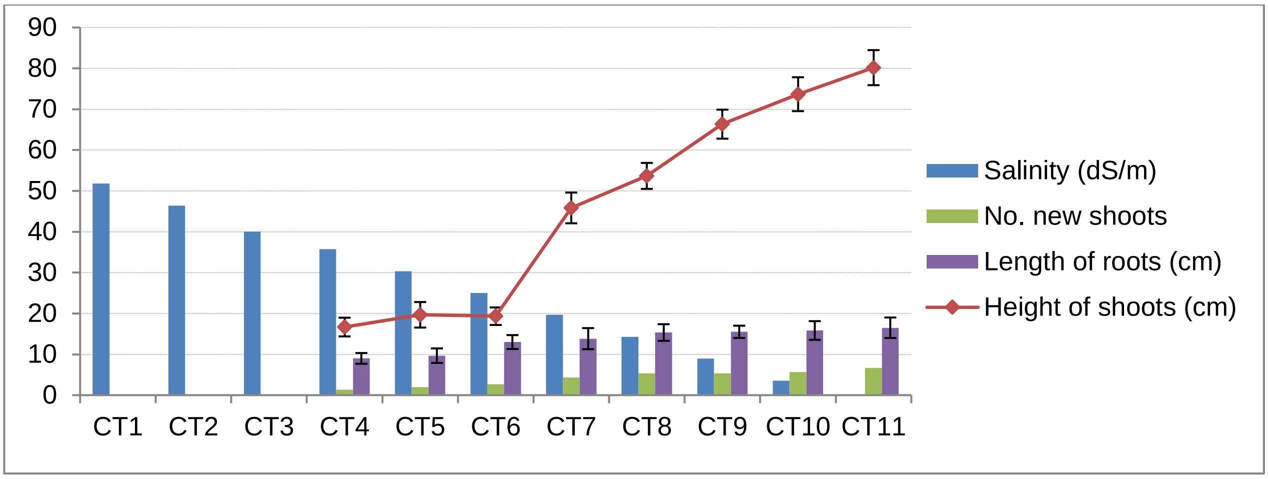
Roots of various treatments of ocean salt in ppm



To date studies on salt tolerance of vetiver have been conducted mainly in the soil environment, and very limited efforts in an aquatic environment. A trial conducted by Cuong et al (2015) to study the effect of sea water salinity on vetiver growth showed that vetiver grass can grow sea water salinity up to 19.64 dS/m (0-11 ‰).

Vetiver grass after 30 days, with increased level of salinity (L - R)

Effects of salinity on the growth of vetiver grass after 30 days



Plant roots under Aluminum stress: Toxicity and Tolerance

Aluminum Toxicity and Acid Soils **(**Gregory, 2006)

Aluminum toxicity is the primary problem limiting agricultural production in acid soils. The reason for this is because aluminum is one of the most prevalent minerals in the Earth's crust, and high acidity in soil can render aluminum soluble, thereby poisoning the water taken up by the roots. In addition to the natural factors that affect weathering, agricultural farming processes such as the excessive supply of inorganic fertilizers or removal of cations by harvest lower the soil pH. Furthermore, the acidity of environment is gradually increased due to environmental pollution and acid rain. Acid soils are infertile because they lack the basic nutrients, such as Ca^{2+}, Mg^{2+}, and K^{+} and are also characterized by high content of toxic elements such as Al, Mn, and Fe or deficiency of P. Most acid soils have low cation exchange capacity, leading to loss of essential minerals and to poor crop production.

Occurrence and Chemistry of Al

Exchangeable Al, next to Mn, is the major toxic elements in most acid soils. Al exists as insoluble alumino-silicates or oxides in the neutral or weakly acidic soil. In neutral solution, $Al(OH)_3$ precipitates as gibbsite that re-dissolves in basic solutions owing to formation of tetrahedral Al(OH)4− as aluminate anion. The nadir of aqueous Al solubility is at pH 6.2, with free Al ion concentration at pH 4, 5, 6, and 7 being ~50 mM, 50 µM, 0.05 µM, and 0.05 nM, respectively.

Inhibition of Root Elongation by Al

Inhibition of root elongation is the first visible symptom of Al stress. In most plant species, root elongation is markedly inhibited by Al^{3+} at the µmol level in a simple solution containing Ca^{2+} alone. Inhibition of root elongation of Al-sensitive maize occurred within 30 min of Al treatment. The root apex (root cap, meristem, distal transition zone, and elongation zone) accumulated most of the Al and played a major role in the Al-perception mechanism. Indeed, only the apical 2–3 mm of maize and pea roots should be exposed to Al for the inhibition of root elongation to take.

Morphological Changes of Intact Roots and Root Cells under Al Stress

A number of studies have shown that the rhizodermis of roots may rupture or crack when exposed to Al. Morphological changes in wheat roots were characterized by cracks on the root surface.

Inhibition of Cell Division. *Cell division in root meristems of several plants is inhibited by Al. Cell division accounts for only 1%–2% of the overall root elongation, and cell cycle in plants takes about 1 day. However, the primal phenomena of Al toxicity are the inhibition of root elongation that occurs within hour(s) of Al treatment. Thus, attention has been largely paid to the inhibition of root cell elongation as the primary site of Al toxicity. However, the inhibition of cell elongation at the elongation zone is not fatal for plant growth as long as the cells can divide at the meristematic zone, suggesting that the lethal cause of Al toxicity might be inhibition of cell division.* **(**Gregory, 2006)

Site of Al Toxicity

Cell Wall: Although there is disagreement with regard to the site of Al toxicity–namely, symplastic or apoplastic – many investigators have stated that 30%–90% of the absorbed Al is localized in the apoplast. Aluminum toxicity recovery of root elongation completely stops in the presence of excess level of toxic Al, and plants begin dying without recovery. The root elongation in acid soil might not be uniform, and the inhibition and elongation and/or re-elongation including the recovery process might occur simultaneously.

Calcium: Ca^{2+} *is an essential element for root growth, and much work was devoted to its role in Al toxicity. Al-induced changes in cell physiology, occurring in the cytoplasm and at the plasma membrane, might be caused by the disruption of* Ca^{2+} *homeostasis. An inhibitory effect of Al was observed not only on* Ca^{2+} *uptake but also on its translocation from the apical region.*

Aluminum Tolerance

Al toxicity is a major factor inhibiting plant growth in acid soil. So far, two mechanisms of Al tolerance have been proposed. One is the exclusion, or external tolerance mechanism, which was defined in case where Al was prevented from entering the plant cells, and the other is internal tolerance mechanism which was defined in case where Al enters the cells and tolerance is achieved by detoxification processes. Recent research indicated that exclusion mechanism at the molecular level is most important.

Exclusion Mechanism.

Since higher plants cannot move away from the acid soil, they have developed ways to reduce this edaphic stress. An effective strategy to reduce the stress is to chelate the toxic Al^{3+} *with exuded organic anions in the rhizosphere rendering less toxicity of* Al^{3+}*. Exudation of organic acids is an important factor, the release of organic acids occurs mainly at the root apex where the toxicity symptoms appear under Al stress. The exudation of organic acids may also contribute to the utilization of Al phosphate in the soil. It was generally accepted that the amount of organic acids at the root tip was similar in Al-tolerant and Al-sensitive cultivars.*

***Internal Al-tolerance Mechanism*.**

The strategy of exclusion mechanism is to exclude the toxic Al into roots by the chelation of Al with exuded organic acids and other substances. However, some crops can retain a large amount of Al inside the cell without any growth reduction. This means that the cell has a mechanism for detoxifying Al after uptake. Amelioration of Al toxicity is induced by the formation of an alumino-silicate compound in the root apoplast.

pH regulation in the rhizosphere:

Solubility of Al depends strongly on pH suggesting that high-solution pH may reduce the solubility and toxicity of Al. An increase in the pH of dilute nutrient solution from 4.5 to 4.6 caused a 26% decline in soluble Al concentration. This suggests that even a slight pH change can affect the toxicity of Al.

General Conclusion

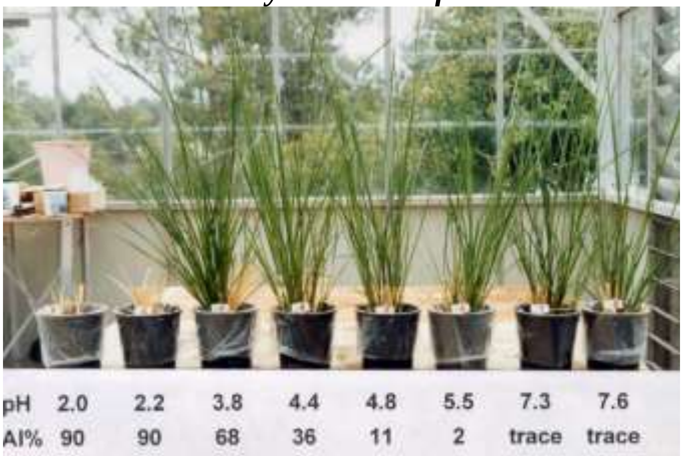
Al is the major element in the soils, and solubilized Al^{3+} at pH lower than 4.5–5.0 inhibits root elongation markedly. Al^{3+} has a strong affinity to the cell constituents, and various cell functions including plasma membrane, cell wall, Ca, signal transduction, and cell division are adversely affected by Al toxicity. Exclusion or extracellular mechanism plays an important role in Al tolerance. **(**Gregory, 2006)

Vetiver Root Development under Aluminium Stress

Acid sulfate soils (ASS) exist globally but are most common in tropical and subtropical regions. These soils share common characteristics: extreme acidity with pH between 2.0 and 3.0 in the dry season, and high levels of Al, Fe, and SO_4^2. In soil with high clay content, such as the Mekong Delta of Vietnam, the soil will crack as it dries. Because of this cracking, very few endemic plants can establish and survive during the dry season, including those considered to be locally tolerant species (Du and Truong, 2002).

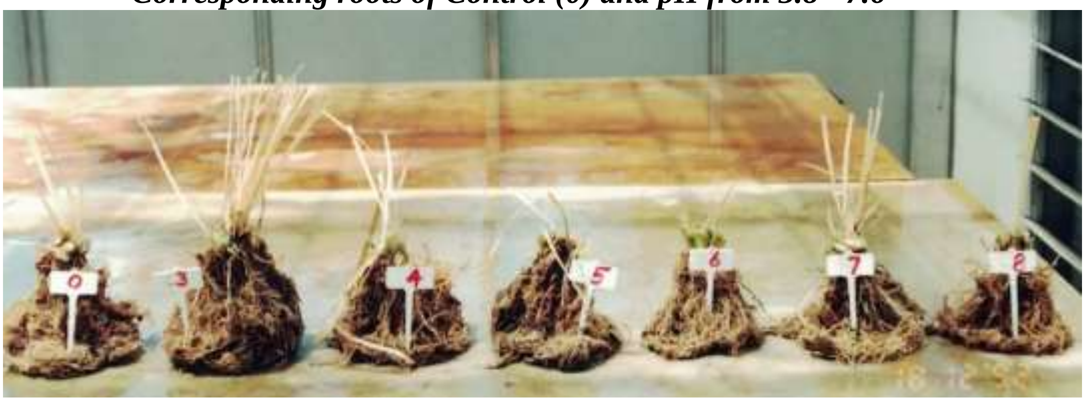
Extensive research conducted by the author in Australia and Vietnam show that vetiver can be established on acid sulfate soils for erosion control due to its tolerance to extremely acidic soils. The following table shows that Al threshold (expressed as *Saturation Percentage)* for vetiver is between 68% - 90%. D. Miller (per com.) found vetiver growing in a soil with Al saturation of 87%. Thus, vetiver is more tolerant to Al toxicity than some of the most tolerant crop and pasture species such as rice (<45%), corn (30%), wheat (30%), soybean (20%), lucerne (15%) and cotton (10%) (Fageria *et al.*, 1988).

Aluminium toxicity tolerance of vetiver in Australia

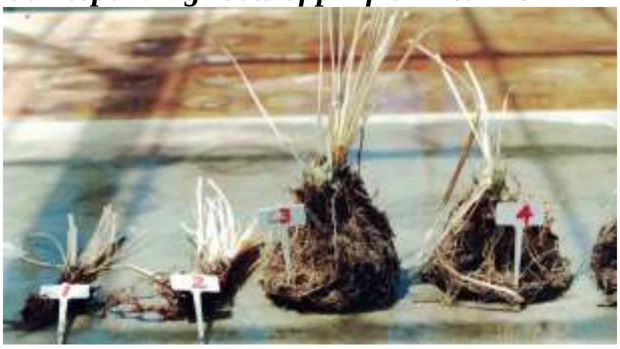


Note: ***Al% is Saturation Percentage (% of soluble Al/CEC)*.**

Corresponding roots of Control (0) and pH from 3.8 - 7.6



Corresponding roots of pH from 2.0 - 4.4



Corresponding roots of pH from 4.8 – 7.6

It was noted that under Al stress, vetiver developed finer roots than under the Control

Finer roots of Al 68%

More major roots in the Control

In tropical Australia ASS have severely affected sugar cane growth in north Queensland. Erosion of drainage channel banks of ASS on an old cane farm near Babinda was persistent and severe. Several attempts in the past to stabilise these banks with various plant species have failed due to plant death and the land holder resorted to using rocks to stabilise the most severe erosion on the banks. Vetiver grass was planted along the banks of these channels late in 1995 and after 8 months, although it was not fully mature, vetiver successfully stabilized these banks during the wet season. Soil analyses show pH of 5-15cm topsoil pH was 3.5, Total Actual Acidity 36moleH^+/T, Total Potential Acidity 272moleH^+/T, Al 2.4meq/100g and Al/CEC21%. These measurements indicate that an actual ASS exist here. Vetiver was successfully established on these soils without any fertilizer and reached a height of 80cm after 8 months. Much more vigorous growth occurred when DAP (300kg/ha) was applied at planting.

Highly erodible ASS with pH 3.5

One year after planting

In the Mekong Delta of Vietnam Du and Truong (2002) reported that the establishment of drainage and irrigation channels in ASS is the most important technique for improving agricultural and fishery productivity. However, the embankments of these channels are highly erodible because they are weak physically and exist under extremely acidic conditions, where very few plants can survive, especially during the dry season leading to severe and costly bank erosion problem. Three trials were conducted to determine the tolerance level of vetiver grass under extreme ASS conditions and its effectiveness in reducing bank erosion. The following table shows the characteristics of the ASS at the three experimental sites.

Trials	pH_{H2O}	pH_{KCl}	EC (mS/cm)	Al^{3+} (meq/100g)	SO_4^{2-} (meq/100g)	Active Fe (ppm)
1	2.80	2.50	3.93	32.4	7.56	882
2	2.50	2.35	3.14	31.0	6.25	1170
3	3.13	2.97	0.88	0.93	2.33	114

Note: the ratio of soil/water (solution) for pH and EC measurement was 1/2.5

The following table shows the levels of some toxic elements over time in the vetiver

Elevated levels of some toxic elements in vetiver grass

Days after transplanting	**Plant part**	**Toxic concentration**		
		Al (ppm)	Fe (%)	SO_4 (%)
70	Leaf	568	0.58	8.36
	Root	**557**	**1.87**	**7.98**
105	Leaf	663	0.44	8.27
	Root	**646**	**2.82**	**10.26**
270	Leaf	660	0.50	9.00
	Root	**600**	**0.55**	**11.00**

It was concluded that:

- Planting of vetiver grass greatly improves bank stability and reduces bank erosion;
- Only a small quantity of lime is needed to neutralize the high acidity around vetiver roots at transplanting time. Once established vetiver plant can survive higher acidity;
- Vetiver grass stopped growing in the dry season, but it re-grew vigorously with as little as 20mm of rainfall; and
- Vetiver grass could improve the quality of water in drainage channels by removing considerable amount of some toxic elements leaching from the ASS embankment.

In Australia due to its abundant natural resources, mining is a major industry, hence mining waste rehabilitation is a priority to protect the environment. The task is extremely difficult to rehabilitee mine tailings, particularly for gold mines.

A particularly challenging project was carried out by the author to revegetate an old gold tailings waste dam with high sulfur content, extremely acidic (pH 2.5-3.5), high in heavy metals and low in plant nutrients. Revegetation of these kinds of tailings is very difficult and often very expensive and the bare soil surface is highly erodible. These tailings are often the source of contaminants, both above ground and underground to the local environment.

Field trials were conducted on two old (8 year) gold tailings sites, one had a soft surface and the other had a hard crusty surface layer. The soft surface site had a pH of 3.6, sulphate at 0.37% and total sulfur at 1.31%. The hard surface site had a pH of 2.7, sulphate at 0.85% and total sulfur at 3.75%.

Both sites were low in plant nutrients. Results from both sites indicated that when adequately supplied with nitrogen and phosphorus fertilizers (300kgha^{-1}of DAP). Excellent growth of vetiver was obtained on the soft surface site (pH=3.6) without any liming. But the addition of 5tha^{-1} of agricultural lime significantly improved vetiver growth. On the hard surface site (pH=2.7) vetiver survived without liming, but the addition of lime (20tha^{-1}) and fertilizer (500kgha^{-1} of DAP) greatly improved vetiver growth.

Old gold mine tailings in Australia with extreme acidity

Good growth with initial moderate lime and fertilizer applications

Six months after planting ***Two years later*** ***Ten years later no fertilizers***

ROOT RESPONSES TO TRACE METALLIC ELEMENTS (Gregory, 2006)

Classifications of Trace Metallic Elements

The term "heavy metals" often applies a group of metals and metalloids associated with environmental pollution. Legal regulations still refer to "heavy metals" as a list of 11 elements with a density higher than five, soluble at physiological conditions, and relevant in the environmental context: arsenic (which is actually a metalloid), cadmium (Cd), chromium, cobalt (Co), copper (Cu), lead (Pb), mercury (Hg), nickel (Ni), tin (Sn), vanadium (V), and zinc (Zn).

The term "trace elements" refers to chemical elements that occur in the earth's crust in amounts less than 0.1% in mass. Since this review focuses on root responses, those metals that are micronutrients will also be considered, they are present as trace amounts in plant organs (<0.01% plant dry weight and fulfill biological functions.

The term Trace Metallic Elements – TME- refers to both "heavy metals" and "trace elements"

- *Essential elements: Of the 92 known elements, 17 elements are classified as essential because they are required to complete plant's life cycle. Among those, Cu, Fe, Mn, Mo, Ni, and Zn are metallic micronutrients. Essential elements play many different biological roles in plants.*
- *Beneficial elements can promote growth or may be essential to particular taxa but not to all plants*
- *Nonessential elements are those that are neither essential nor beneficial. The evolution of mineral elements as essential or nonessential depends on chemical properties but also on their abundance in the Earth's crust.* (Gregory, 2006)

Occurrence of TMEs in Soils and Strategies to Address Human Health and Environmental Issues

Pedogenetic processes and anthropogenic activities can modify the occurrence of TMEs in natural and agricultural systems. Some lands naturally contain relatively high TME levels. Over the past 200 years, emissions of toxic TMEs by human activities have significantly exceeded those from natural sources for practically all metals. Extensive mining, agriculture, and industry have released enormous amounts of TMEs into the environment, which create worldwide environmental and health concerns. For instance, Cd contamination is widespread, particularly in soils containing waste materials from mining industry and in sludge-amended and Cd-rich phosphatic fertilized soils. Farmland may also contain elevated quantities of essential TMEs, due to the continuous spreading of Cu-rich manure and Cu-based fungicides, or scattering of Zn-rich sewage sludge.

To address environmental and human health issues related to contaminated ecosystems, phytoremediation (use of plants and their associated microbes) has attracted much attention during the last decades. The many possible ways in which plants can achieve this include phytoextraction, phytovolatilization, detoxification, and sequestration. The two first processes are particularly attractive in that their use has the potential of moving the contaminants from the local ecosystem altogether. In phytoextraction, plants take up the contaminant of interest from soil, sediment, or water and accumulate it. Phytoextraction is possible within a reasonable time frame in weakly to moderately degree of pollution, by using plants with efficient metal root-to-shoot translocation and high accumulation capacity in the shoot. In such a case, harvesting the aboveground biomass succeeds in removing the contaminant from the local ecosystem. Phytovolatilization is relevant to those trace element contaminants that can be metabolized to a volatile form (e.g., Hg, which is volatile in its elemental form). For more heavily contaminated soils, phytostabilization with tolerant plants aims at stabilization of the contaminated site and at lowering the risk of pollutants leaching.

Hyperaccumulators can be directly used for phytoremediation strategies. However, most hyperaccumulators do not develop sufficient biomass, even in the absence of high concentrations of heavy metals to be useful in phytoextraction, and/or are not amenable to agronomic practices (mechanical harvest). To create suitable plants for phytoremediation, one possibility is the selection of high-biomass plants with the highest tolerance and accumulation capacities. An opportunity is also sought in the association of metal-tolerant microorganisms to enhance tolerance. Finally, genetic manipulation can improve accumulation, tolerance, and detoxification capacities of high-biomass and rapid-growing plants in order to optimize the phytoextraction process. Genetic determinants of those traits can be identified through the study of hyperaccumulators.

Soil Factors Influencing the Absorption of Trace Metallic Elements by Plants

TMEs present in the soil can be divided into different fractions: metals within the matrix of soil minerals, metal precipitates, metals sorbed to clays and organic matter, and soluble metals in the soil solution. Plants cannot access the total TME pool in soil but only the latter fraction. Soil factors as various as pH, redox potential (Eh), organic matter, clay, water regime, redox conditions, Cation Exchangeable Capacity, and microbiological activity determine the proportion of metals in the soil solution. Nonetheless, not all dissolved TMEs but only the bioavailable fraction, which *mainly depends on metal speciation (chemical forms that an element takes in solution), can be directly absorbed by roots. Hence, soil solution pH strongly affects TME speciation and solubility.* (Gregory, 2006)

Uptake Systems of Trace Metallic Elements in Plants

Plants possess highly effective uptake systems for TMEs in the soil solution, which are usually present in the submicromolar to low nanomolar concentration range. For most of the elements, the transport is not specific, enabling nonessential metals to compete for the absorption and enter the root cells as well. When essential TMEs become scarce in soil, or when the mineral balance is unfavorable, plants frequently modify their absorption through coordinated regulation of mineral uptake transporters both at the transcriptional and posttranscriptional levels. (Gregory, 2006)

Transport and Circulation of Trace Metallic Elements in Plants

After crossing the plasma membrane of epidermal cells through a metal uptake system, TME ions, as intermediate to soft Lewis acids, can bind carboxylic, amino, and thiol groups. As a result, free TME concentrations will be very low (they have been calculated at less than one Cu or Zn free ion per cell.

Importance of rhizosphere activity

The rhizosphere encompasses the millimeters of soil surrounding a plant root. Very complex interactions occur between roots and the rhizosphere, in which root exudates (ions, free oxygen and water, enzymes, mucilage, and a diverse array of carbon-containing primary and secondary metabolites) play an important role. Some bacteria, known as "plant growth–promoting rhizobacteria" (PGPR), can increase root hair production and root surface area, thereby affecting TME absorption capacity. Different mechanisms have been identified that can explain that effect: production of plant growth regulators that cause root cell elongation like IAA (indole-3-acetic acid) or production of enzymes degrading ethylene (which inhibits root growth). Furthermore, some bacteria can acidify the rhizosphere and produce high concentrations of chelating agents that can enhance the solubility of metals like Fe. (Gregory, 2006)

Response of Roots to Trace Metallic Elements

Plants respond to fluctuations in mineral nutrient concentrations by altering the physiology and morphology of their roots. Essential TMEs are required by plants in balanced proportions. Because they are needed in much smaller amounts than macronutrients, the concentration window between deficiency and toxicity is narrow. Nonessential TMEs can rapidly become toxic at even lower concentrations. Deviation from equilibrium can result in drastic nutritional disorders and profound root morphological adaptation.

In an exhaustive review Heavy Metal Stress and Some Mechanisms of Plant Defense Response, Abolghassem Emamverdian et al. (2015) pointed out that contamination of soil and water by Heavy Metal (HM) in changing environment poses a serious threat to public and food safety and is now emerging as a major health hazard to humans and plants. This has become more accentuated and prominent as human-made disturbance of biological resources of the planet has accelerated the occurrence of many abiotic stresses by HM. As a consequence, plants are now exposed to this toxicity more than any time in their history since the beginning of their terrestrial life on planet earth. This necessitates making more efforts to deepen our appreciation of HM and the way plants respond to their ever-growing presence.

HM belong to group of non-biodegradable, persistent inorganic chemical constituents with the atomic mass over 20 and the density higher than 5 g/cm^3 that have cytotoxic, genotoxic, and mutagenic effects on humans or animals and plants through influencing and tainting food chains, soil, irrigation or potable water, aquifers, and surrounding atmosphere

There are two kinds of metals found in soils, which are referred to as essential micronutrients for normal plant growth (Fe, Mn, Zn, Cu, Mg, Mo, and Ni) and nonessential elements with unknown biological and physiological function (Cd, Sb, Cr, Pb, As, Co, Ag, Se, and Hg). Both underground and aboveground surfaces of plants are able to receive HM. The essential elements play a pivotal role in the structure of enzymes and proteins. Plants require them in tiny quantities for their growth, metabolism, and development; however, the concentration of both essential and nonessential metals is one single important factor in the growing process of plants so that their presence in excess can lead to the reduction and inhibition of growth in plants. HM at toxic levels hamper normal plant functioning and act as an impediment to metabolic processes in a variety of ways. In addition to their effects on vegetation, HM are also major pollutants to both territorial and aquatic environments.

They reported HM at toxic levels have the capability to inflict serious morphological, metabolic, and physiological anomalies in plants ranging from chlorosis of shoot to lipid peroxidation and protein degradation. In response, plants are equipped with a repertoire of mechanisms to counteract HM toxicity. The key elements of these are chelating metals by forming phytochelatins or metallothioneins metal complex at the intra- and intercellular level, which is followed by the removal of HM ions from sensitive sites or vacuolar sequestration of ligand-metal complex. Another important additive component of plant defense system is symbiotic association with arbuscular mycorrhizal fungi, which can effectively immobilize HM and reduce their uptake by host plants via binding metal ions to hyphal cell wall and excreting several extracellular biomolecules. Additionally, this fungi can enhance activities of antioxidant defense machinery of plants.

The authors selected six metals, Cu, Cr, and Mn that are known for taking part in redox reactions in plants and three non-redox active metals, Ni, Zn, and Al for review in detail to show how they impinge on plants despite possessing different redox states. (Gregory, 2006) [−3]

Aluminum (Al). *Al is known as an inhibitory element for the growth of plants, especially in acidic soils with pH values as low as 5 or 5.5 where the most phytotoxic form of Al (Al^{3+}) is prevalent. Although there is still no known or proven biological role for aluminum in plants, some reports demonstrate that Al at low concentrations may lead to the stimulation of plant growth. A 2-3 ug/g aluminum threshold in soils with a pH below5.5 is considered to be hazardous to most plants. The primary target of Al toxicity is roots of plants where the accumulation of Al inflicts the inhibition of root growth in the space of minutes or hours. It can increase the thickness of lateral roots and change their color to brown. Reduction of root respiration and disturbances in the enzymatic regulation of sugar phosphorylation are also caused by Al toxicity.*

The symptomatic effects of Al-induced stress on shoots, which are similar to phosphorus deficiency, may be stunting of leaves, purple discoloration on stems, leaves, and leaf veins followed by yellowing and dead leaf tips, and those that resemble calcium deficiency can be curling or rolling of young leaves and death of growing points or petioles. The other visible indications of Al toxicity are the appearance of small necrotic spots on the border of young leaves and chlorosis in the margins and center of older leaves. The reduction in stomatal aperture and decreased photosynthetic activity are also reported to be caused by Al toxicity

Chromium (Cr). *It is well documented that Cr is a toxic agent for the growth and development of plants. In addition, it is known as one of the causes of environmental pollution. In plants, Cr is found in the forms of trivalent $Cr^{3+}$and hexavalent Cr^{6+}species, where the former is of lower toxicity than the latter. Cr is transported and accumulated via carrier ions such as sulfate or iron and is not directly absorbed by plants. The highest concentration of Cr occurs in the root rather than other parts of plants. Immobilization of Cr in vacuole of plant root cells is suggested as a main reason for the excessive accumulation of this metal in roots. Cr drastically reduces seedling dry matter production and hampers the development of stems and leaves during plant early growth stage. Chromium toxicity inhibits the cell division and elongation of plant roots, thus shortening the overall length of roots. As a consequence, water and nutrient absorption processes are severely restricted, which can lead to the decreased shoot growth.*

Manganese (Mn). *Mn is an essential micronutrient that plays a pivotal part in many metabolic and growth processes in plants including photosynthesis, respiration, and the biosynthesis of enzymes. Furthermore, manganese is involved in carbohydrate and nitrogen metabolism, synthesis of fatty acid, acyl lipids, and carotenoid as well as hormonal activation.* Mn^{2+} *is the most stable and soluble form of manganese in the soil environment. However, lower soil pH, less soil organic matter, and decreased redox potential increase the availability or toxicity of Mn to plants. Contrary to some elements such as aluminum or copper, there is a tendency for manganese to easily translocate form roots to the upper parts of plants. This mobility is the reason why symptoms of Mn toxicity are first visible in aerial organs of plants. The appearance of visual features in plants affected by Mn toxicity varies with the type of plant species, plant age, temperature, and light level. The symptoms may include crinkled leaves, darkening of leaf veins on older foliage, chlorosis and brown spots on aged leaves, and black specks on the stems.*

Nickel (Ni). *Ni is a micronutrient that is required by both higher and lower plants in very small amounts but its phytotoxicity is deemed to be more important than its shortage. Ni has various oxidative states but its divalent state (*Ni^{2+}*) is the most stable type in the environment and biological systems. Although the role of Ni in metabolic processes of plants has not been identified as extensively as other elements such as Mn or Cu, it is a key factor in the activation of enzyme urease, which is needed for nitrogen metabolism. Moreover, it plays a part in seed germination and iron uptake. The concentration level representing Ni toxicity in plants varies greatly from 25 to 246 ug/g dry weight of plant tissue, depending on the plant species and cultivars. Ni at excess competes with several cations, in particular,* Fe^{2+} *and* Zn^{2+}*, preventing them from being absorbed by plants, which ultimately causes deficiency of Fe or Zn and results in chlorosis expression in plants . Excess nickel adversely affects germination process and seedling growth traits of plants by hampering the activity. Ni, especially at high concentrations, can readily move through phloem and xylem vessels, thereby translocating smoothly from the root to the upper parts of plants.*

Several studies in plants including maize and cowpea indicated that Ni toxicity can result in inhibited lateral root formation and development. Moreover, the agglomeration of Ni in root apex greatly hampers mitotic cell division in this organ, which ultimately results in growth reduction.

Copper (Cu). *Cu is an essential micronutrient that participates in many vital physiological functions of plants including acting as a catalyzer of redox reaction in mitochondria, chloroplasts, and cytoplasm of cells or as an electron carrier during plant respiration. However, Cu becomes toxic when its concentration in the tissue of plants rises above optimal levels. Cu exists in many states in soils but is mainly taken up by plants in the form of* Cu^{2+}*. The concentration of copper in soil is typically between 2 and 250ug/g and healthy plants can absorb 20–30ug/g of dry biomass. But copper availability depends greatly on soil pH and its phyto-availability increases with declining pH. In addition, uptake of Cu by plants and its toxicity are contingent on nutritional condition of plant,* Cu^{2+} *concentration in soil, length of exposure, and genotype of a species. A plethora of research studies on a number of species indicate that copper has a propensity for the accumulation in the root tissues with little upward movement towards shoots. Therefore, the initial characterization of Cu toxicity is the hindrance of root elongation and growth. The subsequent symptoms include chlorosis, necrosis, and leaf discoloration*

Zinc (Zn). *Zn is an essential trace metal that despite having no redox activity is particularly involved in many vital physiological events in plants. Zinc is an indispensable component of special proteins known as zinc fingers that bind to DNA and RNA and contribute to their regulation and stabilization. Moreover, it is a constituent of various enzymes, and plays a role in the formation of carbohydrates and chlorophyll and root growth. Zinc, in divalent state (*Zn^{2+}*), is the most pervasive form found in soil and acquired by plants. Zn bioavailability/phytoavailability is dependent on various variables including the total Zn concentration in soil, lime content and organic matter of soil, clay type, and presence of other HM, soil's pH, and the amount of salt in the substrate.*

Of these, pH is the most important factor influencing Zn availability and higher pH is generally associated with the decreased absorption of Zn by plants. Zn at high soil concentrations (150 to 300ug/g) is strongly toxic and its phototoxicity, in addition to the bioavailability factors, depends on plant type and plant development stage. Visual signs of trouble in plants as a result of Zn toxicity are reported to be chlorosis in young leaves due to iron or manganese deficiency and appearance of purplish-red color in leaves due to phosphorus deficiency, which indicate that Zn^{2+} *in excess can easily supersede other metals, especially those with similar ionic radii in the active sites of enzymes or transporters. Moreover, necrotic spotting between the veins in the blade of mature leaves and inward rolling at leaf margins are attributed to Zn toxicity.*

Growth parameters and structure of plant parts are shown to be negatively affected by Zn toxicity, by decreasing the length of root and shoot as well as leaf area. (Gregory, 2006)

TOLERANCE AND HYPERACCUMULATION OF TRACE METALLIC ELEMENTS

Definition of Tolerance and Accumulation Strategies (Gregory, 2006)

1. *When considering heavy metals (HM) concentration in shoots as a function of the soil HM concentration, three general accumulation strategies can be distinguished in plants:*
2. *Exclusion, which is characterized by mechanisms acting to minimize metal accumulation in aboveground tissues;*
3. *Indication with a positive correlation between shoot and soil element concentrations, in that case proportional relationships exist between metal levels in the soil, uptake and accumulation in plant parts;*
4. *Accumulation, described by high metal concentration in the shoot, even when soil concentration is low.*
   a. *Based on the concentration of metals in aboveground plant parts, which can be further distinguished as accumulators or hyperaccumulators. Hyperaccumulators can accumulate exceptional concentrations of trace elements in their aerial parts without visible toxicity symptoms, usually reflected by a root-to-shoot ratio of metal content above 1. Foliar concentration thresholds for hyperaccumulation have been arbitrarily set to about 100 times the element mean concentrations in other plants grown in the same environment but are currently subject to revision.*

Difference in Root Structure between Hyperaccumulators and Non-hyperaccumulators

Difference in root structure between tolerant and sensitive plants can provide some answers to the question of how the root system has adapted under the selective pressure of high in HM and TMEs. Hyperaccumulation can also be associated with modification of the endodermis. Observations suggest anatomical adaptation underlying efficient uptake by the root and translocation to the shoot in hyperaccumulator. (Gregory, 2006)

VETIVER ROOT DEVELOPMENT AND TOLERANCE UNDER HEAVY METAL STRESS

Vetiver tolerance to Heavy Metals

Early research on the use of vetiver for the phytoremediation of heavy metal contaminated soils indicated that it can survive in a soil environment containing high concentrations of a wide range of heavy metals and can accumulate these metals into roots and shoots. The potential of vetiver in absorbing heavy metals has been studied through a wide range of experiments involving individual heavy metals or their combinations both in field and glasshouse conditions.

A literature search indicated that most vascular plants are highly sensitive to heavy metal toxicity and most plants were also reported to have very low threshold levels for arsenic, cadmium, chromium, copper and nickel in the soil. Results shown in the table below demonstrate that vetiver is highly tolerant to these heavy metals. For arsenic, the toxic content for most plants is between 1 and

10 $mgkg^{-1}$, for vetiver the threshold level is between 21 and 72 $mgkg^{-1}$. Similarly for cadmium, the toxic threshold for vetiver is 45 mg $kg^{-1}$and for other plants between 5 and 20 $mgkg^{-1}$. An impressive finding was that while the toxic thresholds of vetiver for chromium is between 5 and 18 $mgkg^{-1}$ and that for nickel is 347 $mgkg^{-1}$, growth of most plants is negatively affected at the content between 0.02 and 0.20 $mgkg^{-1}$ for chromium and between 10 and 30 $mgkg^{-1}$ for nickel. Vetiver had similar tolerance to copper as other plants at 15 $mgkg^{-1}$.

Threshold levels of heavy metals to vetiver growth

Heavy metals	Threshold to plant growth ($mgkg^{-1}$)		Threshold to vetiver growth ($mgkg^{-1}$)	
	Hydroponic level	**Soil level**	**Soil level**	**Shoot level**
Arsenic	0.02-7.5	2.0	100-250	21-72
Cadmium	0.2-9.0	1.5	20-60	45-48
Copper	0.5-8.0	NA	50-100	13-15
Chromium	0.5-10.0	NA	200-600	5-18
Lead	NA	NA	>1 500	>78
Mercury	NA	NA	>6	>0.12
Nickel	0.5-2.0	7-10	100	347
Selenium	NA	2-14	>74	>11
Zinc	NA	NA	>750	880

Distribution of Heavy Metals in Vetiver Plant

Research conducted by the author as shown in the table below indicates the distribution of heavy metals in a vetiver plant can be divided into three groups:

- Very little of the arsenic, cadmium, chromium and mercury absorbed were translocated to the shoots (1% to 5%)
- A moderate proportion of copper, lead, nickel and selenium were translocated (16% to 33%) to the shoots; and
- Zinc was almost evenly distributed between shoots and roots (40%).

Distribution of Heavy Metals in Vetiver Shoots and Roots.

Metals	Soil ($mgkg^{-1}$)	Shoot ($mgkg^{-1}$)	Root ($mgkg^{-1}$)	Shoot/Root %	Shoot/Total %
Arsenic (As)	959	9.6	185	5.2	4.9
	844	10.4	228	4.6	4.4
	620	11.2	268	4.2	4.0
	414	4.5	96	4.7	4.5
	605	6.5	124	5.2	5.0
Average				**4.8**	**4.6**
Cadmium (Cd)	0.67	0.16	7.77	2.0	2.0
	0.58	0.13	13.60	1.0	0.9
	1.19	0.58	8.32	7.0	6.5
	1.66	0.31	14.20	2.2	2.1
Average				**3.1**	**2.9**
Copper (Cu)	50	13	68	**19**	**16**
Chromium (Cr)	50	4	404	1	1
	200	5	1170	<1	<1
	600	18	1750	1	1
Average				**<1**	**<1**
Lead (Pb)	13	0.5	5.1	10	9
	91	6.0	23.2	26	20

Metals	Soil (mgkg^{-1})	Shoot (mgkg^{-1})	Root (mgkg^{-1})	Shoot/Root %	Shoot/Total %
	150	13.2	29.3	45	31
	330	41.7	55.4	75	43
	730	78.2	87.8	87	47
	1500	72.3	74.5	97	49
Average				**57**	**33**
Mercury (Hg)	0.02	BQ	0.01	-	-
	0.36	0.02	0.39	5	5
	0.64	0.02	0.53	4	4
	1.22	0.02	0.29	7	6
	3.47	0.05	1.57	3	3
	6.17	0.12	10.80	11	6
Average				**6**	**5**
Nickel (Ni)	300	448	1040	**43**	**30**
Selenium (Se)	0.23	0.18	1.00	53	15
	1.8	0.58	1.60	36	27
	6.0	1.67	3.60	46	32
	13.2	4.53	6.50	70	41
	23.6	8.40	12.70	66	40
	74.3	11.30	24.80	46	44
Average				**53**	**33**
Zinc (Zn)	Control	123	325	38	27
	100	405	570	71	42
	250	520	490	106	51
	350	300	610	49	33
	500	540	830	65	39
	750	880	1030	85	46
Average				**69**	**40**

BQ Below Quantification

The important implications of these findings are that when vetiver is used for the rehabilitation of sites contaminated with high levels of arsenic, cadmium, chromium and mercury, its shoots can be safely grazed by animals or harvested for mulch as very little of these heavy metals are translocated to the shoots. As for copper, lead, nickel, selenium and zinc their uses for the above purposes are limited to the thresholds set by the environmental agencies and the tolerance of the animal concerned.

In addition, although vetiver is not a hyper-accumulator it can be used to remove the some heavy metals from the contaminated sites and disposed of safely elsewhere, thus gradually reducing the contaminant levels. For example vetiver roots and shoots can accumulate more than 5 times the chromium and zinc levels in the soil.

Subsequent extensive studies carried out by the author have evaluated the ability of vetiver to remove heavy metals such as arsenic, lead, copper, zinc, cadmium, mercury etc., from contaminated soils. While naturally contaminated soils were used to investigate the ability of vetiver to deal with combinations of several heavy metals, artificially contaminated soils were used to investigate the removal of the following single heavy metals.

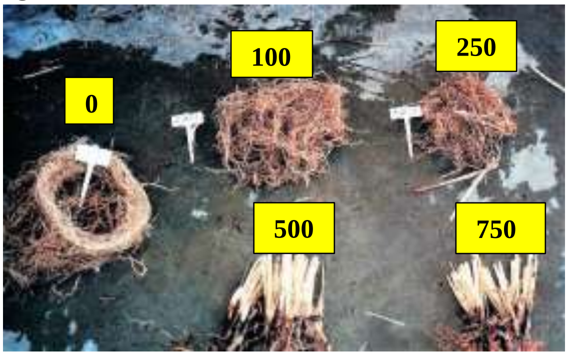
- ***Arsenic.*** Vetiver is tolerant to high concentrations of arsenic (As) in the soil, it survived in soils containing 414 – 959 mg^{kg-1}. In subsequent studies, vetiver survived and showed noticeable growth when planted in soil amended with 450 mgkg^{-1} and 500 mgkg^{-1}. Arsenic distribution to the various parts of vetiver is quite low as it accumulates mostly in the roots. When vetiver was cultivated in soil contaminated with 125 mgkg^{-1}, the concentration of As in the root system after 45 days of experiment was 9.78 mgkg^{-1} dry weight compared to 0.53 mgkg^{-1} in shoots. In China when it was cultivated in soils containing 100 $mgkg^{-1}$ dry soils for 3 months, about 3 $mgkg^{-1}$ were in the shoots, whilst roots contained about 13 $mgkg^{-1}$.

The potential arsenic uptake of vetiver can be significantly improved by using organic amendments or chelating agents. Vetiver grew well in soil containing up to 500 $mgkg^{-1}$ as dairy waste. After six months the total arsenic accumulated in vetiver reached 286 $mgkg^{-1}$in which the amount of arsenic accumulated in the roots (185.4 $mgkg^{-1}$) was higher than that in leaves (100.6 $mgkg^{-1}$).

Results from a Chinese study in 2007 showed that among three tested chelating agents (EDTA, HEDTA and oxalic acid) oxalic acid had the most significant effect on As accumulation in the aboveground parts of vetiver with a threefold increase compared with the control. Although, vetiver is not a As hyper-accumulator, it can be used to remove heavy metals from contaminated sites and can be disposed safely elsewhere, thus gradually reducing the contaminant levels.

Threshold for Arsenic is between 250-500mg/kg

Vetiver roots at various As level

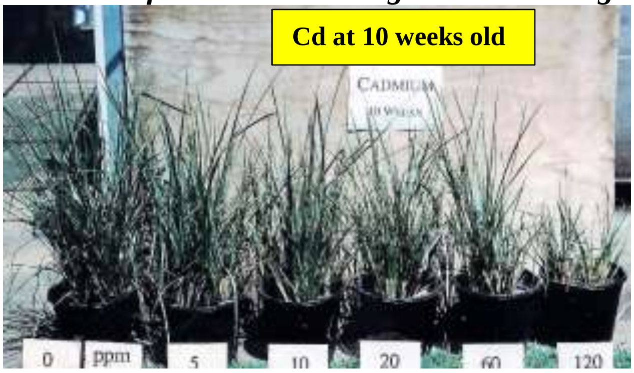


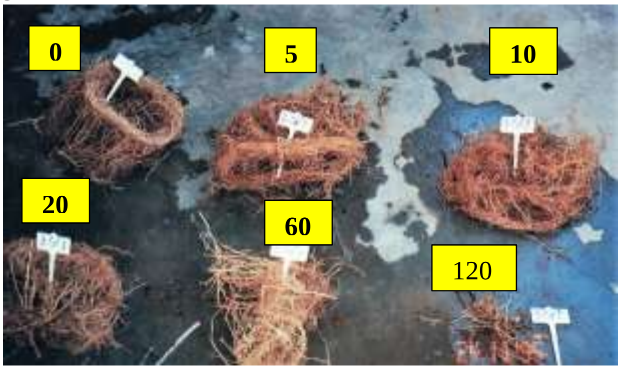
- ***Cadmium.*** Cadmium is very toxic to plant growth. The general threshold level of Cd in soil to plant growth is about 1.5 $mgkg^{-1}$. However, vetiver is tolerant to high Cd concentrations in soil, it has a cadmium threshold level of up to 60 $mgkg^{-1}$. It survived in mine tailing soils containing 32 $mgkg^{-1}$. Moreover, it grew in soils spiked with 10, 20 and 40 mg/kg Cd and showed satisfactory development with 100% survival. The accumulation of Cd in vetiver roots and shoots increased with the increase in the concentration of Cd in the soil and exposure time. At 70 days exposure time, Cd in the roots and shoots of vetiver increased from 0.1417 to 0.2252 $mgkg^{-1}$ and 0.1114 to 0.1522 $mgkg^{-1}$, respectively as Cd concentration in soils increased from 10 to 40 $mgkg^{-1}$. The same pattern was found at exposure times of 30 and 50 days. The results also indicated that longer cultivation times on Cd contaminated soils corresponded to higher concentrations of Cd in vetiver roots and leaves. The Cd level in roots and shoots of vetiver grown in soils amended with 40 $mgkg^{-1}$ was 0.2252 and 0.1522 $mgkg^{-1}$, respectively at 70 days of treatment compared to 0.0909 $mgkg^{-1}$ in roots and 0.0753 $mgkg^{-1}$ in shoots at 30 days of treatment.

The accumulation of Cd in vetiver roots was much higher than in shoots. Vetiver grown in soils contaminated with 0.58 – 1.66 $mgkg^{-1}$ accumulated 7.77 – 14.2 $mgkg^{-1}$ Cd in the roots and very little Cd in the leaves (0.13 – 0.58 $mgkg^{-1}$). In addition, Cd was not detected in the shoots of vetiver grown on mine tailing soils contaminated with 32 $mgkg^{-1}$ of Cd whilst roots had 4.98 $mgkg^{-1}$ after a growth period of 20 weeks. In other studies conducted on shale oil tailings and contaminated farm lands the ratio between the Cd content in vetiver roots and in vetiver shoots varied between 2 and 3, respectively. The highest concentration of Cd in vetiver shoots cited in the literature is nearly 25 $mgkg^{-1}$.

The application of EDTA did not effectively increase concentration of Cd accumulated in roots and shoots. The Cd content of vetiver roots (8.1 $mgkg^{-1}$) and shoots (3.9 $mgkg^{-1}$) under treatment of 6 mmol/kg was nearly equal to value obtained in roots (10 $mgkg^{-1}$) and shoots (3.7 $mgkg^{-1}$) under treatment without addition of EDTA. The addition of organic matter did not decrease Cd uptake in vetiver.

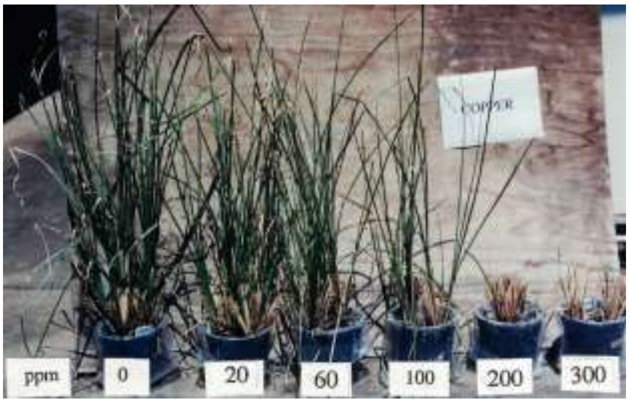
Threshold for Cadmium is higher than 120mg/kg *Vetiver roots at various Cd level*

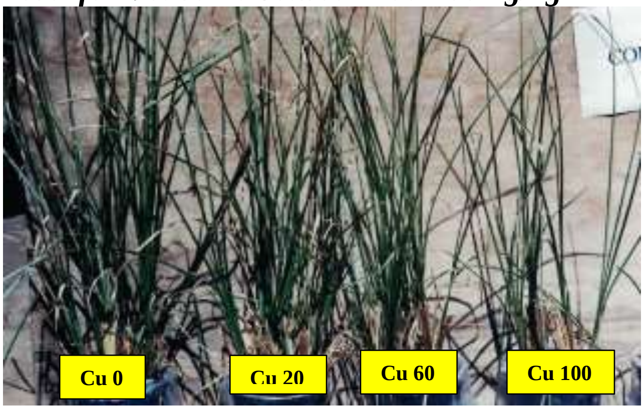




- ***Copper.*** Copper is an essential plant nutrient but becomes toxic to plants at high levels in the soil, level between 35-50 mgkg^{-1}are toxic to most plant**.** Results indicate that the critical soil level for vetiver is between 50 and 100 mgkg^{-1} which is very high compared with the threshold of between 0.5 and 8.0 mgkg^{-1}. Reasonable growth continued at a soil Cu level of 100 mgkg^{-1}.

Cu is highly toxic to plant, the Cu toxic level for Vetiver is between 100-200mg/kg





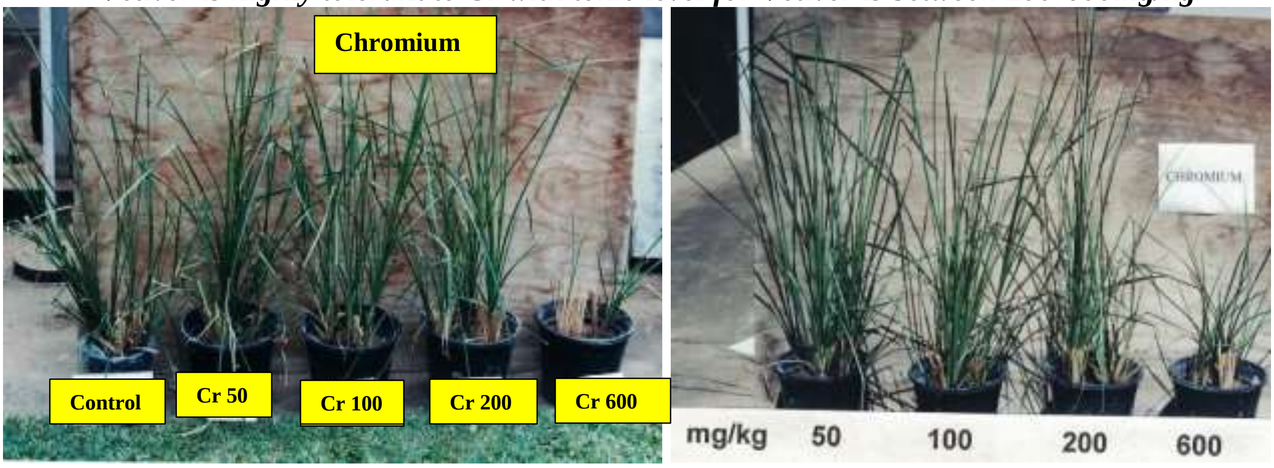
Vetiver tolerates high concentrations of Cu in soils as it survived in soils contaminated with up to 1762 mgkg^{-1}. In addition, vetiver survived and grew on Cu mine tailing soils that contained concentration of Cu as high as 1084 mgkg^{-1} and under daily irrigation with 250 ml of 190 mgkg^{-1} Cu solution for 30 days. In Chile, at the La Africana mine vetiver could grow on copper tailings with total Cu level at 3921 mgkg^{-1}and at the Anglo American mine (altitude 2000m) with 2600 mgkg^{-1} of total Cu in the tailings. The accumulation of Cu in vetiver roots and shoots is quite low. Vetiver grown on Pb/Zn mine tailing soils containing 35 mgkg^{-1} showed Cu uptakes in roots and shoots equal to 58.7 and 3.7 mgkg^{-1}, respectively, after 20 weeks of cultivation. After nine months of growth on firing range soils, vetiver accumulated 820.6 and 39.3 mgkg^{-1} of Cu in roots and shoots, respectively. Vetiver grown on Cu mine tailing soils accumulated 330 mg kg^{-1} Cu in roots and 10 mg/kg in shoots after 4 months of cultivation. Results at the Anglo American El Solado mine showed that 4 month old vetiver plants had 69 mgkg^{-1} in the shoots and 371 mgkg^{-1} in the roots. Ten month old vetiver plants had 65 mgkg^{-1} in the shoot and 953 mgkg^{-1} in the roots indicating that older plant retained more Cu in the roots. Vetiver irrigated daily with 250 ml of 190 ppm Cu solution for 30 days absorbed about 900 and 750 mgkg^{-1} Cu in roots and shoots, respectively.

The Translocation Ratio of Cu from roots to shoots in vetiver is generally quite low, less than 10%. Cu uptake in vetiver roots and shoots can be improved by application of chelating agents. The influence of different chelating agents (CDTA, EDTA, EGTA, citric acid, malic acid, HEDTA, HEIDA, NTA and DTPA) was tested and the most effective additive was HEIDA, which provided Cu solubilities 2.5 to 34 times higher than the other chelating agents when applied at a rate of 20 nmol/kg.

The addition of HEIDA increased the concentration of Cu in both roots and shoots up to 4 times compared to controls. EDTA was also effective in increasing the content of Cu in roots 2-3 times compared to controls. However, the addition of organic matter such as manure compost and sewage sludge significantly decreased the concentration of Cu in roots and shoots of VZ grown on Pb/Zn tailings in China.

- ***Chromium.*** Vetiver growth becomes significantly affected by soil Cr at 600 mg/kg. Its critical level in the soil is between 200 and 600 $mgkg^{-1}$ which is extremely high compared to the threshold of between 0.5 and 10.0 $mgkg^{-1}$ reported for other plants.

Vetiver is highly tolerant to Cr with toxic level for Vetiver is between 200-600mg/kg



Vetiver was shown to survive on canal sludge containing 2290 $mgkg^{-1}$ that is the highest level for Cr reported in literature with respect to Cr tolerance. It survived and grew well under daily irrigation with 250 ml of 623 $mgkg^{-1}$ Cr for 30 days. The majority of Cr accumulation in vetiver is located in the root system. Vetiver grown in soils containing 50, 200 and 600 $mgkg^{-1}$ accumulated 404, 1170 and 1750 $mgkg^{-1}$ in the roots, respectively; whilst the concentration of Cr in the shoots was very low: 4, 5 and 18 $mgkg^{-1}$ respectively. The time dependency of Cr accumulation in roots and shoots varies. It was observed that whilst Cr level in vetiver roots had a threefold increase over 30 days, Cr uptake in leaves was constant over time.

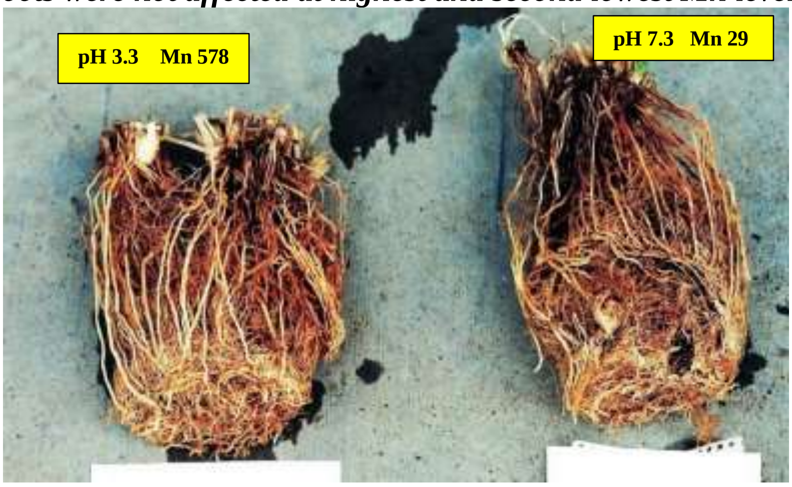
- ***^{137}Cesium and ^{90}Strontium.*** Human activities, such as mining and milling of nuclear fuel, nuclear weapon testing and occasional nuclear disasters (Chernobyl accident) release radionuclides, such as ^{137}Cs and ^{90}Sr, in the soils and water. Radionucleotides from soils and water can enter the food chain and be transferred to humans. The ability of vetiver to remove ^{137}Cs and ^{90}Sr from solutions spiked with individual radionuclide (5×10^3 k Bq L^{-1}) was investigated in India in 2008. After 168 hours of treatment, 61% of ^{137}Cs and 94% of ^{90}Sr could be removed from solutions. As both ^{137}Cs and ^{90}Sr were supplemented together in the solution, 59% of ^{137}Cs and 91% of ^{90}Sr were removed in the same time frame. The locations of ^{137}Cs and ^{90}Sr accumulated in vetiver are different as ^{137}Cs accumulated principally in the roots, whilst ^{90}Sr mostly in shoots. The addition of potassium ion (K^+) and calcium ion (Ca^{+2}) decreased the removal of ^{137}Cs and ^{90}Sr in the presence of vetiver, respectively. ^{90}Sr is an analogue of Ca^{2+} in living organisms, while K^+ ion is a member of the same homologous series to which Cs^+ belongs. When grown in low level nuclear waste solution (7.5×10^4 Bq L^{-1}), vetiver could efficiently remove radionuclides to below detection levels within 15 days. The results indicated that vetiver is a potential candidate plant for the phytoremediation of ^{137}Cs and ^{90}Sr.

- ***Manganese.*** Behind exchangeable Al, Manganese is the major toxic elements in most acid soils. Experimental results from glasshouse studies show that when adequately supplied with nitrogen and phosphorus fertilizers, vetiver can grow in soils with extremely high acidity and manganese levels. Vetiver growth was not affected and no obvious symptoms were observed when the extractable manganese in the soil reached 578 mgkg^{-1}, soil pH as low as 3.3 and plant manganese was as high as 890 mg^{kg-1}. Bermuda grass (*Cynodon dactylon*) which has been recommended as a suitable species for acid mine rehabilitation, has 314 mgkg^{-1}of manganese in plant tops when growing in mine spoils containing 106 mgkg^{-1} of manganese.

***Threshold for Mn is higher than 578* mg^{kg-1}**

Vetiver roots at various Mn levels

Vetiver roots were not affected at highest and second lowest Mn levels



- ***Nickel***. Although nickel is considered a trace element for plants and is a constituent of the important enzyme urease, it is extremely toxic to plants at high concentrations. The toxic threshold level of Ni in the soil has been reported between 7 and 10 mgkg^{-1}for most plants, but in this series of trials, 58% of vetiver growth still occurred at soil concentrations of 100 mgkg^{-1}.
- ***Lead.*** Vetiver is tolerant to extremely high levels of lead (Pb) in soil. Vetiver was shown to have 100% survival rate and had very good growth performance in high Pb concentration soils: 10,750 mgkg^{-1} in artificially contaminated soils and 9,020 mgkg^{-1} in naturally lead contaminated mine

tailings. These values were the highest soil Pb concentrations reported in the literature for Pb tolerance of vetiver. A study in China, showed vetiver survived and had noticeable growth in soils with lead concentration of 5,000 mgkg^{-1}. Similar results were obtained when it was cultivated in mining tailings contaminated with 3,281.6 mgkg^{-1} and 4,164 mgkg^{-1}. Lead concentration in the shoots and the roots of vetiver increased significantly with increasing Pb concentration in the soils. This study also showed the level of Pb in shoots increased from 0.82 to 43.0 mgkg^{-1}, and in the root from 60 to 556 mgkg^{-1} as vetiver was grown in 500, 2,500 and 5,000 mgkg^{-1} Pb amended soils. In Thailand the concentration of Pb increased from 12.5 to 375 mgkg^{-1} in shoots and from 18.7 to 4,940 mgkg^{-1} in the roots as vetiver was grown in soils amended with 0, 100, 1000 and 10000 mgkg^{-1}of Pb.

Experiments in Thailand have proven that vetiver is a lead hyper-accumulator. A plant is a Pb hyper-accumulator if it can accumulate Pb at least 0.1% of biomass equivalent to 1,000 mgkg^{-1} Pb. Most of the absorbed lead in vetiver tends to accumulate in the roots and generally only a small portion of lead is moved to the shoots. Vetiver grown in soils amended with 10,000 mgkg^{-1} accumulated up to 4,940 and 359 mgkg^{-1} Pb in the roots and shoots respectively with root/shoot ratio about 13.8 times. From this information, it can be estimated that vetiver accumulated Pb over 0.22% of its total dry matter.

The accumulation of Pb and its translocation from the roots to the shoots in vetiver can be improved by the addition of chelating agents. Chelating agents increase the mobility and bioavailability of the metal in soils and they also increase its accumulation in the upper parts of plants. Among chelating agents, EDTA has been shown to be the most efficient in mobilizing Pb from various soils. The increase in Pb translocation from shoots to roots by application of EDTA is potentially useful for the phyto-extraction of lead from contaminated soils

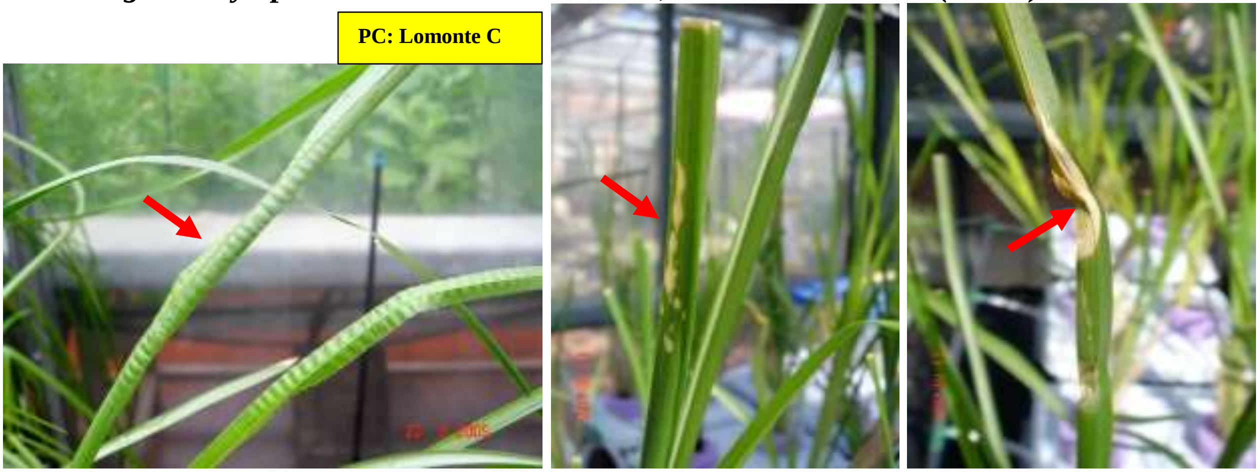
Vetiver growth was not affected at Pb level of 800mg/kg in the soil

- ***Mercury.*** Ines Sepulveda (pers.com.) from Chocó Quibdó, Colombia, subjected vetiver to a “lethal” concentration of mercury chloride, which would kill other species in a few hours, without any nutrients. Observation over the treatment time of 60 days, without water change, with rather low light intensity, and without nutrients noted only a slight yellowing of vetiver shoots. The vetiver roots grew vigorously suspended in the toxic solution and also tyhere was some foliage sprouting from these suspended plants.

Vetiver is highly tolerant to Hg with toxic level for Vetiver is more than 6mg/kg

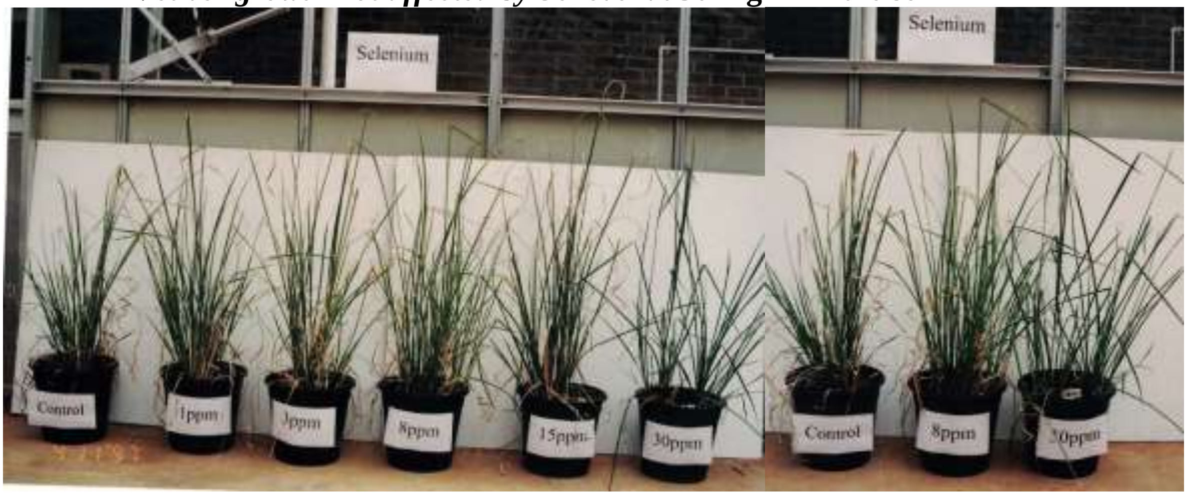
Lomonte et al (2013) of Melbourne University, Australia studied the spatial distribution of Hg in roots of vetiver grass by micro-Proton Induced X-ray Emission (PIXE) spectrometry to gain a better understanding of Hg uptake and its translocation to the aerial plant parts. When tillers of vetiver were grown in a hydroponic culture for 3 weeks under controlled conditions and then exposed to Hg for 10 days with or without the addition of the chelators $(NH4)^2S^2O^3$ or KI, they found that Hg was mainly localized in the root epidermis and exodermis, tissues containing suberin in all Hg treatments. Hg at trace levels was localized in the vascular bundle when plants were treated with a Hg solution only. However, higher Hg concentrations were found when the solution also contained chelators $(NH4)^2S^2O^3$ or KI. This finding is consistent with the observed increase in Hg translocation to the aerial parts of the plants.

Hg toxic symptom on Vetiver leaves – mild, medium and severe (L to R)



- ***Selenium.*** Vetiver is highly tolerant to Se toxicity, while the critical level for most plant is between 7-14 $mgkg^{-1}$ in the soil while, the critical level for vetiver was reported to be more than 74 $mgkg^{-1}$.

Vetiver growth not affected by Se level at 30 mg^{kg-1} ***in the soil***



- ***Zinc.*** The tolerance of vetiver to zinc in soils has been tested in China at contamination levels between 1,583 and 4,377 $mgkg^{-1}$. Vetiver survived at all tested contamination levels and had a good growth in soils containing up to 2,472 $mgkg^{-1}$ of Zn. Vetiver can accumulate high concentrations of Zn in the roots and shoots. Uptakes of Zn higher than 10,000 $mgkg^{-1}$ were recorded in shoots and roots after 30 days of irrigation with a Zn solution.

The heavy metal translocation from roots to shoots was also dependent on the fraction of water soluble Zn. Generally, the majority of Zn accumulated in vetiver is retained in the root system. However, when a high level of soluble Zn in soils was used, translocation ratios as high as 100% of Zn from roots to shoots were recorded. A possible approach to the enhancement of Zn bioavailability is the application of chelating agents. Among the chelating agents tested for the improvement of the water solubility of Zn in soils, nitrilotriacetic acid (NTA) was the most effective. The addition of NTA affected Zn uptake in vetiver. The application of 20 mmol/kg of NTA increased the concentration of Zn in the roots and shoots about 53% and 136%, respectively compared to the control treatment, whilst the use of EDTA did not effectively increase Zn content in the shoots and roots.

Vetiver growth was not affected at Zn level of 180* mgkg^{-1} *in the soil

Zinc levels in vetiver shoots and roots

Soil Concentration (mgkg^{-1})	Shoot Content (mgkg^{-1})	Root Content (mgkg^{-1})	Shoot/root Ratio (%)
118	4	109	**3**
180	25	173	**14**

- ***Multi-heavy metals.*** Glasshouse and field studies have demonstrated that vetiver is tolerant not only to high concentrations of individual heavy metals in soils but also of combinations of several heavy metals. In glasshouse studies, vetiver could survive and grow well on contaminated soils containing total Pb of 1,155 – 3,281.6 mgkg^{-1}, Zn of 1, 18.3 – 1,583 mgkg^{-1}, Cu 68 – 1,761.8 mgkg^{-1}. In field studies, vetiver survived cultivation on mine tailing soils containing total Pb of 2,078 – 4,164 mgkg^{-1}, Zn of 2,472 – 4,377 mgkg^{-1}, Cu of 35 - 174 mgkg^{-1} and Cd of 7 - 32 mgkg^{-1}. Organic matters (domestic refuse and sewage sludge), inorganic fertilizers and especially the combination of organic matters and inorganic fertilizers greatly enhanced the survival rates, growth and biomass increase of vetiver cultivated on soils contaminated by combinations of heavy metals.

Lim (2015) and Hasan et al (2016) conducted a study to assess the removal efficiency of heavy metals (Cu, Fe, Mn, Pb, Zn) using vetiver grass at different root lengths and densities. It also determined metal uptake rates by plant parts (root and shoot) between treatments (low and high concentration). Removal efficiency for heavy metals in water by vetiver is ranked in the order of Fe>Pb>Cu>Mn>Zn. *Results showed that vetiver was effective in removing all the heavy metals, but removals greatly depended on root length, plant density and metal concentration*. Longer root length and higher density showed greater removals of heavy metals due to increased root surface area for metal absorption. Results also demonstrated significant difference of heavy metals uptake in plant parts at different concentrations indicating that the roots have high tolerance towards elevated concentration of heavy metals. However, the effects were less significant in plant shoot suggesting that metals uptake were generally higher in the root than in the shoot. The findings have shown the potential of vetiver in phytoremediation for heavy metals removal in water thus providing significant implications to the ability to purify metal-contaminated water.

Vetiver grass could withstand concentration of heavy metals up to 10 mgkg^{-1}

The order of heavy metal contents in the shoot is: Pb>Fe>Mn>Cu> Zn. Pb has the highest concentration of 2,341 mgkg^{-1} in the shoot at 24 hours (Day 1), instead of Fe with concentration of 2,140 mgkg^{-1} at 168 hours (Day 7). Not only that, Mn content was the third highest with 971 mgkg^{-1} at 0 hour (Day 0), followed by Cu with 655 mgkg^{-1} and lastly Zn with 45 mgkg^{-1} at the early stage of the experiment.

Heavy metal accumulation in plant after 10 days for low concentration (*mgkg^{-1}*)

Plant parts	Cu			Fe			Mn			Pb			Zn		
	10	20	30	10	20	30	10	20	30	10	20	30	10	20	30
Root	1445	1663	1703	2450	3780	3390	0	0	0	293	0	781	224	0	0
Shoot	112	106	142	910	710	650	128	0	0	976	195	195	186	110	0
Translocation (%)	7.19	5.99	7.69	28.8	15.8	16.09	-	-	-	76.9	100	19.98	45.4	-	-

Heavy metal accumulation in plant after 10 days for high concentration (mg/kg)

Plant parts	Cu			Fe			Mn			Pb			Zn		
	10	20	30	10	20	30	10	20	30	10	20	30	10	20	30
Root	835	1233	576	18890	55390	30680	0	0	0	1658	1756	781	76	0	0
Shoot	155	121	161	790	800	900	0	0	0	878	0	98	129	86	0

As in low concentration, metal content was more dominant in the root than in the shoot in high concentration solutions . As the solution mixture has a very high range of concentration, the plants have the ability to take up high amount of heavy metal ions in the roots. Furthermore, it can be seen that Mn and Zn have less or no accumulation in the shoot or root. This is because there was a significant Mn and Zn content in the control plants, which contained an average value of 2,147.3 mgkg^{-1} and 787.67 mgkg^{-1}; and 995.33 mgkg^{-1} and 204 mgkg^{-1} in root and shoot respectively.

Summary and general conclusion of Vetiver under heavy metals stress

From the above data, it is very clear that vetiver is highly tolerant to heavy metal toxicity. It can withstand high concentration of these elements both in the soil or effluent and also internally - within the plant. A very high proportion of these metals are retained in the root. Photographic records show very good root systems even in plants with poor shoot growth.

As the Shoot:Root biomass ratio of vetiver generally varies between 1:1 and 1.2:1.0 in mature and intact plants under field conditions, the general mass of vetiver root can be estimated from the above ratio.

Threshold Levels of Heavy Metals to Vetiver Growth (compared with other species)

Heavy Metals	Threshold Levels in the Soil ($mgkg^{-1}$)		Threshold Levels in the Plant ($mgkg^{-1}$)	
	Vetiver	Other Plants	Vetiver	Other Plants
Arsenic	100-250	2.0	21-72	1-10
Cadmium	20-60	1.5	15-18	5-20
Copper	50-100	na	13-15	1.5
Chromium	200-600	na	5-18	0.02-0.20
Lead	>1,500	na	>78	na
Mercury	>6	na	>0.12	na
Nickel	100	7-10	347	10-30
Selenium	>71	2-14	11	Na
Zinc	>750	na	880	na

na:not available

SOIL MECHANICAL RESISTANCE AND ROOT GROWTH AND FUNCTION

Roots experience mechanical impedance due to the force required to displace soil particles as they elongate. Mechanical impedance (or soil strength) is the most ubiquitous constraint to root growth. Strong soil can be a serious agricultural problem, as the ability of the root system to access water and nutrients from the deeper soil layers is restricted. Soil is a complicated material and its properties are affected by land management and also by the roots themselves. This section describes how roots are able to penetrate strong soils (Gregory, 2006).

Root Penetration and Bending Stiffness

To elongate through soil where existing channels are smaller than the root diameter, roots must exert a growth pressure (which results from turgor pressure and cell wall relaxation) to deform the soil around the root. When roots approach a layer of strong soil, they may either penetrate the layer or may get deflected from their original direction. The mechanisms that determine the outcome of such an encounter are not fully understood. If the root does not have sufficient lateral support, bending of the root may occur above the strong layer. It has long been known that roots of some species are better at penetrating strong soil than others. In particular, dicots have better root penetration than monocots. But as we have discussed, dicots do not have greater maximum axial growth pressures than monocots, which implies that differences in growth pressure do not account for differences in root penetration ability. It has been suggested that species with thicker roots gave better penetration because they were more resistant to buckling.

There are suggestions that good root penetration results not from high growth pressures but from the roots resisting bending as they encounter a hard layer. There is an approximately fourth-power relationship between root diameter and bending stiffness.

It can be concluded that ***e****ven in relatively in well-watered soils, mechanical impedance to root elongation often reduces root elongation rates. In contrast to the common assumption that only compacted soils are strong, we show that even relatively loose soils often become strong as they dry. Soil type (e.g., sand or clay) and condition (e.g., lose or compact) determine the relative balance of water stress and soil strength and the extent that these limit root and crop growth for any given season. Although roots exert surprisingly high pressures, there is little evidence for genotypic variability in these pressures. Instead, arability in root penetration of strong soils is probably linked to variation in the biomechanical properties of individual root tips (e.g., root bending stiffness, root thickening, frictional properties of the root cap). The effects of high soil strength on root growth can be observed in relatively wet soils with little limitation on water availability. Thus the effects of soil strength on root elongation and plant growth are frequently overlooked, despite soil strength often being the major limitation to root growth in agricultural soils*. (Gregory, 2006).

Vetiver Root as a Bio-Engineering Option (Hengchaovanich, and Nilaweera (1998), Hengchaovanich, (1998, 1999).

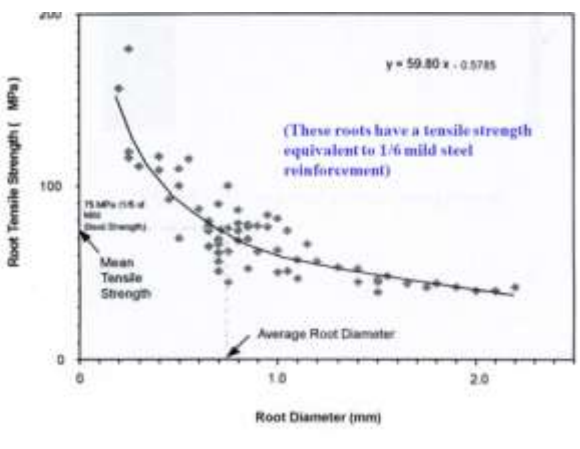
Bioengineering, or strictly speaking soil bioengineering, is a relatively new sub-branch of civil engineering. It attempts to use live materials, mainly vegetation, on its own or in integration with civil engineering works to address the problems of erosion and slope stabilization. In the late 1980s and in the following decade, due to heightened awareness of environmental issues and availability of knowledge and parameters of plants that can aid as well as lend credence to the designs, bioengineering became better known and accepted.

Over the millennia, nature has "designed" vegetation as a means to blanket and stabilize the good earth. In the tropical and subtropical regions, this has evolved into forests of big trees, shrubs and leaf litters covering the organic humus-rich topsoil and offering excellent overall protection. The re-vegetation of slopes can be by means of grassing or leguminous cover crops (for minor surface movement) or the use of fast-growing shrubs and trees for the mitigation of deep-seated erosion in the order of 20-150 cm depths. Tree or shrub roots are able to grip and bind the soils needed to prevent the deep-seated surface slips in the event of heavy and prolonged rainstorms, while normal grasses are unable to do so. This is because roots or "inclusions" impart apparent cohesion (c_r) similar to "soil nailing" or "soil doweling" in the reinforced soil principle, thus increasing the safety factors of slopes permeated with roots vis-à-vis no-roots scenario.

Notwithstanding their virtues, trees and shrubs inherently have several drawbacks in that they are too slow to establish to become effective (even with fast-growing species this process will take about 2-3 years) and are in danger of being uprooted, in cases of heavy storms, typhoons or cyclones. Vetiver, although is a grass, does possess several tree-like features. It therefore becomes an attractive alternative to trees or shrubs when it comes to bioengineering applications. In January 2000, the journal of the International Erosion Control Association (IECA) published an article featuring the attributes of vetiver grass as a bio-engineering option. First, it has to do with the unique characteristics of the grass itself. For the sake of completeness, its main characteristics are reiterated here below:

- *The grass grows upright and is able to form a dense hedge within 3-4 months, resulting in the reduction of rainfall runoff velocity and formation of an effective sediment filter. The hedgerow can adjust itself in tandem with trapped silt by forming new tillers from nodes on the culm of higher branches, thus ensuring that it will never be buried alive.*
- *It has a vigorous, massive and dense subterranean root system that reaches vertically 2-5 m depth depending on soil types.*
- *The roots are very strong compared to other hardwood species as shown in Table below, having average tensile strength of 75 MPa or approximately 1/6th of mild steel.*

Tensile strength of vetiver roots ***Tensile strength of vetiver roots***

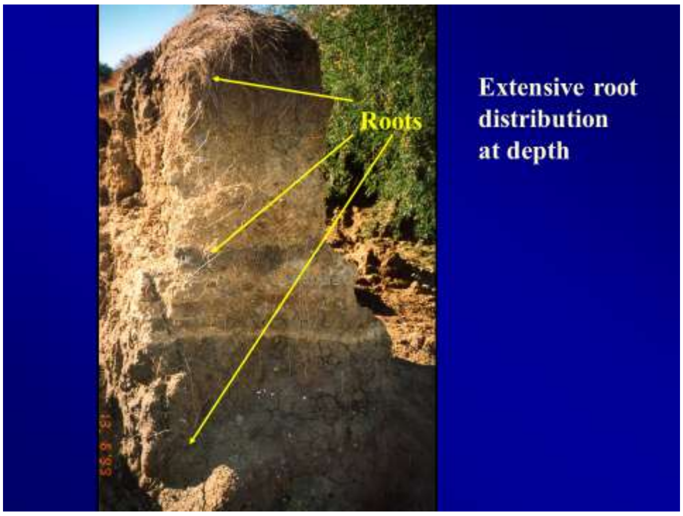


Tensile strength of vetiver roots as compared with other plants

Botanical name	Common name	Tensile strength (MPa)
Salix spp.	Willow	9-36
Populus spp.	Poplars	5-38
Alnus spp.	Alders	4-74
Pseudotsuga spp.	Douglas fir	19-61
Acer sacharinum	Silver maple	15-30
Tsuga heterophylia	Western hemlock	27
Vaccinum spp	Huckleberry	16
Hordeum vulgare	Barley Grass, forbs Moss	15-31 2-20 2-7kPa
Chrysopogon zizanioides	**Vetiver grass**	**40-120 (Average 75)**

Strong vetiver root reinforcement protect this soil column against water erosion

Strong root penetrating several layers of compacted dam wall

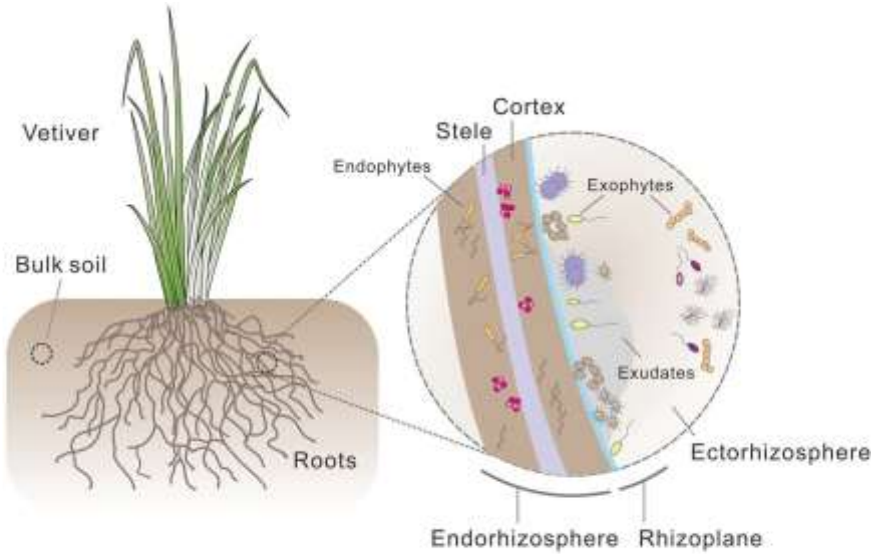


INTERACTIONS OF ROOTS WITH SOIL ORGANISMS

The rhizosphere is a zone that is densely populated with soil organisms, including bacteria, yeasts, fungi, protozoa and insects, all feeding on a wide range of substrates. Much research has now demonstrated that compounds released from roots may act as messengers that communicate and initiate interactions between roots and a wide range of soil-dwelling organisms.

Chen et al (2020) carried out an exhaustive review on the interactions between Vetiver grass and soil microbes for environmental protection. *They stated that the ability of Vetiver in stress tolerance is attributed to its genotype and phenotype, which may be strongly related to the associated microbes, including endophytes and exophytes, feeding on the root exudates and root essential oil. Therefore there is a need to systematically review the rhizospheric microbes associated with Vetiver, in particular their interactions, in fulfilling its economic and ecological capabilities.*

The following Figure shows different zones of the soil-Vetiver system that harbor various endophytic and exophytic communities. The root-soil system consists of microbes within the root (endorhizosphere), on the root surface (rhizoplane), in the rhizospheric soil (ectorhizosphere) and those in the bulk soil. Bacterial communities are mainly governed by soil properties, plant identity and symbiotic fungi.



This article stated that plant-microbe partnerships are essential for the host plant to overcome stress responses. The microbes associated with roots consist of those within the root, on the root surface and those in the bulk soil. Evidence showed that our current concepts of categorizing microbes as pathogenic or beneficial are in its infancy. Bacteria designated as pathogens can be beneficial when confronted with other stresses such as salt, drought and heat. Several plant species such as alfalfa and Arabidopsis require microbial associations for their survival under biotic and abiotic stresses.

The major objectives of this article are to review progress made in the application of Vetiver and associated microbes, under the topics of Phytostabilization, Rhizofiltration, Phytoextraction, Rhizobiodegradation and others. These topics emphasize different strategies in enhancing its efficiency in bioengineering, phytoremediation by extracting toxic metal(loid)s from soil at the sites, purifying organic matters and metal(loid)s from wastewater and acid mine drainage, and dissipation of POPs. Characteristics of Vetiver and the associated microbe subjected to other stresses, including acidity, salinity and drought are also discussed. For each section, studies without involving microbes are reviewed and discussed, followed by those involving microbes. Information about Vetiver-microbe interactions to remediate co-contaminated soils are given. The microbes associated with oil production (potentially related to the degradation of OPs) by Vetiver are also reviewed. Mechanisms *of Vetiver-microbe interactions in abiotic stress tolerance and phytoremediation are subsequently discussed. Finally, a summary and prospects are given*. (Chen et al. 2020)

***Phytostabilization* (*Bioengineering*)**

Only limited information on the effects of vetiver-associated microbes on soil erosion could be found. When inoculating Vetiver roots with fungi taken from the roots of maize, early growth and establishment of Vetiver plants were improved. It is believed that this may be the first published report related to the use of Vetiver and arbuscular mycorrhizal fungi (AMF), for the rehabilitation of severely eroded and degraded sites.

AMF have important contributions to soil remediation. Although there is limited information working on the role of mycorrhizal Vetiver in soil erosion control, root exudates and microbial organic matters (e.g., glomalin from fungi) facilitate the formation of water-stable aggregates. The root and shoot biomass enhanced by mycorrhizal symbiosis could further improve soil structure and prevent soil erosion. More on the role of mycorrhizal fungi (AMF) will be presented in the Interactions with Mycorrhizas section

Rhizofiltration Plant growth-promoting rhizobacteria (PGPR) probably improved Vetiver performance and enhanced its nutrient uptake (ammonium, nitrate and phosphate) in floating islands planted with Vetiver to treat excess nutrients in agricultural and municipal runoffs.

Phytoextraction There are some good examples of raising the phytoextraction coefficiencies and translocation factors by Vetiver when rhizosphere bacteria were involved. For instance, inoculation of Cd-resistant bacteria, Ralstonia sp. and Arthrobacter sp., produced exopolymers which promoted soil Cd solubilization. It subsequently enhanced Cd accumulation in the roots and shoots of Vetiver. Inoculation of Bacillus cereus strain T1B3 increased the concentrations in the roots and shoots of Vetiver.

A pot trial further demonstrated that inoculating AMF (Glomus mosseae and G. intraradices spores) to Vetiver significantly increased the growth and plant P uptake. In soils added with 0 or 10 mg kg^{-1} *of Pb or Zn, inoculation of AMF increased the plant Pb and Zn uptake; whereas in soils added with 100 or 1000 mg* kg^{-1} *of Pb or Zn, the inoculation decreased the uptake of both metals. A subsequent investigation was carried out in the field. Results showed that the addition of refuse compost resulted in biomass three times higher than the control, attributed to the improved soil properties and nutrient supply. Inoculation of AMF also significantly raised the dry mass of Vetiver by 8.1–13.8%, with higher concentrations of N and P in the shoots. Further work was conducted in the field to investigate the effects of compost and AMF on metal uptake of Vetiver. Using AMF alone decreased the Pb uptake of Vetiver; while using AMF and compost decreased the Zn uptake. These two studies demonstrated that AMF could restrict metal accumulation in Vetiver and assist the plant establishment and growth under harsh environments. A pot experiment was conducted to assess the effectiveness of AMF in phytoremediation of Pb-contaminated soil by vetiver. It was observed that mycorrhizal inoculation increased the uptake of Pb and P in the shoots and roots, compared to those without AMF. Furthermore, root colonization increased with AMF inoculation but decreased as Pb levels increased. It was concluded that AMF inoculation enhanced extraction, uptake and translocation efficiencies of Pb. In addition, plants associated with AMF had higher chlorophyll content but reduced levels of low molecular weight thiols, reflecting the ability to better survive under metal- induced stress. It was further noted that Vetiver inoculated with AMF (Glomus sp.) was more effective decontaminating As-contaminated water than soils, by comparing the uptake of As from contaminated hydroponic and soil systems.*

Rhizobiodegradation Rhizobiodegradation is the process in which OPs are degraded or transformed by biochemical reactions mediated by plant roots and the associated microbes. A study showed that Vetiver could potentially maintain the levels of microbial biomass carbon and microbial *biomass N in soil contaminated by diesel fuel. Another study also showed that rhizobacteria associated with Vetiver play a significant role in phenol degradation. Vetiver decreased the toxicity of phenol through rhizobacteria.*

The antibiotic tetracycline from water sources can be removed by Vetiver combined with tetracycline-tolerant bacteria associated with the root. Glutathione-S-transferase (GSTs), as an important group of enzymes in both plant and bacteria, was potentially involved in tetracycline degradation as Arthrobacter sp. strain MCM B- 436, an atrazine-degrading bacterium, was isolated from Vetiver rhizospheric soil. It was also shown that herbicide endosulfan adsorption was more considerable in soils planted with Vetiver than bare soils. The former was accompanied by a higher number of endosulfan-degrading microbes.

Other environmental stresses The role of Vetiver-microbe interactions in remediating acid sulfate, shallow and saline soils has been investigated. Vetiver increased the rhizospheric microbial populations, such as P solubilizing bacteria and mycorrhizal fungi, which significantly correlated with the improved soil properties including nutrient availability (N, P, calcium and sulphur), organic matter and moisture content. Rather than Vetiver, bacteria can benefit other plant species confronting with environmental stresses. Enhanced salt tolerance has been observed in Zea mays inoculated with Rhizobium and Pseudomonas. Enterobacter sp. SA187 produced 2-keto- 4-methylthiobutyric acid (KMBA) which modulates plant ethylene signaling pathway under salt stress.

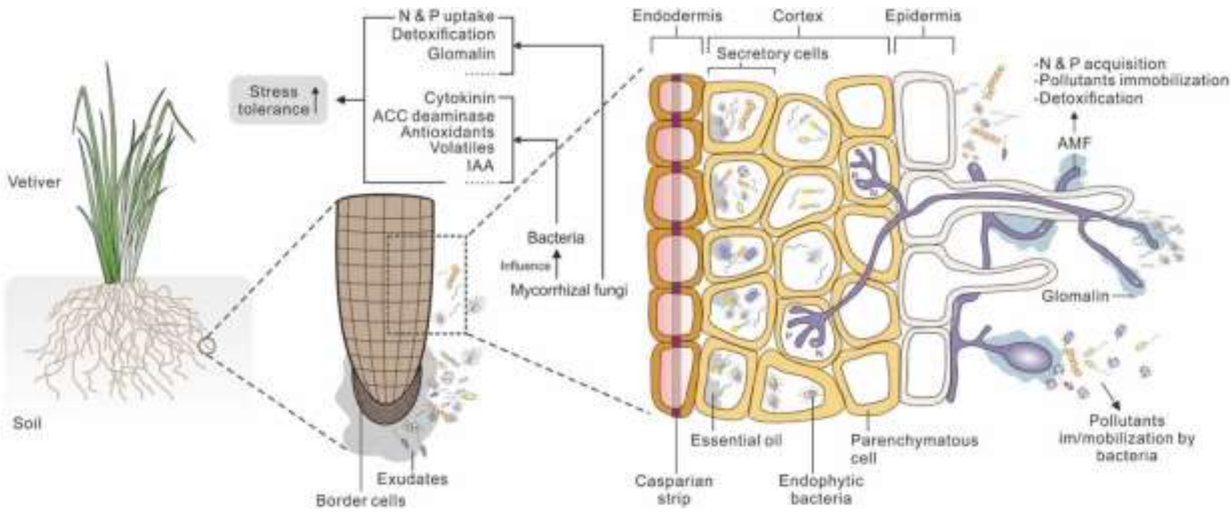
Although the beneficial effects of Vetiver-microbe interactions on phytoremediation have been widely reported, the mechanisms in achieving the benefits were seldom sufficiently illustrated. Based on these limited studies, the role of microbes in affecting performance of Vetiver in soil remediation could be attributed to:

- *Altered nutrients and metals phytoavailability,*
- *Enhanced plant stress tolerance via physical and/or biochemical protection, Promoted plant growth due to bacteria-induced hormones,*
- *Assisted in plant nutrient uptake.*

Highly contaminated soils Pollutants and microbes co-exist in multi factors contaminated soils. The critical soil properties such as pH, soil moisture content, C/N ratio, extractable P are important factors sculpting the microbial community. When Vetiver is involved, critical soil properties can change the chemical composition of the root exudates. For example, the patterns of low molecular weight organic acids (LMWOAs) of root exudates were significantly different among copper concentrations and plant species (Oenothera picensis, Imperata condensata, Lupinus albus and Helianthus annuus). The altered root exudates can attract/stimulate specific microorganisms to improve plant.

Essential Oil production The capability of Vetiver in degrading OPs could be attributed to its ability to produce and metabolize essential oil with the participation of endophytic bacteria. Vetiver has a unique ability among grasses to produce an essential oil in the root. The oil is a complex mixture of sesquiterpene alcohols and hydrocarbons, which is difficult to be reproduced synthetically. The quality of the oil depends on genetic, environmental and technological factors. The oil is produced in secretory cells in the first cortical layer outside the endodermis. Intriguingly, the bacteria within the cortical parenchymatous cells play essential roles in complex oil production. Endophytes of Vetiver roots were a-, b-, and c-proteobacteria, high -G C-content gram-positive bacteria, and those belonging to Fibrobacteres/Acidobacteria. Most of them could use oil sesquiterpenes as carbon sources.

The amount and composition of oil compounds produced by Vetiver varied and much depended on the bacterial community associated with its roots. Bacteria, including Pantoea sp., Acinetobacter sp., Pseudomonas aeruginosa, Bacillus sp., and Lysinibacillus sphaericus, contain ketosynthase gene, which is involved in Vetiver oil production. Isolation and selection of bacterial strains for promoting the growth of Vetiver, leading to improving its essential oil production would be feasible



Schematic illustration on Vetiver root-microbe interactions with the involvement of essential oil, arbuscular mycorrhizal fungi (AMF) and endophytic and exophytic bacteria. Oil generated within root secretory cells is metabolized by endophytes, which can possibly degrade aromatic compounds/pollutants. Rhizospheric bacteria, such as plant growth-promoting rhizobacteria (PGPR), produce indole acetic acid (IAA), cytokinin and other metabolites that change root morphology and transporter activities. These processes increase plant nutrient uptake. Mycorrhizal fungi and the sculpted bacterial community structure improve plant P and N uptake, detoxify and immobilize pollutants, e.g., arsenic and improve soil water retention due to generation of glomalin. Bacterial community is affected by soil type, plant identity and associated mycorrhizal fungi. Nutritional interactions among plants, bacteria and fungi involve a number of trophic events, which combinedly affect the performance of Vetiver in soil remediation.

Summary and Prospects.

Vetiver is a versatile plant which contributes significantly due to its diverse applications, for producing oil and other paramedical products. It also offers sustainable opportunities for carbon sequestration, because it grows fast, with higher biomass, and able to overcome a wide range of environmental stresses. The involvement of soil microbes often enhances the mitigation processes. With this microbial association the efficiencies for pollutant degradation, extraction, and stabilization will be further improved.

Future studies should focus on the interactions of Vetiver with rhizospheric microbes, to further enhance its ability to grow on various manmade habitats with highly stressed environmental conditions (laden with high concentrations of HMs and POPs). There is ample evidence showing AMF and bacteria confer tolerance to Vetiver against HM contamination, by improving stabilization of HMs in soil or enhancing uptake and transfer of such metals to plant, and increasing plant biomass for enhancing their removal; and also bacteria which can degrade organic contaminants effectively. There seems to be a need to identify suitable bacterial and AM fungal strains/species associated with Vetiver to deal with different contaminated substrates, which posed diverse problems.

EFFECTS OF HYDROPONIC MEDIUM ON MORPHOLOGY AND PHYSIOLOGY OF VETIVER ROOTS.

Jones (2014) mentioned that root size, measured in terms of length and extent of branching as well as color, is a characteristic that is affected by the nature of the rooting environment. Normally, vigorous plant growth is associated with long, white, and highly branched roots in hydroponic *medium. Normally, hydroponic plant roots do not have root hairs. Root hairs will be almost absent on roots exposed to a high concentration (100 $^{mgL-1}$) of NO^{3-}. High P in the rooting medium will also reduce root hair development, whereas changing concentrations of the major cations, K^{+}, Ca^{2+}, and Mg^{2+}, will have little effect on root hair development.*

Vetiver root development conforms to the above reference, in size, mass, length and extent of branching. The average length of its roots in hydroponic medium is between 60-80cm depending on temperature and aeration (dissolved O_2). Vetiver root can grow in very low nutrient levels in the tap water for example, and also flourishes in nutrient-rich polluted medium.

Vetiver root grown in tap water ***Polluted water***

PC: Moraes B

PC: TVNI Archive

Vetiver roots in nutrient solution. Note no root hair and darker in colour with age

Vetiver roots in sewage effluent treatment trial, colour changed with age

Sequence of vetiver root growth in a fish tank over time

PC: Millwood E

Vetiver pontoons for algal bloom control in swimming pond at a community pond

Five week old roots, note the proportion of shoot and root growth

Vetiver floating pontoons for sewage effluent treatment in Australia (L) and piggery wastewater pond in Vietnam (R)

Some other things that happened to vetiver roots

In Australia vetiver root on pontoons in sewage effluent treatment ponds was often eaten by eels, native fish and turtles. But its shoot growth was not greatly affected. It was reported that diving wild ducks also ate vetiver roots from pontoons in Argentina.

Roots eaten by aquatic wild life in Australia (fish, turtle and eels)

The following are the results of Vetiver floating pontoons for sewage effluent treatment in Australia

Effluent quality before and after vetiver treatment

Tests	***Sewage Effluent (inlet)***	***Discharged Effluent (2002/03)***	***Discharged Effluent (2004)***
PH (6.5 to 8.5)*	7.3 to 8.0	9.0 to 10.0	7.6 to 9.2
Dissolved Oxygen (2.0 minimum)*	0 to 2 mg/l	12.5 to 20 mg/l	8.1 to 9.2 mg/l
5 Day BOD (20 - 40 mg/l max)*	130 to 300 mg/l	29 to 70 mg/l	7 to 11 mg/l
Suspended Solids (30 - 60 mg/l max)*	200 to 500 mg/l	45 to 140 mg/l	11 to 16 mg/l
Total Nitrogen (6.0 mg/l max) *	30 to 80 mg/l	13 to 20 mg/l	4.1 to 5.7 mg/l
Total Phosphorous (3.0 mg/l max) *	10 to 20 mg/l	4.6 to 8.8 mg/l	1.4 to 3.3 mg/l
* License requirements			

Participating in the cultural event "Plantón Mobil" in Lima, December 2014 –

CHAPTER FOUR

APPLICATIONS OF VETIVER ROOT UNIQUE AND SUPERIOR ATTRIBUTES TO ENHANCE VETIVER SYSTEM TECHNOLOGY

VETIVER ROOT UNIQUE AND SUPERIOR ATTRIBUTES

The above cited literature and research results as well as field experience show that vetiver roots have two unique and superior attributes.

- **Physical attributes**: Vetiver has long, massive, penetrating and high tensile strength root system. When planted close together on contour, it forms a dense hedge which spreads and reduces flow velocity, hence erosion, and traps sediment in runoff water.
- **Physiological attributes**: It has extremely high tolerance levels to pollutants both on land and in aquatic sttings, including heavy metals which most plants cannot tolerate. Vetiver's deep and massive root system is also drought and water logging tolerant. Furthermore with its special shoot/leaves architecture, vetiver has a very high transpiration rate under wet conditions, hence reducing a given volume of polluted water.

VETIVER AS A BIOENGINEERING TOOL

The use of vegetation as a bioengineering tool for erosion control and slope stabilisation have been implemented for centuries but its popularity has increased in the last decades. This is partly due to the low costs of bio-engineering techniques, partly to the 'soft' vegetative approach and partly due to the increased knowledge and information on vegetation that is now available for application in engineering designs.

The factors contributing to slope stability of slope using vegetation are root reinforcement and evapotranspiration; the soil pore pressure being reduced; and soil shear strength being increased. These all contribute to the apparent soil cohesion and internal friction angle of soil particles.

Vetiver, a very fast growing grass, possesses some unique features of both grasses and trees by having a profusely growing, deep penetrating root system that can offer both erosion prevention and control of shallow movement of surfacial earth mass. Vetiver contributes to erosion control and slope stability by slowing down runoff, evapotranspiration and its root reinforcement of the soil. Vetiver roots are very strong with high mean tensile strength about 75 MPa at 0.7- 0.8mm root diameter. It was obvious that the penetration of vetiver roots in a soil profile increased the shear strength of the soil significantly. These attributes are applicable to soil under both dry and wet or inundated conditions. (Hengchaovanich, 1998, 1999; Hengchaovanich and Nilaweera, 1998)

Under Dry Land Conditions

With the high nutrients soils, vetiver slips can develop a completely dense hedgerow in the rainy season that can be effective after 4 months and its hedgerows act as a living wall which stands against and slows down runoff, and the eroded soil is deposited behind the vegetative barrier. Vetiver hedgerows have been found to be able to resist the scouring of water flow 0.028 m^{3s-1}. Under the vetiver hedgerows the root system interact with the soil forming a new composite material comprising roots with high tensile strength and adhesion embedded in a matrix of lower tensile strength (Hengchaovanich and Nilaweera 1998).

Vetiver stabilizes soil slopes not only by root reinforcement but by getting soil to dry out by evapotranspiration. In water saturated soil or on slopes with a high water table, vetiver's massive and deep roots could reduce moisture in the soil thus lowering pore water pressure. This situation will have beneficial effects on slope stability, especially for the 1-2 m depth soil layer that is prone to slippage (shallow-seated failure) or flows (Van and Truong, 2015).

Suched Likitlersuang et al (2020) demonstrated the effect of the vetiver root matrix on the soil slope focusing on the mechanical reinforcement. The vetiver grass specimens grown under one year were used in this study. The investigation program includes root observations and direct shear tests.

According to the root observation and the direct shear test results, they indicated that the vetiver root system can increase the shear strength of the soil by mechanical reinforcement. These results confirm a significant contribution of vetiver root to slope stability. The conclusions of this study are as follows:

- The roots significantly increase the shear strength of soil. The shear strength of the root-reinforced soil depends on the root length and the root area ratio.
- The growth rate of vetiver roots is relatively high compared to other plants. The maximal depth development of the vetiver root system could go up to 200 cm in the first year and the average daily increment of the roots is approximately 10 mm.
- The direct shear test results indicate that the vetiver roots significantly enhance the soil shear strength especially on the cohesion in the order of 1 to 6 kPa. However, the increases of cohesion are different depending upon the type and age of vetiver as well as the type of tested specimen.
- The increase of shear strength from vetiver root can be alternatively explained using an electron microscope technique. The root hairs are of the order of micron level and their interfacial area contributes significantly to the friction due to their increased surface area. This mechanism provides adhesion between the root and the soil during shear events which could be directly linked by the cohesion term in Mohr-Coulomb failure criterion framework.

Nguyen et al (2018) reported on the bioengineering method using vegetation is an ecological approach for slope stabilisation. However, due to a large variability of vegetation root patterns, a precise quantification of root reinforcement is relatively difficult, leading to a reluctance to use such a technique in practice. This study presents a probabilistic framework for slope stability analysis considering the spatial variability of root reinforcement. A residual soil slope under a heavy rainfall event was used to model the seepage and stability analysis. The effect of root reinforcement was considered through an additional soil shear strength or root cohesion. Typical characteristics of the root reinforcement of vetiver grass (*Chrysopogon zizanioides*) in Thailand were used in the analysis. A probabilistic analysis was performed considering both stationary and non-stationary random fields of root cohesion.

The results indicated that the failure of a vegetated slope could occur when the variance coefficient of the root cohesion was more than a critical value (a critical cov = 0.45 for the uniformly distributed root cohesion case and a critical cov = 0.32 for the case of linear decrease of root cohesion in this particular slope). In practice, the efficiency of the bioengineering method can be improved by controlling the variation of root cohesion within such limits.

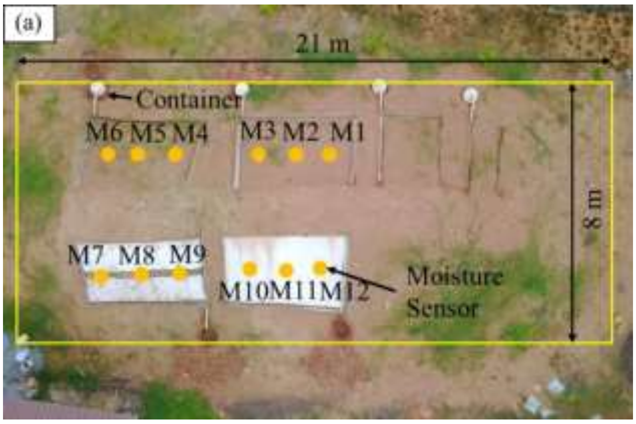
Suched Likitlersuang et al (2017) presented the results using a geotechnical centrifuge and numerical modeling study of root-reinforced systems on soil slopes. The centrifuge models were designed to simulate a soil slope reinforced by vetiver grass root system commonly grows in Southeast Asian countries. Unreinforced and root-reinforced soil slope models were subjected to heavy rainfall using a rainfall simulator designed for a geotechnical centrifuge. Results of the study show that the vetiver root system causes a reduction of the rainwater infiltration rate, a delay in the response of the groundwater table, and an increase in soil shear strength. The results were validated by comparing centrifuge modeling test results with numerical modeling analyses based on limit equilibrium and finite element methods. The transient seepage analysis results were employed in the slope stability analysis. The numerical analysis shows a good correlation with the observations from centrifuge modeling tests wityh respect to failure mechanism.

Suched Likitlersuang et al (2020) investigated the role of different land surface covers on soil erosion caused by rainfall, particularly in mountainous areas, causing landslide hazards on these terrains. In a field experiment where several types of land covers were placed on a full-scale embankment for erosion control. An 8m wide, 21m long, and 3m high embankment with a 45º side-slope on a lateric soil. The soil was compacted under a relative compaction of 70% to simulate a natural soil slope. Two sides of the embankment were divided into six land cover areas, with three different areas of bare soil, and one each of a geosynthetic cementitious composite mat (GCCM), vetiver grass (planted at the recommended 15-cm spacing) and a combination of GCCM and vetiver. Soil erosion and moisture levels were monitored for each land cover area during six natural rainfall events encountered over the experimental period. Field results were compared with a numerical simulation and empirical soil loss equation. The results revealed that the GCCM gave the best erosion control immediately after installation, but vetiver grass also exhibited good erosion control six months post construction.

The area covered by vetiver did not show any substantial resistance against erosion in the short term, but exhibited a high degree of erosion protection in the long-term. This is due to the immaturity of vetiver, it needs time to grow to be effective. However, by the time of the 4th storm (441 days post construction) the vetiver had grown enough to reduce soil loss significantly. On the contrary, the GCCM cover prevented soil erosion during the early period. When GCCM and vetiver were combined to prevent soil erosion, their combined performance was slightly less effective than with GCCM alone and so the combination approach can still be considered effective for preserving soil compaction during rainfalls and storms. Furthermore, during the heaviest storm event, the bare soil exhibited the highest amount of soil erosion.

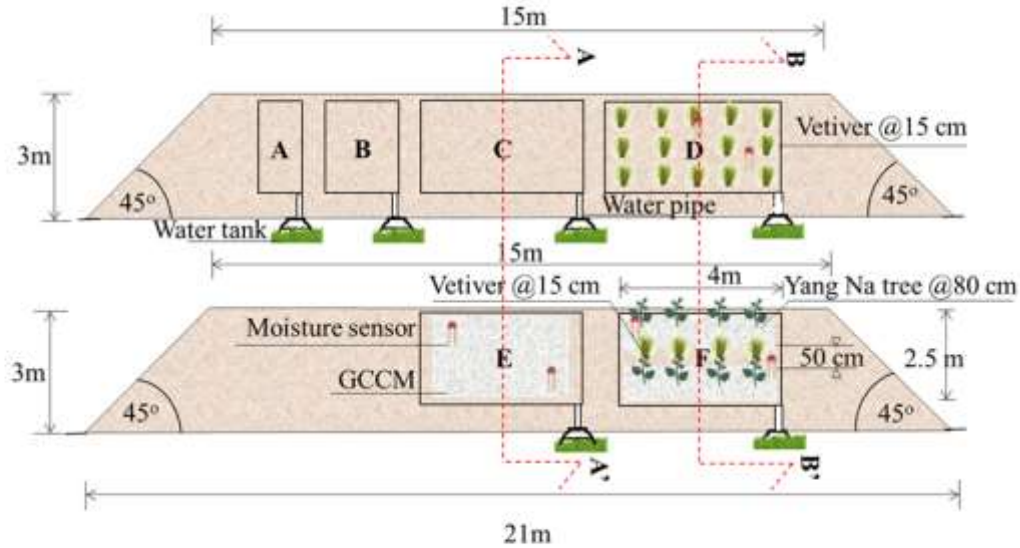
Embankment testing site in Chachoengsao Province to the east of Bangkok, Thailand

Aerial photo of the studied area



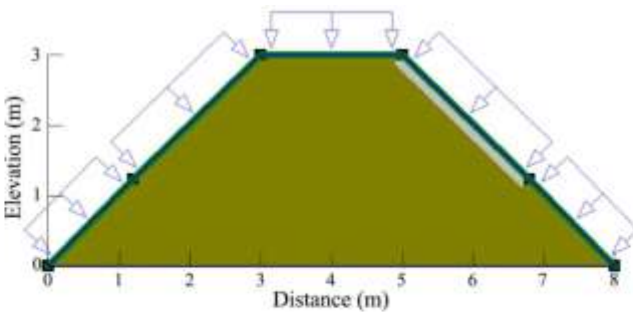
Embankment with soil and water run-off collection drums

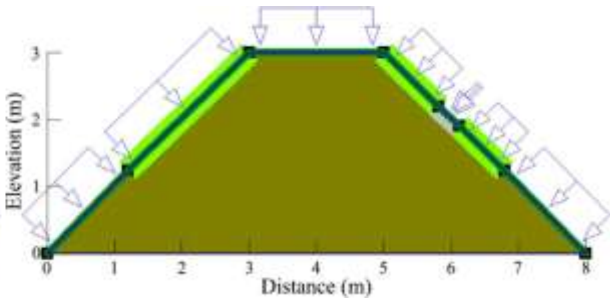
Schematic diagram showing the embankment and its coverage. (A), (B), and (C) are bare soil areas, (D) area planted by vetiver grass, (E) area totally covered by GCCM, and (F) area partially (80% area) covered by GCCM and planted with vetiver and Yang Na Trees *Dipterocarpus alatus* (20% area).



Seepage analysis modeling of the embankment.

Section A-A': Bare soil and GCCM ***Section B-B': Vetiver and GCCM+vetiver***





Most natural slope failures are induced by seepage and/or rainfall. Soil bioengineering is an environmentally friendly method which employs vegetation to reinforce the soil in sloping terrain. The vegetation can contribute to slope stability in two ways, mechanical and hydrological. Kreng Hav Eaba et al (2015) demonstrates the effect of a vegetation root matrix on a soil slope and focuses on mechanical reinforcement using an example of vetiver grass, which had been grown for under a year. The investigation programme includes root observations, direct shear tests and centrifuge model tests. The growing rate of the vetiver roots and the root area ratios were observed during the tests. The cohesion and angle of internal friction of root-reinforced soils were determined from a standard direct shear apparatus and a large direct shear apparatus. A series of centrifuge tests was carried out to demonstrate the effect of vegetation on seepage- and rainfall-induced slope failures. The results indicate that the vetiver roots showed rapid growth within a year and that the shear strength of the root reinforced soil was significantly increased by the bundle of roots. The results also reveal that the bundle of root fibres in the centrifuge model tests helped to reduce the deformation of the soil slope due to instability by increasing the shear strength of the slope.

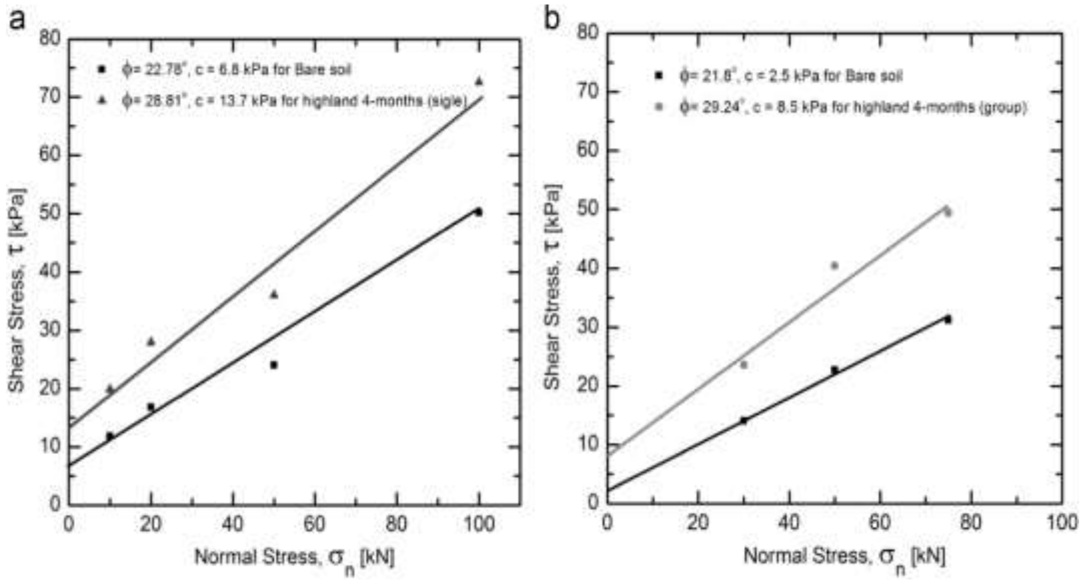
The authors concluded that based on vetiver grass results vegetation roots can increase the shear strength of soil by mechanical reinforcement, thus contributed significantly to slope stability. A series of centrifuge model tests on a slope whose surface is reinforced by model roots was conducted to understand the mechanism of the vegetation reinforcement against seepage and rainfall-induced shallow failure. The conclusions of this study are as follows:

- *The results from direct shear tests confirmed that roots significantly increased the shear strength of the soil. The shear strength of the root-reinforced soil depended on the root length and the root area ratio.*
- *The centrifuge model tests illustrated that slope failure due to seepage and heavy rainfall is triggered by the rising of the water table and starts around the toe of the slope. The rise in the water table causes the effective stress to decrease and results in lowering the shear strength of the soil.*
- *It was confirmed from the centrifuge model tests that the presence of root fibres on the slope surface helps prevent cracking on the soil slope.*
- *The centrifuge model with rainfall simulator tests demonstrated an important role of vegetation roots in slope. Soil displacement calculated from ACCs:*
    - *unreinforced slope under seepage;*
    - *20-mm root-reinforced slope under seepage;*
    - *unreinforced slope under rainfall; and*
    - *40-mm root-reinforced slope under rainfall.*

Results of direct shear tests of four and six month old vetiver roots

Test	Speciment	*Shear strength parameters*	*Increase in cohesion (kPa)*
Standard direct shear test	**Bare soil**	**c=6.8kPa; Φ=22.8^0**	**6.8**
	4-Month-old single vetiver	**c=13.6kPa; Φ=29.7^0**	
Large direct shear test	**Bare soil**	**c=2.5kPa; Φ=21.8^0**	**6.0**
	6- Month-old group vetiver	**c=8.5kPa Φ=29.2^0**	

Shear strength of roots with soil: (a) 4-month-old single vetiver from direct shear tests and (b) 6-month-old group vetiver from large direct shear tests.



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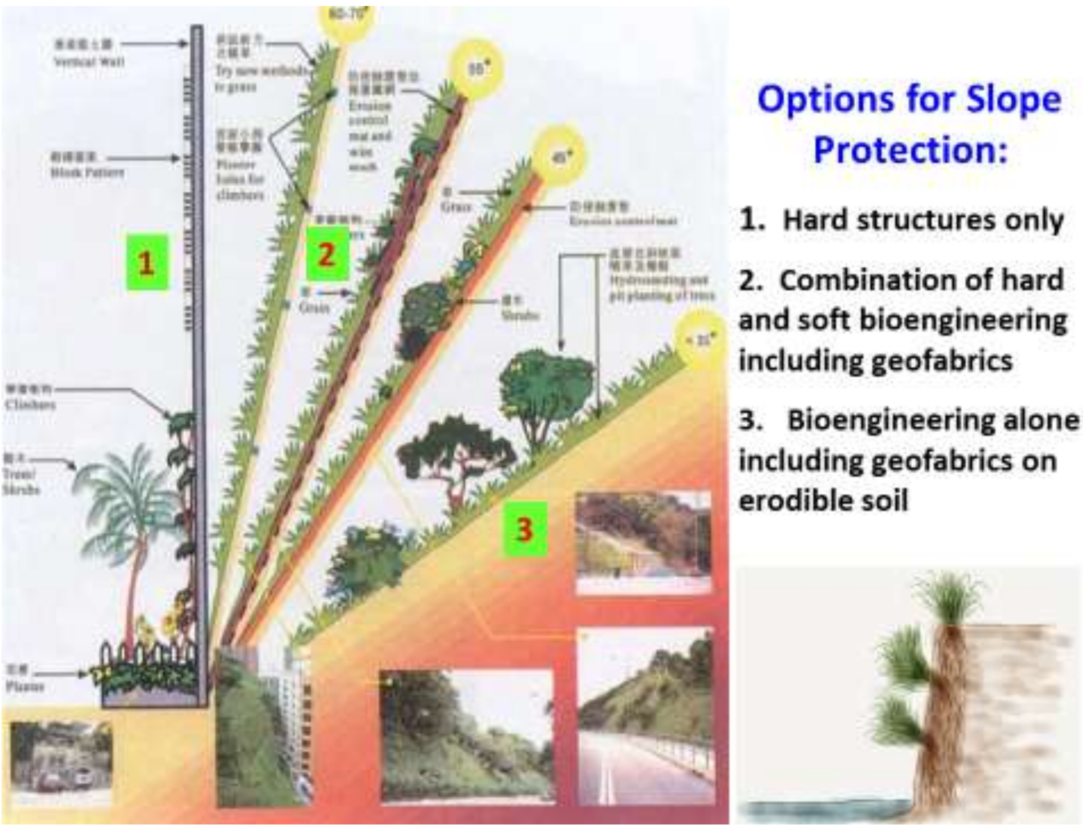
Research in soil moisture competition in crops in Australia indicated that under low rainfall condition this depletion would reduce soil moisture up to 1.5m from the hedges thus lowering pore water pressure and increasing water infiltration in that zone leading to the reduction of runoff water and erosion rate. From a geotechnical perspective, these conditions will have beneficial effects on slope stability. On steep slopes (30-60 degrees) the space vetiver between rows at 1m VI (Vertical Interval) is very close, thus moisture depletion would be greater g8iven this row spacing, therefore it further improve the slope stabilisation process.



Highway fill slope stabilized with vetiver in Fujian Province China

Highway cut slope (L) and fill slope (R) stabilized with vetiver in Thailand

Principles for successful slopes stabilisation with Vetiver System Technology (VST)

The following are main principles for successful and sustainable application of VST for slopes stabilization:

***Appropriate designs and techniques*:**

In bioengineering as with other engineering applications, appropriate design is the key to success. The application of VST for slope stabilisation requires the understanding of biology, soil science, hydraulic principles and hydrological principles.

The slope has to be designed and constructed to the standard that it is structurally and sustainably stable on its own right first. In general, VST will protect the slope from shallow slips by providing structural and hydraulic improvement to the soil profile down to its root depth. VST has been found to be effective even under extreme conditions. It should be stressed that VST is a new technology. As any new technology it has to be learnt and applied appropriately for best results. Failure to do so will bring disappointing outcomes and sometimes adverse results.

Failures of VST in most cases can be attributed to bad design and poor applications rather than the grass itself or the technology recommended. For example in one instance when vetiver was used to stabilise batters on a new highway, the results were very disappointing and failures to establish or to stabilise the slopes were common. It was later found out that from the engineers who specified the vetiver, the nursery personnel which supplied the planting materials to the field supervisors and laborers, who planted the vetiver, had no previous experience or training in the use of VS for steep slopes stabilisation.

Proper implementation:
Even with very good design, the success of the project cannot be ensure if it is not carefully implemented. The followings are main points to be carefully considered:

- Timing, to make the most of rainfall and to avoid or reduce the impact of extreme weather.
- Planting material quality and availability are extremely important.
- There is adequate fertilization and weed control; and
- Staff training is necessary to ensure good establishment to reduce recurrent costs.

<u>*Vigorous maintenance program*</u>***:*** Similar to hard engineering structures, bioengineering structure have to be properly maintained to ensure their sustainability. However, with hard structures, maintenance costs increase as the structures physically deteriorate over time. On the other hand, VST maintenance decreases as the plants mature and endemic species establish themselves between the vetiver rows.

Some examples of VST for extreme slope stabilisation around the world

MADAGASCAR Lavaka

PC Y Coppin

Landscape

PC Y Coppin

MADAGASCAR Railway Batters

USA, Hawaii Island

VIETNAM Ho Chi Minh Highway

PC P Truong

Although badly designed (no benching), it still worked!

PC P Truong

Ho Chi Minh Highway – Central Vietnam

GUINEA (West Africa): Road Batters

PC R Noffke

Hydraulic borer used to plant vetiver on slope with very hard subsoil

Advantages and Disadvantages of Vetiver System Technology

Advantages

The major advantage of VST over conventional engineering measures is its low cost. For steep slope stabilization in China, the saving is in the order of 85-90%. Similar saving could be expected elsewhere as the saving was based on the relative costs of the two technologies used locally. In Australia it was estimated that savings of between 65-75% for various structures on highway in southern Queensland. This saving would be higher in low labour cost countries

As with other bio-engineering technologies, VST provides a natural and environment friendly method of erosion control and land stabilisation which 'softens' the harsh look often associated with conventional engineering measures such as concrete and rock structures. This is particularly important in urban and semi-rural areas where the visual degradation of the environment caused by infrastructure development is often a major concern of local population.

VST maintenance costs are low especially in the long term. In contrast with conventional engineering structures, the efficiency of bio-engineering technology improves with time as the vegetative cover matures. VST requires an adequate maintenance program in the first few years after planting, but once established it is virtually maintenance free in the long term. VST can be used on all scales from small urban individual backyards to large ones such as 1000km long highway or coastline.

Disadvantages

The main disadvantage of VST applications is the time lag between implementation and full effectiveness. Vetiver's growth rate is very slow when fully shaded particularly in the initial establishment phase. Even partial shading will reduce growth rates hence it is not suitable for fully shaded areas. In addition another disadvantage is that it is also temperature sensitive and thus not suitable for temperate climates that have freezing winters.

Based on the above, it is clear that the advantages of using the VST as a bio-engineering tool outweigh its disadvantages particularly when the vetiver plant is used as a pioneer species.

Under Wet or Inundated Conditions

In many parts of the world, embankment failure in rivers, canals, waterways, estuaries and beaches are a major concern for engineers as well as for environmental reasons. Erosion results in the loss of land along these geographic features. Not only is the land lost but the current dikes and levees protecting the embankments are also threathened by erosion. These problems tend to become even bigger due to a lack of effective erosion controls, an ever increasing intensity of boat traffic erode embankments as well. There is also a change in hydraulic boundary conditions.

With VST's inherent superior attributes proven in dryland situations as shown above plus its tolerance to submergence, prolonged inundation and salinity, VST has been used successfully for flood and stream bank erosion control, and estuary stabilization in Australia, Asia and Africa.

With its stiff and erect stems Vetiver can stand up to strong water flow, reduce flow velocity and trap sediment found in runoff water. Research at Delft University of Technology, Netherland (Jaspers-Focks, and Algera, 2006) provided the following results:

***Soil types*:**

Cohesive soil (clay) reduced the growth rate of Vetiver grass by approximately 50% compared to a non-cohesive soil (loam), which was a very significant result. Furthermore, a decrease in phreatic level of 0.17 m resulted in significant higher growth rates as differences were found in the order of 10-20%.

***Vetiver grass as bank protection against vessel-induced loads*:**

The influence of Vetiver grass on small scale mass failure was tested using a physical model test. The drawdown caused by passing ships was reproduced with the use of a wave flume. The amount of eroded material of cohesive soil (clay) was approximately 8-10 times smaller using Vetiver grass. The erosion of non-cohesive soil was also reduced drastically.

The use of Vetiver grass as an armour layer on a dike under wave attack: A single hedge of Vetiver grass planted on the outer slope of a dike can reduce the wave run-up volume by 55%, in contrast with sod-forming grasses that give no reduction. Planting multiple hedges along the contour of the outer slope might result in even more reduction. The application of Vetiver grass on existing dikes may provide a substantial reinforcement of these dikes.

In addition Verhagen et al (2008) reported that Vetiver grass is a sustainable and innovative solution for the protection of banks. It was shown that Vetiver grass was able to establish a full-stop of bank erosion caused by rapid drawdown, therefore it provided them with strong indications that it is highly suitable as an anti-erosion measure. A combination of cohesive soil and Vetiver grass provides the best protection against erosion, which implies that it is highly suitable for banks in delta areas, which consist pre-dominantly of cohesive soil. *A single hedge of Vetiver grass planted on the outer slope of a dike can reduce the wave run-up volume by 55%, in contrary with sod-forming grasses that give no reduction*. Planting multiple hedges along the contour of the outer slope might result in even more reduction. The application of Vetiver grass on existing dikes may provide a substantial reinforcement of these dikes.

The advantages of Vetiver grass above conventional methods with the use of stone are numerous as stated below.

Vetiver grass is sustainable. Vetiver hedges of over 100 years old have been found. Vetiver grass is not invasive and no significant diseases are known. Vetiver grass will, contrary to traditional conventional methods, increase in strength in time.

Vetiver grass is an economic attractive solution. In most countries in South-East Asia Vetiver grass can be planted for less than $3 per meter, while solutions consisting of stone and concrete are expensive in delta areas. Vetiver grass allows people to protect their own property. Since the costs are low and it is easy to use local initiatives can be easily achieved.

Van Tai Tang et al (2018) investigated the performance of ecological revetments (vetiver grass used to stabilize gabions and riprap) implemented on the bank of the Cua Tien River in Vinh city, Vietnam. Based on the ecological, topographical, and hydrological conditions of this river, the gabion and riprap models were introduced to investigate the effect of ecological revetment on the slope stability and ecological restoration characteristics. The effect of prevailing climatic indicators, such as temperature, precipitation, sunlight hours, and humidity were investigated to ascertain the characteristics of weather conditions on the subtropical area. On the surface soil layer of the gabion and riprap, the nutrient indicators of soil organic matter (SOM) and available nitrogen (AN) increased in the spring, summer, and winter, but decreased in autumn, and available phosphorus (AP) did not show an obvious change in the four seasons. The biomass growth rate per Vetiver plant on the gabion and riprap revetments was found to be the highest during the summer, at 15.11 and 17.32 g/month, respectively.

The root system of Vetiver and other native plants could increase the cohesion of soil. After 6 and 12 months, the shear strength of the soil behind the gabion vetiver-stabilized revetment increased by 59.6% and 162.9%, while the shear strength of the soil under the riprap also increased by 115.6% and 239.1%, respectively. The results also indicated that the gabion and riprap revetments could improve the river water purification effect and increase the ecological diversity in the region. In the current study, 26 floral and 9 faunal species were detected in the riprap revetment, whereas 14 floral and 5 faunal species were detected in the gabion revetment, respectively. Through high sequencing technology, the number of bacterial species in that study was found to be 198, 332, and 351 in the water, gabion, and riprap samples, respectively.

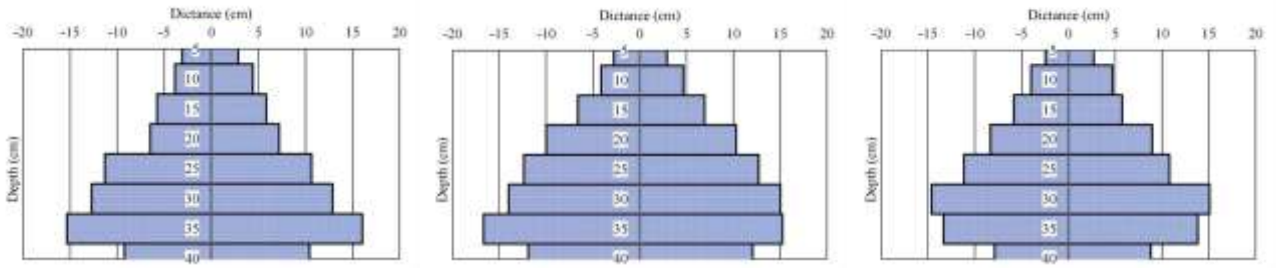
Sriwati *et al* (2018) reported that Bioengineering is a technology thst attempts to maximize the use of vegetation components along riverbanks to cope with landslides and erosion of river cliffs and another riverbank damage. This study aimed to analyze bioengineering using Vetiver as a surface layer for soil erosion control on slope with gradient of 10^0, 20^0, and 30^0. This study wss conducted with three variations of rain intensity (I), at 103 mm/hour, 107 mm/hour, and 130 mm/hour by using a rainfall simulator tool. In addition, the USLE (Universal Soil Loss Equation) method was used in order to measure the rate of soil erosion. In this study, there are a few USLE model parameters that were used such as the rainfall erosivity factor, the soil erodibility factor, the length-loss slope and steepness factor, the cover management factor, and the support practice factor. It was concluded that:

- Without Vetiver, erosion rate increases, in contrast, samples with Vetiver decreases erosion rate.
- The results of the average of reduction of erosion rate on Vetiver, under 3 rainfall levels, at a rainfall intensity of 103 mm/hr it reduced the erosion rate by 84.971%, at a rainfall intensity of 107 mm/hr it reduced the erosion rate by 86.583%, and at a rainfall intensity of 130 mm/hr it reduced 65.851%.

Streambank stabilisation

Vegetation has been used as a bioengineering tool for erosion control and slope stabilization for centuries, however the efficiency of different plant species for this purpose is not the same. In a paper entitled *Enhancement of river bank shear strength parameters using Vetiver grass root system* Hamidifar et al. (2018) investigated the morphological properties of the Vetiver grass root system including root area ratio (RAR), root diameter ratio (RDR), Root diameter and density ratio (RDDR) and root length density (RLD) in a clayey soil. Also, investigated were the effects of morphological characteristics of Vetiver grass root system on the soil shear strength parameters including soil cohesion (C) and soil internal friction factor (φ). The results showed that RAR, RDDR and RLD decrease as the soil depth increases. Also, RDR was found to be correlated to the soil depth. The maximum RAR value was found to be 7.99% which is much higher than those reported by previous researchers for other plants used for soil protection. The maximum RDR, RDDI and RLD values were 72.7, 4.4 and 0.1%, respectively. The results show that among the four root morphological traits studied, RAR and RLD are better correlated to C and φ, respectively. Furthermore, it was found that the plant density is not a significant parameter in the soil reinforcement in the range of densities studied. Moreover, Vetiver grass roots can increase the soil cohesion and soil internal friction factor up to 119.6% and 81.96%, respectively. Based on regression analysis, some empirical equations are presented for the calculation of soil shear strength parameters as functions of the morphological characteristics of Vetiver grass root. They concluded that these findings can be used by ecologists for better management of natural waterways by means of a low-cost environmentally friendly technique

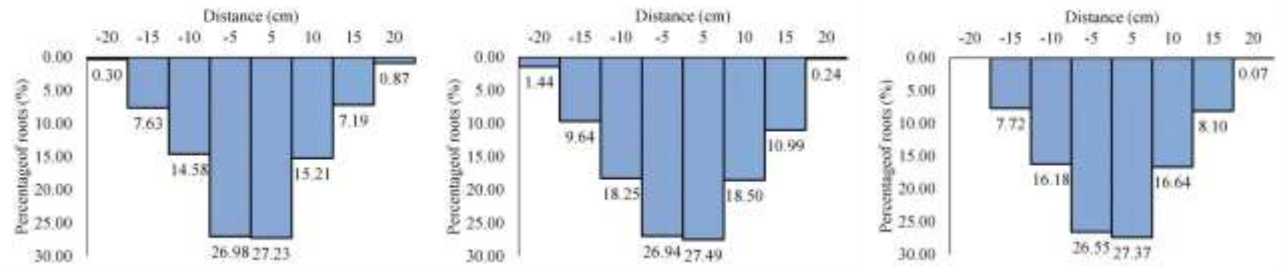
The vertical and radial distribution of the vetiver grass root system are shown below: a) for low, b) for medium and c) for high planting density.



a **b** **c**

The maximum lateral extension of the roots for different planting densities was about 15 cm occurring at the depth 30-35 cm below the ground surface, i.e. above the hard pan layer.

The percentage of the lateral distribution of the Vetiver grass root system for a) low, b) medium and c) high planting density are shown below. These show the variations of the percentage of the root distribution across the centerline of the Vetiver grass plant for various densities.



a **b** **c**

These figures show that more than half of the roots are located within the 5 cm from the plant centerline. Also, as the vegetation density increases, i.e. the distance between plants decreases, the roots tend to move toward the plant centerline. This may be due to spacing, as the plants come closer to each other the competition between them increases and the roots tend to move vertically rather than horizontally.

Principles of the Vetiver System for Stream Bank Stabilisation

In flood erosion control and riverbank stabilisation vetiver relies on its deep and high tensile root system to reinforce the bank slopes and its dense and stiff stems to spread and reduce flow velocity.

- To stabilise the steep bank gradients, horizontal rows planted on approximate contour lines are used.
- To reduce flow velocity of the strong current therefore preventing scouring from the strong flow, planting of cross rows is needed in addition to contour lines.

- For maximum effect, the cross rows are orientated at right angle to the flow direction.
- The spacing of both horizontal and cross rows varies with slope gradient and length, soil type, flow velocity and depth.

Stream Bank Stabilisation in Australia

Planting pattern showing both horizontal and cross rows

Five years after planting

Planting pattern showing both horizontal and cross rows

Three years after planting

Bridge abutment stabilisation in Australia

Riverbank stabilisation in Australia

Riverbank protection against wave action in Mekong Delta, Vietnam

Beach erosion control in Sao Paolo, Brazil, before and after vetiver planting

Stabilisation of the Ganges Riverbanks in West Bengal, India

In India, the Mahatma Gandhi NREGA (MGNREGA) Scheme, Nadia District of West Bengal took the initiative to protect riverbank erosion by planting vetiver only and or in combination with stone boulders/sand bags. The objectives were to protect its people from riverbank landslides and flooding, which occur frequently, as in 2015 and the devastating flood in 2000 where a large part of the district was submerged with the loss many lives and properties. This project named by the Honorable Chief Minister of West Bengal, Mamata Banerjee as "SABUJAYAN" and launched on 23-11-2015. Now, 50 nurseries have been established and planting has started on some areas along the riverbanks.

The main sites are on several tributaries, Bhagirathi, Churni, Mathabhanga, Jalanji, Padma and Ichhamati of the Ganges with one originating in Bangladesh. The total length of the main sites on the river in the district is 743.97 km, ie approximately 1,488km bank on both sides. The project total cost is USD500M and was shared by the Indian and West Bengal State governments. *Currently 22 million vetiver slips are being raised in 55 nurseries along the rivers, making it the world's biggest Vetiver project.*

Erosion along the banks of the Ganges

Nursery along the banks of the Ganges

Vetiver planting along the banks of the Ganges

The Brahmaputra River, Assam. India

The Brahmaputra is probably the most difficult river to control in the entire world. National as well as international team of experts have studied this river for the last 50 years without being able to give any solution to control the erosion of the mighty Brahmaputra. Erosion here is due to:

- Fluvial scouring occurs naturally during the rainy season/flood season in the summer
- Sloughing takes place in the winter when the water level goes down and the bank collapses.

Brahamaputra River Banks Stabilization with Vetiver

All the sites described below are on river banks of the Brahamaputra River and its tributaries. The issues that arise here are:

- The highly erodible alluvial silts;
- The large range of the river level between high and low water (often greater than 5m),
- The high silt load in the river; and
- The scarcity of hard rocks in certain areas needed for rock riprap.

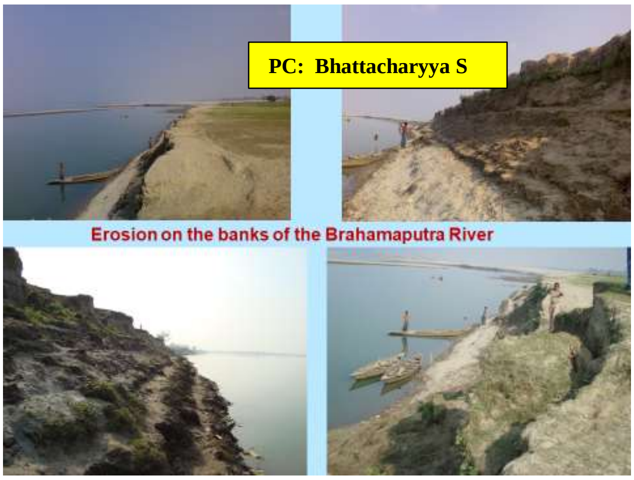
Hard engineering works to prevent erosion

- have been tried over decades
- incurring large financial and environmental costs
- with limited success.

This in combination with limited State budgets means that

- finding an alternative, low cost, environmentally sustainable solution is imperative.
- The Vetiver System shows considerable promise

The following images show the results to date of a series of VST applications in the stabilization of riverbanks and steep slope erosion protection in the region


PC: Bhattacharyya S
Erosion on the banks of the Brahamaputra River


Rock Riprap
Ineffective traditional erosion control measures
Wooden or concrete Porcupines


Major problem
Undercut at toe of banks
Solution
Sandbags and nettin
Sand deposit after floods


Vetiver works along Kapili River
Before and after vetiver planting on a very steep bank of the Kapili River


Majuli River, before and after vetiver planting, note grid pattern
Na- Dihing River, before and after vetiver planting , note silt deposition occurred at the toe of the bank


Before and after vetiver planting along Na- Dihing Rivers
Before and after vetiver planting on a very steep bank of the Kolong River including undercut toe erosion

Planting pattern showing both horizontal and cross rows

Streambank stabilisation in other countries

Streambank in Thailand (L) and estuary erosion control in the Philippines

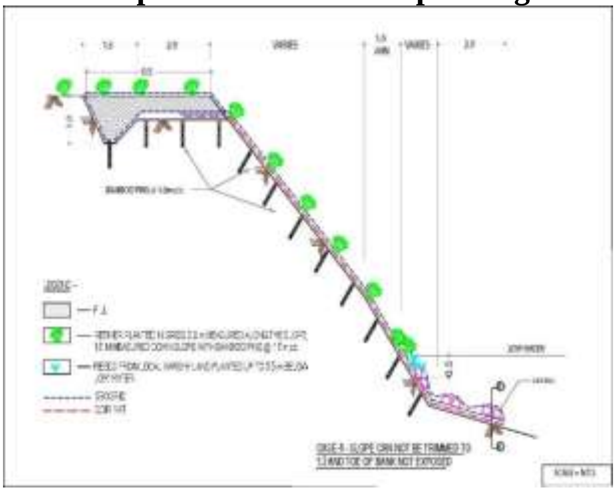
Lake shore stabilisation in Guangdong, China

Very steep river banks stabilization with Vetiver

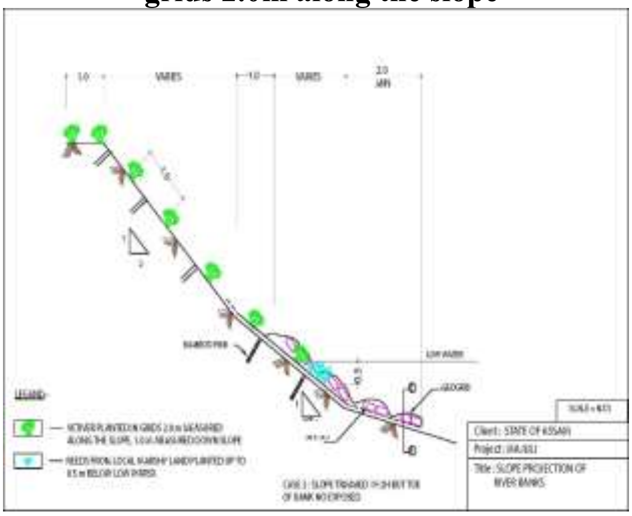
On a visit to Assam in October 2009, Oliver Hawes a Consultant Geotechnical Engineer from

Jacobs Engineering Fairbairn House Ashton Lane Sale Manchester. UK, recommended the use of vetiver in combination with conventional engineering structures to stabilise the eroding very steep Brahmaputra river bank.

Slope can not be trimmed to 1:2 and toe of bank not exposed, has to use fill to build up a platform for vetiver planting



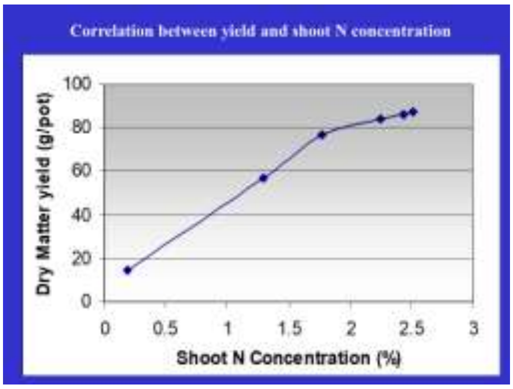
Where slope can be trimmed to 1:2 and toe of bank exposed, Vetiver (green) planted in grids 2.0m along the slope

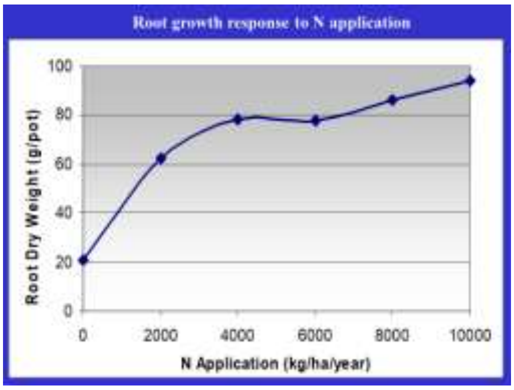


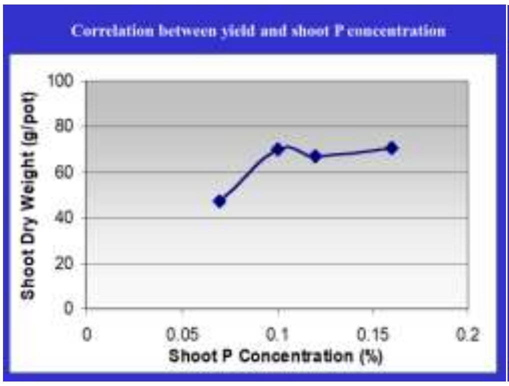
VETIVER AS A PHYTOREMEDIATION TOOL

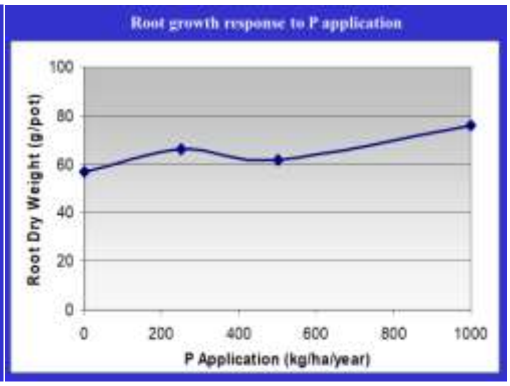
In addition to its extremely high tolerance level to pollutants, particularly N and P, in both land and aquatic media, it also tolerates heavy metals which kill most plants. Vetiver's deep and massive root system is also drought and water-logging tolerant. Furthermore with its special shoot/leaves architecture, vetiver has a very high transpiration rate under wet conditions, hence reducing the volume of polluted water. The combination of these features makes vetiver an ideal plant for phytoremediation of both contaminated land and polluted water.

The following figures show the correlations between N and P on shoot and root growth







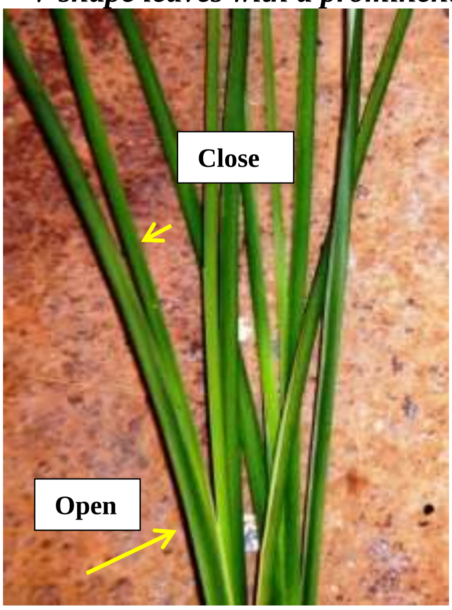


Special Vetiver shoot/leaves architecture,

In addition to its high tolerance and high uptake of pollutants, vetiver high effectiveness in phytoremediation of wastewater also relies on its very high transpiration rate under wet conditions to reduce the volume of polluted water.

Vetiver has an unusual leaf and shoot architecture. Unlike most of other grasses, Vetiver has a V shaped leaf with a prominent mid rib, which can control the opening or closing of the leaf blades. Under moist or wet conditions, the leaves open up, resulting in higher transpiration rate, and Liao et al (2003) found that *leaf blades of vetiver grown in wetlands became thinner and the density of stomata increased* – an ideal combination for wastewater disposal. But under dry conditions, the leaves close up resulting in a lower transpiration rate to conserve moisture, so it is very drought tolerant.

V shape leaves with a prominent mid rib which can open or close leaf blade



Vetiver Canopy

The stiff and erect shoots form a dense funnel shape canopy with leaf angles varying between 45° and 135°, not flat or horizontal like broadleaf plants or most grasses such as Lemon grass and Panicum spp. This shoot architecture has several important implications: the longer sunlight interception of individual leaves as the sun moves from east to west; and sunlight interception from both sides of the individual leaf, exposing most of the leaves simultaneously to sunlight, with minimal shading of leaves within the canopy as is in the case most other plants.

Another unique feature of vetiver leaves is that it only bends almost at the tip, unlike other grasses, such as lemon grass, the whole leaf bends down shading lower leaves. Consequently, larger leaf surfaces of Vetiver are exposed to sunlight over a longer time for increased photosynthesis, leading to better growth as compared to other plant species.

Stiff and erect shoots with 45°-135° angle (left) and its bent tips (centre), forming a thick hedge when planting close together (right)

Lemon grass canopy, note internal shading caused by upper and outer leaves.

Side view ***Top view***

Panicum grass canopy, note internal shading caused by upper and outer leaves.

Mature plant

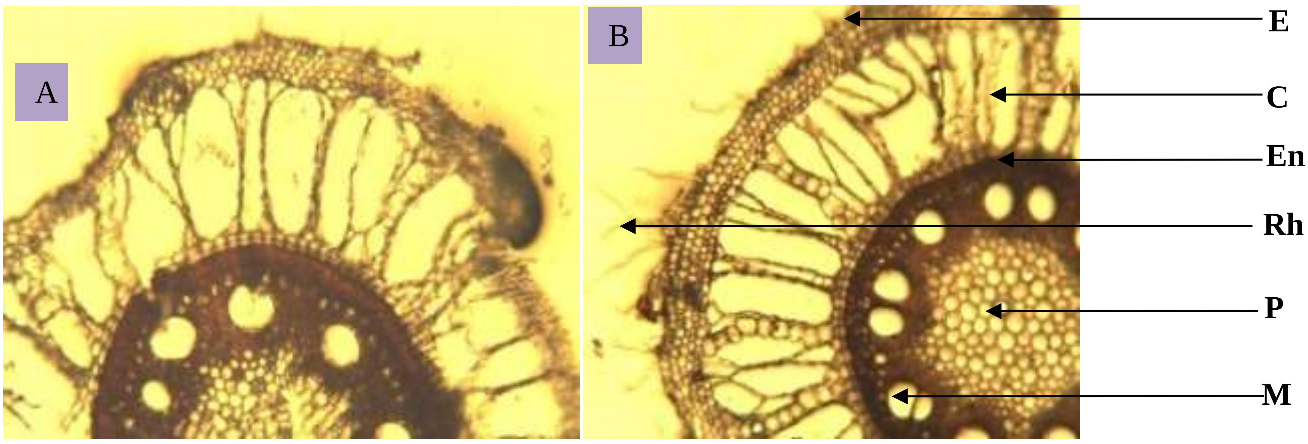
Side view of young plant

Top view of young plant

Anatomical Differences between African and South Indian Vetiver Species as an Indicator of its Phytoremediative Potentials

Catherine Nnamani and Itam Micheal, botanists from the United Nations University in Nigeria studied the anatomy of the two Vetiver species: African *Chrysopogon nigritana* and South Indian *Chrysopogon zizanioides* to demonstrate the superior phytoremediative potentials of the South Indian species. They reported that the anatomy of Vetiver roots and shoots is ones possible indicator of its ability in cleaning up wastewater. (Oku et al, 2016)

Plant roots may respond to physical and chemical changes in the environment by developing certain physical structures such as aerenchyma. Aerenchyma which enable dryland plants to grow deep into the soil and resist drought, also gives buoyancy to plants growing in waterlogged conditions. As an adaptation to waterlogged conditions, Chinese scientist reported that aerenchyma in Vetiver increased in size and number when transplanted from land to water. Aerenchyma also play a leading role in the remediation of polluted soil and water by releasing oxygen into polluted media, this may oxidize toxic pollutants resulting in compounds which are less harmful to the environment. Transverse section of roots of Vetiver as shown below show the presence of aerenchyma in root cortex and air cavities in pits of both the *C. nigritana* (African species) and *C. zizanioides* (South Indian species).



Transverse Sections of Roots of both Species

A: Root of *C. nigritana* x 40

B: Root of *C. zizanioides* x 40

Legend:

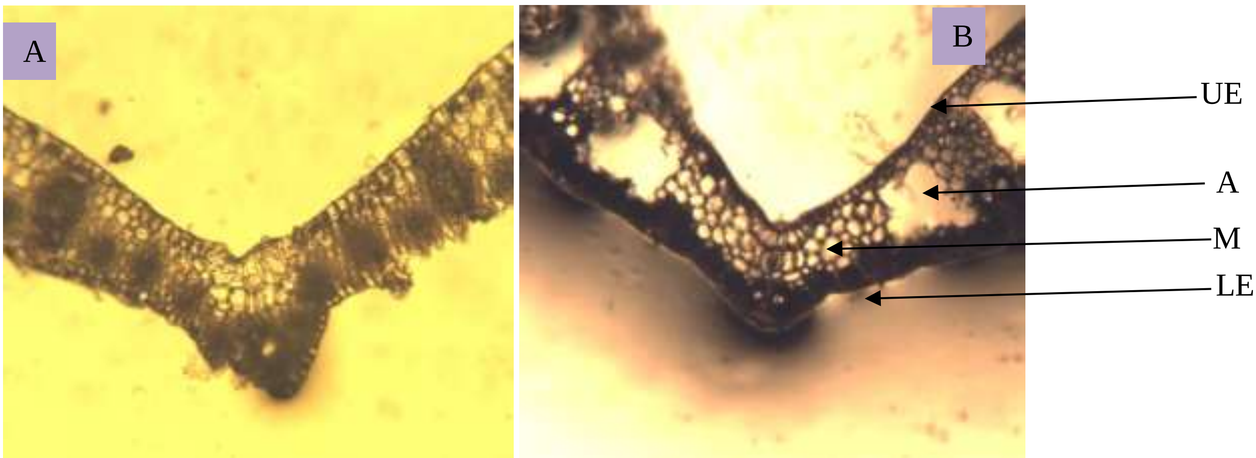
E = Epidermis, C = Cortex with Aerenchyma, En = Endodermis, Rh = Root Hair, P = Pith and M = Metaxylem Lacunae.

Root hairs were observed in C. zizanioides whereas, it was absent in C. nigritana. The presence of root hair in the South Indian species tends to give it comparatively additional contaminant absorbing capacity, though the difference were not statistically significant in most case. An average of fourteen (14) metaxylemlacunae was observed to occur in pairs forming a ring within the endodermis of C. zizanioides. A comparative count in C. nigritana gave ten (10) single metaxylemlacunae also found to be in the ring.

Vetiver shoots also have an important role in determining its effectiveness in phytoremediation, particularly in wastewater treatment, as it reduces the volume through transpiration. The most important components of its shoot in this role are its leaves and culms.

Leaves

The leaves of vetiver are tough and slender, 1-1.5m long and 5mm wide at maturity. The edges are rough due to the presence of tiny barbs. These barbs are often more concentrated towards the tip. Transverse sections of the leaves of both species of Vetiver reveal large intercellular spaces and "V" shaped contours.



Transverse sections of leaves of both species

A: Leaf of *C. nigritana* X 40

B: Leaf of *C. zizanioides* X40

Legend:

UE = Upper Epidermis, A = Air Cavities, M = Mesophyl and LE = Lower Epidermis

Stomata were abaxially placed (at the back side of leaves) in *C. zizanioides*, but were found on both sides of leaves in *C. nigritana*. This suggests that the African species could be more suitable in environments with abundance of water including hydroponic systems, while *C. zizanioides* may be more suitable in arid conditions. This perhaps reveals one reason why *C. zizanioides* is well known for its suitability for drought prone areas or related conditions. The stomata of Vetiver may also play a role in wastewater cleaning. In an experiment to compare the anatomical features between *C. zizanioides* grown in dryland and in a constructed wetland in China, Chinese researchers reported that air chamber density and stomatal apparatus increased in the wetland Vetiver. In another study it was reported that the area of stomatal apparatus in Vetiver isreduced when there is reduction in water supply.

PHYTOREMEDIATION OF CONTAMINATED LAND.

There has been increasing concerns in Australia and worldwide about environmental contamination caused from by-products of mining and manufacturing industries. The majority of these contaminants contain high levels of heavy metals which can negatively affect flora, fauna and humans living in these areas, as well as in the vicinity or downstream of the contaminated sites. The table below shows the maximum levels of heavy metals tolerated by environmental and health authorities in Australia and New Zealand.

Investigation Thresholds for Contaminants in Soils (ANZ, 1992)

Heavy Metals	Thresholds (mg/kg) Environmental *	Health *
Antimony (Sb)	20	-
Arsenic (As)	20	100
Cadmium (Cd)	3	20
Chromium (Cr)	50	-
Copper (Cu)	60	-
Lead (Pb)	300	300
Manganese (Mn)	500	-
Mercury (Hg)	1	-
Nickel (Ni)	60	-
Tin (Sn)	50	-
Zinc (Zn)	200	-

*Maximum levels permitted, above which investigation is required.

Mining wastes - overburden and tailings – and industrial solid wastes are the main sources of contaminated lands.

Mining wastes: Overburden and tailings.

Concerns about the spreading of these contaminants have resulted in strict guidelines being set to prevent the increasing concentrations of heavy metal pollutants. In some cases industrial and mining projects have been stopped until appropriate methods of decontamination or rehabilitation have been implemented at the source. Methods used in these situations have been to treat the contaminants chemically, burying or removing them from the site. These methods are expensive and at times impossible to carry out, as the volume of contaminated material is very large, such as in the case of gold and coal mine tailings.

If these wastes cannot be economically treated or removed, off-site contamination must be prevented. Wind and water erosion and leaching are often the causes of off-site contamination. An effective erosion and sediment control program can be used to rehabilitate such sites. Vegetative methods are the most practical and economical. However, revegetation of these sites is often difficult and slow due to the hostile growing conditions present which include toxic levels of heavy metals.

Vetiver grass, due to its unique morphological and physiological characteristics mentioned above is an ideal plant to treat and rehabilitate the mining industry wastes. The followings are some examples of this application globally.

Gold Mine Tailings in Australia

***Heavy Metal Contents of a Representative Gold Mine Tailings in Australia*.**

Heavy Metals	Total Contents (mg/Kg)	Threshold levels (mg/Kg)
Arsenic	1 120	20
Chromium	55	50
Copper	156	60
Manganese	2 000	500
Lead	353	300
Strontium	335	NA
Zinc	283	200

NA Not available

Coal Mine overburden in Australia

The overburden of open cut coalmine is generally highly erodible. These spoils are usually sodic and alkaline. Vetiver has established successfully on these soils and stabilized the spoil dump with a slope higher than 45^0 and promoted the establishment of other sown and native pasture species.

This coal mine waste dump was barren for 50 year. Before and after vetiver planting for erosion control

Bentonite Tailings in Australia

Sodium bentonite mine tailings (reject) are extremely erodible as they are highly sodic with Exchangeable Sodium Percentage (ESP) values ranging from 35% to 48%, high in sulphate and extremely low in plant nutrients. Revegetation on these tailings was very difficult as sown species were often washed away by the first rain and what was left could not thrive under these harsh conditions. With an adequate supply of nitrogen and phosphorus fertilisers vetiver established readily on these tailings. The hedges provided erosion and sediment control, conserved soil moisture and improved seedbed conditions for the establishment of indigenous species.

Bauxite Redmud

Vetiver could be established on modified bauxite red mud and residue sand which are highly caustic with pH level between 11and 12.

Vetiver grew well on modified bauxite red mud (L) and residue sand (R)

Phytoremediation of Agrochemicals in Agricultural Lands.

Cotton farming requires more agrochemicals than any other large-scale field crops. It needs a large number of both pesticides and herbicides to control insect pests and weeds. As all these agrochemicals are applied either on the crop (pesticides) on soils surface (herbicides) sprays, they accumulate on the surface soil. On sloping land, unless it is controlled, the topsoil is eroded by runoff water, which carry these agrochemicals causing serious offsite pollution to areas downstream of the field.

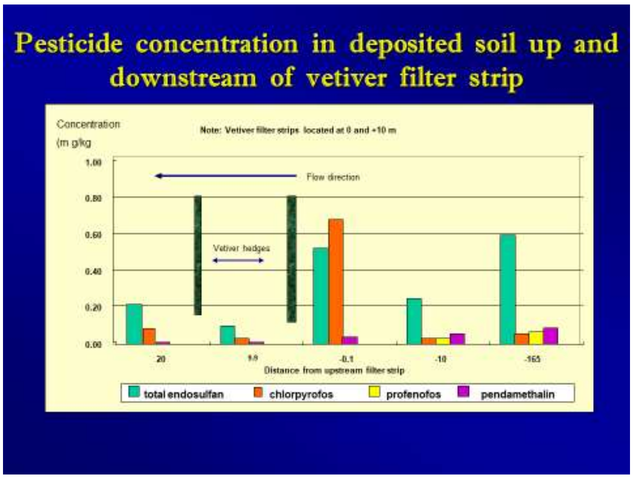
A trial was conducted in Australia to test the effectiveness of vetiver in trapping the agrochemicals in the sediment. On this farm four pesticides ***(Endosulfan, Chlopyrofos, Pronofos***

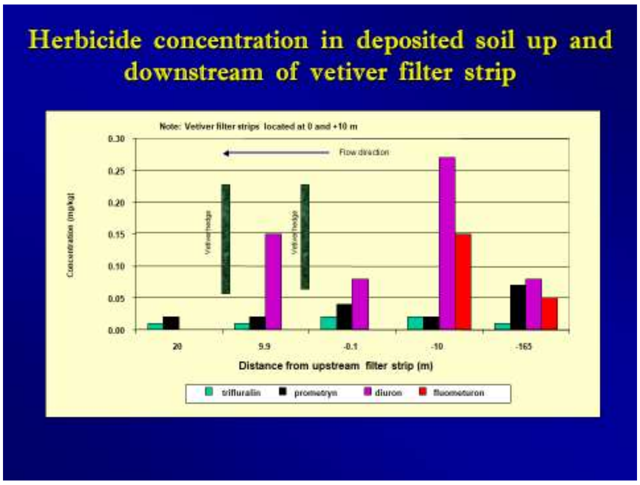
and Pendamethalin) and four herbicides ***(Trifluralin, Promotryn, Diuron and Fluometuron***) were used

Vetiver filter strips installed on a drain on a cotton farm in Australia

Sediment trapped upslope of Vetiver filter strips

Effectiveness of Vetiver filter strips in trapping pesticides and herbicides on a drain on a cotton farm in Australia





PREVENTION AND TREATMENT OF POLLUTED WATER

VST prevents and treats contaminated water in the following ways:

- Eliminating or reducing the volume of wastewater by: seepage control, land irrigation and wetland; and
- Improving the quality of wastewater and polluted water by: trapping sediment and particles, tolerating and absorbing pollutants including heavy metals and detoxification of industrial, mining and agrochemical wastes.

In addition, one of the most significant advancements recently is the use of computer modelling to predict volume uptake using vetiver grass for on-site industrial wastewater disposal. On-site disposal often means eliminating the use of diesel-burning tanker trucks for hauling wastewater to distant treatment plants, a corresponding reduction in greenhouse gas emissions (along with sequestration of CO_2 by vetiver), reduced liability for the generator, and additional benefits for the community at large. The disposal potential has been carefully studied with the model, and field evidence has helped to calibrate and validate the computer modelling. The use of this model allows designers to properly size the area necessary for waste disposal by vetiver.

In the modelling process, not only all known aspects of vetiver physiological and morphological attributes, but also its potential were carefully studied and analyzed in the calibration process. The results again establish and confirm our admiration for this unique plant.

Extensive research in Australia, China and Thailand has established vetiver's tolerance to elevated and sometimes toxic levels of salinity, acidity, alkalinity, sodicity as well as a whole range of heavy metals and agrochemicals. The latest research also shows its extraordinary ability to absorb and to tolerate extreme levels of nutrients, to consume large quantities of water in the process of producing a massive growth. These attributes indicate that vetiver is highly suitable for treating contaminated and polluted wastewater from industries as well as from domestic discharge. The results again establish and confirm great admiration of vetiver for its ability to cost-effectively solve environmental problems.

In a keynote presentation at ICV3, *Clean Water Shortage, an Imminent Global Crisis How Vetiver System can Reduce its Impact* (Truong, 2003) pointed out that fresh water scarcity is predicted to become the greatest single threat to international stability, human health, global food supply and even the spectre of war over water. According to the World Resources Institute, within 25 years, more than half of the world population will be suffering from severe fresh water shortages.

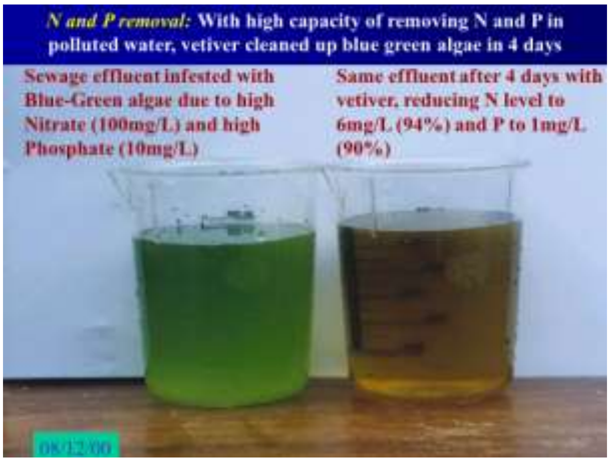
This publication covers past and current research and applications of Vetiver System in treating wastewater, including:

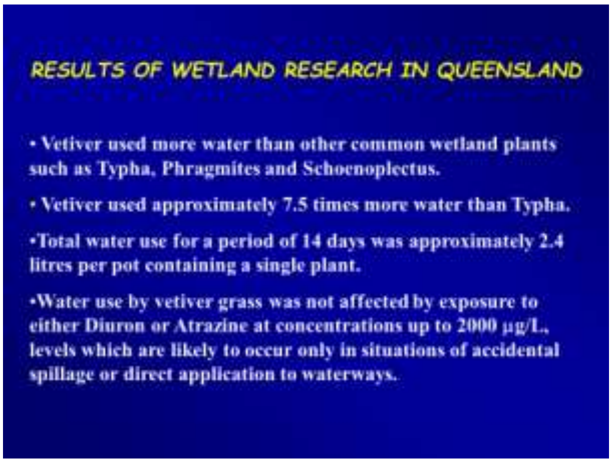
- Wastewater volume or quantity by: seepage control, land irrigation and wetland; and
- Wastewater quality by: trapping sediment and particles, tolerating and absorbing pollutants, and heavy metals and detoxification of industrial, mining and agrochemical wastes.

In a review entitled *Effectiveness of Vetiver Grass versus other Plants for Phytoremediation of contaminated Water* Darajeh *et al* (2019) reported that the reduction of contaminants is strongly affected by plant growth rate and hydraulic retention time. They found that *C. zizanioides* is either equally and often more effective in treating highly contaminated wastewaters than other Vetiver genotypes (*C.nigritana and C. nemoralis*) and other commonly used macrophytes including several Cyperus species (*C. papyrus*, *C. alternifolius), Phragmites* species (*P.karka, P. mauritianus, P. cyprus, P. australis),* Typha species (*T. angustifolia, T. orientalis, T. latifolia)* and another 14 plant species, including water hyacinth (*Eichhornia crassipes), Canna indica and Canna iridiflora*. It was shown that *C. zizanioides* is the most effective species among the top three, including *Phragmites australis* and *Cyperus alternifolius*.

Floating Platform or Pontoon

The combination of vigorous root growth under hydroponics conditions, high capacity for nutrient absorption and water use rate, make vetiver very effective in treating nutrient loaded wastewater. As a result, vetiver pontoon treatment method is beeing used successfully around the world. However one disadvantage of this system is the high initial establishment and potentially high maintenance costs.



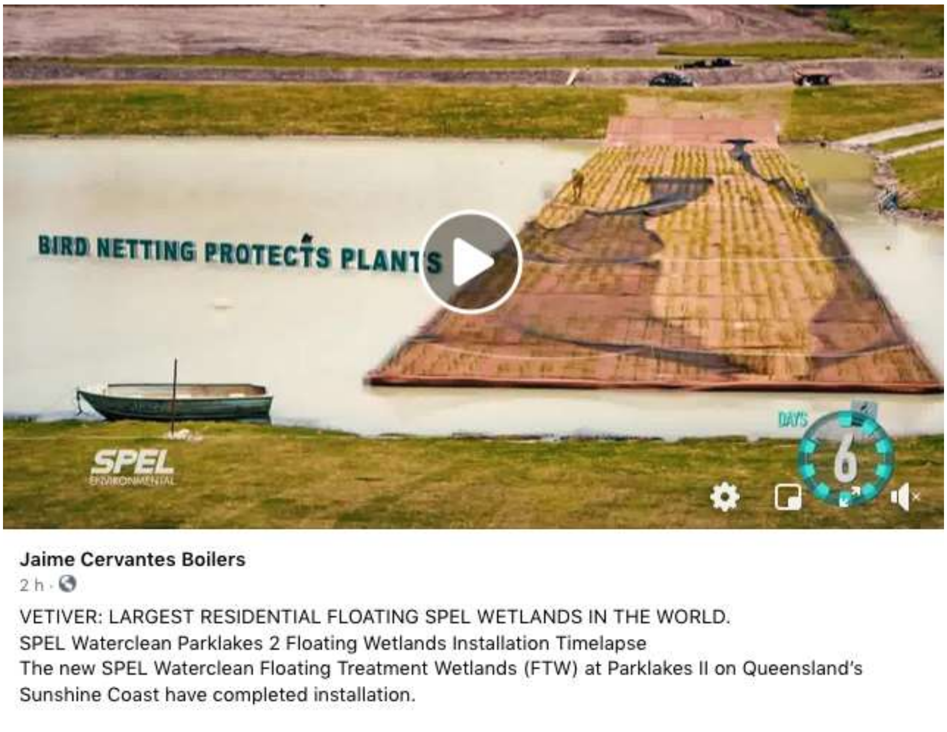


The followings are some examples of highly effective applications of vetiver pontoon treatment around the world.

Vetiver pontoon treatment in effluent ponds in Australia



SPEL WATERCLEAN (www.spel.com.au) installed this floating wetland at Parklakes 2, Agnes Pt, Bli Bli in Queensland.




Vetiver pontoon treatment in industrial wastewater ponds in Australia


Vetiver pontoon treatment in industrial wastewater ponds in Malaysia (L) and China


Vetiver pontoon treatment in industrial wastewater ponds in Vietnam (L) and Madagascar

Vetiver pontoon treatment of municipal wastewater ponds in West Bengal, India

Floating Billboard treating polluted water in Manila, Philippines by Noah Manarang

Wetlands

Natural and constructed wetlands have been shown effective in reducing the amount of contaminants in runoff from both agricultural and industrial lands. The use of wetlands for the removal of pollutants involves a complex variety of biological processes, such as microbiological transformations and physio-chemical processes, e.g. adsorption, precipitation or sedimentation. Vetiver wetlands have been successfully used in Australia for sewage disposal, in China for reducing both the volume and high nutrient loads of piggery effluent, in Thailand and Vietnam for industrial wastewater and for sewage effluent.

Natural wetlands

On shallow wetlands vetiver can be planted directly in the ponds. On deeper wetlands vetiver can be planted around the edges. In both cases large mature plants should be used to ensure faster growth when initially established.

Shallow wetlands

Deep wetlands

Constructed wetlands

Constructed wetlands can be built above ground or in ground, the following are some successful examples of different construction methods.

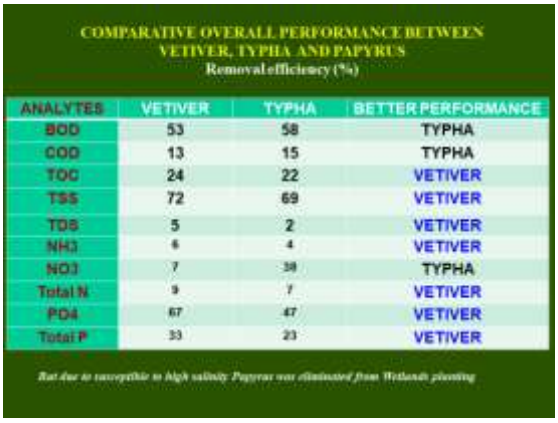
Concrete above ground constructed wetlands in Rio de Janeiro, Brazil

To evaluate Vetiver efficiency in removing nutrients present in domestic sewage, a vertical flow constructed wetland was establshed. Samples were collected from the sewage effluent before and after passing through the treatment modules. Ammonia nitrogen and total phosphorus, as well as BOD, COD, total coliforms and Escherichia coli were determined to calculate the efficiency of their removal, with a hydraulic retention time of 1.97days. Results obtained over the period of 12 months from planting are shown below.

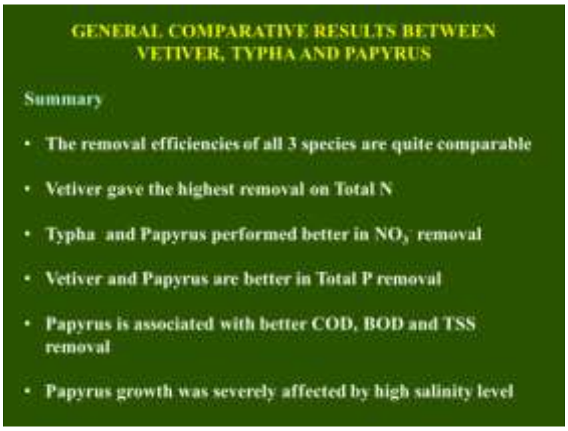
Effectiveness of vetiver treatment in reducing different analytes

	Analytes						
	COD	**BOD**	**Total N**	**Total P**	**Coliform**	**Turbidity**	**E.coli**
Reduction levels (%)	90	91	71-93	80	99	63	83

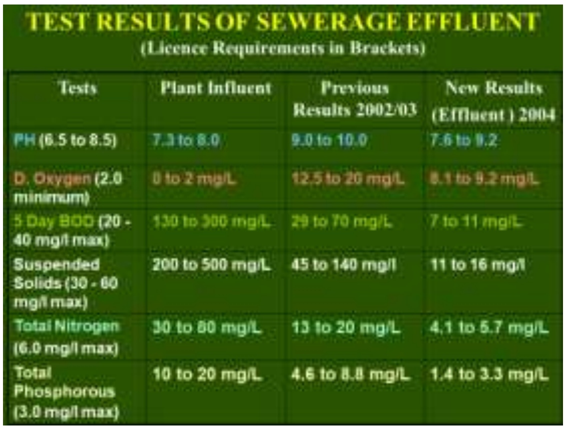
Earthen bank above ground constructed wetlands in Singapore


COMPARATIVE OVERALL PERFORMANCE BETWEEN VETIVER, TYPHA AND PAPYRUS
Removal efficiency (%)

ANALYTES	VETIVER	TYPHA	BETTER PERFORMANCE
BOD	53	58	TYPHA
COD	13	15	TYPHA
TOC	24	22	VETIVER
TSS	72	69	VETIVER
TDS	5	2	VETIVER
NH3	6	4	VETIVER
NO3	7	38	TYPHA
Total N	9	7	VETIVER
PO4	67	47	VETIVER
Total P	33	23	VETIVER



Ephemeral Wetland in Australia

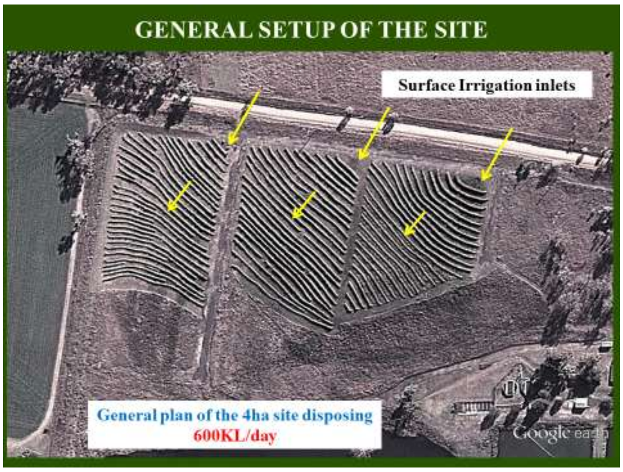


TEST RESULTS OF SEWERAGE EFFLUENT
(Licence Requirements in Brackets)

Tests	Plant Influent	Previous Results 2002/03	New Results (Effluent) 2004
PH (6.5 to 8.5)	7.3 to 8.0	9.0 to 10.0	7.6 to 9.2
D. Oxygen (2.0 minimum)	0 to 2 mg/L	12.5 to 20 mg/L	8.1 to 9.2 mg/L
5 Day BOD (20 - 40 mg/l max)	130 to 300 mg/L	29 to 70 mg/L	7 to 11 mg/L
Suspended Solids (30 - 60 mg/l max)	200 to 500 mg/L	45 to 140 mg/l	11 to 16 mg/l
Total Nitrogen (6.0 mg/l max)	30 to 80 mg/L	13 to 20 mg/L	4.1 to 5.7 mg/L
Total Phosphorous (3.0 mg/l max)	10 to 20 mg/L	4.6 to 8.8 mg/L	1.4 to 3.3 mg/L

Municipal Sewage effluent treatment

This plant serves a small rural town in Queensland, Australia, with the capacity to treat 600, 000L of sewage effluent per day. Vetiver Phytoremediation Technology was adopted to reduce both construction and maintenance costs in upgrading this plant to comply with new Australian EPA regulations.



Landfill leachate

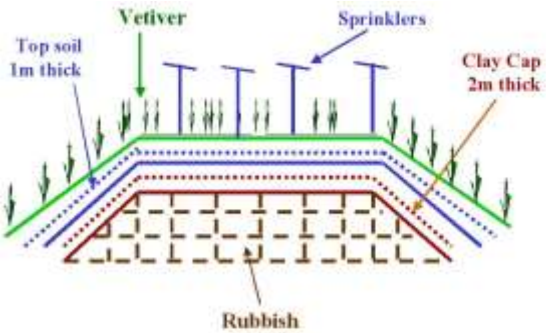
Stotts Creek Landfill is a major waste depot in NSW, Australia. Disposal of leachate is a major concern as the landfill site is close to agricultural areas. An effective and low cost leachate disposal system was needed, particularly during summer high rainfall season. Leachate quality at this landfill was low in heavy metals but relatively high in salts and nutrients. Currently leachate and runoff from the landfill site are stored in ponds at the foot of the mound. During dry periods the leachate is irrigated onto the top of the completed waste mound where it evaporates or transpires into the atmosphere. During heavy rainfall the leachate overflows into a system of wetlands and then to a local creek.

Following capping and top soiling, vetiver wasen planted on the surface of the completed waste mound and irrigated with leachate from the collecting ponds. An area of 3.5ha was planted with vetiver in January 2003 and was increased to about 6ha in total late in 2004.

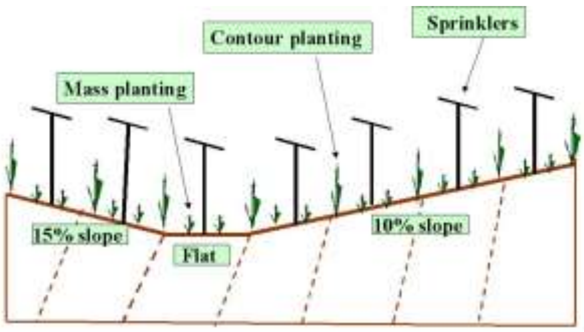
Results to date have been excellent, in the second year thick and tall, 3m high, vetiver growth was recorded. The growth was so vigorous that during the dry period, there was not enough leachate in the ponds to irrigate both the old and new plantings.

The design was based on the pump capacity of delivering 1,300L/minute, and the irrigation frequency of 1 hour twice every day in summer and once a day in winter. The first planting of 3.5ha effectively disposed of 4 ML a month in summer and 2 ML a month in winter.

Cross section (left) and longitudinal section (right) of the Stotts Creek landfill mound



Diagrammatic cross section of the mound at Stotts Creek Landfill, Muwillumbah



Diagrammatic longitudinal section of the mound at Stotts Creek Landfill, Muwillumbah

Overhead sprinkler irrigation

One month after planting

Vetiver tolerates leachate

Sub surface Irrigation

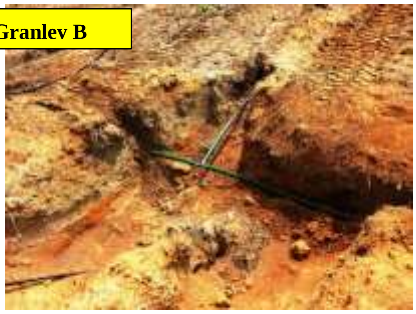
Although overhead (sprinkler) irrigation is more economical, in certain counties with very strict environmental protection laws, subsurface irrigation is required to dispose of landfill leachate.

Leachate Management Specialists (LMS) of Denver, Colorado, USA, is a major wastewater management company that specializes in landfill leachate disposal in subtropical Gulf States of the USA, using Vetiver Phytoremediation Technology. LMS has undertaken 13 projects in Alabama, Florida, Louisiana, Mississippi, and Texas in the mainland US and also in Puerto Rico and Mexico (PASA) (Granley and Truong, 2012). Due to strict USEPA standards for leachate disposal, all leachate has to be disposed via subsurface methods in most states. Therefore, underground drip irrigation was used. The following is an example of a project at the Gulf Pines Landfill, Biloxi, Mississippi. This landfill is a closed facility that accepted municipal solid waste as well as construction and demolition debris beginning in 1988 and concluding in 1996 or 1997.

The depth of refuse is between 20-40 feet. Soil cover was between 3 to 6 feet. The soil types consist predominantly of sand, low on nutrients. Leachate disposal has averaged approximately 270,000 gallons per month (3.2 million gallons per year), which is equivalent to 36,000L/day); and planning for 45,000L/day (4 million gallons per year) later. *The Leachate Total N value of 172mg/L* *is medium, but total P value of 0.2mg/L is very low. All other parameters are well below the tolerant levels of vetiver grass.*

The vetiver system was installed in 2011 and won a National Engineering Excellence Award in Washington D.C. through the American Academy of Environmental Engineers in 2012 for environmental innovation.

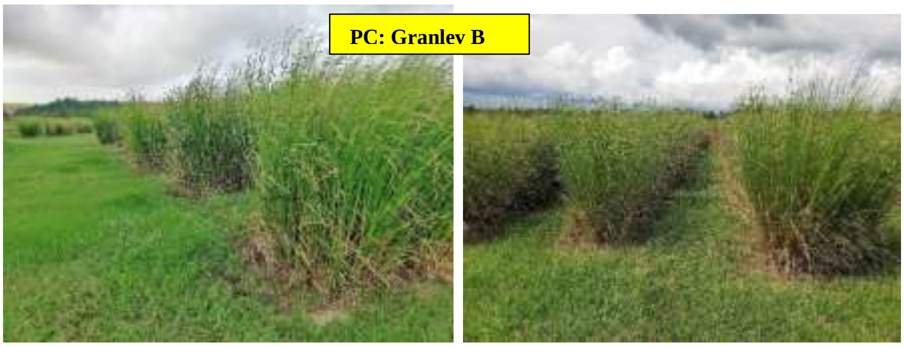
Leachate storage tanks ***Laying subsurface irrigation pipe***



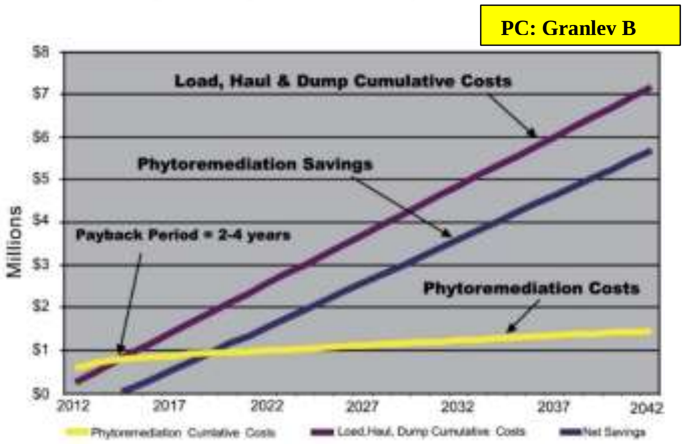
Vetiver planting on either side of subsurface irrigation pipe

Planting (L) and three months after planting

Nine years after planting (Aug 2020), four month old regrowth following annual harvest in April



Typical Cost Savings using Vetiver Phytoremediation at landfills



Ecological Compound Micro-Circulation (ECM) Wastewater Treatment Technology in Southern China.

Miniaturizing sewage treatment technology is a worldwide challenge, especially in communities and rural areas where land is densely populated, land resources are scarce, and climates vary with locations. Land, facilities and maintenance costs are the greatest obstacle for miniaturizing sewage treatment technologies. Although the traditional sewage treatment technology varies, the process of miniaturizing the technology faces the challenges of insufficient treatment capacity, high treatment cost, and low treatment efficiency. Past research and the mechanisms involving the use of Vetiver Phytoremediation Technology (VPT) applications to treat sewage have resulted in many successful results. VPT is particularly important where land availability is a limiting factor. If VPT is combined with other technologies to form an ecological based "miniaturized sewage treatment" system, not only will construction and maintenance costs be reduced, but less land, will be needed.

Feng (2020) developed Ecological Compound Micro-Circulation (ECM) wastewater treatment technology for rural China due to land shortages. This miniaturized system (ECM) of wastewater treatment technology is another economic and practical method for using VPT to treat sewage. Its application is now being adopted in 5 provinces of China at an accelerated pace.
The following illustrations summarize some specific examples for rural China.

Niujiao Village (Puding County)

Qixing Community (Xixiu District)

Ma'anshan (Zhenning County)

Caixin Village (Puding County)

Baiyun Village (Pingba District)

90-day Root System of Qixing Community

90-day Root System of Caixin Village *90-day Root System of Baiyun Village*

Construction started *Caixin Village original construction site*

Construction started *Nursery block*

Construction site completed

Landscaping after completion

Landscaping after completion

Vetiver final polishing ponds

Vetiver floating island technology is used at the end of wastewater treatment in farm

Highlight and Summary of ECM

- ***Energy Consumption*:**

The average total energy consumption was 0.16kw / m3 (already in the category of microcirculation). When wind-solar energy is used, only 1/3 - 1/2 of the external paid grid power consumption is needed, and the processing cost is less than 0.02 Yuan/m3. This achieves low energy consumption environmental standards.

- ***Operating Costs***:

Due to the use of improved technology of biochemical reactors and anaerobic granular sludge and the comprehensive application of ecological plants and vetiver technology, there is no sludge removal and transportation process, so the maintenance costs are extremely low, with only a few simple inspections and winter plant pruning each year, the cost can be limited to the range of 6,000-8,000 Yuan. Its low-cost, high-efficiency operating model has achieved the requirements of sustainable development.

- ***Processing Effect***:

The four environmental monitoring data sets show that the treatment of domestic sewage under normal conditions can fully meet Standard A of First Class of National Urban Sewage Comprehensive Discharge Standard (G18918-2002) by using the ECM technology treatment with removal rates of 90%, compared to the high-concentration of contaminants under traditional conditions, meeting the design standards.

- ***Ecological and Environmental Protection:***

Retaining walls, access roads and handicrafts (including the use of used tires to reduce the amount of backfill earthwork), combined with the ecologically friendly garden landscape form a unique scenic effect. The reuse of water reflects the harmonious unification of urban and natural. The ecological and environmental protection aspects that include sunlight (solar energy), air (wind power generation), water (sewage treatment), microorganisms (anaerobic granular sludge), and trees, irrigation, and grass (including aquatic plants and Vetiver) are fully realized.

- ***Economic Benefits**:*

After the project was completed and in operation, its ecological measures and the scenic effect formed by the garden landscape created employment opportunities for local residents as well as the promotion of local tourism resources. Various aquatic plants and Vetiver, after use were sold, to generate revenue.

- ***Demonstration Effect**:*

The project not only treated rural domestic sewage, but also demonstrated the effects of energy conservation, emission reduction, and ecological poverty alleviation. More importantly, it demonstrated and promoted the wider application of the technology through visits and study from local authorities interested in sewage treatment. The Vetiver floating island technology was introduced under the project. The application of Vetiver floating island technology in Guizhou has been further promoted to other locations in China.

ECM impact in southern China

As of December 2019, there were more than 80 ECM projects designed by Feng (2020), mainly concentrated in Anshun City and Liping County in Guizhou Province. Of these 45 were under the guidance of Feng during construction. In 2020, Guangzhou, Yunnan, Hunan, Hainan, and Jiangxi provinces were adopting ECM projects. More importantly, many projects that originally adopted the "anaerobic pond + aquatic plants" treatment technology model now include the "Vetiver Floating Island" technology to enhance the effluent filtration as a result of the successful demonstration of the Feng’s ECM based projects.

Conclusion

"ECM" wastewater treatment is a combined multi-functional technology with special inclusion of the "Vetiver Floating Island Technology" and clean renewable energy, compared with traditional treatment methods. ECM benefits include: reduced land requirements; significant and long-lasting sewage treatment; reduced operating and maintenance costs; sale of byproducts; odorless emissions; an improved landscape; and a place for recreation and community interaction. ECM is an important technical and replicable approach to sewage treatment that has ecological, health and economic benefits. The application and design standards of the Vetiver component in "ECM" can be further developed through trial and experimentation.

Supplementary instructions for using ECM

The "ECM" scheme can also be applied to industries such as "farm sewage treatment", agricultural product processing wastewater and medical wastewater with high COD, BOD and permeate concentrations. However, in the treatment of wastewater in different industries, the "ECM" program is mainly designed and modified in two aspects: the microcirculation biochemical reaction system and the vetiver application system. For example, in the Pig Farm Wastewater Treatment case,

The wastewater is urinary foaming manure, water flushing manure or scraper-type dried manure, the concentration of COD, BOD and SS in the wastewater far exceeds that of ordinary domestic sewage as shown in Table below. If vetiver is planted directly to this wastewater, the survival rate of vetiver is basically zero. Therefore, the initial wastewater concentration must be reduced first, and secondly, the design and configuration of vetiver usemust be adjusted.

Quality of wastewater from breeding areas

Wastewater category	**pH**	**COD** mg/L	**BOD** mg/L	**SS** mg/L	**NH3-N** mg/L	**TP** mg/L	***Escherichia coli*** (a/L)
Swine wastewater	6-9	15,000	10,000	3,000	1,500	400	150,000

When the COD concentration in the wastewater reaches 2,000mg/L, almost all of the vetiver was killed, but when the COD concentration is 700-1000mg/L, the adsorption capacity of vetiver is the strongest. In China, the discharge standards for treating aquaculture wastewater are shown below

Control item	**COD** (mg/L)	**BOD** (mg/L)	**SS** (mg/L)	**N-NH4** (mg/L)	**TP** (mg/L)	***Escherichia coli*** (a/L)
Standard value (within)	400	150	200	80	8.0	10000

Therefore, when adopting the "ECM" scheme to treat wastewater from a pig farm, the following steps must be taken:

- Carrying out solid-liquid separation;
- Strengthening the biochemical reaction rate of the microcirculation system by adjustment of anaerobic granular sludge and biological carrier;
- Designing and adjusting the area of vetiver floating islands and density according to the requirements of the treatment time; and
- Managing the transpiration amount by adjusting the planting area according to geographical conditions and sewage discharge standards of the farm.

Effects of Methane Leaks on Vetiver Growth

When landfills are covered with a geosynthetic cap liner, a thick impermeable membrane to collect biogas (methane) over time the cap liner often cracks, and the trapped gas leaks and affects vetiver planting nearby. These leaks are fairly common on old landfills as they tend to start and stop and start and shift randomly around the site. The following photos show the effect of methane leaks on Vetiver growth at a 15-year old landfill - Judy Holt - in Redland Shire near Brisbane, Australia. The membrane (blue) in between the clay cap and topsoil cover for vetiver planting. Vetiver was planted for erosion and seeping leachate control on this site.

In general the damages caused by the leaks are fairly temporary and has little impact on vetiver growth. Even in severe cases, vetiver was not killed and could recover slowly.

- In mature plants it causes discoloration (yellow) in vetiver leaves. When the leak stops, vetiver leaves return gradually to its normal green colour
- In newly planted plants, the effect is more severe and plant growth is badly affected, but not killed. Growth will resume when the leaks stop

The following slides show the effects of methane leaks on vetiver growth on an old landfill in Brisbane, Australia. Vetiver was planted for erosion and seeping leachate control.

Typical symptoms of a methane leak on mature vetiver plants. Chlorosis normally start on young shoots at ground level, where the gas comes out

Even in severe case, gradual recovery occurs after 3-4 months after the leak stopped

In less affected case, faster recovery

Young vetiver plants are more susceptible to methane leaks, but gradually resume growth when the leaks stop

CHAPTER 5

OTHER IMPORTANT APPLICATIONS

Landslide Rehabilitation due to Climate Change

Due to the outstanding attributes of Vetiver Grass such as extremely deep and massive finely structured root system, with high tensile and shear strength, form dense hedges when planted close together and tolerance to extreme climatic variation it is ideal for landslide rehabilitation . With the advance of climate change the VST has been widely used and proven around the world for its effectiveness in alleviating and rehabilitating damages from disasters and extreme events such as landslides and flood erosion. The followings are some examples of this outstanding achievement.

Eboli, J. and Vieira, C. (2013) reported an outstanding success in the rehabilition of a phenomenal landslide in Itaipava, Rio de Janeiro, Brazil. The following photos show the progress and successful result of this project

Greenhouse gas and Carbon sequestration

Although the focus of much plant and soil science has been on the return of leaves to the soil both as a stock of C in the soil and as a substrate for soil organisms, root returns to soil are larger than shoot returns in several regions. For example, early work by ecologists such as Weaver *et al* (1935) in the USA demonstrated that several grasses produced more organic matter below ground than above ground. This interest in carbon inputs to soils has been re-ignited with the current debate over sequestration of C by vegetation in an attempt to mitigate the greenhouse effect induced by rising CO_2 concentration of the atmosphere.

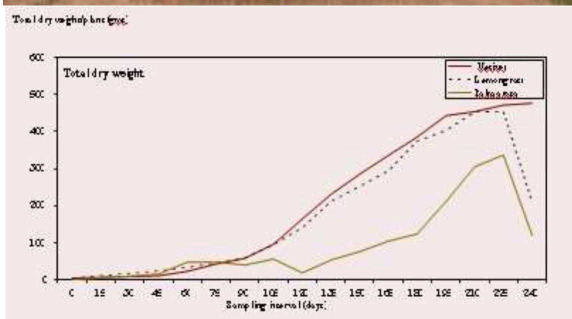
Fisher et al. (1994) pointed out that estimates of the global carbon dioxide balance have identified a substantial 'missing sink' of 0.4-4.3 Giga tons per year. It has been suggested that much of this may reside in the terrestrial biosphere as a result of a study on the carbon stored by pastures based on deep-rooted grasses which have been introduced in the South American savannas. Although the deep-rooted grasses were chosen principally for agricultural reasons, they also sequester significant amounts of organic carbon deep in the soil. If these study sites are representative of similar pastures throughout South America, this process could account for the sequestration of 100 - 507 Mt carbon per year - a substantial part of the 'missing sink'. Thus, although some land-use changes (such as burning tropical rainforests) contribute to the atmospheric CO_2 burden, the authors conclude that the introduced pastures studied here help to offset the effect of anthropogenic CO_2 emissions.

The plants used in this study were perennial grasses *Andropogon gayanus* and *Brachiaria humidicola and* are of African origin, the former is tall-growing with a tussock habit, whereas the latter forms swards, which are very similar to Vetiver in their root development.

Singh et al (2011) in an essay titled A Strategy for Sustainable Carbon Sequestration using Vetiver (Vetiveria zizanioides (L.)): A Quantitative Assessment over India pointed out that sequestration of atmospheric carbon is one of the mitigation measures for countering the anthropogenic climate change due to emission of greenhouse gases. However, such sequestration measures need to be sustainable and significant, without conflict between groups with diverse priorities. Identification of a system for efficient land-based carbon sequestration therefore requires a quantitative estimate on a regional setting. Based on a field experiment and analysis, it is shown that vetiver, an economically viable crop due to its essential oil and medicinal properties, also provides an efficient carbon sequestration system. A comparison of normalized (to 12-month growth cycle) carbon sequestered by different trees and crops with that by vetiver shows it to be more efficient. Similarly, the carbon sequestered is found to be the highest by vetiver in comparison to two other aromatic grasses viz. lemongrass (Cymbopogon flexuosus) and palmarosa (Cymbopogon *martinii var. motia). Our study also provides first time estimates of differential carbon sequestered by roots and shoots; once again vetiver is found to be significantly more efficient. It is further pointed out that plantation of vetiver as an inter-crop in short rotation forest plantations and in agro-forestry systems can provide significant lift to the rural economy without any adverse effect on the eco-system. Based on a number of estimates it is suggested that utilization of waste and degraded lands and social forestry systems for vetiver plantations can provide multiple benefits including significant carbon sequestration in India.*

C-sequestered by Vetiver

Vetiver System	C-sequestered(Tg year^{-1})	% of emissions (2009) India	World
Biomass	150	34.6	1.8
Soil	50	11.5	0.6
Total	200	46.1	2.4



Comparative Economics of three aromatic plants and rice

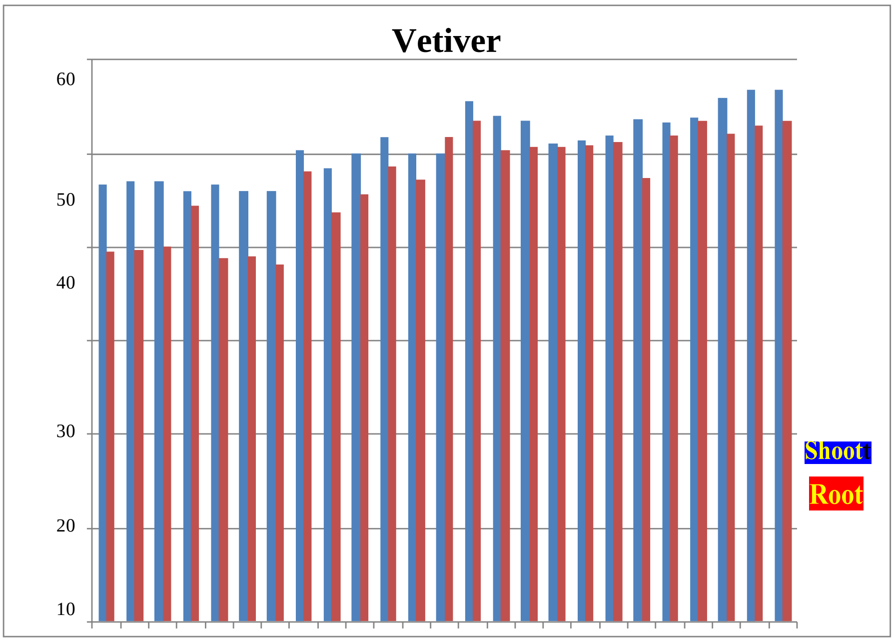
Crop	Fresh biomass (Mg ha^{-1} year^{-1})	Essential oil (%)	Cost (Rs ha^{-1})	Net returns (Rs ha^{-1} year^{-1})
Vetiver	4.25 (roots)	0.8	100 000	230 000
Lemongrass	27.7	0.8	42 800	84 550
Palmarosa	30.0	0.5	40 000	110 000
Rice			15 400	30 200

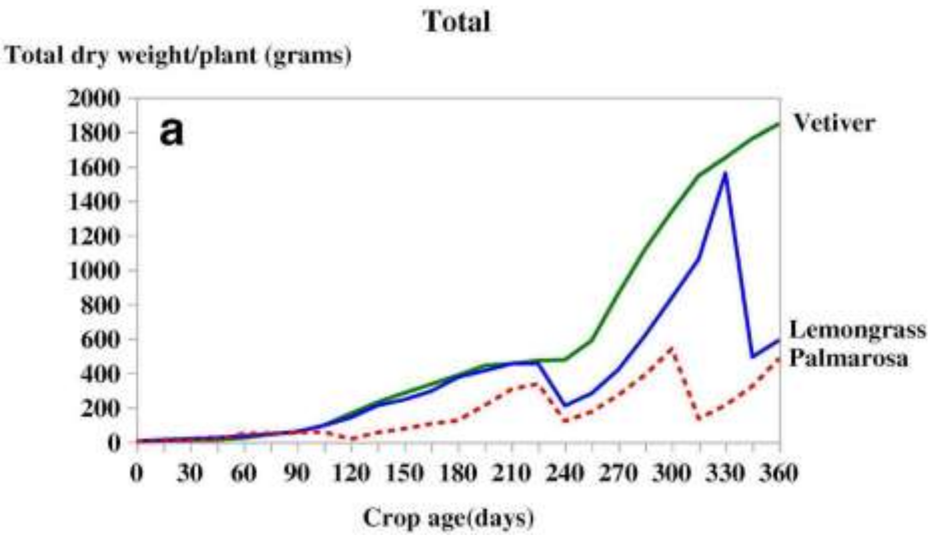
CO_2 (Carbon) Emission by India and World

Total CO_2 emissions ($TgCO_2$)					
Country	**Year**				
	2005	2006	2007	2008	2009
India	1183.28 (323.0)	1282.68 (350.2)	1368.38 (373.5)	1463.3 (399.4)	1591.13 (434.3)
World	28366.15 (7743.9)	28939.22 (7900.3)	29724.51 (8114.7)	30400 (8299.2)	30313.25 (8275.5)

Rao (2016), in a paper entitled Vetiver Systems for Carbon Sequestration and Economic Returns, presented at the Vetiver Workshop at Tamil Naidu Agriculture University in April 2016, the former Head, Central Institute of Medicinal and Aromatic Plants, Research Centre, Bengaluru, India, reported the Organic Carbon Sequestered of the followings three aromatic crops: Vetiver, Lemon grass and Palmarosa. The author found that dry matter yield for vetiver was 28.62 and 1.56t/ha/year for shoot and root respectively, those for lemon grass were 10.50 and 1.57t/ha/year and for Palmarosa were 11.11 and 0.65t/ha/year respectively, *resulting in C sequestered for Vetiver was 15.24t/ha/year, 5.38 for lemon grass and 6.14t/ha/year for Palmarosa*.

Dry matter yield of Vetiver, Lemongrass and Palmarosa over time





Estimated C sequestration by Vetiver in degraded lands in India

Total waste lands (mha)	Carbon sequestered (t/yr) in India for 10mha degraded soil	Emissions (2009)	
		India (t/yr)	% of World
107.83	200	46.1	2.4

Organic Carbon sequestered by Vetiver, Lemongrass and Palmarosa shoot and root

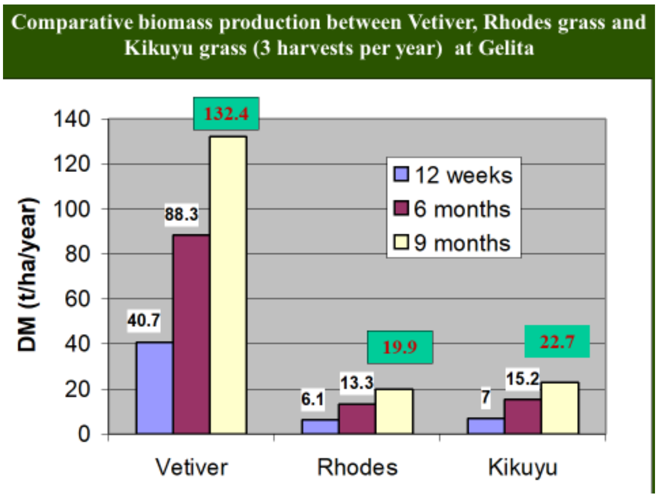
Crop	Carbon (%)		Dry Matter (t/ha/yr)		C sequestered (t/ha/yr)		Total
	Shoot	Root	Shoot	Root	Shoot	Root	
Vetiver	50.53	50.27	28.62	**1.56**	14.46	0.78	**15.24**
Lemongrass	44.45	48.14	10.50	1.57	4.83	0.55	5.38
Palmarosa	52.77	43.49	11.11	0.65	5.86	0.28	6.14

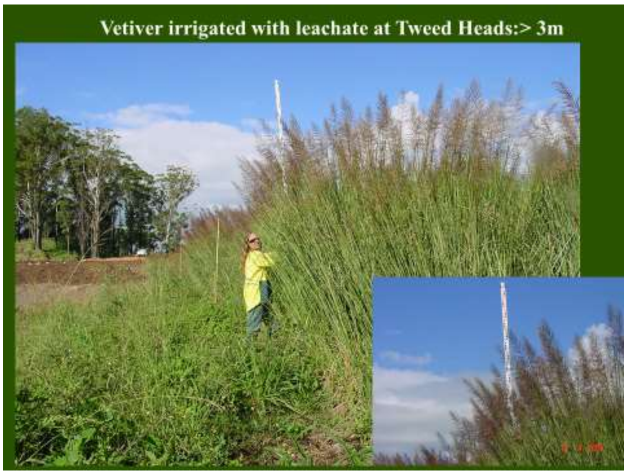
The high level of CO_2 concentration in the atmosphere was found to be effective in promoting the growth of Vetiver and difference among ecotypes was also found in respects to the amount of enhancement and the partition of biomass between above- and below-ground tissues. A field study of the emission gas discharged from power plant as a supply of CO_2 fertilizer indicated higher amount of discharge of the emission gas supported high rate of growth in the order of 8 times of gas discharge to 2.24 times of vetiver growth (Chen Tsai et al (2006).

Biofuel

Advantages of using Vetiver Grass for biofuel production:

- Faster growth and higher biomass under favorable conditions than trees and other grasses.
- It is a perennial crop, no replanting needed after each harvest; and
- Very low incidence of disease or pest attacks





Gueric Boucard, (Texarome Inc.) (pers.com.) from the Dominican Republic established a very large plantation of Vetiver for essential oil production (from the root). He uses both shoot and root biomass as fuel for his boiler to produce steam to distil essential oil from the roots, instead of using petroleum or coal. The followings are his calculation of energy sources:

1 pound of petroleum produces	18,000 BTU
1 pound of high-grade coal produces	14,000 BTU
1 pound of dry Vetiver grass produces	7,000 BTU

Comparatively, vetiver biomass produces roughly a third of the energy of petroleum. Therefore, it would take 3 tons of vetiver to replace 1 ton of petroleum.

Based on the output of his farm over the last 15 years, Boucard established the production cost of Vetiver fuel is US$15/ton. At the current prices for coal of US$45/ton and crude petroleum US$595/ton, then the cost to produce one Million BTU from:

- Vetiver fuel: **US$1.07**
- Coal fuel: **US$1.60**
- Crude petroleum: **US$16.5**

Boucard concluded that there is at least one immediate Energy Alternative for the Dominican Republic. BIOMASS FUEL, from the amazing Vetiver plant. Electricity can be produced at a very low cost, while providing thousands of jobs in agriculture, plus a great number of environmental benefits. Valuable foreign currencies formerly used to import petroleum fuels, would be saved and put to use in other areas of urgently needed infrastructure development.

Shoot biomass harvesting and stockpile at Texarome Inc farm

PC: Boucard G

Root biomass stockpile ***Furnace burning vetiver shoot biomass***

PC: Boucard G

Animal Fodder

Due to its fast growth Vetiver grass has a very high shoot biomass- up to 132t/ha under favorable conditions. This biomass has been used extensively for animal feed around the world in both fresh as green pasture and green cut fodder and hay.

One of the major uses of Vetiver grass is for environmental protection purposes due to its high tolerance levels of pollutants including heavy metals in the soil or water. There are concerns that the heavy metals absorbed by the plant will contaminate the feed. Extensive research in Australia, China, India and Thailand has shown that a very high proportion of metals uptake by Vetiver remained in the roots, for example only 1% to 5% of arsenic, cadmium, chromium and mercury absorbed were translocated to the shoots. A slightly higher percentage for lead and zinc, and these translocation rates are similar to other pasture and crop plants. The Vetiver Network International is confident and highly recommends the use of vetiver biomass for animal feed.

Chinese research on digestibility of the nutrient contents in Dongshan goat showed that digestible of gross energy, dry matter, crude protein, ether extract, crude fiber, calcium, phophorus and nitrogen free extract in Vetiver grass hay were 29.65%, 46.09%, 23.15%, 28.79%, 46.44%, 61.00%, 66.60% and 36.25% respectively. 1 kg dry matter of Vetiver grass hay could provide 1.47 Mcal digestible energy, 13.4 g digestible crude protein and 4.17 g ether extract, which indicated that Vetiver grass is a promising feed resource for goats and other domestic animals. The following Table shows the nutritional values of Vetiver, Rhodes and Kikuyu grasses grown in Queensland, Australia.

Analytes	**Units**	**Vetiver grass**			**Rhodes**	**Kikuyu**
		Young	***Mature***	***Old***	***Mature***	***Mature***
Energy (Ruminant)	**kCal/kg**	**522**	**706**	**969**	**563**	**391**
Digestibility	**%**	**51**	**50**	**-**	**44**	**47**
Protein	**%**	**13.1**	**7.93**	**6.66**	**9.89**	**17.9**
Fat	**%**	**3.05**	**1.30**	**1.40**	**1.11**	**2.56**
Calcium	**%**	**0.33**	**0.24**	**0.31**	**0.35**	**0.33**
Magnesium	**%**	**0.19**	**0.13**	**0.16**	**0.13**	**0.19**
Sodium	**%**	**0.12**	**0.16**	**0.14**	**0.16**	**0.11**
Potassium	**%**	**1.51**	**1.36**	**1.48**	**1.61**	**2.84**
Phosphorus	**%**	**0.12**	**0.06**	**0.10**	**0.11**	**0.43**
Iron	**mg/kg**	**186**	**99**	**81.40**	**110**	**109**
Copper	**mg/kg**	**16.5**	**4.0**	**10.90**	**7.23**	**4.51**
Manganese	**mg/kg**	**637**	**532**	**348**	**326**	**52.4**

Zinc	**mg/kg**	**26.5**	**17.5**	**27.80**	**40.3**	**34.1**
Palatability	**Horse, dairy cows, cattle, sheep, buffalo, rabbits, goat, kangaroo, wallaby.**					

Cattle grazing in Australia


Buffalo grazing in China and cattle in India


Cows, pig and buffalo in Vietnam



Horse in El Salvador

Thatching

Due to its relatively high silica content, vetiver leaves are more durable than other grasses and straw commonly used for thatching purpose. This is one of the main reason that Indian immigrants brought vetiver with them when they migrated to other countries. One good example is Fiji where it has been used extensively for both roof and wall thatch.

Roof thatch in Fiji

Vietnam

China

Vetiver roof and bamboo structure, Bali, Indonesia



Roof thatch in Fiji

Handicraft

Traditionally, only vetiver roots were used for Handicraft in India, before or after essential oil extraction.

Various Handicrafts made from vetiver roots in Kerala, South West India

In addition to the above uses of vetiver shoots mentioned above, its leaves have increasingly become a very important commodity in the making of Handicrafts. This important "discovery" was initiated by HM The King of Thailand and developed and promoted by the Thai Chaipattana Foundation, under the guidance of Princess Maha Sridihorn, Patron of The Vetiver network International.

This applications has been now spread globally, but the Thai products are very intricate and refine, up to jewelry standard, where the leaves or only the leaf blades are specially treated and dyed. Whereas the African and Latin American products use mostly whole leaves. The followings are some examples of Vetiver Handicrafts.

Thailand

Africa



Brazil



China



Indonesia



Venezuela

Socio-economic

In addition to the extraordinary impacts on Agriculture, Infrastructure Stabilisation and Environmental Protection, application of the Vetiver System Technology also has a significant impact on the development and poverty alleviation of the local community, where it is carried out. The following case studies have demonstrated this enormous impact:

Ekoturin Foundation's East Bali Poverty Project (EBPP) was established in 1998 by David Booth (Coordinator Indonesian Vetiver Network)**,** as a non-profit organisation, with the specific goals of reducing poverty and promoting culturally sensitive sustainable social economic development, by prioritizing children, in Desa Ban, the most impoverished and isolated mountain village in Bali. In March 2000 EBPP introduced VS to the most isolated and impoverished region of Bali where 19 scattered communities had never seen the outside world. This facilitated for the first-time, all weather access roads through steep and sandy volcanic mountain terrain. The project demonstrated different VS uses that were rapidly accepted by this impoverished community. All this was a direct result of the close partnership established with the whole community at the outset and farmers seeing for themselves the benefits of the VS. In a paper, *"Vetiver Improving Lives of Impoverished Indonesian Subsistence Farming Mountain Communities, Led by Children"* Booth and Adinata (2006) reported that by bringing profitability to the rural sector, the economy of the whole area benefited. Community cooperatives are now developing to sell vetiver slips commercially from well-established mountain nurseries.

Children preparing vetiver for planting to protect potato and carrot crops

Mountainous rural roads protected by VST

Flood mitigation in the Mekong Delta, Vietnam (Le Viet Dung et al. 2006).

This region is known as the floating rice area, it is next to the Cambodian border, and is characterized by annual flooding, averaging 2-3m deep and occasionally up to 5-6m deep. The soil is deep alluvial, silty loam in texture and highly erodible when wet. In the past 15 years, a regional policy aimed at increasing the rice production by constructing a dike and canal system, thousands of kilometers long, surrounding rice-growing areas. The VST will provide an effective and cheap method to stabilize dyke banks and stop soil erosion during flood season.

A very large multiplication program was initiated to rapidly increase planting materials for this massive project. The Faculty of Agriculture, Cantho University produced about 3,000,000 slips by tissue culture. These plantlets were supplied to various villages in the region to put in polybags and they looked after them until they were ready to plant out. This operation provided a unique opportunity for local people, particularly women and children, to earn an income without leaving the village, and the men jobs in planting and maintaining the VST.

One of the community nurseries preparing vetiver for this 3 million plant project

Poverty Reduction and Resource Protection in China

A project entitled: *Poverty Reduction and Resource Protection in a Guangxi Province Minority* was initiated in 2008. The main aim was to apply the VST for soil and water conservation on the farms and to improve its productivity and income

Female local workers preparing vetiver slips and planting on the riverbank and male workers planting on the steep slope



The second aim was to provide employment and income to rural people, particularly women and children by producing Vetiver Handicrafts (Xu, 2009).

Handicraft training, production and marketing



Poverty alleviation and rural employment in Madagascar.

Madagascar, the fourth largest island in the world is one of the poorest countries on earth with approximately 80% of the population being engaged in subsistence farming where only 4% have access to potable water. Life expectancy is 52 years and the infant mortality rate is 89 per 1,000. Economic isolation of many communities resulted from the deterioration over the past 25 years of the regional road network and in 2004, 80% was then impassable for year round.

Hydromulch (Pty) Ltd, a vegetation restoration and environmental contracting company from the Republic of South Africa provided rehabilitation and vegetation restoration of approximately 40 hectares of exposed side slopes. This project necessitated the propagation of approximately 4,000,000 Vetiver slips. (Noffke, 2015, 2017). Hydromulch initiated a Vetiver nursery program utilizing members of the local community surrounding the entire project, who would be able to "sell" their production crop. Fifteen communities were initially approached during December 2006, which expanded to 32 communities by August 2008. These communities had been involved in the propagation of Vetiver plants at their respective villages, 133 families jointly became involved in these programs. It was important to first establish the Vetiver source close to the site so that progressive stabilisation of the civil works on the construction site could take place.

Farmers Vetiver nurseries provide secure income and full family employment at home

This community based “Vetiver propagation program” generated approximately (US$250,000.00) collectively to the respective communities within the construction period. This excludes the establishment cost paid for by Hydromulch for plant material, potting bags, fertiliser, watering cans & loose tools.

Poverty Reduction and rural employment in Venezuela

Although Vetiver has been in Venezuela for more than one hundred years, it is in the last 20 years that it has reached a very important height, and its presence has been expanded to many places of the country. In 2006 VFEPP, the “Fundacion Empresas Polar” Project started the Vetiver Project with the following general objectives:

- To alleviate social disparities;
- To promote the making of vetiver handicraft;
- To increase participation of the poor population; and
- To develop markets for vetiver handicraft in Venezuela

The innovative approach of the VFEPP was aimed at raising interest in vetiver handicraft in participants: women, young people and children, and later to the whole family. The activities begin through the conference *Why vetiver? An economic, ecological and social project.* Shortly after that the training for vetiver handicrafts begin and connected the participants to the market, setting basic principles for quality, which occured very quickly (in some cases in a month). It provides extra income to the families, and the community developeds a great interest in the Project. It was designed to demonstrate the reasons why the planting of vetiver with the intention of producing fiber is promoted, on sites close to the houses where erosion, and contaminated waters are the common ecological problems. The social aspect was focused on the principle of motivating motivating community members.

From 2001-2006 VFEPP was implemented in wide and diverse geographic zones, having trained approximately 11,000 people. (Luque et al. 2006).

Participation of women and children at home in Handicraft production and marketing

These Handicrafts have brought significant income to the remote subsistent farming communities

The above illustrations clearly demonstrate that vetiver projects can contribute to an integral community development, and poverty alleviation through generating short term economic resources, which motivates participants to reach ecological and social objectives that are effective and easy to implement. The integral contribution of this impact is to benefit the quality of life of the participating communities, particularly the long term effect of vetiver handicraft production to communities - especially in the indigenous and remote area. This is a solution that can supplement ancestral customs.

CONCLUSION

This book with a focus on the unique characteristics of vetiver grass roots, provides a detailed technical back drop to the author's more than 30 years of work that he has devoted to this plant. As the sharing of information with others is vital in promoting VST by providing both scientific and anecdotal information to those interested in the plant and its applications, some useful information that many wereperhaps completely unaware of.

Most people never give a second thought to the hidden parts of a plant comprising its roots and the microflora and fauna and soil nutrients that those roots interact with. It can be exciting to think that vetiver roots are so unique, allowing the plant to multifunction in so many different ways - a compendium of applications that no other plant can to the same extent emulate. Vetiver's root structure gives it the strength of 1/6 of mild steel and are stronger than those of trees. The roots contain many complex chemicals that are able, with some help from symbiotic arbuscular mycorrhiza, to process and absorb toxic chemicals and metals at levels that other plants cannot. Its root cells are able to change and adapt under different conditions - it can survive under extreme drought and yet survive more than three months of total submergence in water, it can survive and even flourish in extreme acid, alkaline, and saline soils. Those same roots create chemical pheromones that attract the stem borer moth to lay its eggs on vetiver leaves in preference to crops like maize and rice, and then another chemical that is able to destroy the digestive enzyme of the larva of that moth - truly an amazing plant. Most importantly these roots, and those above ground visible parts, provide the means for communities of all sizes and cultures an affordable tool that can be used in a sustainable manner to combat the current threat of climate change including the very destruction and health of the soils that we depend on for our survival.

Amongst many applications, perhaps the most important one was the discovery of the value of the plant for phytoremediation of contaminated water and land - thus allowing fairly low cost remediation of contaminated mining and industrial sites and the treatment of polluted water. The latter is a major problem particularly in developing countries and vetiver provides an excellent low-cost solution when designed and applied correctly.

This book will undoubtedly enhance the Vetiver Grass Technology, a technology brought to our attention by John Greenfield 35 years ago and since nursed and further developed by all vetiver associates around the world to create a better environment for tens of thousands of people across the tropical and sub-tropical world via its non-invasive nature, cost-effectiveness, self-resilience, all of which are vital considerations in developing nations where capital resources are scarce and land and water degradation problems are acute and increasing.

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APPENDICES

APPENDIX 1

Reported Vetiver growing at different latitudes

Continent	Country	Location	Latitude	Notes
Oceania	Australia	Geelong, Victoria	38º S	
		Hamilton, Victoria	37º 44S	
		Adelaide, South Australia	34º 55S	
	New Zealand	Gisborne	38º 28S	
		Auckland South	36º 35S	
		Kerikeri	35º 15S	
America North	USA	Yolo County, North California	38º 44N	
		Fort Bragg, North Carolina	35 08N	
		Clemson, South Carolina	34º 41N	
		Mt Vernon, Alabama	31º 1N	
America South	Chile	Chillan	36 º 36S	
Asia	China	Zhejiang Province	N32°20'N	Temperature 38.8° to –12.5°C
	Iran	Teheran	35 º 44N	
	Japan	Tokyo	35 º 42N	
	Nepal	Kathmandu	28º 07N	
Africa	South Africa	Cape Town	33º 55N	
Europe	Germany	Eastern Germany coal mine	51º N	Along river valley
	France	Montpellier	43º 46N	

	Italy	Tuscany, Florence	43º 45	
		Cagliari, Sardinia	39º 11N	
	Portugal	North Portugal	41º N	
		Algarve	37 º 12N	
	Spain	Valencia	39º 28N	
	Latvia	Riga	56º 57N	Floating island in lake in summer and hot house in winter

APPENDIX 2

Phytoremediation of polluted water.

Fresh water scarcity is predicted to become the greatest single threat to international stability, human health, global food supply and even the spectre of war over water. According to the World Resources Institute, within 25 years, more than half of the world population will be suffering severe fresh water shortages.

The VST can reduce the impact of this imminent global crisis in two ways by:

- Reducing or eliminating the unwanted volume of contaminated water
- Improving the quality of wastewater and polluted water

Darajeh et al (2019) clearly showed the effectiveness of Vetiver Phytoremediation Technology versus several macrophytes in treating a wide range of industrial and domestic wastewater, polluted river and lake water by hydroponics and constructed wetlands treatment methods, it was found that Vetiver grass is either equally and often more effective in treating these polluted wastewaters than two other Vetiver *genotypes and* other commonly used macrophytes such as various *Cyperus* species, various *Phragmites* species, various *Typha* species and another 14-plant species.

Plant Species	**Wastewater Types**	**Removal Performance (%)**						
		BOD	**COD**	**TSS**	**TN**	**NH4-N**	**NO3**	**TP**
*Chrysopogon zizanioides***	Aquaculture Effluent	_	_	_	_	0-67	_	0-75
Chrysopogon zizanioides	Piggery Effluent	74*	70		87*	_	_	83
Chrysopogon zizanioides	Sewage Effluent	96	90		49	94	_	89
Chrysopogon zizanioides	Landfill Leachate	67	69	_	_	_	43	56
Chrysopogon zizanioides	Fertilizer Processing	74	64	_	_	_	94	78
Chrysopogon zizanioides	Pinora Juice Effluent	94	95	82	_	41	10	85
Chrysopogon zizanioides	Palm Oil Proc. Effluent	51	10	71	_	40	6	19
Chrysopogon zizanioides	Biogas Effluent	91	82	95	_	42	99	35
Chrysopogon zizanioides	Sewage Effluent	92		92	_	_	_	87
Cluysopogon zizanioides	Pig farm WW	_	_	_	75	_	_	15-58
Cluysopogon zizanioides	Septic tank	_	_	_	99	_	_	85
Cluysopogon zizanioides	River Water	_	_	_	71	_	_	99
Chrysopogon nigritana	Landfill Leachate	66	67	_	_	_	59	79
Chrysopogon nigritana	Fertilizer Processing	69	60	_	_	_	93	80

Phragmites karka	Sewage Effluent	90		91		_	_	86
Scirpus spp	Piggery Effluent	76	80		85	_	_	65
Cluysopogon zizanioides /Cypress tenuifolius	Pig farm WW	68	64	_	_	20	_	_
Chrysopogon zizanioides/Phragmites mauritianus	Textile WW	67.47	46.2	81.49	_	_	_	_
Typha angustifolia	Piggery Effluent	81	84		88	_	_	65
Eichhornia crassipes	POME	_	50	_	88	_	_	64
Typha angustifolia/phragmites	Domestic WW	66	48.15		_	66	_	61
Typha latifolia/Typha angustifolia	Dairy WW	_	_	_	9	21	_	94
Canna/Phragmites cyprus	Municipal WW	93.6	92.2	94	_	_	_	
Phragmites australis	River Water	15.4	17.9	70	83.4	_	_	96
Phragmites australis	Oil Produce WW	88	80		10.2	_	_	18.5
Phragmites australis	Greywater/Secondary	70.3	65.9	82.2	36	_	_	32.4
Phragmites australis	Black Water/Secondary	86.4	83.5	89	69.3	_	_	56.2
Phragmites australis	Municipal Sludge/Tertiary	90	72	81	67	_	_	75
Phragmites australis	Tannery WW/Secondary	98	98	55	_	86	_	87
Phragmites australis	Municipal WW /Secondary	22	56	84.15	39.3	_	_	_
Phragmites australis/Typha orientalis	WW /Industrial	70.4	62.2	71.8	_	40.6	_	29.6
Phragmites australis/Zizania aquatica	River Water	90.5	73.5	92.6	10.6	10.5	_	30.6
Phragmites australis/ Iris australis	Municipal WW/Secondary	_	_	_	91.33	91.2	88.79	_
Typha angustifolia/Scirpus grossus	Municipal WW/Secondary	68.2	71.9	_	74.7	50	19	_
Typha angustifolia	Lake Water		16.5	_	19.8	22.8	34.2	35.1
Typha angustifolia	Municipal WW /Secondary	80.78	65.18	_	58.59	95.75	_	66.5
Typha angustifolia	Lake Water	_	36.9	_	52.1	32	65.3	65.7
Typha angustifolia	Lake Water	_	40.4	_	51.6	45.9	62.9	51.6
Typha latifolia/Phragmites mauritianus	Municipal Sludge/Tertiary	_	60.7	_	_	23	44,3	_
Typha latifolia	River Water	_	35	_	64.85	71.25	_	61.24
Typha latifolia/Canna indica	Municipal WW /Secondary	89.3	64.15	85.25	50.55	61.2	67.76	59.61
Typha latifolia/Phragmites australis	Municipal WW /Secondary	52	68	79	_	_	_	14
Typha angustifolia/Canna iridiflora	Municipal WW	65.5	_	_	_	81.6	50	88.5
Cyperus alternifolius	Sewage Effluent	99	93		57	98	_	91
Cyperus alternifolius	Aquaculture Effluent	_	_	_	_	0-67	_	42-71
Cyperus papyrus	Sugar Factory WW	_	_	76	_	36	_	29
Cyperus papyrus	Municipal W\V /Secondary	52.98	43.89	72.91		17.13	22	57.14
Cyperus papyrus	Tannery WW /Tertiary	_	_	_	72.48	75.43	60.87	83.23
Cyperus papyrus	Tannery WW /Tertiary	_	_	_	89.7	89.3	_	84.53

Cyperus alternifolius	Municipal WW /Secondary	90	70		46	50	_	60
Cyperus alternifolius	Municipal WW /Secondary	_	83.6	99	64.5	71.4	_	68.1
Cyperus alternifolius	Municipal WW /Secondary	_	84.1	99.6		79.6	_	84.5

*** COLOR CODES**

70-84.9%	
>85%	

****ACHIEVEMENTS FROM 70 - >85%**

Chrysopogon zizanioides	**28**
Phragmites australis	**18**
Cyperus alternifolius	**12**

APPENDIX 3

Paul Truong Vetiver Propagation and Training Centre

The Paul Truong Vetiver Propagation and Training Centre was established by a NGO **Group for Rural Alternative Movement (GRAM)** in Kolkata, under the leadership of Mr Manik Mondal, Founder Trustee / Secretary of GRAM. The author was invited to the ceremony to lay the foundation plague and initiate the program at Katwa, West Bengal in April 2016.

Welcoming party

Welcoming sign along the road

Installing the plaque

Vetiver Propagation and Training Centre

Vetiver Propagation and Training Centre Workshop

Future participants

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