ADDIS ABABA UNIVERSITY SCHOOL OF GRADUATE STUDIES

POTENTIAL OF WATER HYACINTH (*Eichhornia crassipes* (Mart.) Solms) FOR THE REMOVAL OF CHROMIUM FROM WASTEWATER IN ARTIFICIAL POND SYSTEM

BY
DANIEL WOLDEMICHAEL

MARCH, 2009

Potential of Water Hyacinth (*Eichhornia crassipes* (Mart.) Solms) for the Removal of Chromium from Wastewater in Artificial Pond System

A Thesis Submitted to the School of Graduate Studies Addis Ababa University in Partial Fulfillment of the Requirements for the Degree of Master of Science in Environmental Science

By
Daniel Woldemichael

March, 2009

ACKNOWLEDGMENTS

First and foremost I would like to glorify and gratitude the Almighty God, who allow me to join and complete this program. I would also like to express my deepest appreciation to my research advisors Dr Feleke Zewge, Dr Seyoum Letta and Dr Seyoum Mengistu, Addis Ababa University (AAU) for their consistent and encouraging advice, valuable suggestions, constructive criticism and critical reading of the manuscript throughout the study.

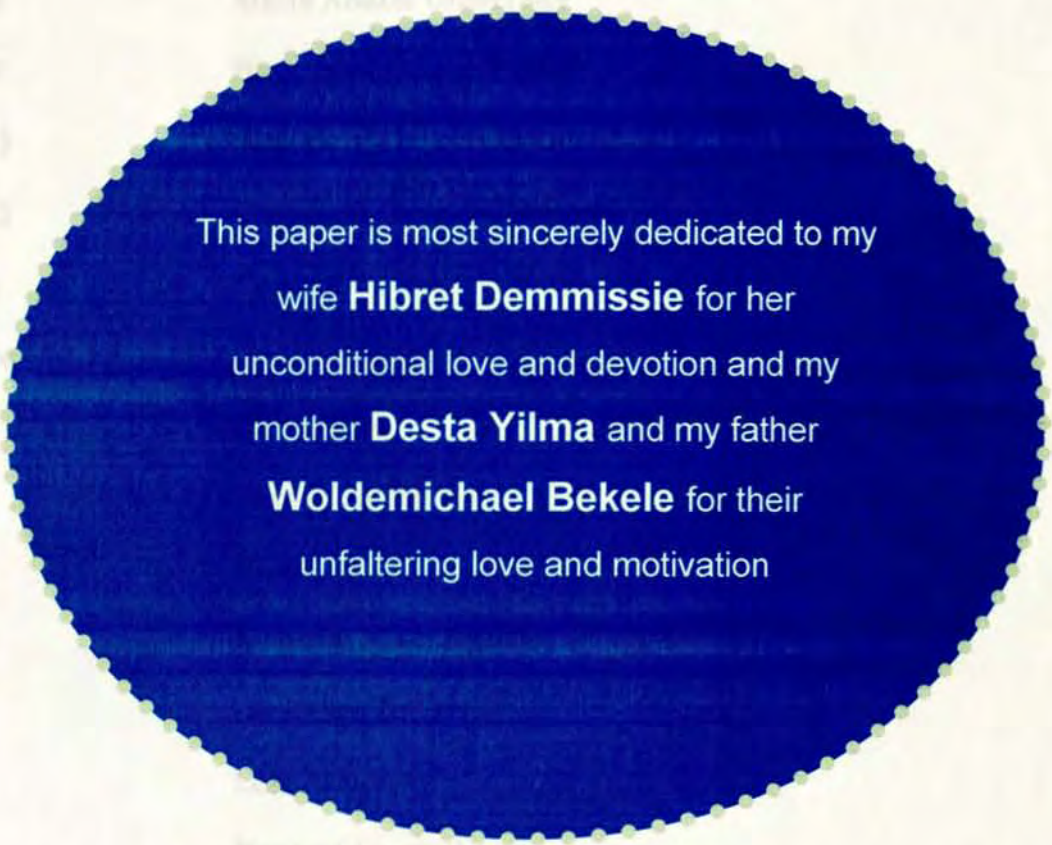
I would like to thank the Horn of Africa Environmental Network and Ethiopian Institute of Agricultural Research (EIAR) for the financial support. I would also like to transfer my real appreciation and gratitude to EIAR managements especially, to Ato Rezene Fessehaaie, National project coordinator and Dr Taye Tessema, vice-coordinator.

I also thank Professor Masresha Fetene, AAU for providing me necessary materials and allow me to use greenhouse facilities. I am thankful to Dr Wallelengh, Hawassa University for his encouragement and extraordinary support.

My deepest heart-felt gratitude goes to my caring wife and classmate for her encouragement, deepest love and patience she bore in managing the family. I am also grateful to my father Woldemichael Bekele, my mother Desta Yilma and my brothers and sisters, who provided me consistent support and encouragement throughout this study. So many thanks to my friends (in alphabetical order) Birhanemeskel Kumbe, Enqubahery

Habtamu, Getu Assefa, Hailamariam Assefa, Isayas Fanataye, Mamush Lelesa, Dr Wondwosen Tsegaye and others for their encouragement and support they endowed.

I am indebted to Environmental Science Program, Faculty of Science AAU, for supporting me with necessary materials. I am also indebted to chemistry department and ecophysiology laboratory, AAU for providing me facilities.


This paper is most sincerely dedicated to my
wife **Hibret Demmissie** for her
unconditional love and devotion and my
mother **Desta Yilma** and my father
Woldemichael Bekele for their
unfaltering love and motivation

LIST OF ACRONYMS

AAEPA	Addis Ababa Environmental Protection Agency
AAS	Atomic Absorption Spectrophotometer
AAU	Addis Ababa University
BAF	Bioaccumulation Factor
BOD	Biological Oxygen Demand
COD	Chemical Oxygen Demand
Con	Control
Conc.	Concentration
Cr	Chromium
E	Transpiration Rate
h	Hour
IAS	Invasive Alien Species
LAI	Leaf Area Index
LA	Leaf Area
ln	Natural Logarithm
mA	Milliampere
min	Minute
$mM/m^2/s$	Millimole per Meter Square per Second

RGR	Relative Growth Rate
RH	Relative Humidity
RTI	Root Tolerance Index
SE	Standard Error
sq cm	Square Centimeter
sq m	Square Meter
sq km	Square kilometer
TF	Translocation Factor
TIR	Tolerance Index of the Root
WMTI	Wet Mass Tolerance Index

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF PLATES

ABSTRACT

Among several invasive alien species (IAS), water hyacinth (*Eichhornia crassipes* (Mart.) Solms) is considered as an emerging issue in the world. This aquatic plant is one of the important, valuable and most promising plants of different purposes. On the other hand due to its fast spread and congested growth, the plant is considered as one of the ten top noxious weeds of great treat to the ecology and economic well being of the planet. Therefore, the present study demonstrates the potential of water hyacinth for the removal of Cr from wastewater and its limitation in natural wetland. This experiment was performed using healthy and young acclimatized water hyacinth collected from Awash river. Cr concentrations of 3, 5, 7, 10 and 20 mg/L were added to five different polyethylene tanks, containing 40 liter tap water cultured with Hoagland's solution and the 6th polyethylene tank as a control group (without Cr). Six similar water hyacinth plants of equal wet mass (wet mass of each plant 12.5 ± 2 g), shoot length (11 ± 2 cm) and root length (6 ± 2 cm) were transferred into each tank and allowed to grow in greenhouse for 42 days. Water hyacinth of similar mass and length were also transferred into 7th polyethylene tank for transpiration data measurements. From each tank plants were harvested randomly each week. Shoot and root length; wet biomass and dry weight of the shoot and root were measured. Relative growth rate (RGR), tolerance index of the root (TIR), tolerance index of wet mass (TIWM), bioaccumulation factor (BAF) and translocation factor (TF) were analyzed. After all the water hyacinth plant samples had been harvested from each tank the water level of the tanks were adjusted to the original level and water samples were collected for the determination of Cr remained in each tank. After 42 days of plant growth transpiration (water loss) of the plant (*Eichhornia crassipes)* were also conducted from the 7th tank using portable Porometer on wet and dry seasons. The RGR of the plant during 42 days of Cr exposure showed decreasing trend in increasing Cr concentration and the growth were inhibited above 15.3 mg/L Cr concentration. The maximum accumulation of Cr 2.52 X 10^3 µg/g in water hyacinth was noted in the plant exposed to 20 mg/L Cr solution. The highest BAF, 506 was obtained in plant

treated in 3 mg/L Cr solution. The root part of the plant accumulates 2.42 to 3.82 times higher than the shoot part. Maximum removing potential 91% was recorded for the plant grown in 3 mg/L Cr containing solution. The maximum transpiration of water hyacinth 9.26 ± 0.26 $mM/m^2/s$ (millimole per meter square per second) was measured at dry season. By considering the LAI of the plant at Aba Samuel wetland, the plant loses (transpires) 18.57 and 12.33 mm of water per day in dry and wet seasons, respectively. The evaporation in dry and wet seasons were 5.32 and 2.54 mm per day, respectively. Therefore, transpiration of water hyacinth exceeds evaporation 3.49 to 4.85 times in dry and wet season, respectively.

Keywords/phrases: Water hyacinth, Wastewater treatment, Cr, Bioaccumulation factor, Translocation factor, Root tolerance index, Wet mass tolerance index, Transpiration, Aba Samuel wetland

1. INTRODUCTION

1.1. Exotic invasive alien species

An exotic species is a non-native plant, animal or microbe deliberately or accidentally introduced into a new habitat. Such species that are able to reproduce and survive outside their habitats where they evolved are also referred to as alien, introduced, invasive, non-native, or non-indigenous (Elton, 2000).

Invasive alien species (IAS) is a species introduced outside its natural distribution; that out-competes native species for available resources, reproduce prolifically and dominate regions and ecosystem because they often arrive in new area unaccompanied by their native predators. The threat to biodiversity due to invasive alien species is considered second only to that of habitat loss. They cause negative economic, environmental and/or social impacts (Mooney and Hobbs, 2000).

The spread of IAS is now recognized as one of the greatest threats to the ecological and economic well-being of the planet. Globalization, with increasing trade, travel and transport of goods across borders, facilitates the spread of IAS (Bolenz *et al.,* 1990). These alien species are causing enormous damage to biodiversity and the valuable natural agricultural systems upon which we depend. They affect the native biodiversity, by transforming the structure and species composition of the ecosystems and by repressing or excluding native species, either directly by out-competing them for resources or indirectly by changing the nutrient cycle. The damage to nature is often irreversible (Julien *et al.,* 1999).

1.2. Water hyacinth

Among several IAS, water hyacinth is an aquatic plant species that is considered as an emerging issue in the world. It is one of a rapidly growing and very productive free-floating aquatic weed, originated in the Amazon South America, where it was kept under control by natural predators (Bolenz *et al.,* 1990). Outside its native range, South America, it has invaded many waterways and quickly grows to very high densities (over 60 kg m^{-2}) thereby completely clogging water bodies (Julien *et al.,* 1999).

Water hyacinth is the eighth fastest growing plant on earth. Its explosive growth is considered unfavorable in natural water bodies and causes undesirable taste and odor to the water (Lockley and Turner, 1961). Due to its fast growth and toughness of its seeds, water hyacinth has caused major problem in the whole world (Aweke, 1993). It is now widely recognized as one of the top ten weeds in the world (Holms *et al.,* 1977).

Water hyacinth was introduced into many countries during late 19th and early 20th centuries, where it spread and degraded aquatic ecosystems. It is still rapidly spreading throughout Africa, where new infestations are creating life-threatening situations as well as environmental and cultural upheaval (Cock *et al.,* 2000).

1.2.1. Plant biology

Scientific name: *Eichhornia crassipes* (Mart.) Solms; Common names: Water hyacinth (English) (Thorne, 1992), Bochae (Ethiopia) (personal communication with the local community), Organism type: Floating aquatic plant (Plate 1).

"A" (Thorne, 1992)

"B" (Source: Daniel, this study)

Plate 1 "A" and "B" water hyacinth on water body

1.2.1.1. Flower

Water hyacinth has attractive bluish purple or lilac colored flower with a yellow spot of two-three inches in size (Plate 2). The flowering period lasts for about fifteen days. When flowering period ends flower stalk bends and the

pike is now under the water surface and seeds are released directly into the water (Thorne, 1992).

Plate 2 Flower of water hyacinth

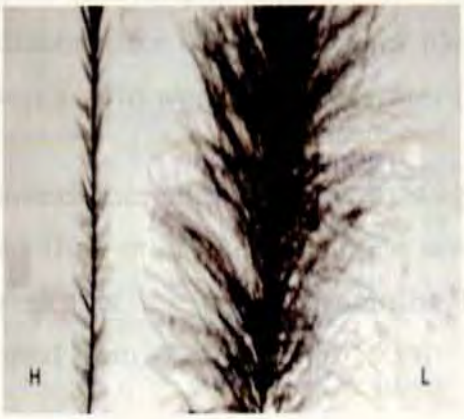
1.2.1.2. Leave

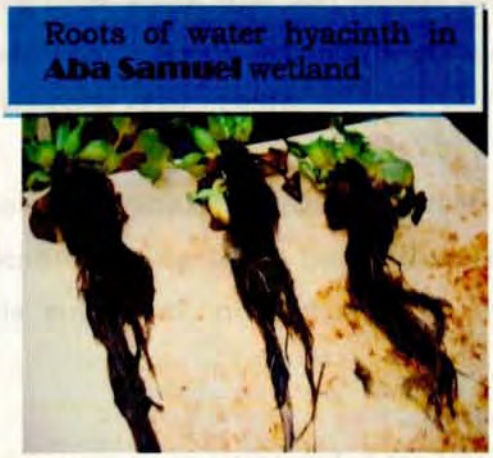
Water hyacinth is a floating plant with round to oval leaves up to ten inches in diameter, although smaller leaves are common. The leaves are bright green, shiny, and held upright; so they act like sails, which facilitates distribution of the plant (Plate 3). The leaf is spongy and thick and helps to keep the plant buoyant (Barrett, 1980).

Plate 3 Leaves of water hyacinth

1.2.1.3. Root

As much as 50% of water hyacinth's biomass is root. Roots are adventitious and fibrous, 10-300 cm in length (Plate 4 "B"). As many as 70 lateral roots per cm give the roots a feathery appearance (Plate 4 "A"). They are dark violet to bluish or pinkish violet (though whitish if grown in total darkness) and contain soluble pigments, including anthocyanins that may protect the root from herbivores (Barrett, 1980).





"A" "B" (Source: Daniel, this study)

Plate 4 "A" and "B" roots of water hyacinth

1.2.1.4. Reproduction

Water hyacinth reproduces sexually by seeds and vegetatively by budding and stolon production. For rapid spreading, the vegetative propagation is more important (Verma *et al.,* 2003). Daughter plants sprout from the stolons and doubling times have been reported 6–18 days (Barrett, 1980). Under favorable conditions of temperature and high nutrient availability, the vegetative propagation is very fast and the edge of mat can even enhance by 60 cm/month. Whether sexual reproduction occurs in the water hyacinth had been a mystery for long time as most of the early investigators could not observe seeds/seedlings in nature. Seed capsules normally contain fewer than 50 seeds each (Barrett, 1980). Each inflorescence can produce more than 3000 seeds and a single rosette can produce several inflorescences

each year (Barrett, 1980). The small, long-lived seeds sink and remain viable in sediments for 15 to 20 years (Gopal, 1987). Seeds germinate on moist sediments or in warm shallow water (Hitchcock *et al.,* 1950).

Population increases mainly by vegetative means. However, observations showed that even after complete eradication of vegetative parts, there was recurrence of the water hyacinth. This suggested, new plant must have developed from seeds (Parija, 1934).

Holm *et al.* (1969) found that two parent plants were surrounded by 300 off-springs in 23 days and by 1200 after 4 months. A single inflorescence will have about 20 flowers and each flower produces 3000 to 4000 seeds. The seeds germinate only when the water recedes 3-4 cm. After germination, the seedlings remains attached to the mud.

1.2.2. Environmental requirements

Water hyacinth grows best in neutral pH, high macronutrients, warm temperature and high light intensity. It tolerates pH levels from 4.0 to 10.0 (Haller and Sutton, 1973). Prolonged cold kills the plant, but reinfestation from seed follows during later warmer periods (Penfound and Earle, 1948). Growth inhibited at water temperature above 33 °C. Salinity is the main obstacle for growth of water hyacinth (Knipling *et al.,* 1970).

1.2.3. Plant origin and geographical distribution

Water hyacinth is South American plant. Outside its natural environment it was first appeared in North America, at the end of 19th century (1884) about the same time it was spotted in Egypt (Ghabbour *et al.*, 2004; Gopal, 1987). It was introduced into many countries during the late 19th and early 20th centuries, where spread and degraded aquatic ecosystems. It is still rapidly spreading throughout Africa, where new infestations are creating life-threatening situations as well as environmental and economic upheaval (Cock *et al.*, 2000). Water hyacinth was first reported in lake Victoria in 1989 and quickly spread around the lake margins (Twongo, 1991). Water hyacinth now flourishes in all continents but Europe, where it exists, but does not flourish as a result of climatic conditions. Water hyacinth grows abundantly throughout the tropical and subtropical regions of the world (Cock *et al.*, 2000).

In Ethiopia this weed was officially reported in the year 1956 in Koka dam and Awash river. Although the infestation was small the earliest observation of water hyacinth was reported in Dugda Bora district between the year 1949 to 1958 (Stroud, 1994). The point of introduction and the primary source of infestation were assumed to be Aba-Samuel wetland that is enriched by Akaki river, in which almost all types of wastes of Addis Ababa city have been damped. Farmers in the area reported that foreign inhabitants residing near the wetland introduced water hyacinth to the water body (Senait *et al.*, 2007). Except lake Elen (8 km North of Alemtena town) and Koka dam, other rift valley Lakes: Ziway, Langano, Abijata, Shala

and Awassa are proved to be free from water hyacinth, but the risk (potential) for their infestation is still there (Rezene, 2005). During 1962 the plant succeeded in infesting the whole stretch of the White Nile from Juba to Jebel Aulia dam; the whole length of the Sobat river from its mouth eastwards up to Baro and Gillo rivers and south wards from Pibo river to Akobo (Rezene, 2005).

1.2.4. Negative impacts

Water hyacinth is considered as one of the world's worst aquatic plants. It can double its size in a week time and a mat of medium sized plant may contain 2 million plants per hectare that weigh 270 to 400 ton (Epstein, 1998). Water hyacinth poses serious socioeconomic and environmental problems for millions of people in riparian communities (Howard, 2003).

The rapid growth of water hyacinth forms expansive colonies of tall and interwoven plants. It blankets large water bodies, creating impenetrable barriers and obstructing navigation (Zeiger, 1962). The dense mats of water hyacinth loses water from a body of water, reduce light to submerged plants, prevents oxygenation of water and the establishment of phytoplankton and much of the zooplankton, making areas unsuitable for fish feeding and fish breeding (Holms *et al.*, 1977; Howard, 2003; Barret, 1980; Annushee, 2005). The resultant lack of phytoplankton alters the composition of invertebrate communities, ultimately affecting fisheries, decreased fish population destroying native plants and wildlife habitat (Hansen *et al.*, 1971). The plant interferes with navigation, recreation, irrigation, power

generation and enhances reduction of biodiversity. They also create good breeding conditions for mosquito vectors of malaria (Epstein, 1998; Mailu, 2001).

1.2.4.1. The water loss (transpiration) impact of water hyacinth

Transpiration is the evaporation of water in the vascular system of plants through leaf stomata. Opening and closure of stomata is controlled by their guard cells. Hence, transpiration is a bio-physical process, because it involves tissues of living organisms. Liquid water extends through the plant from the soil or water to the leaf cell surfaces where it is converted from a liquid into gas through the process of evaporation (Salisburg and Wadsworth, 1992; Noble, 1991).

The hydrological impact of water hyacinth is increased transpiration rates. The increased transpiration due to dense mates of water hyacinth can have serious implications where water is already scarce for increasing human demands, feeding and breeding for fish, birds and other organisms (Noble, 1991). For example in Texas, USA water hyacinth transpiration rates are estimated to be three to six times normal evaporation. Immersed and floating plants, such as cattail and water hyacinth, because of their structure and leaf area, transpire more water than would evaporate in the same area. Therefore, lakes filled with immersed and floating plants will lose more water to the atmosphere than will open water lakes having few plants (Maarguarite and Rawlik, 1993).

In all wetlands evapotranspiration causes major loss of water (Winter, 1992). Despite evaporation and transpiration causing the largest water loss from wetlands, aspects of transpiration characteristics of wetland vegetation and their role in wetland hydrology are not well understood (Winter, 1992). Studies of transpiration are inconclusive, for it is not clear whether the presence of wetland vegetation increases or decreases water losses from a body of water. Obviously, the presence of vegetation retards evaporation from the water surface, but the question is whether the transpiration of water through the plants matches or exceeds the difference (Kadlec, 1989).

Evaporation and transpiration are enhanced by meteorological conditions, such as solar radiation or surface temperature that increase the value of the vapour pressure at the water surface (Mitsch and Gosselink, 1993). Despite adequate moisture, certain plants can also physiologically limit transpiration through the closing of leaf stomata during a period of stress (Mitsch and Gosselink, 1993). It is known that environmental variables can condition plant physiology markedly, thereby regulating transpiration (Snyder and Boyd, 1987; Van der Weert and Kamerling, 1974). As pointed out by Lee (1967) transpiration seems to be controlled by stomata rather than by environmental conditions.

1.2.5. Potential uses of water hyacinth

Although water hyacinth is often seen as a weed responsible for many of the problems outlined above, there are other schools of thought that advocate

useful applications of the plant. These groups of people strongly agree that *Eichhornia crassipes,* is one of the most productive and most promising aquatic plants that could serve different purpose in different part of the world (Nevena, 2006; Abbasi and Ramasamy, 1999). Fast growth is a feature valued in crops grown by man. The water hyacinth would therefore, have a great potential if it is seen as raw material for industries or if incorporated into agricultural practice (Anushee, 2005). Water hyacinth contains more than 95% water but due to its fibrous tissue and a high energy and protein content, it can be used for a variety of useful applications, such as: wastewater treatment, alcohol production, biogas production, compost, animal fodder and a number of other uses some of which have been developed and others are still in their infancy (Tchobanoglous *et al.,* 1989; Woomer *et al.,* 2000; Stocker and Haller, 1999; Nigam, 2002; Van Der Meer and Verdegam, 1996; Kumar, 2005).

1.2.5.1. Water hyacinth for the application of wastewater treatment

Water hyacinth (*Eichhornia crassipes* (Mart.) Solms) is quite versatile plant as far as phytoremediation capability is concerned. Natural wetland systems colonized by water hyacinth could serve as "nature's kidney" for proper effluent treatment to preserve the earth's precious water resource from getting polluted (Tchobanoglous *et al.*, 1989).

Eichhornia crassipes, has attracted considerable attention because of its ability to grow in heavily polluted water together with its capacity for metal ion accumulation (Annushee, 2005). The plant has been used for decontaminating inorganic nutrients, persistent organic pollutants, as well as toxic metals (Tchobanoglous *et al.,* 1989).

Water hyacinth can serve for the reduction of BOD, COD, total nitrogen as well as other nutrient-rich wastes (Trivedy and Thomas, 2004; Sinha and Sinha, 2000). The plant can also be used for the application of phosphorus removal, agricultural runoff, dairy effluents, pulp and paper mill, textile and electroplating effluents and for the treatment of recalcitrant organic chemicals such as herbicides (Roy and Hanninen,1994; Debusk *et al.,* 2001; Reddy *et al.,* 1982; Tripathi and Upadhyay, 2003; Singhal and Ray, 2003; Trivedy and Gudekar, 1987).

In terms of growth output as well as nutrient reduction from wastewater, water hyacinth has been found to perform better than other plants such as pennywort (*Hydrocotylea umbellat*) and water lettuce (*Pistia stratiotes*) (Sooknah and Wilkie, 2004).

1.2.5.1.1. Heavy metal removal capability of water hyacinth

In the last few decades, significant progress in bioremediation of metals and radionuclide has been made. Aquatic plants are known to accumulate metals from their environment and affect metal flux through ecosystems. Water hyacinth has exceptionally high affinity and accumulation capacity for several metals (Zhu *et al.,* 1999). Particularly roots of water hyacinth have high capacity not only for nutrient uptake but also for trace elements uptake (Guo and Zhang, 2002).

Water hyacinth has been used for treatments of various metals: for example, for the application of silver recovery, for removal of lead, copper, zinc, aluminum, and also showed highest removal capacity for cadmium,

chromium, arsenic and nickel from low external concentrations (Zhu *et al.*, 1999; Guo and Zhang, 2002; Liao and Chang, 2004; Roldan, 2002; Mukherjee Mondal, 1995).

1.2.5.1.2. Application for chromium removal from industrial wastes

In recent years, contamination of the environment by Cr, especially Cr(VI) has become a major area of concern. Cr is used on a large scale in many different industries, including metallurgy, electroplating, production of paints and pigments, tanning, wood preservatives, Cr chemical production and pulp and paper production are among others (Guo and Zhang, 2002; Mukherjee Mondal, 1995). Due to its huge industrial use, this metal is one of the important contaminant released into the environment (Nriagu and Nieboer, 1988). Often wastes from such industries are used as a fill material at numerous locations to reclaim marsh lands, and for backfill at sites following demolition. At many such sites, leaching and seepage of Cr(VI) from the soils into the groundwater poses a considerable health hazard. The tanning industry is an especially large contributor of Cr pollution to water resources (Salunkhe *et al.,* 1998).

In Addis Ababa Ethiopia, there is increasing concern about water pollution from industries. According to Addis Ababa environmental protection agency (AAEPA) more than 80% of industries in the city discharge their effluent into the environment without any appropriate treatment (AAEPA, 2004). Of all the discharged contaminates, the total Cr discharge of paint manufacturing

industries in the year 2000 alone exceeds 2000% above the industrial effluent discharge standard limit of the country (AAEPA, 2004).

Moreover, treatments of this wastewater is expensive; so that many poor countries only employ an initial treatment and this treatment still leaves chromium above the legal discharge limit for surface as well as for ground waters (Alves *et al.,* 1993). Therefore, in the case of Cr, further treatment is often mandatory such as: ion exchange resin, reverse osmosis and electrolysis have been investigated as methods of further purification (Kocaoba and Akcin, 2002; Vlyssides and Israilides, 1997; Hafez *et al.,* 2002). However, these methods are expensive and are often not considered cost effective for small sized industries.

Currently there is considerable interest in the use of bioremediation and phytoremediation to treat Cr contaminated soils, sediments, and waters (Chandra *et al.,* 1997). Phytoremediation of Cr pollution can be achieved by extraction of the metal from polluted soils and water into harvestable plant tissues (Terry and Banuelos, 2000). The choice of appropriate method of remediation is governed largely by the mobility, distribution and speciation of Cr in soils and plants. Knowledge of the physical, chemical, and biological processes affecting the mobility, distribution, and chemical speciation of Cr in the environment is therefore, essential in order to devise cost-effective and efficient remediation strategy (Chandra *et al.,* 1997).

Therefore, in view of the seriousness of Cr pollution, considerable efforts have to be made to develop suitable methods for the remediation of

chromium contaminated soils, sediments and water (Nriagu and Nieboer, 1988). Water hyacinth, whose increased appetite for nutrients and high explosive growth rate could be harnessed to remove Cr from the environment (Salt *et al.,* 1995).

Lots of researches have been done to show the potential of water hyacinth to accumulate heavy metals such as: silver, lead, cadmium, zinc, iron and mercury. There are very few plant species such as: *Sutera fodina, Dicoma niccolifera* and *Leptospermum scoparium* which have been reported to accumulate Cr(VI) in their tissues (Peterson, 1975). However, study on the accumulation potential of water hyacinth to Cr is scarce, particularly here in Ethiopia. Study on the potential of water hyacinth is key for Cr removing from wastewater due to the fact that the plant is already introduced into the environment and the central unit of the treatment engine is driven by photosynthesis, thus the process is sustainable, and cost efficient.

Therefore, the present study was conducted to assess the potential of water hyacinth to remove Cr(VI) from wastewater and evaluate the water loss impact of this exotic alien invasive plant, due to transpiration.

2. OBJECTIVES

2.1. General objective

To study and compare the potential use of water hyacinth for wastewater treatment with its limitation

2.2. Specific objectives

To:

- determine the effect of chromium on the growth of water hyacinth and estimate the toxicity level;
- evaluate the chromium removing potential of water hyacinth from chromium contaminated water;
- assess the chromium bioaccumulation factor and the chromium localization of the plant part (tissue);
- estimate the amount of water loss from the surface of a water body covered by water hyacinth, and
- compare water loss from covered water surface (transpiration) by water hyacinth with uncovered water surface (evaporation).

3. MATERIALS AND METHODS

3.1. Description of the experimental sites

Experiments were conducted in a greenhouse at Addis Ababa University, over a period of 7 weeks between May and July 2008. The average temperature and relative humidity of the greenhouse were 26.87 ^{0}C and 22.07%, respectively. The average water temperature and pH were 22.43 ^{0}C and 6.4, respectively.

The plant material, *Eichhornia crassipes* were obtained from Awash river located near Wonje town.

The transpiration potential of the plant was determined by evaluating the leaf area index (LAI) of the plant at Aba Samuel wetland. The wetland is found 20 km South-East of Addis Ababa. It was chosen based on the local and global biodiversity importance of the site and the severity of the invasion of water hyacinth. Evaporation from the open water surface (free from vegetation) and transpiration from the total vegetation cover (water hyacinth) of the projected area were also assessed from Aba Samuel wetland.

3.2. Experimental conditions

During the experiment the following instruments have been used: Polyethylene tanks were used for water hyacinth sample collection from the field and for plant growth in the greenhouse. Digital analytical balance (METTLER Toledo, Model AT250, Switzerland) for weighing plant samples (wet and dry biomass). Portable pH meter, for measuring the pH and

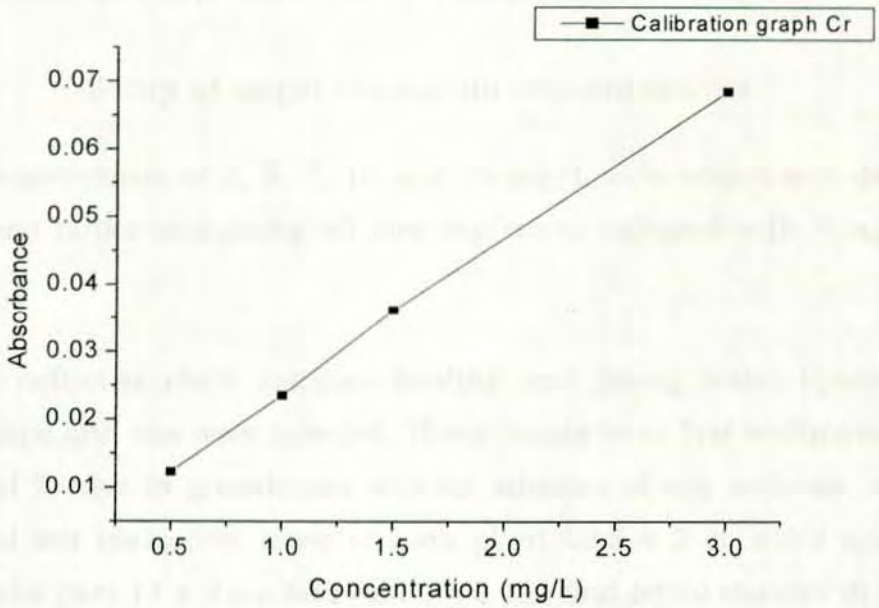
temperature of the water. Oven (J.P.SELECT, Spain) was used for drying the wet plant samples. Ceramic mortar and pestle were employed for grinding and homogenizing plant samples. Quick-fit round bottom flask (150 mL) fitted with reflux condenser were used in Kjeldahl apparatus hot plate to digest the samples. (BUCK SCIENTIFIC MODEL 210VGP East Norwalk, USA) Atomic Absorption Spectrophotometer (AAS) equipped with deuterium ark was used for analysis of Cr. Leaf area meter (ADC, AM 100 Analytical Development Company Limited EN110AQ, 1997, UK) was used for measuring plant leaf area. Quadrate (50 cm x 50 cm) made of timber was used for measuring leaf area index. Portable analytical balance was also used for measuring water hyacinth mass in the field. Porometer (ADC, Bio-Scientific Ltd, Hoddeson, EN110DB, 2002, UK) was employed for measuring transpiration of water hyacinth.

3.2.1. Setup of instruments and calibrations

AAS standard solution containing 1000 mg/L (Buck Scientific) were used for preparing intermediate standard (10 mg/L). The working standard solutions were prepared freshly by appropriately diluting the intermediate standard of Cr with de-ionized water in the range of 0.5-3 mg/L (Table 1). The calibration curve (Figure 1) of the working standard solution was made after optimizing maximum signal intensity and sensitivity of the instrument (Table 2).

Table 1 Working standard for determination of Cr in plant and water samples

Conc. (mg/L)	Absorbance
0.5	0.01228
1	0.02351
1.5	0.03614
3	0.06836



$R^2 = 0.999$

Figure 1 Calibration graph of Cr standard solution

Table 2 Instrumental operating conditions for determination of Cr

Element	Wave length (λ) (nm)	Slit width	Lamp energy (mA)
Cr	357.9	0.7	2

A steady state Porometer was used to measure water loss (transpiration) of water hyacinth. This method measures the flow rate of dry air necessary to maintain a constant relative humidity (nulled to ambient RH) inside a cuvette which has been clamped onto a transpiring leaf. The Porometer consists of a cuvette with a broadleaf aperture (6.25 cm^2) which permits precise measurements of water loss by transpiration.

3.2.2. Setup of target chromium concentrations

Cr(VI) concentrations of 3, 5, 7, 10 and 20 mg/L were added to 5 different polyethylene tanks containing 40 liter tap water cultured with Hoagland's solution.

From the collected plant samples healthy and young water hyacinth of similar shape and size were selected. Those plants were first acclimatized for a period of 7 days in greenhouse without addition of any nutrient. After 7 days equal wet mass (wet mass of each plant 12.5 ± 2 g), shoot and root length (aerial part 11 ± 2 cm and root 6 ± 2 cm) and equal number (6 plants in each pot) were transferred into round polyethylene containers of 60 cm depth and 50 cm width containing various concentrations of Cr and tap water cultured with Hoagland's nutrient solution. One control group was prepared in which the same volume of water cultured with Hoagland

solution in a similar way but Cr was not added. The total volume of the solution in each container was kept constant by adding tap water cultured with Hoagland's solution every 5 days for water lost through plant transpiration and evaporation. Each experiment was performed in triplicates.

The same procedure for plant growth described above was maintained, for measuring the transpiration aspect of water hyacinth but Cr was not added in the solution.

3.3. Reagents and nutrient medium

During the experiment the following chemicals have been used: 69-72% HNO_3 and H_2O_2 were used for digestion of plant samples. Analytical reagent $K_2Cr_2O_7$ was used for Cr stock solution (1000 mg/L) preparation. Hoagland's nutrient solution was prepared from macro and micro nutrients. De-ionized water was used for preparation of Hoagland solution, sample dilution, and cleaning (rinsing) purpose. Tap water was used for water hyacinth growth medium. 3% (v/v) HNO_3 used for cleaning purpose.

3.3.1. Chromium stock solution preparations

Mass of 3.734 g $K_2Cr_2O_7$ (dried at 100 ^{0}C for 1 h) were measured and transferred to 1000 mL volumetric flask containing 100 mL de-ionized water to that 5 mL 1:1 HNO_3 was added to dissolve the salt and de-ionized water was added to the mark and the solution was kept in polyethylene bottle and stored in refrigerator less than 4 ^{0}C (Hashitani *et al.*, 1987).

3.3.2. Preparation of Hoagland's nutrient solutions

Plant nutrient solution was prepared by Gamborg and Wetter (1975) method.

Solution "A"

280 mg H_3BO_3

340 mg $MnSO_4 \cdot H_2O$

10 mg $CuSO_4 \cdot 5H_2O$

22 mg $ZnSO_4 \cdot 7H_2O$

10 mg $NH_4Mo_7O_{24} \cdot 4H_2O$

The volume was adjusted to 100 mL with de-ionized water. Stored at 4 ^{0}C.

Solution "B"

0.5 mL Conc. H_2SO_4

The volume was adjusted to 100 mL with de-ionized water. Stored at 4 ^{0}C.

Solution "C"

3.36 g Na_2EDTA

2.79 g $FeSO_4$

The volume was adjusted to 400 mL with de-ionized water.

The solution was heated to 70 ^{0}C while stirring until the color turns yellow brown cooled down, the volume adjusted to 500 mL and stored at 4 ^{0}C.

Hoagland's stock solution

4.7 g $Ca(NO_3)_2 \cdot 4H_2O$

2.6 g $MgSO_4 \cdot 7H_2O$

3.3 g KNO_3

0.6 g $NH_4H_2PO_4$

5 mL Solution "A"

0.5 mL Solution "B"

The volume was adjusted to 500 mL with de-ionized water. Stored at 4 ^{0}C.

Hoagland's nutrient solution

100 mL stock solution

5 mL solution "C"

The volume was adjusted to 1000 mL with de-ionized water prepared just before use.

3.4. Sampling

To determine Cr accumulated in the plant parts, water hyacinth samples were randomly harvested each week, from 9 to 11 A.M for consecutive six weeks (42 days) from all polyethylene tanks of different Cr concentrations (0 (control), 3, 5, 7, 10 and 20 mg/L).

Percentage removal capacity of the plant of each treatment tanks (3, 5, 7, 10 and 20 mg/L) were determined after all the plant samples (water hyacinth) had been harvested and volumes of the water in the plant growth tank were adjusted to the original level. Water samples (500 mL) were collected from all tanks and filtered with Whatman filter paper No 41 (0.45 µm pore size).

After 6 weeks of the plant growth, transpiration potential of the plant was conducted using portable Porometer. Sampling was conducted for consecutive 6 sunny and 6 rainy days. May 1 to 6 "Bega" (sunny season) and June 3 to 14 "Kiremt" (rainy season) at hourly intervals from sunrise to sunset (7:00 to 18:00 standard time) in the year 2008. To avoid bias of the analysis by stomatic variability, three simultaneous measurements on each of three different leaves of the plant with different orientations were conducted. Care was taken during sampling to ensure that all leaves were completely dry before reading.

The LAI, the area of which this plant has covered the water surface was assessed from (50 cm x 50 cm) quadrant measured four times at three randomly selected sites of Aba Samuel wetland: April, May, June and September 2008.

3.5. Sample preparation

The harvested plants were washed three to four times with tap water to remove any adsorbed materials on the plant external part. After the separate part (shoot and root) were oven dried separately at 85-90 ^{0}C for 24 h the samples were weighed, ground and homogenized separately.

Well powdered and homogenized plant parts of 0.25 g were weighed and transferred into a round bottom flask (150 mL) to this 5 mL HNO_3 and 2 mL H_2O_2 had been added and underwent wet digestion overnight. The wet digested solutions were then fitted to micro Kjeldahi digestion apparatus by setting the temperature dial to 6 (150 ^{0}C) for 3 h and 30 min. The digested

solution was then cooled for 15 min at room temperature. After cooling, the solution was filtered into 50 mL volumetric flask fitted with Whatman filter paper (0.41 µm) and the samples were diluted with de-ionized water to the mark.

From each collected water samples, 100 mL were transferred into 250 mL round bottom flask containing 2 mL HNO3 and 1 mL HCl. The flask was placed on hot plate covered with an elevated watch glass. The temperature was adjusted to approximately 85 ^{0}C. After 2 h and 30 min the volume of the sample was reduced to 20 mL. The flask was allowed to cool for 15 min and transferred to 50 mL volumetric flask, the volume was adjusted to the mark with de-ionized water (Martin *et al.*, 1994).

The blank solution was prepared by digesting the mixture of reagents (69-72% HNO_3 and H_2O_2) following the same digestion and dilution procedures.

The samples were kept in the refrigerator below 4 ^{0}C, until levels of Cr in the sample solution were determined by AAS.

3.6. Measurement and analysis

3.6.1. Biometrics

Every week the shoot and root length of the harvested plant samples from all tanks were separated and measured using ruler. Wet and dry masses of the shoot and root part were measured using analytical balance.

3.6.1.1. Tolerance index (TI)

Root tolerance index (RTI) of root length and wet biomass tolerance index (WMTI) of the total fresh weight are commonly used to quantify plant metal tolerance (Benicassa, 1988). The higher the TI, the better the tolerance.

$$RTI = \frac{\text{Root growth in metal containing solution (cm)}}{\text{Root growth in control group (cm)}}$$

$$WMTI = \frac{\text{Wet biomass in metal containg solution (g)}}{\text{Wet biomass in control group (g)}}$$

3.6.1.2. Relative growth rate (RGR)

Plant growth rate was evaluated according to Radford (1967).

$$RGR = \frac{\ln dm2 - \ln dm1}{t2 - t1}$$

Where: dm_1 and dm_2 = Initial and final total dry mass

t_1 and t_2 = Time interval between two samplings (days)

3.6.2. Determination of chromium

The accumulation of metal in plant part is expressed in microgram (μg) of metal per gram (g) of dry matter. To determine the original Cr concentration in the plant part (before exposed to different chromium solutions), 6 plants were randomly chosen, their shoot and root parts were separated and Cr concentrations were analyzed.

The prepared sample solutions were aspirated in to the AAS instrument and concentration or absorbance readings of total chromium concentrations of the separate plant part (shoot and root) and water samples were examined. Average values of three replicates were taken for determination of Cr in each measurement. Blanks were also analyzed in the same manner. The concentration reading from AAS was directly used and those reading above detection limit of the instrument, absorbances were recorded. All absorbance readings were converted into concentrations using the calibration curve and Origin 6 software.

The recorded concentrations from AAS and the converted absorbance from Origin software was changed into actual concentration of metal in the sample using the equation:

$$\text{Conc. } (\mu g/g) = \frac{\text{AAS reading } (\mu g/g) \text{ x Dilution volume (mL)}}{\text{Digested sample mass (g)}}$$

3.6.2.1. Bioaccumulation factor (BAF)

The BAF provides an index of the ability of the plant to accumulate the metal with respect to the metal concentration in the external solution, which is the ratio of particular metal concentration in the plant to concentration in external solution. This parameter is a useful parameter to evaluate the potential of the plant in accumulating metals from soil/water (Zayed *et al.*, 1998).

$$BAF = \frac{\mu g/g \text{ of Cr in plant body (dry mass)}}{\mu g/g \text{ of Cr in external solution}}$$

3.6.2.2. Translocation factor (TF)

Movement of metal containing sap from the root to the shoot termed as translocation. TF gives the root/leaf chromium concentration and depicts the ability of the plant to translocate the metal species from roots to shoot at different concentrations (Zayed *et al.*, 1998).

$$TF = \frac{\text{Cr content of the root } \mu g/g}{\text{Cr content of shoot } \mu g/g} \times 100$$

3.6.3. Determination of transpiration

3.6.3.1. Porometric measurement

Transpiration potential of water hyacinth was measured in millimole per square meter per second ($mM/m^2/s$). The transpiration rate of this plant at each hour was obtained from the mean of the two successive hour Porometer readings (E). Measurements of transpiration and leaf conductance before sunrise and after sunset have shown that the stomata of this species close in the absence of light (Groeneveld, 1986). To set the zero points on the parabolic diurnal transpiration curve, the sunrise and sunset time were assumed to be at 6:00 and 19:00 standard time.

3.6.3.2. Leaf area index (LAI) and plant growth

LAI were calculated from total leaf area per surface (ground) area ($cm^2\ cm^{-2}$). Simple LAI is used to describe the average number of leaf layers covering a given site (Brown and Blaser, 1968).

From the laid quadrates all leaves were cut, collected and counted. Representative leaf samples from each quadrant were separated into 10 types (by leaf size). Then the leaf samples were subsequently transported to the laboratory and leaf area was measured in two replicates. The area covered by the plant in a quadrate was calculated by multiplying total number of leaves with the mean of the representative leaf samples. Individual leaves were also examined for dead and other non-photosynthetic (brown) areas, which were removed.

Plant growth is described in two ways: one by reporting the percentage of water surface covered by plant species and second and more useful method is by reporting the plant density; wet plant mass per unit of surface area (US EPA, 1988). In this study, the water hyacinth growth in Aba Samuel wetland was assessed by both methods: by the plant density and percentage of the water surface covered.

3.6.3.3. Determination of volume of water transpired

Volume of water transpired from water hyacinth was calculated from the diurnal mean transpiration of the 12 hour of the sunny and rainy days, separately. To convert quantity of water transpired from mM m^{-2} s^{-1} to mL m^{-2} s^{-1} and progressively to m^3 m^{-2} s^{-1} the following formula was used.

Volume of water transpired (mL m^{-2} s^{-1}) =

$$\frac{\text{(E)Transpiration in mole x Molecular mass of water } \left(18 \frac{g}{mole}\right)}{\text{Density of water } \left(1 \frac{g}{mL}\right)}$$

1mole = 1000 mM

3.6.3.4. Additional data source

In addition to the primary data generated during this study, evaporation data was collected from National Meteorology Agency (NMA) for the year 2000-2006.

3.7. Data analysis

ANOVA, regression, mean and standard error of the mean (± SE) of the shoot length, root length, wet biomass, dry biomass, relative growth, Cr concentration, RTI, WMTI, BAF, TF, percentage removal and volume of water transpired were calculated using SAS 8.2 (2001) program, Origin 6 software and Microsoft excel (2007). ($P<0.05$) was regarded as statistically significant.

4. RESULTS AND DISCUSSION

4.1. Growth of water hyacinth in chromium containing wastewater

4.1.1. Shoot and root length

Results of mean shoot, root length (cm) and the root tolerance index (RTI) of water hyacinth after exposure to 3, 5, 7, 10 and 20 mg/L Cr for 42 days are given in Table 3. It was noted that the shoot and root length of the water hyacinth decreased with increasing Cr concentration. However, there is no significance difference ($P<0.05$) between the plants grown in 3 and 5 mg/L Cr containing solution and the control group. The shoot and root length of the plant exposed to 7, 10 and 20 mg/L Cr solution significantly decreased ($P<0.05$) compared to the control group.

Phytotoxicity symptoms are usually quoted as a percentage growth inhibition (Cervantes *et al.*, 2001). Figure 2 shows the shoot and root length growth of water hyacinth exposed to 3, 5, 7, 10 and 20 mg/L Cr containing water solutions. The shoot length of the plant exposed to 20, 10 and 7 mg/L Cr concentration decreases by 43, 25 and 23% and the root length decreased by 44, 36 and 32% compared to the control group. It was reported that at higher Cr concentration (plants grown in 25 to 50 mg/L), plants were stunted and had narrow and brownish red leaves with small necrotic areas with poorly developed root system (Hunter and Vergnano, 1953). Growth change is often the first and most obvious reactions of plants under heavy metal stress (Hagemeyer, 1999). In this study wilting and plasmolysis

on water hyacinth exposed to 10 and 20 mg/L Cr solutions were observed. It was also reported that Cr can affect roots of plants causing wilting and plasmolysis (Roy *et al.*, 1992). The finding of this study partly agrees with Hunter and Vergnano (1953) that they observed chlorotic leaves and normal roots at low Cr concentrations (5–10 mg/L).

Table 3 Mean shoot and root length and root tolerance index of water hyacinth after 42 days exposure to Cr

Conc. (mg/L)	Mean length (cm)				RTI
	Shoot	± SE	Root	± SE	
3	12.23	1.07	9.25	1.09	0.78
5	11.14	0.60	8.8	0.84	0.75
7	10.05	0.68	8.02	0.44	0.68
10	9.83	0.70	7.62	0.58	0.64
20	7.36	1.02	6.57	0.20	0.56
Con	13.02	1.11	11.83	1.55	

Con = Control; RTI = Root Tolerance Index; SE = Standard error

RTI of the plant decreased with increasing Cr concentration. This is in agreement with Shewry and Peterson (1974); they observed that the first toxic effect of Cr was inhibition of root growth. Shoot growth is only being affected at higher levels. This might be due to degradation of protein in plants which results in the inhibition of nitrate reductase activity (Panda and Choudhury, 2004). High production of H_2O_2 and O_2^{1-} radicals were reported in many plant species exposed to Cr and the metal has been implicated in the generation of oxidative stress (Roy *et al.*, 1992; Dixit *et al.*, 2001). It is also known that Cr is a non-essential heavy metal, and has inhibitory effects on plant growth.

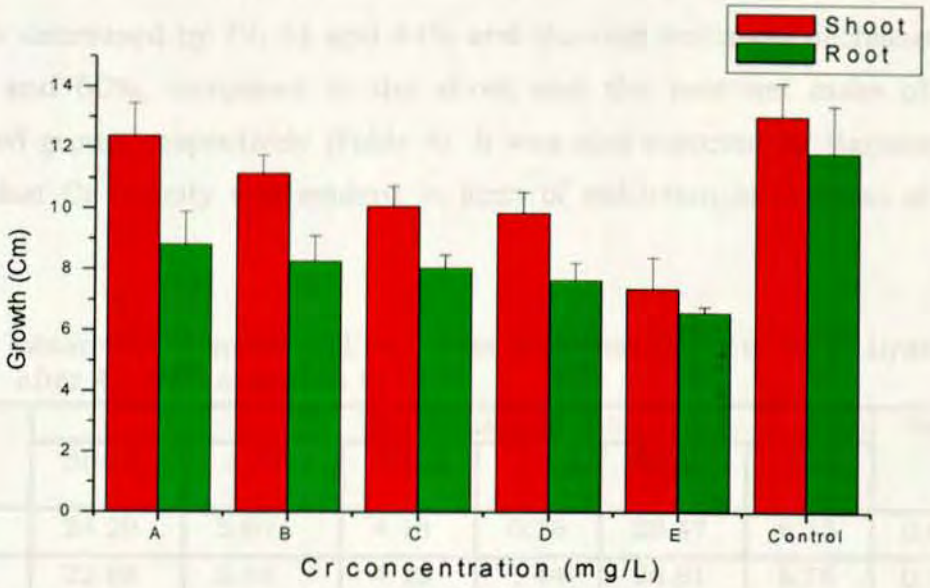


Figure 2 The mean length of shoot and root (cm) of water hyacinth after 42 days exposure to Cr solution (A=3; B=5; C=7; D=10; and E=20)

4.1.2. Mean wet biomass and wet mass tolerance index

The mean wet biomass of the shoot, the root and WMTI of the water hyacinth grown in various Cr concentrations for 42 days are given in Table 4. As it is shown from the Table, both the shoot biomass and the root biomass decreased with increasing Cr concentration. However, the shoot and root biomass of the plant grown in 3 and 5 mg/L Cr solution did not significantly decreased ($P<0.05$) but, the plant exposed to 7, 10 and 20 mg/L Cr solution significantly decreased ($P<0.05$) compared to the controlled

group. The shoot wet mass of the water hyacinth exposed to 20, 10 and 7 mg/L Cr decreased by 72, 51 and 44% and the root wet mass decreased by 73, 64 and 60%, compared to the shoot and the root wet mass of the controlled group, respectively (Table 4). It was also reported by Hagemeyer, (1999) that Cr toxicity was evident in form of reduction of biomass of the plant.

Table 4 Mean wet biomass and wet mass tolerance index of water hyacinth after 42 days exposure to Cr

Conc. (mg/L)	Wet biomass (g)						WMTI
	Shoot	± SE	Root	± SE	Total	± SE	
3	24.29	5.80	4.18	0.76	28.47	6.52	0.69
5	22.68	5.46	4.13	1.44	26.81	6.78	0.65
7	19.55	4.08	4.05	1.02	23.60	5.07	0.53
10	17.17	3.43	3.65	0.30	20.82	3.73	0.45
20	9.82	0.48	2.72	0.23	12.54	0.52	0.27
Con	35.18	10.72	10.23	3.13	45.42	13.56	

WMTI = Wet Mass Tolerance Index

The WMTI of the plant decreased with increasing Cr concentration (Table 4). Visible damage symptom like wilting and chlorosis was observed on plants exposed to 10 and 20 mg/L Cr solutions. It was reported that the initial symptoms of Cr toxicity appeared as severe wilting and chlorosis in water hyacinth, as confirmed by Turner and Rust (1971).

As a result of root damage, water and nutrient uptake is diminished and this is evidenced by wilting and reduced wet biomass and by visual symptoms, like mineral deficiencies (e.g., Fe deficiency chlorosis) in leaves. Reduced

shoot wet biomass probably results in reduced leaf expansion. This was observed in this study, in higher Cr concentrations (10 and 20 mg/L). Turner and Rust (1971) proposed that chlorosis appeared in the upper leaves of the plant, as an indirect effect of Cr, probably due to the retardation of Fe and Zn translocation.

In this study, the following sequence of sensitivity of the symptoms of Cr toxicity was observed: root wet mass > shoot wet mass > root length > shoot length. It agrees with Hauschild (1993) that he observed the following sequence of sensitivity of symptoms of Cr toxicity: root growth > visible damage symptoms > leaf growth.

4.1.3. Pattern of relative change in wet mass growth with time

Results of the total biomass growth pattern of water hyacinth exposed for 42 days under different Cr concentrations are shown in Figure 3. In this study the growth pattern of the plants exposed to 3, 5, 7, 10 and 20 mg/L Cr containing water solution increased with time, regularly up to 21st day and sudden increment was observed after the 21st day (3rd week) except the plant in 20 mg/L. However, the growth of all treatments was almost in a regular pattern after 28th day (4th week). The maximum wet mass growth was measured at the 42th day (6th week) for all treatment groups, except for 20 mg/L Cr concentration (Figure 3). This agrees with Jayaweera and Kasturiarchchi (2004) who documented optimum growth of water hyacinth was observed when the plant attained 4 to 6 weeks of age.

The wet mass of the plant in 20 mg/L Cr treatment decreased after 21st day and the maximum biomass growth was observed at 21st day (3rd week). After the 35th day of Cr exposure, the wet mass of the plants in 3 and 5 mg/L was similar (Figure 3).

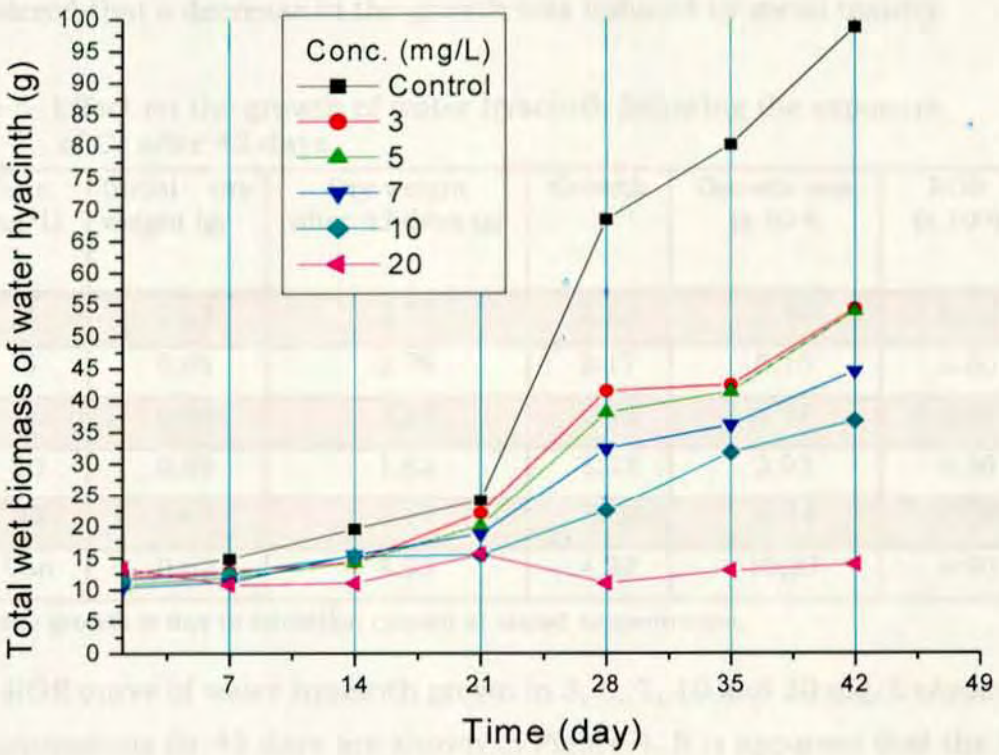


Figure 3 The wet biomass growth pattern for all portion of water hyacinth exposed to different Conc. of Cr containing water solutions

4.1.4. Relative growth rate

The effect of chromium on the growth rate of water hyacinth is given in Table 5. From the Table it was noted that the RGR decreased with increasing Cr concentration and at exposure of 20 mg/L of Cr the growth was negative. The negative value of RGR may be taken to indicate zero growth. It was considered that a decrease in the growth was induced by metal toxicity.

Table 5 Effect on the growth of water hyacinth following the exposure of Cr after 42 days

Conc. (mg/L)	Initial dry weight (g)	Dry weight after 42 days (g)	Growth	Growth rate ($\times 10^{-2}$)	RGR ($\times 10^{-2}$)
3	0.53	2.78	2.25	5.35	4.20
5	0.61	2.78	2.17	5.15	3.50
7	0.56	2.19	1.63	3.87	0.90
10	0.59	1.82	1.23	2.93	0.30
20	0.67	0.61	-0.06	-0.14	-0.20
Con	0.62	4.93	4.32	10.27	4.90

Negative growth is due to inhibition caused at stated concentration.

The RGR curve of water hyacinth grown in 3, 5, 7, 10 and 20 mg/L chromium concentrations for 42 days are shown in Figure 4. It is apparent that the RGR curve intercepts the concentration axis approximately at 15.32 mg/L. This indicates that the growth stopped after this concentration, due to the metal toxicity (Figure 4). It was reported for toxicity level of Cd to water hyacinth at 3.3 mg/L (Hasan *et al.*, 2006). This shows that Cr is less toxic than Cd.

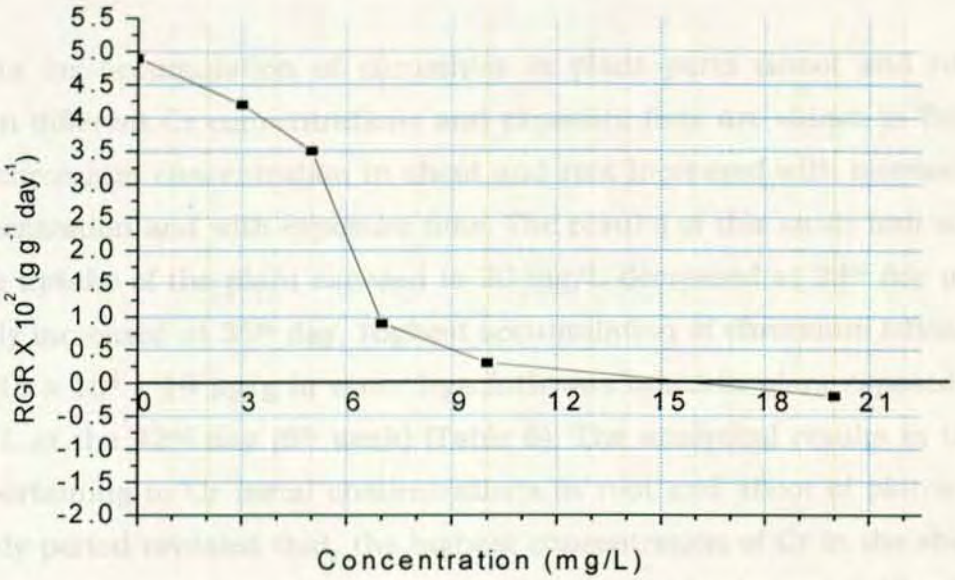


Figure 4 Relative growth rate of water hyacinth exposed for 42 days in different Conc. of Cr containing water solutions

In this study, despite the fact that, growth of the plant decreased with increasing Cr concentration but the plant was healthy up to 10 mg/L. The ability of the plants to stay healthy and therefore continue to grow is an important factor in the choice of plants for wastewater treatment. A plant will only take up the metal to any great extent if it is growing, and it will only grow if it can tolerate the concentration of metal in the media in which it is growing. Therefore, the plant in 20 mg/L was unable to tolerate the toxicity level of Cr and the growth retarded after 21st days of exposure (Figure 3).

4.2. Accumulation of chromium in plant tissue

The data for accumulation of chromium in plant parts (shoot and root) grown in different Cr concentrations and exposure time are shown in Table 6. The chromium concentration in shoot and root increased with increasing Cr concentration and with exposure time. The results of this study indicates that the uptake of the plant exposed to 20 mg/L decreased at 28th day and suddenly increased at 35th day. Highest accumulation of chromium (shoot + root) 4.18 x 10^3 ± 19 µg/g in water hyacinth was noted in plant exposed to 20 mg/L at the 42nd day (6th week) (Table 6). The analytical results in this study pertaining to Cr metal concentrations in root and shoot of plants in the study period revealed that, the highest concentration of Cr in the shoot accumulated 1.01 x 10^3 ± 82 µg/g at the 35th day and in the root 3.17 x 10^3 ± 78 µg/g at the 42nd day in the plant grown in 20 mg/L Cr solution. The results of the present study suggested that the plant accumulates more Cr in its root than shoot.

Similar result was reported with other metals, that water hyacinth grown in water containing Cd and Zn showed higher accumulation of these metals as a result of increasing metal concentration in growing medium and exposure period (Hasan *et al.*, 2006).

Table 6 Cr uptake of water hyacinth (μg/g) exposed to different Cr solutions

Conc. (mg/L)	Plant part	Days of treatment (day)						
		Initial	7	14	21	28	35	42
3	S	6 ± 1	175 ± 1	302 ± 32	366 ± 33	437 ± 27	414 ± 25	507 ± 71
	R	16 ± 1	751 ± 9	1,028 ± 176	1,390 ± 109	1,536 ± 72	1,721 ± 234	1,985 ± 135
	T	22 ± 1	926 ± 10	1,329 ± 144	1,756 ± 78	1,972 ± 48	2,134 ± 259	2,491 ± 206
5	S	5 ± 0.14	241 ± 5	385 ± 17	260 ± 26	493 ± 5	531 ± 22	661 ± 281
	R	14 ± 1	918 ± 18	846 ± 70	1,647 ± 81	1,814 ± 129	2,148 ± 39	2,292 ± 70
	T	19.49 ± 1	1,158 ± 15	1,232 ± 86	1,907 ± 55	2,307 ± 123	2,679 ± 29	2,953 ± 21
7	S	6 ± 0.42	456 ± 7	473 ± 12	613 ± 32	893 ± 37	905 ± 35	941 ± 21
	R	17 ± 1	1,144 ± 22	1,193 ± 34	1,507 ± 158	2,017 ± 19	2,173 ± 67	2,306 ± 91
	T	23 ± 1	1,600 ± 14	1,666 ± 25	2,120 ± 129	2,910 ± 41	3,078 ± 39	3,247 ± 78
10	S	6 ± 0.47	427 ± 21	471 ± 30	707 ± 33	841 ± 31	865 ± 45	948 ± 39
	R	14 ± 0.39	1,140 ± 70	1,302 ± 46	1,592 ± 120	2,391 ± 136	2,502 ± 95	2,571 ± 92
	T	20 ± 0.65	1,567 ± 85	1,773 ± 68	2,299 ± 148	3,232 ± 106	3,367 ± 56	3,519 ± 68
20	S	6 ± 0.12	142 ± 10	616 ± 38	945 ± 51	825 ± 52	1,014 ± 83	1,011 ± 98
	R	16 ± 0.30	1,152 ± 47	1,665 ± 67	2,663 ± 58	1,756 68± 102	3,000 ± 63	3,173 ± 78
	T	22 ±0.18	1,294 ± 37	2,281 ± 29	3,608 ± 6.35	2,582 ± 50	4,014 ± 20	4,184 ± 20
Con	S	5 ± 0.33	9 ± 0.35	10 ± 0.27	17 ± 0.33	12 ± 0.09	15 ± 1	16 ± 1
	R	15 ± 0.00	8 ± 0.43	15 ± 0.43	37 ± 0.59	35 ± 0.14	67 ± 0.29	55 ± 0.87
	T	20 ± 2	17 ± 3	25 ± 4	54 ± 4	47 ± 5	82 ± 1	70 ± 2

S = shoot, R = root, T = total, Con = Control

4.2.1. The mean accumulation of chromium in plant tissue

Table 7 presents the mean Cr accumulation in plant shoot, root and the whole portion after exposed to 3, 5, 7, 10 and 20 mg/L Cr solution for 42 days. The mean Cr accumulation increased with increasing Cr concentration in both the shoot and root part. Deducting Cr content of the plant in the control group, the plant in 20, 10, 7, 5 and 3 mg/L Cr containing water solution accumulates 2.52 x 10^3, 2.2 x 10^3, 2.04 x 10^3, 1.7 x 10^3 and 1.47 x 10^3 µg/g of Cr from first to fifth rank, respectively.

Table 7 Mean Cr accumulation in shoot (top), root and whole portion of water hyacinth exposed to different Cr Conc. for 42 days

Conc. (mg/L)	Shoot (x 10^2 µg/g)	± SE	Root (x 10^3 µg/g)	± SE	Total (x 10^3 µg/g)	± SE
3	3.15	65	1.20	252	1.52	317
5	3.68	83	1.38	310	1.75	386
7	6.12	128	1.48	300	2.09	428
10	6.09	125	1.52	351	2.25	475
20	6.51	159	1.92	425	2.57	576
Con	0.11	2	0.031	9	0.044	10

Con = Control

In the present study, accumulation of water hyacinth to Cr shows better than other aquatic plants. For example, Sarital *et al.* (2001) reported that *Alternathera sessilis* (rooted emergent), accumulated 1.21 x 10^3 µg/g; and *Najas indica* (submerged) 473 µg/g of Cr, when exposed to 8 mg/L Cr solution for 9 days.

Uptake of metals by plants is affected by several parameters; for example pH, temperature and chemical constituents. However, this study has taken only two important parameters in detail: exposure time and concentration of Cr to which the plant was exposed.

4.2.2. Cr uptake versus time of exposure

The pattern of Cr uptake of water hyacinth with respect to exposure time and exposure level (concentration) is shown in Figure 5. The uptake increased with time and took place in three stages (Figure 5). The first uptake took place, up to the 14th day (2nd week), except the plant grown in 20 mg/L followed by a second stage, with enhanced uptake up to the 28th day (4th week), and the third stage, up to 42nd day (7th week), as it is observed in Figure 5. The result of this study agrees with the result reported by Jayaweera and Kasturiarchchi (2004), they noted that water hyacinth would optimize phytoremediation during the period of optimum growth. In this study, the maximum growth of water hyacinth was observed on 35th and 42nd day of the exposure time. During this period the maximum accumulation of Cr for all treatment groups were noted and the plant in 20 mg/L accumulates best in this study period, $4.18 \times 10^3 \pm 19$ µg/g at the 42nd day. The plant in 20 mg/L Cr solution showed a sharp peak uptake of $3.60 \times 10^3 \pm 6$ µg/g on the 21st day; this is due to the peak biomass growth encountered during this period (Figure 3). Plants with higher biomass accumulate higher metals Soltan and Rashed (2003). However, on the 28th day falling down to $2.58 \times 10^3 \pm 50$ µg/g showing saturation of the cell with Cr, reflecting domination of hindering processes due to reduced biomass at the 28th day. The third enhanced uptake of the plant in 20 mg/L Cr solution

was the maximum peak encountered on the 35th and 42nd days of exposure were 4.01 x 10^3 ± 19 µg/g and 4.18 x 10^3 ± 20 µg/g respectively. This might be due to high exposure time and optimum biomass growth.

The calculated regression coefficient between Cr uptake and its exposure time were found to be 0.919, 0.945, 0.883, 0.901 and 0.841, for the plant grown in 3, 5, 7, 10 and 20 mg/L Cr containing water solutions, respectively. The accumulation versus time of exposure showed linearity between all exposure levels. The linearity trend could be established with highest confidence at 3 and 5 mg/L exposure levels (Figure 5). Thus, this observation showed that the extent of Cr uptake of the plant in 3 and 5 mg/L Cr containing solution was highly dependent on exposure time than exposure level (Conc.) and the uptake of the plant in 7, 10 and 20 mg/L Cr containing solution dependent on exposure time too and also other factors.

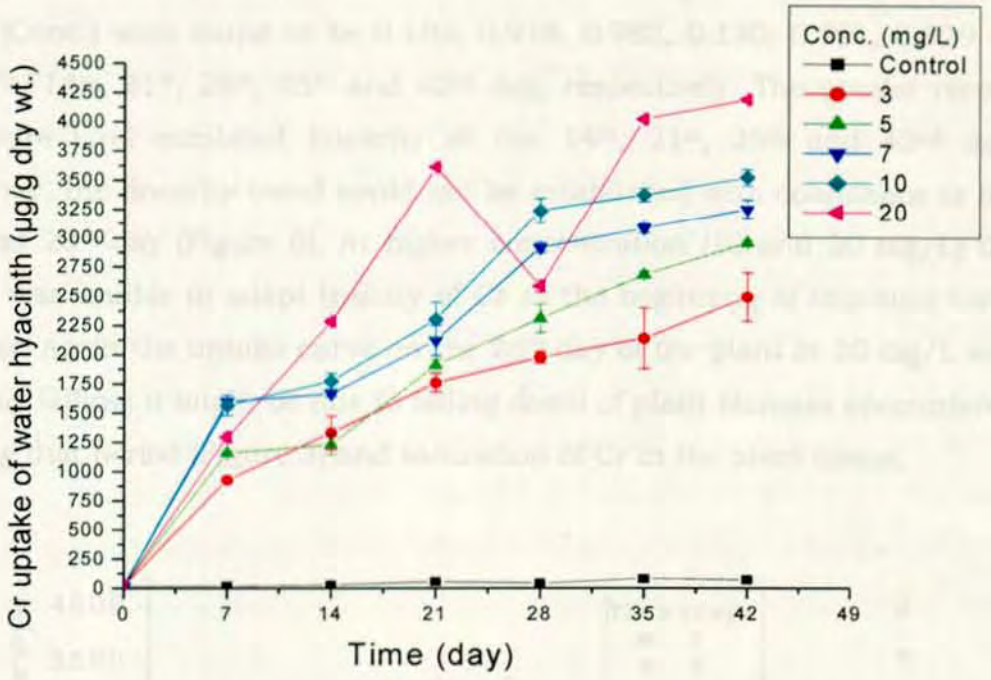


Figure 5 Concentration of Cr in all part of water hyacinth treated under 3, 5, 7, 10 and 20 mg/L Cr solution for 42 days

4.2.3. Cr uptake versus exposure level (concentration in medium)

The uptake patterns of water hyacinth grown in different concentration of Cr containing water solution are given in Figure 6. The uptake increased with increasing concentration of Cr in aqueous solution (3, 5, 7, 10 and 20 mg/L) as exhibited in the Figure 6.

The calculated regression coefficient between Cr uptake and its exposure level (Conc.) were found to be 0.102, 0.918, 0.985, 0.130, 0.891, 0.929 on the 7th, 14th, 21st, 28th, 35th and 42nd day, respectively. The uptake versus exposure level exhibited linearity at the 14th, 21st, 35th and 42nd day. However, the linearity trend could not be established with confidence at the 7th and 28th day (Figure 6). At higher concentration (10 and 20 mg/L) the plant was unable to adapt toxicity of Cr at the beginning of exposure time, 7th day. Again the uptake curve on the 28th day of the plant in 20 mg/L was sharply falling; it might be due to falling down of plant biomass encountered during that period (Figure 3) and saturation of Cr in the plant tissue.

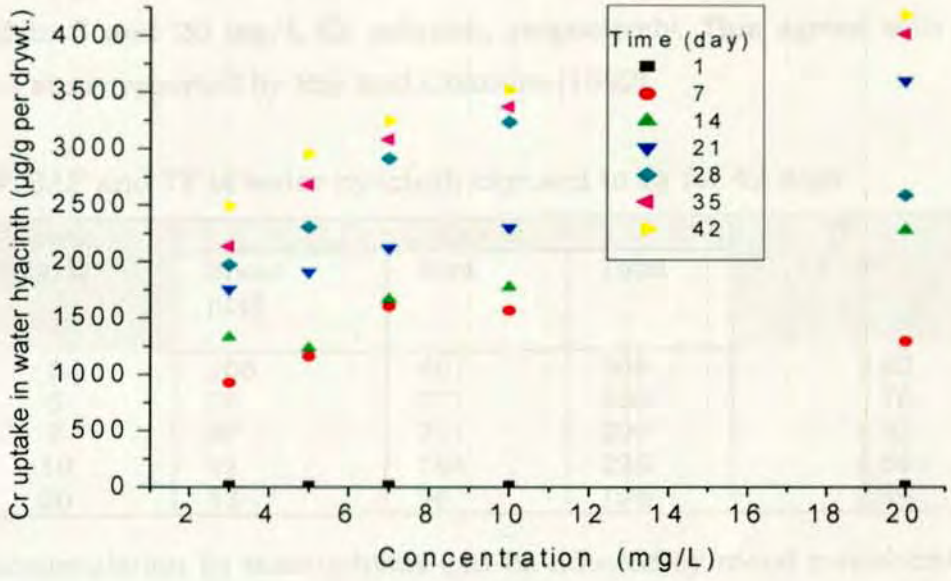


Figure 6 Cr accumulation of the plant exposed to 3, 5, 7, 10 and 20 mg/L Cr containing water solution

4.3. BAF of water hyacinth to chromium

Data on BAF and TF of water hyacinth resulting from exposure of 42 days to various concentration of Cr containing water solutions (3, 5, 7, 10 and 20 mg/L) are shown in Table 8. The BAF of water hyacinth decreased with increasing Cr concentration (Table 8). The BAF of the shoot ranged: from 33-1.05 x 10^2 and the root part 96-4.01 x 10^2. An assessment of the data given in Table 8 showed that the accumulation of Cr in root part was higher than the shoot part and this might be the direct contact of surface of the root tissue to the metal ions and strategy of the plant not to accumulate harmful ions to the shoot part. The maximum and the minimum BAF recorded for the whole part in this study were 5.06 x 10^2 and 1.28 x 10^2, from the plants exposed to 3 and 20 mg/L Cr solution, respectively. This agrees with the previous study reported by Ray and Chandra (1992).

Table 8 BAF and TF of water hyacinth exposed to Cr for 42 days

Cr conc. (mg/L)	BAF			TF (x 10^2)
	Shoot (top)	Root	Total	
3	105	401	506	3.82
5	74	277	350	3.76
7	87	211	299	2.42
10	61	164	225	2.50
20	33	96	128	2.94

Metal accumulation by macrophytes can be affected by metal concentration in water or soil medium as indicated by Zayed *et al.* (1998). The ambient metal Cr in water was the major factor influencing the metal uptake efficiency of the plant. When the metal concentration in water increases, the

amount of metal accumulation in plants increases, whereas the BCF values decreases (Sarital *et al.,* 2001).

Sarital *et al.* (2001) found that the BAF of water hyacinth were very high for Cd, Cu and Cr at a lower concentration, and decreased as the external concentration increases. Similarly, in the present study high BAF value was obtained when concentration in growth medium was low (3 mg/L).

It has been observed that water hyacinth showed far better accumulation capacity than other aquatic plants. For example, after 60 days study in 5, 10 and 20 mg/L Cr concentration, *Pharagmytes karka* (in hydroponic) has shown the BAF of 185, 182 and 115 respectively (Sarital *et al.* 2001).

4.4. TF of water hyacinth to chromium

An additional study was carried out to find out the pattern of metal mobility in root, top and whole plant.

In this study it was noted that TF of Cr decreased with increasing Cr concentration up to 7 mg/L and increased again in 10 and 20 mg/L. High TF implies a poorer translocation capability (Zayed *et al.,* 1998). Maximum TF, 382 (poor translocation capability) was observed from the plant exposed to 3 mg/L Cr containing water solution. In this study, water hyacinth absorbed Cr into the root and translocates 20 to 29% into the shoot. The maximum translocation capacity (29%) which has the lowest TF (242) in this study was obtained from the plant exposed to 7 mg/L Cr containing water solution. The translocation capability of 20, 21, 23 and 27% was recorded for the water

hyacinth grown in 3, 5, 20 and 10 mg/L Cr containing water, respectively. This result moderately agrees with the report made by Soltan and Rashed (2003) that water hyacinth accumulated heavy metals mostly to the roots and translocated only 6 to 25% to the shoots. Fine lateral roots of the water hyacinth reduce high toxic Cr(VI) to the less toxic Cr(III), and then translocate relatively non-toxic Cr(III) to leaf tissues (Lytle *et al.*, 1998). Lower accumulation of metals in shoot than root can be associated with protection of photosynthesis from toxic levels of trace elements (Baker, 1981; Landberg and Greger, 1996). This mechanism of partitioning from root to shoot is a common strategy of the plant that concentrates harmful ions in the roots in order to prevent toxicity to the leaves, site of photosynthesis and other metabolic activities (Sarital *et al.*, 2001).

Plants must have the ability to translocate Cr from the root to the shoot, or to compartmentalize it, in order to continue absorption of Cr from the external solution. Better translocation is advantageous to phytoextraction; because, it can reduce Cr concentration and thus reduce toxicity potential to the root (Baker, 1981). The differential localization of metals within the plant tissues may also be important in determining how well the metals may be bound and released from the plants (Suren, 1989).

Translocation of trace elements from roots to shoots could be a limiting factor for the bioaccumulation of elements in the shoots.

4.5. Removal efficiency of water hyacinth to Cr containing solutions

Table 9 presents the Cr removing potential of water hyacinth treated in 3, 5, 7, 10 and 20 mg/L Cr containing water solution for 42 days. From the Table, it is apparent that removing potential of water hyacinth decreased with increasing Cr concentration.

At lower Cr containing water solution (3 mg/L) the removal efficiency of the water hyacinth reached about 91%. The removal efficiency of the plant exposed to 5, 7 and 10 mg/L Cr containing water solutions was appreciable (about 80%). Similar result have been reported, with removal efficiency of 70% from the water hyacinth exposed to 7 mg/L Cr concentration, for a period of 17 days, (Cyle *et al.*, 2006).

Table 9 Chromium removing potential of water hyacinth in percent (%) from different concentrations of Cr containing water solution during 42 days

Cr Conc. in growth medium (mg/L)	Mean Cr Conc. in growth medium after 42 days (mg/L)	Removing potential in percent (%)
3	0.26	91
5	0.72	85
7	1.35	80
10	1.94	80
20	5.69	69

The removal efficiency of the plant in 20 mg/L Cr containing water solution decreased to 69%. As the concentration increased, the plant was not able to take up as much percent as the low concentration level of Cr, but the amount that the plant was able to take up was still significant. This might be the, Cr concentration in water exceeded the tolerance limit and the plant tissue may be injured.

Hence, the result in the present study shows that Cr was efficiently removed (80%) from wastewater containing Cr concentration up to 10 mg/L. This is an indication that high removal efficiency could be achieved at a diluted Cr solution. Maine and Durate (2001) reported that water hyacinth effectively removes appreciable quantity of heavy metals (Cd, Co, Cr, Cu, Mn, Ni, Pb and Zn) from wastewater, especially at low concentrations.

4.6. Limitation of water hyacinth for application in wetland

Loss of water (transpiration impact) by plant is affected by several parameters; like soil water potential, humidity and aerodynamic conditions. However, this study has taken two parameters in detail: plant factor such as, stomatal conductance and LAI and seasonal factor (dry and rainy seasons). Factors such as soil water potential, humidity and wind speed can be regarded as less important in environment with permanently waterlogged soil such as wetlands, as pointed out by Lee (1967) rather transpiration seems to be controlled by stomata rather than by aerodynamic conditions.

Hourly mean cycles (diurnal time) of transpiration of water hyacinth in dry and wet seasons are shown in Table 10. Leaf transpiration rate of this plant in dry and wet season increased regularly up to 13 (standard time) and decreased again after 13 (standard time) in the same pattern. The minimum and maximum mean transpiration of water hyacinth (± SE) measured in sunny day and rainy day was 2.1 ± 0.16, and 9.26 ± 0.26 ($mM/m^2/s$);and 1.99 ± 0.19 and 5.42 ± 0.19 ($mM/m^2/s$), respectively. The maximum transpiration of water hyacinth was measured in dry season at 13, standard time (Table 10).

Table 10 Hourly mean transpiration of water hyacinth

Standard time	Dry season		Wet season	
	Mean E ($mM/m^2/s$)	± SE	Mean (E) ($mM/m^2/s$)	± SE
7.00	2.10	0.16	1.99	0.19
8.00	3.00	0.27	2.34	0.12
9.00	4.23	0.34	3.46	0.09
10.00	4.82	0.37	3.51	0.19
11.00	5.69	0.05	4.43	0.14
12.00	7.49	0.34	4.60	0.29
13.00	9.26	0.26	5.42	0.19
14.00	6.00	0.37	3.02	0.32
15.00	5.04	0.05	2.33	0.21
16.00	4.17	0.14	2.65	0.13
17.00	3.71	0.11	2.61	0.11
18.00	2.14	0.07	2.11	0.15

± SE = Standard error

The mean hourly leaf transpiration curve of water hyacinth at dry and wet seasons is shown in Figure 7. The diurnal transpiration curve of water hyacinth tends to form downward opening. The curve peaked at 13.00 standard time with transpiration value of 9.26 ± 0.26 and 5.42 ± 0.19 mM

m^{-2} s^{-1}, in both dry and wet seasons, respectively (Figure 7) but the daily leaf transpiration courses of most species showed one peak at different points during the day depending on the period of the season (John *et al.*, 2005). This curve suggests that transpiration processes for this species is controlled largely by the temperature gradient induced by the solar radiation received by the leaves. In most cases, solar radiation explained more than 75% of the variance of transpiration, while relative humidity and temperature only explained residues (Salvador *et al.*, 2001). In this study, the rainy season transpiration was significantly reduced than sunny season, owing basically to the reduction of light and lower solar incidence.

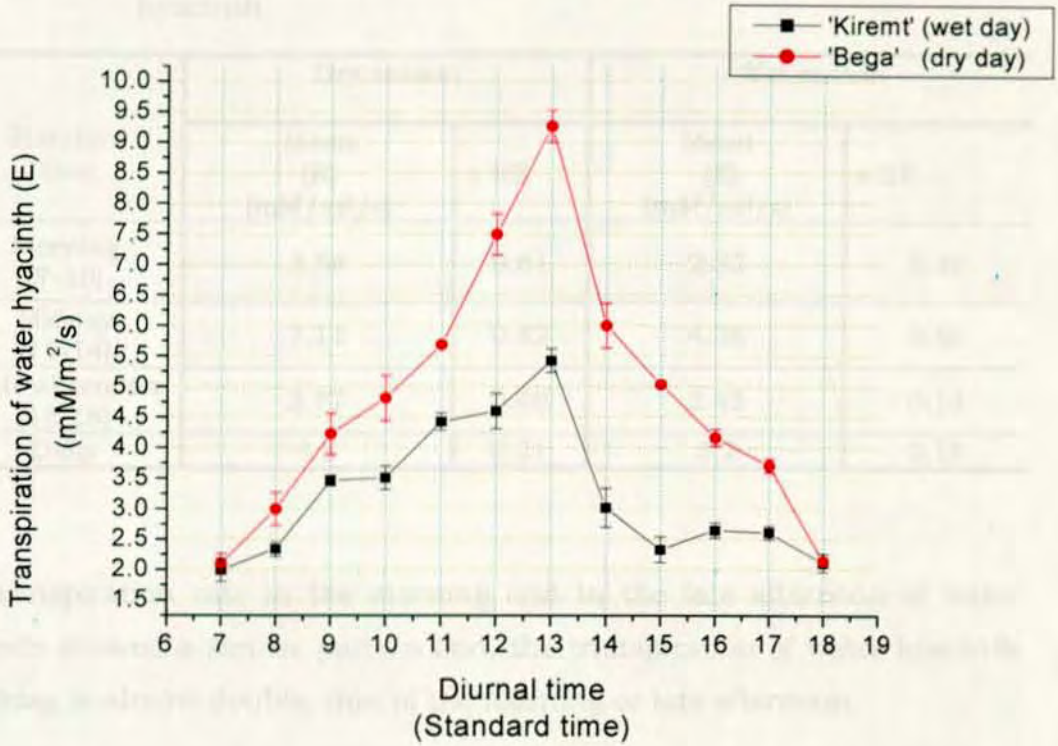


Figure 7 Seasonal variation (wet and dry season) of diurnal transpiration rates (mM m^{-2} s^{-1}) of water hyacinth measured in greenhouse at AAU

The rates of morning, midday and late afternoon diurnal transpiration of water hyacinth in dry and wet seasons are shown in Table 11. The maximum transpiration rates of water hyacinth in dry and wet seasons were measured at midday.

Table 11 Morning, midday and late afternoon transpiration (E) of water hyacinth

Standard time	Dry season		Wet season	
	Mean (E) ($mM/m^2/s$)	± SE	Mean (E) ($mM/m^2/s$)	± SE
Morning (7-10)	3.54	0.61	2.83	0.39
Midday (11-14)	7.11	0.82	4.38	0.50
Late afternoon (15-18)	3.77	0.60	2.43	0.13
Daily	4.8	0.21	3.2	0.18

The transpiration rate in the morning and in the late afternoon of water hyacinth showed a similar pattern and, the transpiration of water hyacinth at midday is almost double, that of the morning or late afternoon.

4.6.1. LAI in Aba Samuel wetland

Results of LAI and plant density of water hyacinth at vegetation stand in Aba Samuel wetland is given in Table 12. In this study, the LAI and density of water hyacinth in Aba Samuel wetland was 4.95 and 35.3 ± 1.39 kg/m^2, respectively.

Table 12 LAI and density of water hyacinth on vegetation stand at Aba Samuel wetland

Vegetation	Leaf number per area	Average LA (cm^2)	LAI ($cm^2 cm^{-2}$)	Wet biomass (kg m^{-2})
Water hyacinth	161 ± 7.31	76.95 ± 14.3	4.95	35.3 ± 1.39

Reddy and Sutton (1984), reported that under normal condition, loosely packed water hyacinth can cover the water surface at relatively low plant density (10 kg/m^2 wet mass) and it can reach maximum density of 50 Kg/m^2. This showed the growth of water hyacinth in Aba Samuel wetland has arrived nearly in a severe condition as shown in Plate 5.

Plate 5 LAI of water hyacinth in Aba Samuel wetland
(Source: Daniel, this study)

4.6.2. Transpiration potential in Aba Samuel wetland

Once curves of expected transpiration are developed for a species, this curve (Figure 5) may be adapted to other areas where these plants grow by using the same leaf conductance. However, it should be noted that this technique is generally applicable only for areas where the growing seasons and climatic conditions tend to be similar (David *et al.*, 1986).

Therefore, transpiration potential of water hyacinth of the projected area (LAI x E) in dry and wet seasons, per time (second, minute, hour and day), are given in Table 13. LAI for each species was the basis for estimating land-surface transpiration because transpiration was measured and modeled in terms of unit leaf area (David *et al.*, 1986).

Table 13 Transpiration rate (potential) of water hyacinth of the projected area

Atmospheric condition	Transpiration rate of the projected area per time (E x LAI) (mM/m^2/time)			
	Second	Minute ($\times 10^3$)	Hour ($\times 10^5$)	Day ($\times 10^6$)
Sunny season	48	2.87	1.71	2.06
Rainy season	32	1.90	1.14	1.37

Therefore, from an area of square meter of water surface covered by water hyacinth in Aba Samuel wetland, 18.57 liter (18.57 mm) and 12.33 liter (12.33 mm) of water per day will be lost (transpired) in dry and wet seasons, respectively (Table 13).

4.6.2.1. The estimated total volume of water transpired from Aba Samuel wetland

According to Tassew (2005) from the total area of the wetland (13.2 sq km), about 48% (6.35 sq km) was covered by water hyacinth. Mats of water hyacinth may double their size in as little as 6-18 days (Mitchell, 1976). Accordingly, it can be estimated that currently the mat of water hyacinth in Aba Samuel wetland proliferated more than double of wetland coverage reported in 2005 (Tassew, 2005). Therefore, currently more than 90% (11.88 sq km) of the Aba Samuel wetland have been covered by water hyacinth (Plate 6).

From the wetland surface area, covered by water hyacinth total of 220, 624 m^3 and 146, 480 m^3 of water per day will be lost (transpired) during dry and wet seasons, respectively.

It was reported by Mitchell (1976) that an acre (4, 000 sq m) of water hyacinth transpires 87 m^3 of water per day. In this study, a single acre of water hyacinth in Aba Samuel wetland transpires 61.8 m^3 of water per day. The discrepancy between the results, here and that reported by Mitchell (1976) may be due to different vegetation density and climatic regions.

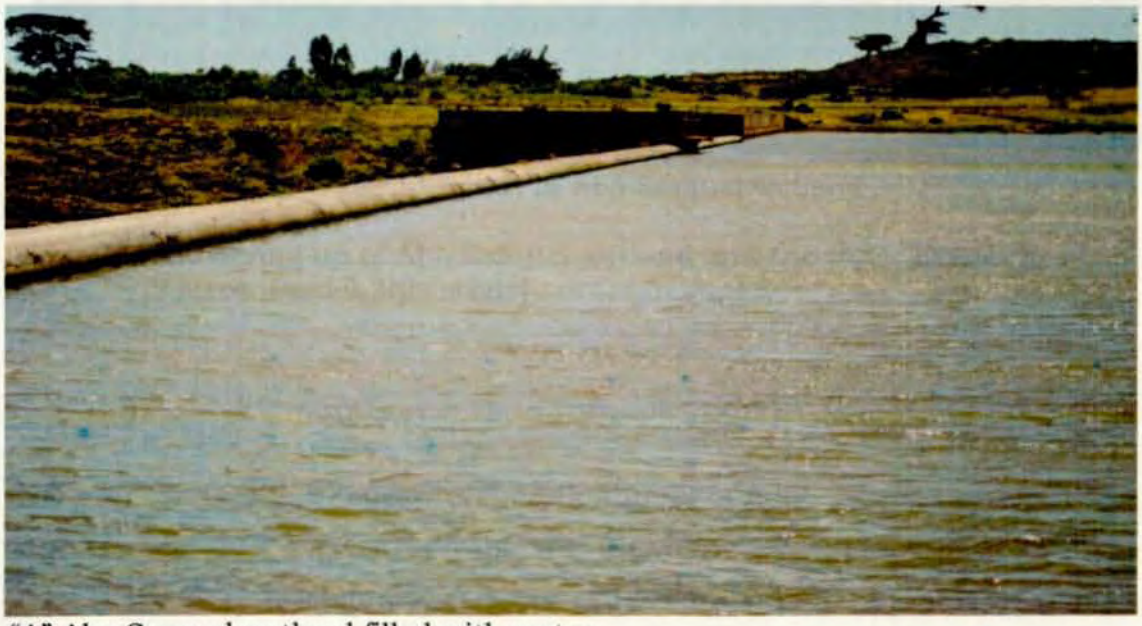
It was reported by Reddy and Sutton (1984) that an acre of water lettuce transpired 33.3; salvinia transpired 32.05; and azolla transpired 28.65 m^3 of water per day. In comparison, water hyacinth, in this study transpires (loses) fresh water 1.85 to 2.15 times more than salvinia and azolla. Hence, as it was forecasted by Zeleke (1992) that, water hyacinth will make Aba

Samuel wetland dry in the long run, and therefore, the problem has already started (Plate 6) and (Plate 7B).

"A" Dense mat of water hyacinth in Aba Samuel wetland

"B" Surface area of Aba Samuel wetland covered by water hyacinth

Plate 6 Aba Samuel wetland covered by water hyacinth
(Source: Daniel, this study)



"A" Aba Samuel wetland filled with water

"B" Ruminants of Water hyacinth in Aba Samuel wetland

Plate 7 The drying up of Aba Samuel wetland and the water hyacinth (Source: Daniel, this study)

4.6.3. Evaporation versus transpiration in Aba Samuel wetland

The year 2000 to 2007 mean evaporation (mm) of Addis Ababa city and the surrounding area is given in Table 14. The daily mean evaporation of dry season (January to May and October to December) is 5.32 mm and of the wet season (June to September) is 2.54 mm.

Table 14 Mean evaporation in (mm) around Addis Ababa city

Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
127.72	152	144.72	151.98	170.2	99.62	62.74	61.8	81.14	172.08	178.5	180.6

(Bole station) (NMSA, 2008)

Therefore, transpiration of water hyacinth exceeds evaporation 3.49 times in dry season and 4.85 times in wet season, respectively.

Knowledge of the quantity of the amount of water loss through transpiration by aquatic plants must be taken into account when planning wastewater treatment in a natural wetland. As indicated in this study significant amount of water has been lost from the wetland each second because of the presence of water hyacinth in high density in the Aba Samuel wetland. It has a serious implication where water is already scarce for increasing human demands, feeding and breeding for fish, birds and other biodiversities.

5. CONCLUSIONS

Water hyacinth (*Eichhornia crassipes* (Mart.) Solms) due to its fast growth (eighth fastest growing plant on earth) and its ability to grow in heavily polluted water body can be successfully used for the removal of Cr from aqueous solution. In this study, the plant showed high accumulation of Cr in its tissue and accumulation increased with the concentration of Cr in external solution.

The growth of the plant was inhibited due to Cr toxicity above 15.34 mg/L Cr concentration in water solution. The ability of plants to stay healthy and therefore continue to grow is an important factor in the choice of plants for wastewater treatment. A plant will only take up the metal to any great extent if it is growing, and it will only grow if it can tolerate the concentration of metal in the media in which it is growing. Therefore, the application of water hyacinth for Cr removal will be sustainable if the concentration of Cr in wastewater not exceeding approximately 15.34 mg/L.

In this study water hyacinth accumulated 2.52×10^3, 2.20×10^3, 2.04×10^3, 1.70×10^3 and 1.47×10^3 µg/g from 20, 10, 7, 5 and 3 mg/L Cr containing water solutions in decreasing order and with BAF of 128, 225 299, 350 and 506 with increasing value, respectively. As the concentration increased, the plant was not to be able to bioaccumulate as much percent as the plant in low concentration Cr solution, but the amount was still significant.

More Cr was accumulated in the root than the shoot; the translocation factor of water hyacinth to Cr was 382, 376, 242, 250 and 294 for the plants in 3, 5, 7, 10 and 20 mg/L Cr solution, respectively. A larger number of translocation ability implies a poorer translocation capability. The poor translocation of Cr from root to shoot is a major hurdle in harvesting the plant after accumulation of Cr in plant. Therefore, the translocation capability of water hyacinth to Cr was in the plant exposed to 7, 10, 20, 5 and 3 mg/L Cr containing water solution in the decreasing order of translocation capability.

The removal efficiency of water hyacinth in 3 mg/L reached up to 91% and those in 5, 7 and 10 mg/L Cr containing water solutions were 85, 80 and 80%, respectively.

Considering all the above criteria water hyacinth can be a promising candidate for the removal of Cr from wastewater efficiently and in a sustainable way at 7 to 10 mg/L Cr containing water.

Water hyacinth is a plant with such an advantage however, it has one major limitation: it is one of the most invasive aquatic weed and can destroy precious aquatic ecosystems, by transpiring (losing) a significant quantity of fresh water which can lead to serious problems on the lively hood of the community and aquatic ecosystem. Therefore, natural wetland is not an appropriate place for phytoremediation of Cr using water hyacinth. The plant may be applied for the removal of Cr in a constructed wetland or ponds.

6. RECOMMENDATIONS

After approximately six weeks a complete harvesting is recommended, to optimize the Cr removing potential. Therefore, the application of water hyacinth for Cr removal shall be coupled with other applications like biogas and alcohol production from the harvested biomass however, further study is recommended because the harvested biomass contains high amount of Cr. Furthermore, the basic problem of disposal of the plant after Cr treatment is unsolved. Therefore, it is recommended to be studied for other utilization options that can ensure mass consumption of the water hyacinth biomass.

The role of microorganisms associated with the plant's potential to remove Cr from waste water is recommended to be studied for better yield. Further study on the adsorption capacity of the dried biomass of water hyacinth to Cr is also recommended to compare with the living potential of the plant.

7. REFERENCES

(AAEPA) Addis Ababa Environment Protection Agency, A. A. (2004). Addis Ababa situation assessement standard. Addis Ababa: (unpublished).

Abbasi, S., & Ramasamy, E. (1999). Biotechnological methods of pollution control. Hyderabad. Orient Long man: Universities press India.

Alves, M., Gonzalez Beca, C., Guedes, D., Carvalho, R., & Castanheria, (1993). Chromium removal in tannery wastewater . *J Water Res* (27), 1333-38.

Anushee, M. (2005). Environmental challenge vis a vis opportunity: the case of water hyacinth. *J Environmental Qual* (32), 122-38.

Aweke, Gebru. (1993). The water hyacinth (*Eichhornia crassipes*) in Ethiopia. *J of Academic Sci* (39), 399-404.

Baker, A. J. (1981). Accumulators and excluders strategies in response of plants to heavy metals. *J of Plant Nutr* (3), 643-654.

Barrett, S. (1980). Sexual reproduction in *Eichhornia crassipes* (water hyacinth): Seed production in natural populations. *J Applied Ecol* (17), 113-24.

Benicassa, M. (1988). Analysis of plant growth . *J. Experimentaal Bot*

Beraatowicz, S., Leszczynski, S., & Tyczynska, S. (1976). The influence of transpiration by emergent plants on the water balance in lakes. *J Aquatic Bot* (2), 275-288.

Bolenz, S., Omran, H., & Gierschner, K. (1990). Treatments of water hyacinth tissue to obtain useful products. *J Biological Wast* (33), 263-74.

Brown, R., & Blaser, R. (1968). Leaf area index and pasture growth. *J Herb Abst.* (1), 1-9.

Carvalho dos santos, M., & Lenzi, E. (2000). The use of aquatic macrophytes (Eicchhornia crassipes) as a biological filter in the treatment of lead contaminated effluents. *J Environmental Tech* (21), 615-22.

Cervantes, C., Campos-Garcia, J., Devars, S., Gutierrez-Corona, F., Loza-Tavera, H., Torres-Guzman, J. (2001). Interactions Of Chromium With Microorganisms and Plants. *J Microbiology* (25), 335-347.

Chandra, P., Sinha, S., & Rai, U. (1997). Bioremediation of Cr from water and soil by vascular aquatic plants. *J* of *American Chem Soc.* (64), 274-82.

Cock, M. R., H. Herren, M. H., M. Julien, P., & Neuenschwander, J. (2000). Harvesters get that sinking feeling. *J Environmental Tech* (21), 213-17.

Collatz, G. J., Ball, J. T., Grivet, C., & Berry, J. A. (1991). Physiological and environmental regulation of stomata conductance, photosynthesis and transpiration: a model that includes a laminar boundary layer. *J Agriculture for. Met.* (54), 107-36.

Cyle, K., Hamid, B., Susan, V., & B.S, B. (2006). Removal of Copper, Chromium and Arsenic by water hyacinth . *36th annual conferrrrence* (pp. 13-18). Missippi: CCA.

David H. wilson, R. J. (1986). Evapotranspiration measurements of native vegetation, owns valley. Water Resource investigation. California, USA: US geological survey .

Debusk, T., Dierbg, F., & Reddy, K. (2001). The use of macrophyte-basedsystem for phosphorus removal: an overview 25 yerrs of research and operational results in Florida. *J Water Sci Tech.44*, 39-46.

Decker, J. P., & Wetzel, B. F. (1957). A method for measuring transpiration of intact plants under controlled light, humidity, and temperature. *J Forest Sci* (3), 350-54.

Delgado, M., Bigeriego, M., & Guardiola, E. (1993). Uptake of Zn,Cr and Cd by water hyacinths. *J Water Res* (27), 269-72.

Dixit, U., Pandey, V., & Shyam, R. (2001). Chromium ions inactivate electron transsport and enhaance superoxides generation. *J. plant cell* (25), 687-693.

Elton, C. S. (2000). The Ecology of invasions by Animals and Plants. Chicago: University of Chicago Press.

Epstein, P. (1998). Weeds brings disease to the East Africa water ways . *J weed Sci (3)*, 351-377.

Felix, K., Noam, H., & Haya, Z. (2007). Evapotranspiration through ponds with floating aquatic plants. *J Environmental Qual (19), 210-2.*

Gamborg, D., & Wetter, L. (1975). plant tissue culture method. Sasskaton Nalt Res.counccil, Canaada.

Gopa, B. (1987). Water hyacinth, aquatic plant studies series. *J Environmental Qual* (11), 264-72.

Grocnevcld, D., Warren, D., Hubbard, P., & Yamashita, I. (1986). *Transpiration proccsses of shallow groundwater shrubs and gasses in the Owens Valley, California.* California State: university press.

Guo, Y. & Zhang, Z. (2002). Function of water hyacinth root eco-system in purification of silver contained wastewater. *J of Wuxi Univ* (5), 506-510.

Hafez, A., El-Manharawy, M., & Khedr, M. (2002). Membrane Removal Of Unreacted Chromium From Spent Tanning Effluen. *J of Desalin* (144), 237-42.

Hagmeyer, J. (1999). Ecophsiology of plant growth under heavy metal stress. J *Springer- Ver. Berlin* , 157-81.

Haller, W. T., & Sutton, S. (1973). Effects of pH and high phosphorus concentrations on growth of waterhyacinth. *J. of Hyacinth Contr.* (11), 59-61.

Hansen, K. L., E. G., R., & R. L., T. (1971). Trophic relationships in the water hyacinth community. *J of the Florida Academy of Sci.* (34), 107-13.

Hashintoni, M., Okumar, M., & Fuginga, K. (1987). Standard solutions preparation method. *J Analytical Chem* (5) , 86-98.

Hassen, S., Talat, M., & Rai, J. (2006). Sorption of cadmium and zinc. *J. Bioresource Tech.* (98), 918-928.

Hassen, S., Talat, M., & Rai, J. (2006). Sorption of cadmium and zinc. *J. Bioresource Tech.* (98), 918-928.

Hauschild, M. Z. (1993). Putrescine (1,4-diaminobutane) as an indicator of pollution-induced stress in higher plants: barley and rape stressed with Cr(III) or Cr(VI). *J Environmental Saftey* (26), 228-47.

Hitchcock, A. E., P. W., Z., H. Kirkpatrick, J., & D T. T., E. (1950.). Growth and reproduction of water hyacinth and alligator weed and their control by means of 2,4-D. *J Environmental Qual* (16), 91-130.

Hitchell, D. (1967). The growth and management of Eichhornia Crassipes and salvinia spp in their native and alive situation. *J aquatic weed*, 399.

Holms, L. P. (1977). *The World's Worst Weed: Distribution and Biology.* (18th Edition. ed.). Hawaii, Honolulu: University Press.

Holms, L., Weldon, L., & Blackburn, R. (1969). *Aquatic weed scie.* (166), 699-709.

Howard, G. (2003). Alien Invasive Species in Africa's Wetlands. *IAS Conference.* Nairobi, Kenya, : IUCN Eastern African Regional Program.

Hunter, J., & Verggnano, O. (1953). Trace element toxicity in oat plants. In the Annals of applied biology. Eds. R W Marsh and I Thomas. Cambridge: University press.

Jackson, L., Rasmusen JB., & Kalff, J. (1994). A mass balance analysis of trance metals in two weedbeeeeds. *J Water Air Soil pollut (22)*, 107-19.

Jayaweera, M., & Kasturiarachchi, J. (2004). Removal of nitogen and phosporus from indusrial wastewaters by water hyacinth. *J Water Sci Technol* (50), 217-25.

John P.A., L., Bart, W., & Yulduz, J. (2005). Evaluation of young and adult tree plantation for biodranage management in the lower Amudarya river, Uzbekistan. *ICI 21 st European Regional Conference*. Frankfurt, Germany.

Julien, M., & Orapa, W. S. (1999). Proceedings of the International Symposium on Biological Control of Weeds. Bozeman, MT, USA: Montana State University press.

Kadlec, R. H. (1989). Hydrologie factors in wetland water treatment. Michigan, USA: Lewis Publishers.

Knipling, R., Rible, R., Kadleck, R., & Reed, S. (1970). Wetlands for wastewater treatments: performance data base in constructed wetlands for water quality improvement. (G. Moshiri, Ed.) Boca Raton: Lewis Publishers CRC press.

Kocaoba, S., & Akcin, G. (2002). Removal And Recovery Of Chromium And Chromium Speciation With MINTEQA2. *J Talanta* (57), 23-30.

Kumar, S. (2005). Studies on efficiencies of bio-gas production in anaerobic digesters using water hyacinth and night-soil alone as well as in combination. *Asian J Chem* (17), 934-8.

Landberg, T. & Greger, T. (1996). Difference in uptake an tolerance to heavy metal in Salix from unpolluted and polluted areas. *J Applied Geochem* (11), 175-180.

Lee, R. (1967). The hydrologie importance of transpiration control by stomata. *J Water Resour Res* (3), 737-752.

Liao, S., & Chang, W. (2004). Heavy metal phytoremediation by water hyacinth at constructed wetlands in Taiwan. *J Aquat Plant Manag* (42), 60-8.

Lockley, G. & Turner, T. (1961). The Control of Water Hyacinth. Department of Agriculture . *Ministry of Agriculture Bulletin, 1–6* , pp. 1-6.

Lytle, C., Lytle, F., Yang, N., Qian, J.-H., & Hansen, D. (1998). Reduction of Cr(VI) to Cr(III) by wetland plants: potential for in situ heavy metal detoxification. *J Environmental Sci Tec* (32), 3087-93.

Mailu, A. (2001). Preliminary assessment of the social, economic and environmental impacts of water hyacinth in the Lake Victoria basin and the status press. *ACIAR Procceding coference*. Nairobe: University press.

Maine, M., & Durate, M. (2001). Cadmium uptake by floating macrophytes. *J Water Res* (35), 2629-2634.

Marguerite S., K., & Rawlik, P. S. (1993). Transpiration and Stomatal conductance of two wetland macrophytes in the subtropical everglades. *American J of botany , 80* (10), 1146-1154.

Martin, T., Creed, J., & Brockhoff, C. (1994). Sample preparation procedure for spectrochemical determination of total recoverable elements-Method 200.2, revision 2.8. *J analytical chemistry.*

Microcal Software, I. (1999). Origin data analysis and technical gaphics software for windows version 6.0. *Microcaloftware, inc* . Northampton, USA.

Mitsch, W., & Gosselink, J. (1993). *Wetlands* (2nd edition ed.). Newyork, USA: Van Nosstrand Reinhold.

Mukherjee, S., & Modale, G. (1995). Removal of lead by water hyacinth. *Indian J Chem Technol* (2), 59-62.

Mukherji, S. & Roy, B. (1978). Characterization of chromium toxicity in different plant materials. *Indian J Experimental. Biol.* (16(9)), 1017-1019.

Nevena, N. (2006). Potential use of water hyacinth (*Eichornia crassipes*) for wastewater treatment. *NWWC* (pp. 12-18). Belgrade: University press.

Nigam, J. (2002). Bioconversion of water hyacinth (*Eichhornia Crassipes*) hemiccccellulose acid hydrollysate to motor fuel ethanol by xylose-fermanting yeast . *J Biotechnol* (97), 107-16.

NMSA. (2008). National Meterological Source Agency. Addis Ababa, Ethiopia.

Noble, P. (1991). Phsicochemical and envirnonmental plant physiology. Sand Diego, CA: Academic press Inc.

Nriagu, J., & Nieboer, O. (1988). Production and use of chromium: chromium in natural and human environment. New york: John wiley press.

Panda, S., & Choudhury, S. (2004). Changes in nitrate reductase (NR) activity and oxidative stress in moss Polytrichum commune subjected to chromium, copper and zinc toxicity. *Braz. J. Plant Physiol* (46), 289-94.

Parija, P. (1934). A note on the reappearance of water hyacinth seedlings in cleared tanks. *Indian J Agric Sci* (4), 1049.

Penfoud, WT. & Earle, P.(1948). Water hyacinth control. 448.

Peterson, P. (1975). Heavy metals in the environment. *International coferance. 2*, pp. 20-39. Toronto: University of Toronto.

Radford, P. (1967). Growth analysis formula their use and abuse. *J. crop Sci* (7), 171-75.

Rai, U., & Chandra, P. (1992). Accumulation of copper, lead, manganesse and iron by field population of Hydrodictyon reticulatum lagerheim. *Sci total Environ* (116), 203-11.

Reddy, K., & Sutton, D. (1984). water hyacinths for water quality improvement and biomass production. *J Environmental Qual* (13), 1-8.

Reddy, K., Campbel, l. K., Graetz, D., & Portier, K. (1982). Use of biological filters for treating agricultural drainage effluents. *J Environmental Qual* (11), 591-5.

Rezene, Fessehaye. (2005). Water hyacinth (*Eichhornia crassipes*): A review of its weed status in Ethiopia. *Arem* (6), 24-30.

Roldan, G. (2002). Treating industrial wastes in Colombia using water hyacinth. *J Waterlines* (11), 6-8.

Roy, S., & Hanninen, O. (1994). Pentachlorophenol: uptake/elimination kinetics and metabolism in an aquatic plant, *Eichhornia crassipes*. *J Environ Toxicol Chem* (13), 763-73.

Salisburg, F., & Wadsworth, C. (1992). *Plant physiology* (4th ed.). Belmont,CA: Wadsworth publishing Co.

Salt, D., Blaylock, M., Kumar, P., Dushenkov, V., Ensley, B., Chet, I., et al. (1995). Phytoremediation: anew strategy for the removal of toxic metals from the environment using plants. *J Biotechnology* (13), 468-74.

Salunkhe, P. B., Dhakephalkar, P. K., & Paknikar, K. M. (1998). Bioremediation of hexavalent Cr in soil microcosms. *J Biotech* (20), 749-751.

Salvador, S., Miguel, A., & Manuel, B. (2001). A simple method for estimating water loss by transpiration in wetland. *J Hydrological Science* (64), 537.

Sarital, S., Rohit, S., & Shraddha, S. (2001). Comparative study on accumulation of Cr from metal solution and tannery effluent. *J. environmental Mon. and Ass.* (17-31).

SAS, Inc. (2001). SAS/Stat Software, version 8.1 . *SAS 8.1* . USA: SAS institute, Inc.

Senait Regasa, A. T. (2007). Socio economic impact and control baseline condition of water hyacinth. Addis Ababa, Ethiopia : EARO.

Senayit, Regasa, Agajie, T., Taye, Tessema., & Adefires, Woldearegay. (2004). Invasive Alien Plant Control and Prevention in Ethiopia. under the PDFB phase of the UNEP GEF. *Pilot Surveys and Control Baseline Conditions EARO, Ethiopia and CABI* . Addis Ababa, Ethiopia: EARO.

Shewry, P. R., & Peterson, P. J. (1974). The uptake and transport of chromium by barley seedlings (Hordeum vulgare L.). *J Experimental Bot.* (25), 785-97.

Singhal, V., & Rai, J. (2003). Biogas production from water hyacinth and channel grass used for phytoremediation of industrial effluents. *J Bioresour Tech* (86), 221-5.

Sinha AK, Sinha, R. (2000). Sewage management by aquatic weeds (water hyacinth and duck weed). *J Environmental Edu* (19), 215-26.

Snydder, R., & Boyd, C. (1987). Evapotranspiration By *Eicchhorniaa Crassipes* and *Typha latifolia. J Aquatic Bot* (27), 217-27.

Soltan, M., & Rashed, M. (2003). Labratory study on the survival of water hyacinth under several conditions of heavy metal concentrations. *J Environmental Res* (7), 321-334.

Sooknah RD, and Wilkie, W. A. (2004). Nutrient removal by floating aquatic macrophytes cultured in anaerobically digested flushed dairy manure wastewater. *J Ecol* (22), 27-42.

Stocker RK, Haller, W. (1999). Residual effects of herbicide-treated *Eichhornia* as soil amendem. *J Hycaldro Biolo* (415), 329-33.

Stroud, A. (1991). Water hyacinth (*Eichhornia crassipes* [Mart.] Solms) in Ethiopia. In: Rezene Fessehaie (ed. *Proceedings of the 9th Annual Conference of the Ethiopian Weed Science Committee. EWSC, Addis* (pp. 7-16.). Addis Ababa, Ethiopi: EARO.

Suren, A. (1989). Histological changes in macrophyte tissue during decomposition. *J Aquat Bot* (22), 27-40.

Tassew, Tessema. (2005). Application of GIS/ RS for study of water hyacinth and water pollution coverage in Aba Samuel wetland. *unpublished* .

Tchobanoglous, G., Maitski, F., Thoson, K., & Chadwick, T. (1989). Evaluation and performance of city of San Diego plot scale aquatic wastewater managemen system using water hyacinth . *J wastewater* (61), 11-2.

Terry, N., & Banuelo, G. (2000). *Phytoremediation of Contaminated Soil and Water.* New York: Lewis Publishers.

Thomson S, E. T. (2002). Control of aquatic weeds through pollutant reduction and weed utilization: a weed management approach in the lower Kafue River of Zambia . *J Physics Chem Earth Sci* (27), 983-91.

Thorne, R. (1992). Classification and geograaphy of flowering plants. *J Botanical rev* (58), 225-348.

Tripathi, B., & Upadhyay, A. (2003). Dairy effluent polishing by aquatic macrophytes. *J Water Air Soil Pollut* (143), 377-85.

Trivedy PK, Gudekar, V. (1987). Treatment of textile industry waste using water hyacinth. *J Water Sci Technol* (19), 103-7.

Trivedy, R., & Thomas, S. (2004). Review of performance data of few pilot and full scale aquatic weed based waste treatment plants from India. *Asian J Microbiol Biotechnol Environ Sci* (6), 157-62.

Turner, M., & Rust, R. (1971). Effects of chromium on growth and mineral nutrition of soybean. J *Soil Sci* (35), 755-758.

Twongo, T. (1991). Status of the water hyacinth in Uganda. In Greeathead, A., P.J (eds.) control of Africa's floating water weeds. *Control of Africa's floating weeds* (pp. 55-57). Harare,Zimbabwe: CAB international.

U.S.EPA. (1988). Design manual-constructed wetlands and aquatic systems for municipal wastewatrer treatment. Cincinnati: U.S. environmental protection agency.

Van Der Meer MB, Verdegam, M. (1996). Comparison of amino acid profiles of feeds and fish as a quick method for selection of feed ingredients: a case study of Colossoma macropomum (Cuvier). *J Aquac Res* (27), 487-95.

Vander Weert, R. & Kamerling, G. E. (1974). Evaporation of water hyacinth (*Eichhornia crassipes*). *J of hydrobiology* (22), 201-212.

Verma, R., Singh, H., & Ganeesha, R. (2003). Assessment of changes in water hyacinth coverage of water bodies in Northern part of Banglore city using themporal remote sensing data . *J Aquatic Sci* , 792-804.

Vesk, P., & Allaway, W. (1997). Spatial variation of copper and lead concentrations of water hyacinth plants in a wetland receiving urban run-off. *J Aquat Bot* (59), 33-44.

Vlyssides, A., & Israilides, C. (1997). Detoxification Of Tannery Waste Liquors With An Electrolysis System. *J Environmental Poll* (97), 147-152.

Winter, T. (1992). A physiographic and climatic framework for hydrological studies of wetlands. *J Environment Canada*, 127–148.

Woomer PL, M. R. (2000). Biological management of water hyacinth waste in Uganda. *J Biological Agri* (17), 181-96.

Zayed, A., Gowthaman, S., & Terry, N. (1998). Phytoaccumulation of trace elements of by wetland plants. *Duckweed. J Environmental Qual* (27), 715-21.

Zeiger, C. F. (1962). *Hyacinth* obstruction to navigation. *J Hyacinth Cont* (1), 16-17.

Zeleke, Tessema. (1992). Effect of water hyacinth on the level of dissolved oxygen in Aba Samuel wetland. *unpublished*

Zhu, Y., Zayed, A., Qian, J.-H., De Souza, M., & Terry, M. (1999). Phytoaccumulation of trace elements by wetland plants: Water hyacinth. *J Environ Qual* (28), 339-44.