

**INTERACTIVE EFFECT ON MORPHOLOGY, PHENOLOGY AND
GROWTH OF THE WILD RICE (*Oryza longistaminata*) ECOTYPES AND
CULTIVATED RICE (*Oryza sativa*) CULTIVARS FROM THE COASTAL
REGION OF KENYA //**

BY

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BIOLOGICAL SCIENCES, IN PARTIAL FULFILLMENT OF A
MASTERS OF SCIENCE DEGREE IN BOTANY**

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DECLARATION

I, Serah Mwingi Njeru, hereby declare that this thesis is my original work and has not been presented for a Master of Science degree in any other university

Signature SMN

Date 18/8/2011

We the undersigned declare that this thesis has been submitted with our approval as the university supervisors

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DEDICATION

This thesis is dedicated to my mother, Retisia Kivutu, for her great dedication towards all my academic achievements. Her efforts are always appreciated and will remain a challenge to me forever. Mama you have done a great job.

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Abbreviations and Acronyms

ANOVA	Analysis of Variance
DAT	Days after Transplanting
DMRT	Duncan's Multiple Range Test
IRRI	International Rice Research Institute
JICA	Japan International Cooperation Agency
MIAD	Mwea Irrigation Agricultural Development
MIS	Mwea Irrigation Scheme
NIB	National Irrigation Board
PVC	Polyvinyl Carbon
RFLP	Restriction Fragment Length Polymorphism
RH%	Relative Humidity %
SSR	Single Sequence Repeats

Definitions

Genus: A taxonomic category ranking used in biological classification that is below a family and above a species level, and includes group(s) of species that are structurally similar or phylogenetically related.

Species: An individual belonging to a group of organisms (or the entire group itself) having common characteristics and (usually) are capable of mating with one another.

Variety: A taxonomic rank below subspecies in botany, which is usually as a result of selective breeding and diverge from the parent species or subspecies in relatively minor ways.

Cultivar: A race or variety of plant that has been created or selected intentionally and maintained through cultivation.

Wild species/population: Group of plants with common characteristics which grow in a natural state and are not cultivated.

Weedy species/population: undesirable group of plants with common characteristics which grow together with the cultivated crops and cause harm to them

Ecotypes: A subdivision of an ecospecies, comparable to a subspecies or geographic race and consisting of an isolated population selectively adapted to a particular set of environmental conditions.

Land race: local variety of a domesticated plant species which has developed largely by natural processes, by adaptation to the natural and cultural environment in which it lives

ABSTRACT

Rice is the third most important cereal crop in Kenya after maize and wheat. Among the major constraints of rice production in Kenya is wild rice which occur in or near the rice fields as weeds or in wild form. One of the important weeds is *Oryza longistaminata* which belongs to the *sativa* complex with AA genome the same as cultivated rice (*Oryza sativa* L). *O. longistaminata* is an economically important weed in Kenya and there is need for the conservation of the species in its natural form. This experiment was therefore, aimed at finding out the effect of interaction between wild rice (*O. longistaminata*) and cultivated rice (*O. sativa*), by studying their morphology, phenology and above ground biomass. Four ecotypes of *O. longistaminata* were collected from the coastal region of Kenya at Kipini, Fioni, Ramisi and Tana Delta. Two cultivars of *O. Sativa* (Pishori and Basmati) were got from the same region. Pishori is a landrace while Basmati is an improved variety. The ecotypes and the cultivars were planted in pure and mixed stands in a screen house. Morphological data which included plant height, number of tillers, flag leaf area and reproductive stages in growth of a rice plant showed significant ($P<0.05$) differences in the wild/weedy populations from the cultivars while the two cultivars did not show any significant ($P>0.05$) difference in their morphological characteristics and phenology. The effect due to interaction between the weedy/wild ecotypes and the cultivars was slightly shown in the Kipini weedy population. The above ground biomass was affected by the cropping system, where, rice plants planted in pure stands had a higher biomass than the ones planted in mixed stands. The wild/weedy populations had a lower biomass than the cultivars. These results suggest that more time is needed for such a study and genetic markers used to detect the possible development of new traits. Interaction of the wild/weedy populations of *O. longistaminata* and *O. Sativa* cultivars may lead to alterations in morphology and phenology of the two.

CHAPTER ONE

1.0 INTRODUCTION

Rice (*Oryza sativa* L) is the world's single most important food crop and primary food source for almost a half of the world's population (Anon, 1994). In Kenya, rice is the third most important staple food after maize and wheat, and forms a large part of human diet in urban populations. The widely used variety is *Basmati 370W*. About ninety five percent of the rice in Kenya is grown under irrigation in paddy schemes. The remaining five percent of the rice is rain fed (Wanjogu, 2004).

Among the major cereal crops grown in the world, only rice and sorghum have their wild related species growing in the same geographical regions (Bhatia and Mitra, 2003). In the coastal region of Kenya, *Oryza. longistaminata* chev. Et Roehr are found growing together with cultivated rice as a weed or near the fields with *Oryza sativa*, alongside river banks and in the wild as wild rice. This close association between weeds and domesticated crops has led some weeds to evolve important characteristics associated with their life history strategies through mimicry of associated crop characteristics such as growth form and maturity dates (Barrett, 1983). These aspects have been facilitated by the unconscious human selection for weed populations along with the repeated sowing and harvest cycles of the crops.

Unintended human assistance has helped the wild relatives of rice expand their ranges to all of the inhabited continents, often becoming serious weeds of both cultivated rice and other crops (Holm *et al.*, 1997). Cultivated rice species are interfertile with certain close wild relatives such as *Oryza rufipogon* Griff and *O. sativa* (Chen *et al.*, 2004).

The fitness of hybrid progeny from crosses between wild and cultivated rice is generally high but hybrids from certain crosses show reduced fertility (Chu *et al.*, 1969, Langevin *et al.*, 1990). Hybrids between cultivated rice and wild species of *Oryza* have been reported in Orissa region in India (Harlan *et al.*, 1973). In Kenya, a hybrid was found between *O. punctata* and an *O. sativa* landrace (locally called sindano bahari). This was in a small farmer's field in a valley bottom at the Kenyan coast (Harlan, 1981). The five hybrid plants were growing as weeds in the cultivated rice field. The potential for a crop to hybridize with a weed is highly dependent on sexual compatibility and relatedness between the parent species, normal embryo and/or endosperm development. Overlap of the flowering period of sexually compatible cultivars in traditional cultivars, landraces, wild and weedy relatives

enhances the chances of out crossing. Most crops have been dramatically altered in their general phenotype following their long history of domestication probably due to hybridization (Conner *et al.*, 2003).

This crop-wild weed hybridization can be of great significance in evolution, because this process may result in crop-wild gene flow that could alter the genetic make-up of both populations (Ellstrand *et al.*, 1999). A great interest in crop-wild hybridization has been generated by the concerns of biosafety for genetically modified crops, because gene flow can be an avenue for transgene escape from genetically modified crops to their wild relatives (Ellstrand and Hoffman, 1990).

Studies carried out by Snow *et al.* (1998, 2001, 2003); Spencer and Snow (2001); Burke and Rieseberg (2003) have all found crop-wild/weed hybrids to be nearly as fit as their wild parents, but Jørgensen *et al.* (1996), reported a decline in fitness among hybrid progeny. This shows genes from cultivated crop might persist in wild populations for some time (Song *et al.*, 2004). As estimated by the performance of hybrids in comparison with their parents, it is essential to identify the possible hybrids in a species in order to predict their fate, and, understand the evolutionary significance of inter specific hybridization (Arnold and Hodges, 1995). This also can serve as an effective approach for assessment of ecological consequences caused by transgene escape through gene flow (Snow *et al.*, 2003; Burke and Rieseberg, 2003). Gene flow from crop taxa can add new genes into wild populations, which then can re-assort into novel combinations and therefore may have a substantial impact on the evolution of wild populations (Arriola and Ellstrand, 1996; Arnold, 1997; Ellstrand *et al.*, 1999; Jarvis and Hodgkin, 1999). This may result in more aggressive weeds or super weeds (*et al.*, 1985; Langevin *et al.*, 1990; Arriola and Ellstrand, 1996), and may lead to extinction of rare and endangered species (Ellstrand and Elam, 1993; Rhymer and Simberloff, 1996). If gene flow from crop to wild plants is common, we would expect that local weeds should be more similar morphologically and genetically to the local cultivars, in the areas they grow in, than to other cultivars grown in other places. Weeds may mimic crops by natural selection based on existing variation in their population, or that they may evolve as a result of gene flow from the local cultivars (Barrett, 1983).

1.1 Rice production in Kenya

Most of the rice produced in Kenya is grown under irrigation. Total national rice production in Kenya is about 70,000 tonnes per annum. Consumption is about 200,000 tonnes signifying a deficit of 140,000 tonnes that is imported (Wanjogu, 2004). In Kenya, the rate of increase in consumption of rice is higher (12%) than in other cereals like wheat (4%). Average unit yield production under irrigation is 5.5 t/ha for aromatic varieties and 7 t/ha for non-aromatic varieties. Rain fed rice is about 8% of the total cultivated rice in Kenya with unit yield of 2 t/ha (Wanjogu, 2004). These low yields are as a result of diseases, for example rice blast and blights, low rainfall, poor varieties which are not adapted to the climate and weeds. One of the major weeds, especially in the rain fed rice growing areas, is *O. longistaminata* and *O. punctata*. *O. longistaminata* grows at the edges of rice fields, and since it is rhizomatous it spreads very fast in to the rice fields suppressing the cultivated rice (Vaughan, 1994) making it to be a very serious weed of rice.

1.2 Problem Statement

In Kenya, *O. longistaminata* is gradually being eroded due to destruction of natural habitats by expanding agricultural activities, overgrazing and changes in land use (NWCMP, 1996; Emerton and Muramira, 1999). This species is an important genetic resource for rice improvement since it is accessible for gene transfer through sexual means (Oka, 1988). The species is highly out-crossing; meaning other genes from cultivated rice can be transferred into it resulting into inter-specific hybrids. This may lead to loss of its important traits and therefore the need to conserve *O. longistaminata* in its natural form as wild rice.

1.3 Justification

Little is known about mimicking ability of *O. longistaminata* on *O. sativa*, although some reports, for example, Song *et al.*, (2004); Ellstrand,(2003); Aggarwal *et al.*, (1997) show that there is gene flow between wild and cultivated rice cultivars. Understanding the mimicking ability of the two cultivars would be important for both genetic conservation of the *O. longistaminata* genetic resources and also for impact assessment of rice especially in this era of genetically modified crops, their transgene escape and the possible ecological consequences. If gene flow is common, we would expect local weeds to be more similar to neighbouring local cultivars, more than they are to other cultivars from another region. The

assumption during this research was that, *O. sativa* is compatible with *O. longistaminata*, a wild relative to form hybrids, since they belong to the same genomic group. The study therefore will try to answer the following questions, 1) does *O. longistaminata* mimic the characteristics of associated cultivated rice species (*O. sativa*), in the areas it grows in? 2) are the four ecotypes of *O. longistaminata* different in morphology, phenology and biomass/seed production? 3) and if they are, are weedy *O. longistaminata* ecotypes more similar to landrace of *O. sativa* and wild *O. longistaminata* ecotypes different from the two cultivars of *O. sativa* ?

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Origin and distribution of *Oryza sativa* and *Oryza longistaminata*

Oryza sativa originated from Asia and it is the most widely grown species. It is grown all over the world and more than 90% of it is grown and consumed in Asia (Khush, 1997). *O. sativa* is mostly grown under irrigated agro-ecosystems in Central, Coast and Western parts of Kenya. *O. sativa* is grown mainly as improved cultivars BW 196, IR 2793, ITA 310, Jasmine and Basmati 217 and 370, which are aromatic and most preferred by consumers (Wanjogu and Mugambi, 2001). These cultivars have been improved mainly to increase their production, resistance against diseases and adaptation to different environments. Landrace *O. sativa* was brought to Kenya from Tanzania more than one hundred years ago. It started being grown along the coastal areas and was first cultivated in Kenya at Ziwani in Kwale District. The most grown *O. sativa* landrace cultivar grown in Kwale is *Pishori*. The cultivar is also grown locally and is an equivalent of the Basmati which is grown in the irrigation schemes in other areas such as central Kenya. Improved Basmati has common characteristics as the landrace *Pishori* in terms of production, maturity time and aroma.

Oryza longistaminata originated from West Africa and later spread to other parts of Africa. It is the most widely distributed wild rice species in Africa occupying 54% of world's wild *Oryza* species (Vaughan *et al.*, 2003). In Kenya, *O. longistaminata* is found in Kwale, Tana River, Kilifi and Lamu districts in the coastal region (Kiambi *et al.*, 2005). This species is a perennial relative of *Oryza glaberrima*.

2.2 Taxonomy and genome of rice

Rice belongs to the grass family Poaceae (Gramineae) and to the genus *Oryza* that includes twenty-two wild species, nine of these wild species are tetraploid. The rest of the wild as well as cultivated species are diploid (Khush, 1997). The genus has ten identified genomes which are assigned based on cytological and biochemical differences. These includes crossing relationships, chromosome number, and chromosome pairing behaviour of F4 hybrids from crosses between species with known and unknown genomic constitution and by evaluation of divergence at the molecular level through total genomic hybridization (Aggarwal *et al.*, 1997). According to their genetic relationship, *Oryza* species are grouped into four complexes

as shown in Table 1. *Oryza sativa* complex with AA genome, *Oryza officinalis* complex with CC, BBCC, CCDD, EE genomes. *Oryza ridleyi* complex with HHJJ genome and *Oryza granulata* complex with GG genome. *Oryza brachyantha* with FF genome and *Oryza schlechteri* with unknown genome are species that have not yet been assigned any complex (IRRI, 1964, 1999; Chang, 1976; Vaughan *et al.*, 2003). *O. sativa* is closely related to the wild species *O. longistaminata* as they share the AA genome and so it is assumed they can hybridize easily. It belongs to the AA Genome Group of the *O. sativa* complex and is diploid ($2n=24$). It is outcrossing and is partially self-incompatible (Kiambi *et al.*, 2005).

Genes from wild rice species can be transferred to *O. sativa* by conventional crossing and back crossing procedures (Brar and Khush, 1997). It has been shown with enough evidence that *O. sativa* and *O. glaberrima*, which is a member of genome group AA and a native of West Africa, naturally hybridize with their wild relatives (WARDA, 1996). Plants appearing to be hybrids or hybrid derivatives of both cultivated rice species and their wild relatives occur frequently in and near rice fields where wild taxa are present, particularly in Asia and Africa (Ellstrand, 2003). Griselda *et al.* (2005) observed hybrids of *O. sativa* and *Oryza rufipogon*. It has also been observed that hybrids between *O. sativa* and *O. rufipogon* have nearly the same fitness as their parental species (Song *et al.*, 2004).

2.3 Habitat and morphology of rice

O. sativa has great diversity of forms and varietal diversity; and is classified into three major groups namely, indica, temperate japonica and tropical japonica. Indica varieties are usually slender, with awnless grains, light green leaves and many tillers. Temperate japonica varieties are usually roundish, pubescent grains, dark green leaves and few tillers while tropical japonicas are usually large, rounded awned, pubescent spikelets and low shattering with few tillers. The species is cultivated on dry-lands as rain fed rice and wetland as irrigated rice. It can also survive deep waters (≥ 4 m deep), as floating rice. It does better at altitudes of 0-3000m above sea level (Vaughan, 1994).

Table 1: Chromosome number, genome and distribution of species in genus *Oryza*. (Vaughan *et al.*, 2003)

Complex and species	2n	Genome	Distribution
<i>O. sativa</i> complex			
<i>O. barhii</i> ^a A.Chev.	24	AA	Sub-Saharan Africa
<i>O. longistaminata</i> ^a Chev.et Roehr	24	AA	Sub-Saharan Africa
<i>O. glumaepatula</i> Steud	24	AA	West Africa
<i>O. meridionalis</i> Ng	24	AA	South & Central America
<i>O. nivara</i> Sharma et Shastry	24	AA	Tropical Australia
<i>O. rufipogon</i> Griff	24	AA	Tropical & sub-tropical Asia
	24	AA	Tropical & sub-tropical Asia & tropical Australia
<i>O. officinalis</i> complex			
<i>O. punctate</i> ^a Kotechy ex Steud	24,48	BB,BBCC	Sub-Saharan Africa
<i>O. eichingeri</i> ^a Peter	24,48	CC	East Africa & South Asia
<i>O. officinalis</i> Wall ex Walt	24,48	CC,CA	Tropical & sub-tropical Asia & sub-tropical Australia
<i>O. rhizomatis</i> Vaughan	24	CC	sub-tropical Australia
<i>O. minuta</i> J.S. Pres. ex C.B Presl.	48	BBCC.	Sri Lanka
<i>O. alta</i> Swallen	48	CCDD	Philippines & Papua New Guinea
<i>O. glandiglumis</i> (Doell) Prod.	48	CCDD	Guinea
<i>O. latifolia</i> Desv.	48	CCDD	South & Central America
<i>O. australiensis</i> Domin	24	EE	South & Central America Tropical Australia
<i>O. meyeriana</i> complex			
<i>O. gramilata</i> Nees et Arn. ex Watt	24	GG	South & South east Asia
<i>O. meyeriana</i> (Zoll.et Mor.ex Steud.) Baill.	24	GG	South east Asia
<i>O. ridleyi</i> complex			
<i>O. longiglumis</i> Jansen	48	HHJJ	Irian Jaya, Indonesia & Papua New Guinea
<i>O. ridleyi</i> Hook.	48	HHJJ	South Asia
Species not yet assigned any complex			
<i>O. brachyantha</i> ^a Chev.et Roehr	24	FF	Tropical Africa
<i>O. schlechteri</i> Pilger	48	*	Papua New Guinea
<i>O. neocaledonica</i> Morat	24	*	New Caledonia

Note: * = Unknown genome

O. longistaminata is found in swampy areas, edges of lakes or ponds, in and at the edges of rice fields, and in irrigation canals; in permanently wet or seasonally dry areas. It can grow in waters 4m deep, but usually 1m or less. The species prefers black cotton soils, as well as alluvial soils. It is found at altitude of 0 - 2000m (an average of 924m) above sea level (Vaughan, 1994). It is an erect, perennial hydrophytic grass with extensive, branched

rhizomes that grows to a height of 2m tall or more. Culms are spongy. The leaf blades are auriculate, 450 mm long and 15 mm wide. The ligule is an unfringed membrane, 15 - 45 mm long and acute or clefted and more than 15 mm wide. The inflorescence is an open or loosely contracted panicle. The spikelets are 4.5 -11.5 mm long, compressed laterally and disarticulating above the glumes. Lemmas are 3 - 9 nerved, entire, with awns 40 - 80 mm long. Palea is present on the glume but is relatively long but narrower than the lemma. The anthers are 1.5 - 8.2 mm long (Gibbs-Russel *et al.*, 1989).

2.4 Weed evolution in rice

Weeds continue to evolve in man-made habitats through a range of strategies (de Wet and Harlan, 1975; Londo and Schaal, 2007). They may evolve from colonizer species by selection for adoption to habitats that are continually being disturbed, from the abandonment of domesticated races or escape of crops from cultivated attributes, from derivatives of hybrids between wild and cultivated races of crop species and from gene introgression between the wild and cultivated races (Conner *et al.*, 2003). The latter two, combined with increased weediness, are important for assessing the possibility of out-crossing.

2.4.1 Mimicking in weeds

Mimicking is a state where the weed resembles the crop at specific stages during its life history and as a result of mistaken identity, evades eradication (Barret, 1983). Mimicking can take place through selection, hybridization and gene flow. Crops can also contribute to weeds mimicking through hybridization, evolution of crops into weeds and crops appearing as unwanted volunteers in the subsequent culture where they are considered to be weeds.

2.4.1.1 Selection

Selective forces imposed by agricultural practices have caused diverse adaptive strategies of weed populations, making these populations develop close resemblance with the crop, thus making the weed difficult to eradicate (Barret, 1983). Most weeds especially in the tropics have been influenced by artificial selection caused by cultivation practices such as mechanization, fertilizer application and irrigation among others, hand weeding, threshing and winnowing. Among the most important selective pressure is herbicide selection in weeds. Wide use of herbicides can lead to broad changes in the spectrum of weeds infesting a

particular crop. Herbicides may exert strong selection forces which modify the growth patterns of weeds resulting into strategies that enable these weeds escape the destructive impacts of the herbicide. Such a case was found in United States of America where annual grasses have increased in abundance in maize; and in soybeans where broad-leaved weeds have replaced grasses as the most serious weeds in some states due to the use of herbicides (Barret, 1983).

Weed populations which are continuously associated with specific agricultural systems may evolve phenological patterns which optimize survival within the most favorable growing period. The more closely a weed species resembles the crop in habit, phenology, and ecological requirements, the more difficult it is to control without causing damage to the crop. In rice, weedy rice is a particularly insidious weed due to its similarity to the domesticated varieties. Many of the easiest ways to kill weedy rice are likely to also harm the crop, making it very difficult to manage the weed. Moreover, if even a small fraction of the weedy plants survive and reproduce, weedy rice is so productive that it can spread and cause major economic damage (Ferrero, 2003). If selection imposed by agricultural practices continues for some span of time and some specific genetic variation occurs, locally adapted weeds races will occur which may be difficult to control (Barret, 1983).

2.4.1.2 Hybridization and gene flow

Hybridization between cultivated species and their wild/ weedy relatives occurs frequently in a wide range of plant species especially grass (de Wet and Harlan, 1975; Kane and Riesberg, 2007). This may have important practical and economic consequences, since it promotes the evolution of more aggressive weeds or “super weeds”, which may be difficult to control (Barret, 1983, Conner *et al.*, 2003). Where such hybridization occurs, gene flow is mainly in one direction, from the cultivated species into the wild/weedy population. Examples of these are seen in sorghum, pearl millet and rice (Langevin *et al.*, 1990). Rice species with the AA genome have relatively high sexual compatibility, complete chromosome pairing in meiosis of F1 interspecific hybrids, and relatively high pollen and panicle fertility of the F1 hybrids (Naredo *et al.*, 1997 and 1998). This indicates the potential for genes in cultivated rice varieties to escape to wild relatives through cross pollination and persist through the survival of intraspecific and interspecific hybrids.

The primary transfer of genetic material is through pollen donation by the cultivated species into the weedy populations (Ladizinsky, 1985). Gealy *et al.*, (2003) reviewed previous studies of gene flow from cultivated to weedy rice and noted that typical rates of

crop – weed hybridization were approximately 0.01 to 1 percent. They also noted that gene flow may occur in either direction, from crop to weed or from weed to crop.

The transfer of characters from the cultivated rice species to wild populations is fostered by the breeding structure of rice populations. The cultivars are predominantly self fertilizing while wild populations are out crossing at varying degrees (Oka and Chang, 1959; Londo and Schaal, 2007). The individuals in the wild rice populations are usually at a relatively low density in comparison to the rice cultivars and are therefore, likely to be cross fertilized by the pollen originating from the cultivated populations, assuming that the flowering periods are roughly synchronous. The extent to which crop genes are passed to weedy/wild populations is lowest when the flowering times of the crop and the weed are partially synchronous, as it occurs in some situations. However, it is possible that the crop itself can establish feral populations (Baki *et al.*, 2000), which could then possess wild genes.

Some characteristics of cultivated rice found in the hybrids include larger grain size, thicker stems, narrower and more compact panicles, higher fertility, and less or even no shattering of panicles (Lu and Snow, 2005). All these characteristics may be as a result of crop – weed hybridization (Song *et al.*, 2004). Just as hybrid vigour is seen when inbred, cultivated lines are crossed to produce “hybrid” rice, and the weedy rice may benefit from hybridizing with the crop and this would result in greater heterozygosity. In Louisiana USA, for example, Langevin *et al.* (1990) reported greater vigour in crop – weed hybrids than in their weedy parents, and frequencies of crop alleles in weedy rice were as high as 52 percent after only two years of contact with the crop. However, in Arkansas in USA, first generation hybrids between cultivated and weedy rice flowered so late that they had much lower fitness than their weedy parents (Zhang *et al.*, 2003).

Over the course of several generations, crop genes that are strongly deleterious to weedy rice, as well as other genes that are linked to deleterious crop genes, are likely to be purged from weedy populations by natural selection and selection pressures from farmers. Conversely, linked genes that are associated with greater survival and reproduction are expected to increase in frequency following episodes of hybridization. Thus crop genes are expected to introgress and persist in weedy rice at rates that reflect both the frequency of hybridization and the selective effects of these genes (Ellstrand, 2003).

2.4.1.3 Evidence of rice hybridization in Africa

Some wild species growing in and along the borders of farmer's fields in Kenya, Malawi and Mozambique were reported to flower almost at the same time with *O.sativa*, leading to production of spontaneous inter-specific hybrids (WARDA, 1997). Most of the hybrids and their derivatives were more vigourous than the wild forms with which they grew. Sometimes they formed separate colonies or populations and in some cases they were found growing together with the cultivated forms as mixed populations (WARDA, 1997).

In Kenya, a hybrid was reported between *O. punctata* and an *O. sativa* landrace (sindano bahari) (WARDA, 1997). This was in a small farmer's field in a valley bottom at the Kenyan coast. The five hybrid plants were growing as weeds in the cultivated rice field. These hybrids were characterized by vigourous vegetative growth, larger seeds than the wild forms, and short and strong whitish awns. In Malawi, there was a hybrid between *O. longistaminata* and a landrace (kalulu) found in farmers field (WARDA, 1997). Several immature populations of both *O. barthii* and *O. longistaminata* were also found on the farm borders and irrigation canals. Two types of hybrids were observed, one had a non-shattering rachis, was rhizomatous and also vigourous with long, soft whitish awns, characteristics of *O. longistaminata*. It was found growing in small populations in 0.3m deep water in which the landrace would not normally survive. The other hybrid had shattering rachis, not rhizomatous with soft whitish awns growing at the side of the lake. Results of the work on inter-specific hybridization between *O. longistaminata* and *O. sativa* by WARDA (1997), showed that the seeds of these hybrids matured earlier than those of the wild species and later than for cultivated rice.

2.5 Weed management in rice

Weeds are plants growing out of place. They are unwanted, prolific, competitive and often harmful to the target environment. Weeds cause many direct damages to a crop. They reduce crop yield and quality by competing for nutrients, water and light. They intensify the problem of disease, insects, and other pests by serving as their hosts and also reduce efficiency in irrigation systems (De Datta, 1987).

Rice fields are the primary habitats for weedy rice and the weed can substantially reduce the crop yields (Baki *et al.*, 2000). Weedy rice can be very difficult to control because

many of its seeds disperse before the crop is harvested and then accumulate in the soil as seed bank. At the same time, undispersed seeds of weedy rice can be collected by farmers and inadvertently planted with the next generation of crop seeds. Competition with the cultivated crop and contamination of harvested seeds with undesirable weedy rice grains make weedy rice a serious problem for rice growers worldwide (Lu and Snow, 2005). Weed competition in crop plants is severe because of the wide range of adaptability of the weeds that are native to an environment (Remison, 1978, Ferrero, 2003). When early competition is not controlled, the rate of growth of the crop is restricted significantly (Dingkuhn *et al.*, 1998). In cultivated rice, the critical period of weed competition is between 4 - 9 weeks after seeding or transplanting and this period coincides with the time between maximum tillering and flowering (IRRI, 1978, 2004; Arraudeau and Vergara, 1988).

Weed competition in rice does not occur during the entire cropping period. Weeds should be controlled in the critical period of competition, which is between vegetative to flowering. Control of weeds by use of one method may not be sufficient and so integrated weed control practices (IWP) are advisable (IRRI, 2004; Rainbolt, 2006; De Datta, 1981; Arraudeau and Vergara, 1988; Parker and Dean, 1976). IWP includes preventive, cultural, manual, mechanical, biological and chemical practices. The widely used practices are preventive, cultural and manual control (IRRI, 2004). Chemical control is efficient but it is detrimental to the environment and more so it is very expensive. For example, use of dalapon and diuron, herbicides with high residual toxicity, necessitates a long fallow period (Parker and Dean, 1976). Another possible weeds control is by use of genetically modified rice that is resistant to herbicides.

2.5.1 Genetic modification as a solution to weed control

Introduction of herbicide resistant rice could bring areas heavily infested with weedy rice and have been abandoned back to rice production, allow longer term crop rotations, reduce consumption of fossil fuels, promote the replacement of traditional chemicals by more environmentally benign products, and provide more rice grain without adding extra land for production. However, there are also concerns about the impact of releasing herbicide-resistant rice. The most concern is the possibility of transfer of the resistance trait to compatible weed species, for example *O. longistaminata*. Development of such herbicide resistant weedy rice populations would substantially increase chemical weed management options by farmers. There could also arise problematic herbicide resistant rice volunteers. This added selection pressure to weed populations could aggravate the already serious weed

resistance problems. If it evolves, weedy *Oryza* species could become resistant to broad-spectrum herbicides such as imidazolinone. Introgression from domesticated rice may allow these weeds to gain advantageous traits such as herbicide resistance from imidazolinone-tolerant strains now widely cultivated (Gealy 2005), or take seed colour or size that mimic those of domesticated varieties, enabling weedy rice to escape screening (Ellstrand *et al.*, 1999).

The high potential for introgression into weedy varieties should be carefully considered when new transgenic genotypes of rice are generated, as problematic transgenes can escape even in early experimental stages of new crop production in some cases (Reichmann *et al.* 2006). Gene flow between different weedy varieties may allow these advantageous traits to spread and combine into potentially more problematic phenotypes. Mitigating measures to the problem have been proposed such as prevention of gene flow, attainable by both conventional breeding and molecular genetics.

2.6 Growth and Development of Rice Plant

Rice is grown from the Equator to 50° North and South at altitudes of 0 - 2,500 meters above sea level. It takes approximately 120 - 180 days from planting to maturity, depending on the variety. Days in vegetative stage differ with variety. Reproductive and ripening phases are constant for most varieties; and panicle formation to flowering takes approximately 30 days (Wanjogu *et al.*, 1995).

2.6.1 Development stages of rice plant

The development of the rice plant may be divided into four phases:

Nursery stage – This stage runs from sowing to transplanting. It takes duration of approximately 1 to 3 months.

Vegetative stage - This stage runs from transplanting to panicle initiation. Duration varies from one and half to three months. The vegetative phase is characterized by active tillering, gradual increase in plant height, and leaf emergence at regular intervals. Tillers that do not bear panicles are called ineffective tillers. The number of ineffective tillers is a closely examined trait in plant breeding since it is undesirable in irrigated varieties but is sometimes an advantage in rainfed lowland varieties, where productive tillers, or panicles may be lost due to unfavourable conditions.

Reproductive stage- This stage runs from panicle initiation to flowering. Duration is approximately one month. The reproductive growth phase is characterized by culm elongation (which increases plant height), decline in tiller number, emergence of the flag leaf (the last leaf), booting, heading, and flowering of the spikelets. Panicle initiation is the stage just before heading when the panicle has grown to about 1 mm long and can be recognized visually or under magnification following stem dissection. Spikelet anthesis (or flowering) begins with panicle exertion (heading). Consequently, heading is considered a synonym for anthesis in rice. It takes 10 - 14 days for a rice crop to complete heading because there is variation in panicle exertion among tillers of the same plant and among plants in the same field. Agronomically, heading is usually defined as the time when 50% of the panicles have exerted. Anthesis normally occurs between 10.00 and 13.00 hours in tropical environments and fertilization is completed within 6 hours. Only very few spikelets have anthesis in the afternoon. Usually anthesis occurs when the temperature is low. Within the same panicle it takes 7 - 10 days for all the spikelets to complete anthesis. The spikelets themselves complete anthesis within 5 days.

Ripening stage – This stage runs from flowering to full maturity of rice grains. Duration is approximately one month. This phase includes grain growth or filling (De Datta, 1981; IRRI, 2004). Ripening follows fertilization and may be subdivided into milky, dough, yellow-ripe and maturity stages. These terms are primarily based on the texture and colour of the growing grains. The length of ripening varies among varieties from about 15 to 40 days. (<http://beaumont.tamu.edu/eLibrary/StudyRiceContest/2006/RicePlantMorphology.pdf>, 2007).

2.6.2 Dormancy in rice

Seed dormancy refers to the low germinability of viable, freshly harvested seed (Jennings *et al.*, 1979). It is an adaptation of the plant, which prevents premature germination and helps to preserve a supply of seed in the soil, an obvious selective advantage in wild plants, especially in environments that undergo favourable and/or unfavourable season cycles (Harlan, 1992). According to Vaughan (1994), the majority of wild rice species possess a strong dormancy.

Japonica seed varieties have little or no grain dormancy whereas many indica varieties have grain dormancy that varies from weak to strong. Germination rates of harvested seeds in rice vary widely in dormancy duration and the degree of breaking the seed dormancy. Non-dormant or weakly dormant grain often germinates insitu (Yoshida, 1981 and Jennings *et al.*, 1979). The duration of dormancy varies according to the biotype and the storage conditions

of the seed after shattering (Naredo *et al.*, 1998). Environmental conditions during seed formation, especially moisture and the storage temperature are considered to be the main factors that can affect the length of dormancy. Seed dormancy has also been shown to have a hereditary component in a number of species, many of which are crops or serious crop weeds (Baskin and Baskin, 1998). Several studies have shown that seed dormancy is a complex character influenced not only by genetic factors but also by environmental (Li and Foley, 1997). For example, in rice, studies have shown that seed dormancy is a quantitative trait governed by polygenes with cumulative but unequal effects, and is also strongly affected by environmental conditions during seed development (Chang and Tagumpay, 1973).

Varying degrees of dormancy have also been reported for seeds of weedy rice. Franco *et al.* (1997) reported that in cultivated rice, depending on the cultivar, seeds present post-harvest dormancy that may persist for 90 – 120 days. The degree of dormancy may also vary considerably if seeds of *O. sativa* are harvested at various stages of ripeness and if stored at different temperatures, losing dormancy more rapidly at warmer temperatures than cooler temperatures (Gu *et al.*, 2003; Cohn and Hughes, 1981).

Several methods to break rice seed dormancy are available which include heat treatment at 50 °C for 4 - 5 days, mechanical dehulling or use of chemicals such as ammonium nitrate. The most and widely used method to treat freshly harvested rice grain is by heat. To achieve quick and full germination of different *Oryza* species for use of germplasm, ideal or standard procedures are necessary for breaking dormancy. However, no single method is recommended (Naredo *et al.*, 1998)

2.6.3 Grain germination in rice

After dormancy is broken, the seed absorbs adequate water for the first 18 hours. This water intake increases the seeds water content to between 25 and 35%. This duration is almost independent of temperature.

The germination process is divided into imbibition, activation and post germination stages. Water absorption at the activation stage is lower than at imbibition stage. The duration is temperature dependent and it increases at lower temperature. Germination occurs as a consequence of very active metabolic changes during the activation stage (Takahashi, 1965). Water content of rice seeds at the onset of germination varies with temperature and it ranges from 30 to 40% (Yoshida, 1981).

2.6.4 Seedling emergence in rice

The first seedling leaf usually emerges 3 days after sowing of pre-germinated seeds. Pre-germination is achieved by soaking the seeds for 24 hours and incubating them for 48 hours. The seedling stage includes the period from emergence until just before the appearance of the first tiller (De Datta, 1981).

2.6.5 Tillering in rice

Tillers are branches that develop from the axils at each unelongated node of the main shoot or from other tillers during vegetative growth (Yoshida, 1981). Each stem of rice is made up of series of nodes and internodes. The internodes vary in length depending on variety and environmental conditions, but generally increase from the lower to upper part of the stem. Each upper node bears a leaf and a bud, which can grow into a tiller. The number of nodes varies from thirteen to sixteen with only the upper four or five separated by long internodes. Under rapid increases in water level, some deep water rice varieties can also increase the lower internode lengths by over 30cm each, fairly quickly (Yoshida,1981). The leaf blade is attached at the node by the leaf sheath, which encircles the stem. Where the leaf blade and the leaf sheath meet is a pair of claw like appendages, called the auricle, which encircle the stem. Coarse hairs cover the surface of the auricle. Immediately above the auricle is a thin, upright membrane called the ligule. The tillering stage starts as soon as the seedling is self supporting and generally finishes at panicle initiation. Tillering usually begins with the emergence of the first tiller when seedlings have five leaves. The first tiller develops between the main stem and the second leaf from the base of the plant. Subsequently when the sixth leaf emerges, the second tiller develops between the main stem and the 3rd leaf from the base. Tillers growing from the main stem are called primary tillers. These may generate secondary tillers, which may in turn generate tertiary tillers. These are produced in a synchronous manner. Although the tillers remain attached to the plant, at later stages they are partly independent because they produce their own roots. Varieties and races of rice differ in tillering ability. Numerous environmental factors also affect tillering including spacing, light, nutrients supply, and cultural practices (<http://beaumont.tamu.edu/eLibrary/StudyRiceContest/Rice/Plant/Morphology.pdf>, 2007).

2.6.6 Panicle initiation and development in rice

Panicle initiation stage begins when the primordium of the panicle has differentiated and become visible. The panicle initiation occurs first in the main culm and follows into the tillers

in an uneven pattern (De Datta, 1981). Panicle development and growth start with the neck-node differentiation and end when the pollen is fully matured (Yoshida, 1981). The spikelets become distinguishable and the panicle extends upwards inside the flag leaf sheath. The total duration of panicle development varies with variety and weather, and range from 27 to 46 days (Yoshida, 1981).

Booting is the latter part of the panicle development stage. It occurs 20 to 25 days before flowering. The base of the leaf sheath bulges. Senescence of leaves and unproductive tillers become noticeable at the base of the plant (Arraudeau and Vergara, 1988; De Datta, 1981). The booting stage is followed by the emergence of the panicle out of the flag leaf sheath, known as heading.

2.6.7 Flowering or anthesis in rice

Anthesis begins with protrusions of the first dehiscing anthers in the terminal spikelets. At the time of anthesis, the panicle is erect in shape. In tropical environment, flowering begin at the top, middle and lower thirds, occurring in the first, second and third day respectively, after panicle exertion (Hernandez *et al.*, 1979). Flowering occurs about 25 days after visual panicle initiation regardless of rice variety. Flowering continues until most spikelets in the panicle have bloomed (De Datta, 1981). It takes around 7 days for all the spikelets in a panicle to open (Arraudeau and Vergara, 1988).

2.6.8 Ripening and senescence in rice

Ripening period is characterized by grain growth or filling, increase in size and weight, changes in grain colour, and senescence of leaves (Yoshida, 1981). Ripening involves milk grain, dough grain and mature grain stages. At the milk grain stage, the contents of the caryopsis are first watery but later turn milky. The top of the panicle bends gently in an arc when held upright. In the dough grain stage, the milky part of the grain turns first into soft and later hard dough. In the mature grain stage, grain colour in the panicles begins to change from green to yellow. The mature grain stage is complete when 90 - 100% of the filled spikelets have turned yellow and the senescence of the upper leaves, including flag leaves, is noticeable. In some varieties, the culm and upper leaves may remain green even when the grains have ripened (De Datta, 1981). During the initial phases of grain filling, the water content of the grains is above 58% and it declines to about 20% or less at maturity (Yoshida, 1981).

2.6.9 Rice harvesting

At maturity, the grains are full-sized and hard, and the panicles bend down (Arraudeau and Vergara, 1988).traditionally, rice harvesting is done by use of a sickle, where the rice is cut at the base when the heads are yellow (IRRI, 2004). After harvesting, the panicles are kept away from moisture. Threshing is done as soon as possible and grains dried to 14% moisture content (Yoshida, 1981; Arraudeau and Vergara, 1988).

2.8 Overall Objective

The overall objective was to study the interactive effect on morphology, phenology and growth of four ecotypes of wild rice (*O. longistaminata*) and that of two cultivars of cultivated rice (*O. sativa*).

2.9 Specific Objectives

- 1) To compare the general morphology and phenology of four ecotypes of *O. longistaminata* with that of two cultivars of *O. Sativa* grown in mixed planting and pure planting systems.
- 2) To estimate the biomass of the wild rice (*O. longistaminata*) ecotypes and that of the cultivated rice cultivars (*O. sativa*) grown in mixed planting and pure planting systems.

2.10 Hypotheses

- 1) Morphology and phenology of *O. longistaminata* ecotypes and those of *O. sativa* cultivars are similar when grown in mixed planting and pure planting systems.
- 2) *O. longistaminata* ecotypes and *O. sativa* cultivars have the same biomass when grown in mixed planting and pure planting systems.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Study Site

The study was carried out at Mwea Irrigation Agricultural Development (MIAD) centre in Central Province, Kirinyaga District (Figure 1). It is a project station within the Mwea Irrigation and Settlement Scheme under the National Irrigation Board (NIB). MIAD is a technical cooperation between the Government of Kenya and the Government of Japan. It was implemented by the Japan International Cooperation Agency (JICA), a Japanese governmental organization responsible for technical cooperation with developing countries.

The Mwea Irrigation and Settlement Scheme is situated approximately 100km, north east of Nairobi on the foot hills of Mount Kenya, at the intersection of longitude 37°20" and latitude 0°40" south at an altitude of 1,159 meters above sea level. Mwea occupies the lower altitude zone of Kirinyaga County in an expansive low-lying area. The mean annual rainfall is 950 mm with maximum amount falling in April/May (long rains) and October/November (short rains). The average maximum temperatures are in the range of 16 to 26.5°C. Relative humidity varies from 52 to 67%.

The Mwea Rice Irrigation Scheme is located in the west central region of Mwea Division and covers an area of about 14,000 acres of paddy rice fields, producing an average of 75 - 80% of total Kenya rice, accounting for 27,000 tonnes annually. The soils are black cotton vertisols (Wanjogu, 2004).

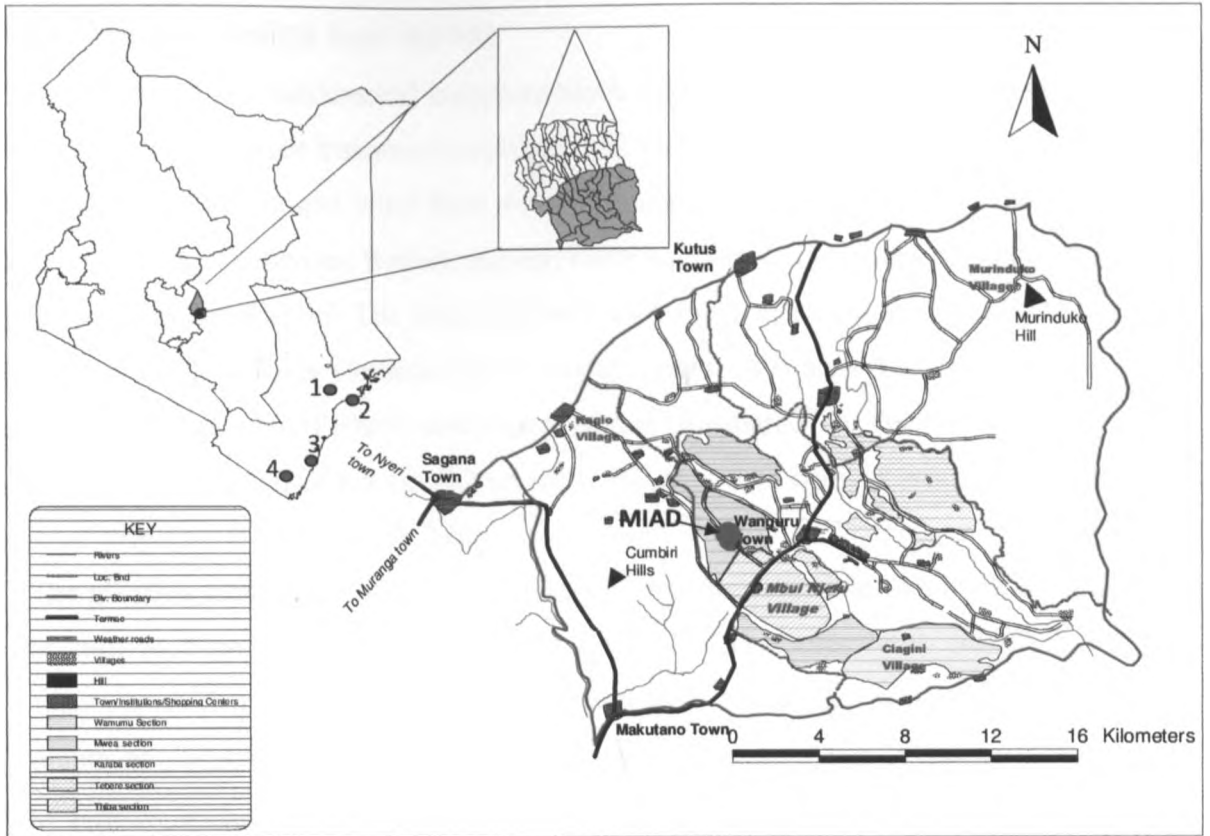


Figure 1: Rice collection sites at the Kenya Coast and location of the MIAD (Mwea Irrigation Agricultural and Development Centre (1 = TDIS or Tana Delta Irrigation Scheme; 2 = Kipini; 3 = Ramisi; and 4 = Fioni).

3.3 Experimental design and layout

The experiment was in a randomised complete block design (RCBD with three blocks each with six treatments and eight treatment combinations. The blocking was due to fluctuation in light intensity, temperature and wind flow within the screen house. The study consisted of four *O. longistaminata* ecotypes; Ramisi, Kipini, Fioni and Tana – Delta, and two cultivars of *O. sativa*; Basmati and Pishori. The four ecotypes of *O. longistaminata* were collected from Kwale, Lamu and Tana River Counties in the coastal region of Kenya. The two cultivars of *O. sativa* were landrace rice (Pishori) and improved rice (Basmati 370). The landrace cultivar was obtained from a farmer in Kwale district while the improved cultivar was obtained from the National Irrigation Board in Tana Delta.

The plantlets from the wild/weed populations were from rhizomes while the seedlings from the cultivated populations were raised from seeds. Rhizomes were collected from mature plants apart from the ones for the population from Kipini that were collected from young plants at vegetative stage. The Tana - Delta population was collected from an irrigation scheme with rice which had been there for six years. Since the two had been growing together for several years, it was assumed that there could be close interactions between the two cultivars. Ramisi and Fioni populations were from fields neighbouring landrace rice. There could have been interactions between the two populations of landrace and the wild rice. Kipini population was obtained from dry land without any pooled or flooded water. This population was collected from a region where rain fed rice had been growing since the introduction of rice at the Kenyan coast until the late 1990's when the fields were abandoned. *O. longistaminata* continued growing together with *O. sativa* until *O. sativa* was eradicated due to competition and the *O. longistaminata* remained in these fields.

Each ecotype and cultivar was grown alone in pure planting as Basmati, Pishori, Ramisi, Kipini, Fioni and Tana - Delta, and constituted six treatments. In mixed planting, each of the four ecotypes of *O. longistaminata* was grown together with each cultivar of *O. sativa* at equal densities in alternating rows. The combinations of the four ecotypes and the two cultivars were; **RP, RB, KP, KB, FP, FB, TP** and **TB** (table 2) and these constituted the eight treatment combinations.

In each block, there were six plots of pure planting for each cultivar and ecotype that served as control and eight plots of mixed planting resulting in fourteen plots per block. The pots were put in a screen house that was 10m in length by 8m width and 4m high. The frames were made of timber and Polyvinyl carbon (PVC) was used for roofing and also to cover the

sides. Chicken wire was used to affirm the PVC on the sides. The poles were made firm by use of cement, ballast and sand. The floor was cemented for easier maintenance. The door was made of timber and wire mesh.

Planting was done in circular plastic pots where every pot represented a plot, (Figure 2). Each pot was 75cm in diameter and 50cm deep and filled with black cotton soils before planting. The soil was collected from the MIS farm. It was disinfected by using furadan, active ingredient carbofuran. The soil was soaked with water and furadan for one week prior to planting. Soil was put in the pots up to 40cm level.

Each pot consisted of 22 rice plants (11 of *O. longistaminata* and 11 of *O. sativa*) mixed planting and 22 rice plants in pure planting at a spacing of 10cm between rows and 10cm between plants. Spacing between one block to the other was 50cm. The treatments were randomized within a block. There was synchronization during planting, where the planting material was at the same leaf stage. Plantlets from the wild/weed populations and seedlings from the cultivated populations were all planted at three to four leaf stages

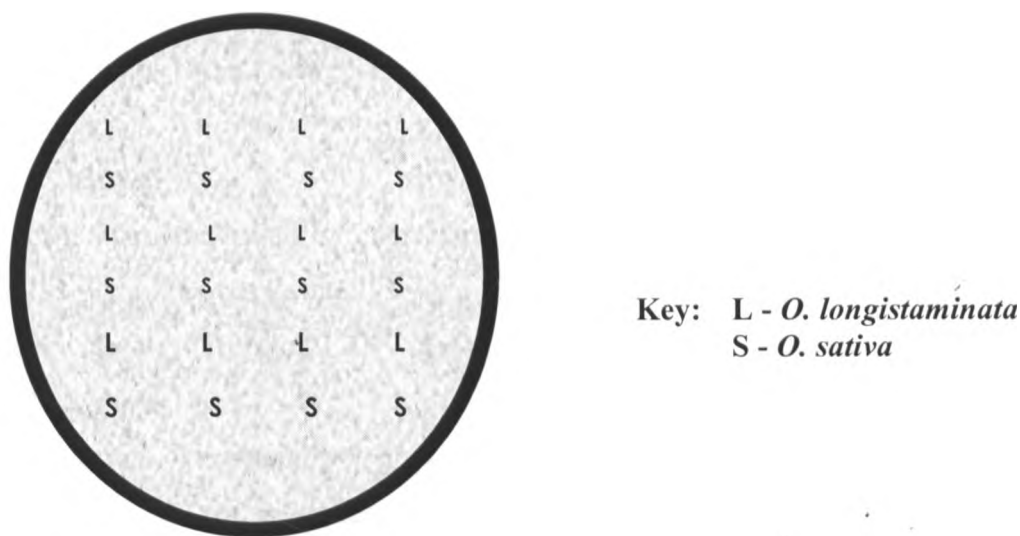


Figure 2: General layout of plantings in a pot showing alternating rows

3.4 Cultural practices in the screen house

Irrigation was done by flooding the pots with water to a depth of 3cm above the soil level during the first two weeks. This was increased up to 5 cm until the fourth week, then to 10 cm until the end of 6th week. The last level of water was up to 15cm which went on until 18th week when irrigation stopped and the water drained since the cultivated rice had shown signs

of physiological maturity. Weed control was done by hand pulling, where weeds were pulled by hands after every two weeks. The main pests were red spider mites, which were controlled by the chemical polyurethane (mitac). Blast disease struck during 4th week and was controlled by use of a fungicide goldazim with active ingredient as carbendazim 50%. Rice stemborers were controlled by use of fenitrothion and esfenvalerate (sumithion).

3.5 Sampling Procedure

Twelve plants were selected randomly from each plot. In the mixed plots, six *O. longistaminata* plants and six of *O. sativa* were selected. In the pure plots, twelve plants of the single crop were selected. These plants were tagged and used for data collection.

3.6 Morphology and growth measurements

3.6.1 Plant height

Height measurements were taken from the 7th day after transplanting. Height was measured by use of a metre rule (to the nearest cm). In young seedlings, height was measured from the base of the plant to the tip of the tallest leaf. In mature plants, height was measured from the base to the tip of the tallest panicle. The measurements were taken after every 7 days until 22nd week.

3.6.2 Tillering

Tillering was determined by counting the total number of tillers produced per hill on the selected plants above. Counting began on the 7th day after transplanting. The counting was done after every 7 days until 22nd week.

3.6.3 Leaf area

Leaf area was determination followed the methods of Yoshida (1981); by measuring the leaf length and width of the flag leaves in 4 plants per hill and calculating the average per plant. This was done at the maturity stage when the flag leaves were fully developed. Leaf areas were calculated using the following equation:-

$$\text{Leaf area (cm}^2\text{)} = k \times \text{length (L) cm} \times \text{Width (W) cm} \dots \dots \dots \text{Eq. 1}$$

Where k is a correction factor used in calculating leaf area in rice leaves (Yoshida, 1981). A correction factor (k) of 0.80 was used.

3.6.4 Reproductive stage characteristics

This is a stage characterized by increase in plant height, decline in tiller number and emergence of the flag leaf. These stages were determined by counting the total number of days to which each stage occurred after transplanting. These four stages were;

- Panicle initiation stage which occurs about 25 days before heading when the panicle has grown to about 1 mm long and can be recognized visually.
- Booting stage which is identified by the panicle bulging out before exertion.
- Heading stage which is the time when 50% of the panicles have exerted.
- Flowering (Anthesis) stage which begins with panicle exertion, where, within the same panicle it takes 7 - 10 days for all the spikelets to complete flowering. Fifty percent flowering occurs when half of the panicle has flowered and the grains start forming. This happens between one and six days after exertion.

3.7 Determination of standing crop biomass

The procedure of determining standing crop biomass by Hall *et al.* (1993) was followed. All vegetative material (alive and dead) within the selected samples was clipped at ground level at the end of 22 weeks. The material was dried to constant weight at 80°C in a well-ventilated oven. The materials were allowed to cool and then weighted on a sensitive electronic balance.

3.8 Data Analysis

Data were analyzed by SPSS version 13.0. Two way Analysis of Variance (ANOVA) was carried out on all data at 0.05 level of significance. The ANOVA was compounded for the whole growth period. New Duncans Multiple Range Test (DMRT) was used to test for the mean differences.

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION

4.1 Morphological characteristics

4.1.1 Height

There were some significant ($P < 0.05$) differences in plant heights between populations in both mixed and pure plantings and in all blocks. The heights of Pishori and Basmati plants were not significantly ($P > 0.05$) different. Mean separation on plant (Table 2), showed that Pishori, Basmati and Kipini populations did not differ significantly ($P > 0.05$) in height. The height for Ramisi population was significantly ($P < 0.05$) different from all other wild/weedy populations and cultivars. The plant heights of Tana Delta and Fioni populations were not significantly ($P > 0.05$) different from each other, but they were significantly ($P < 0.05$) different from the cultivars of Ramisi and the Kipini population. After transplanting there was gradual drop in height on all weed populations, especially the Kipini population (from 51.1 cm in the first week to 37.5 cm in the second week and from 43.1 cm in the first week to 32.9 cm in the second week in mixed plantings and in pure plantings respectively (see appendix 2)). Wild/weedy populations with rhizomes may have suffered transplanting shock more than the cultivars with fibrous root system. This is shown by the drop in height of all wild/weedy populations in both mixed and pure planting during Week 1 of growth except for the Tana Delta population which did not drop in height in both pure and mixed planting.

Table 2: Mean separation for plant heights (cm) for wild/weedy populations and the rice crop cultivars. Means followed by the same letter are not significantly different

Population	Mean Plant Height (cm)
Kipini	134.5a
Tana Delta	138.2a
Fioni	141.7a
Ramisi	176.2b
Basmati	180.4b
Phisori	193.1c

Cultivated rice populations (Basmati and pishori cultivars) continued growing normally. After transplanting, there was increase in height for Basmati plants (from 31.1 cm

in Week 1 to 39.3 cm in Week 2 and 26.5 cm in the Week 1 to 40.8 cm in the Week 2 in mixed and pure planting respectively). Pishori increased in height (from 36.2 cm in Week 1 to 45.9 cm in Week 2 in mixed planting and 34.9 cm in Week 1 to 46.3 cm in Week 2 in pure planting). This was due to their good root system and may be the adaptation to transplanting since rice is first planted in nurseries and later transplanted to the field (Ros *et al.*, 2002). Biologically it is expected that weedy rice plants recover faster than the rice cultivars having been planted from rhizome cuttings, but the root system may have a vital role to play. Das and Ahmed (1989) found under rain fed conditions that removing 98 to 100% of roots of rice seedlings increased percentage of hill mortality, compared to seedlings with intact roots. This effect is possible in wild/weedy rice populations since during transplanting, a large portion of the rhizome was cut. The reduction in growth resulted from the large drain in carbohydrate reserves for new root and rhizome re-growth and reduced water and nutrient uptake during the period when a new root is being established (Ros *et al.*, 2002). The wild rice population from Kipini may have been affected by the transplanting shock due to the early stage at which the rhizomes were obtained. Early shoot or rhizome cutting sufficiently suppress the growth of rice and also reduces the above- and below-ground biomass (Chandra and Tanaka, 2006). The seedling stage ends at Week 4 when a gradual increase in plants height was observed as the populations entered into the vegetative stage.

Results presented in Figures 3 and 4 show that, the populations were growing at almost the same rate except for the Kipini population that had a slower growth rate. This population took more time (18 days in both mixed and pure planting) to recover from the transplanting shock and this too affected the seedling vigour. During recovery of seedlings after transplanting, it does appear that the new root growth may have used more nutrient resources at the expense of the shoot (Ros *et al.*, 2002).

As the plants entered the reproductive stage, there was a slight drop in growth, because the plants had by then started utilizing some of the resources for reproduction. This was observed from Week 10. At Week 13, there was decrease in height from the cultivated populations. At Week 14, the cultivars had reached the reproductive stage while the wild/weeds on the other hand continued to increase in height because they had not attained the reproductive stage yet, hence the straight-line curve observed in Figures 3 and 4.

The height trend could also be dependent on where the populations grew initially. This could explain the close increase in height trend in Kipini population to the cultivars since it had been collected from a field where the two (Kipini and Pishori) had been growing together for a long time. Moreover, this could have been brought by the suppression of the young rhizomes that were collected from Tana Delta, Ramisi and Kipini, and perhaps were not adopted to the flooded soils. Flooded soils have high partial pressure of carbon dioxide (>5Kpa) (Greenway *et al.*, 2006). This infers higher partial pressure in the roots and a carbon dioxide diffusion gradient exists from the roots to the rhizomes and shoots. This could have reduced the growth rate of Kipini population plants (Greenway *et al.*, 2006).

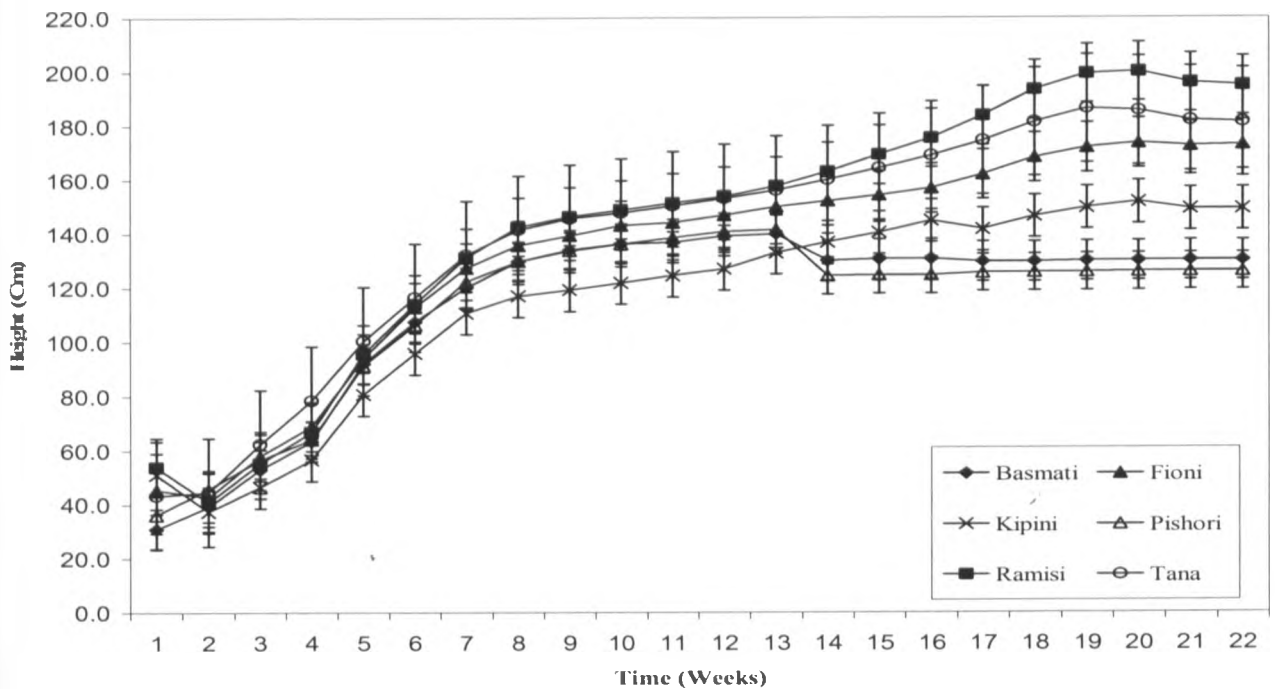


Figure 3: Trend in plant heights (cm) for four ecotypes of *O. longistaminata* and the two cultivars of *O. sativa* through out the growth period (Weeks) in mixed planting. Vertical bars indicate standard error of means

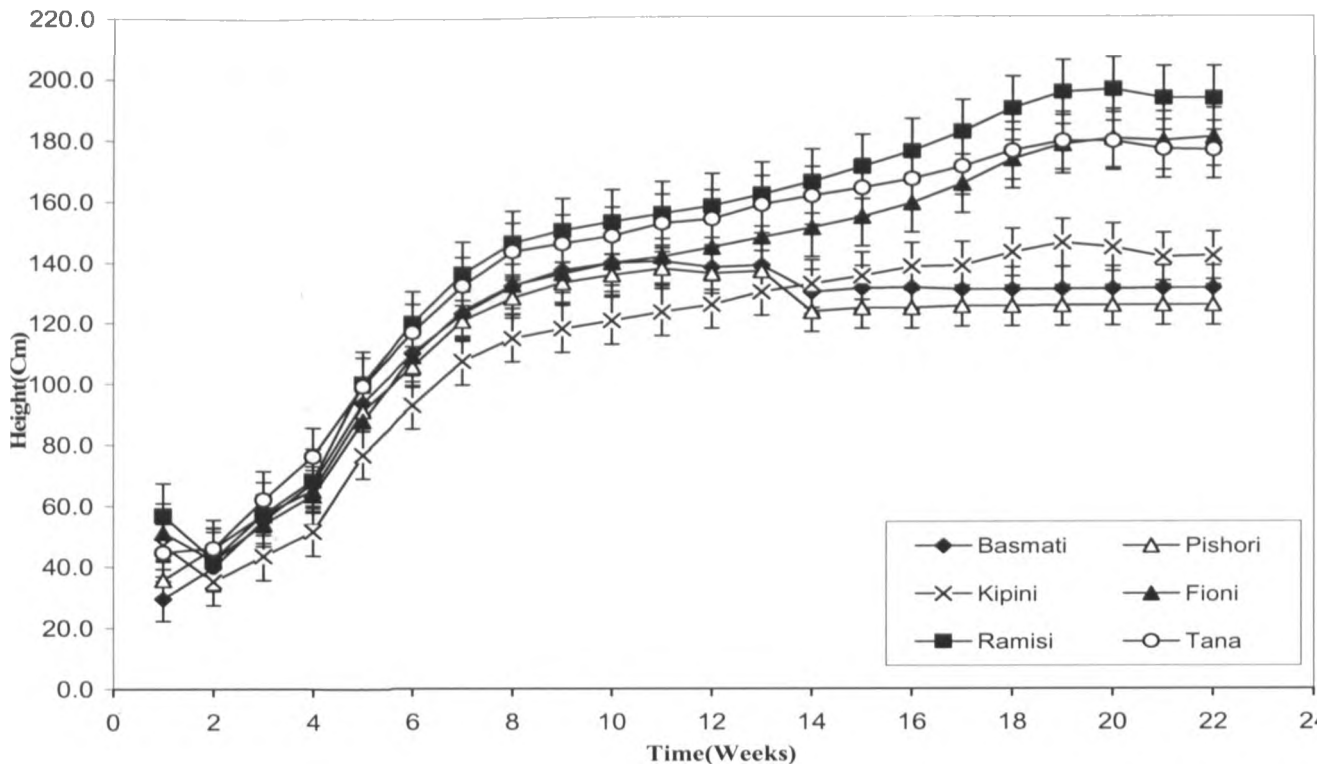


Figure 4: Trend in plant heights (cm) for four ecotypes of *O. longistaminata* and the two cultivars of *O. sativa* through out the growth period (Weeks) in pure planting. Vertical bars indicate standard error of means

As the plants entered vegetative stage (Week 4 to Week 8 for the cultivars and Week 4 to 22 for the wild/weedy populations) there was rapid increase in height. After the crop cultivars reached the reproduction stage (Week 9 to Week 13), they stopped increasing in height while the wild and weed populations continued to grow since they had not attained the reproduction stage. This could be explained by the fact that *O. longistaminata* is a perennial and so requires at least two seasons for it to reproduce. (<http://www.lucidcentral.org/keys/FNW/grasses/html/fact/sheets/Oryza.htm>, 2007).

In addition, the change in growth environment could have affected the wild/weedy populations growth and so it took longer time for them to acclimatize. As a result, the Kipini wild rice population became taller than the crop cultivars which had attained maximum height. This could be a character obtained from the interaction of the wild rice Kipini population with the cultivated Basmati, since Kipini rice population is weedy while Pishori and Basmati rice cultivars are cultivated.

The ecotypes and the cultivars had varied height characteristics, where, Ramisi population attained the greatest height while Pishori had the least height and Kipini population almost at the middle in both pure and mixed plantings. This implies that the significant ($P < 0.05$) differences in heights may have been caused by the differences within the wild/weedy populations and the cultivars. Also, competition for resources may have contributed to these differences.

The Kipini population could have mimicked some of the growth characteristics from the cultivar in which it was growing since there were no significant ($P > 0.05$) difference in height between it and the crop cultivars (of Pishori or Basmati). In addition, there could be hybridization between the population and the cultivars and the hybrids formed might have been inferior to their parents and this could explain the slower growth compared to that of the cultivar; (as Jørgensen *et al.*, (1996) reported that some rice hybrids have declined fitness than their parents).

4.1.2 Tillering

Tillering process differed significantly ($P < 0.05$) for Kipini population compared to all other wild/weedy populations and cultivars. The tiller number for the Basmati and Pishori plants did not differ significantly ($P > 0.05$). Ramisi, Fioni and Tana Delta populations also did not differ significantly ($P > 0.05$) in the number of tillers (Table 3). Tillering in all populations was slow at seedling stage (starting from Week 1 to Week 4) and increased steadily at vegetative stage (starting from Week 5 to Week 10) in both pure and mixed planting. Kipini population plants tillered more (7 tillers in mixed planting and 8 tillers in pure planting) during the vegetative stage than the other three wild populations but less than the rice crop cultivars (10 tillers in Basmati and 12 in Pishori in mixed planting, and 12 tillers in Basmati and 12 in Pishori in pure planting). Tillering appeared to have followed the same trend for all wild/weedy populations and crop cultivars with maximum tillering between Weeks 7 and 10 in both mixed and pure planting (Figures 5 and 6). The two cultivars had equal number of tillers (11) at maturity in pure planting.

The type of planting system did not significantly ($P > 0.05$) affect the number of tillers produced in both pure and mixed planting, Basmati had the highest number of tillers while Tana Delta population had the least number of tillers in both pure and mixed planting. This may be an indication that tillering is not affected by the interactions of the wild/weedy rice with the cultivated rice.

Table 3: Mean separation for tiller number for wild/weedy populations and the rice crop cultivars. Means followed by the same letter are not significantly different

Population	Mean Number of Tillers
Kipini	6a
Tana Delta	7a
Fioni	7a
Ramisi	8b
Basmati	12c
Phisori	13c

It would be expected that wild and weed populations would have higher tillering than the cultivated populations. However, crop cultivars had higher tillering than the wild and weed populations. This may be because rhizomes spread very fast within a non restricted area. The planting was done in pots and it was observed that the rhizomes coiled themselves round minimizing the area in which the tillers are produced. This made the cultivars which have fibrous root system to have an advantage and so they tillered more. These results are in agreement with Sacks *et al.*, (2006), who made similar observation. Tillering of the wild/weedy populations may also have been affected by the climatic conditions at Mwea, which were different from the ones at the coastal region of Kenya where the samples had been collected. Tillering was high during vegetative stage when maximum rate of tillering occurred in all populations. This is a stage that occurs in preparation of the reproductive stage in grasses in order to obtain maximum yields. The more tillers a rice plant produces, the higher the yields (Yoshida, 1981 and Awan *et al.*, 2007). Tana wild rice population had the least tillering (6 tillers in mixed planting and 7 tillers in pure planting). However, this population was severely infected with blast disease at Week 6. Due to this, the population general growth became slow and so its general tillering was less than the one in other wild populations in both mixed and pure planting (Figures 5 and 6). Kipini population had an average number of 8 tillers, and the tiller growth closely resembled the one for the crop cultivars up to Week 6 and for the wild/weedy populations (Figures 5 and 6). It is possible that mimicking on the crop cultivars had occurred on growth during the seedling stage which could not be expressed wholly by the tillering ability.

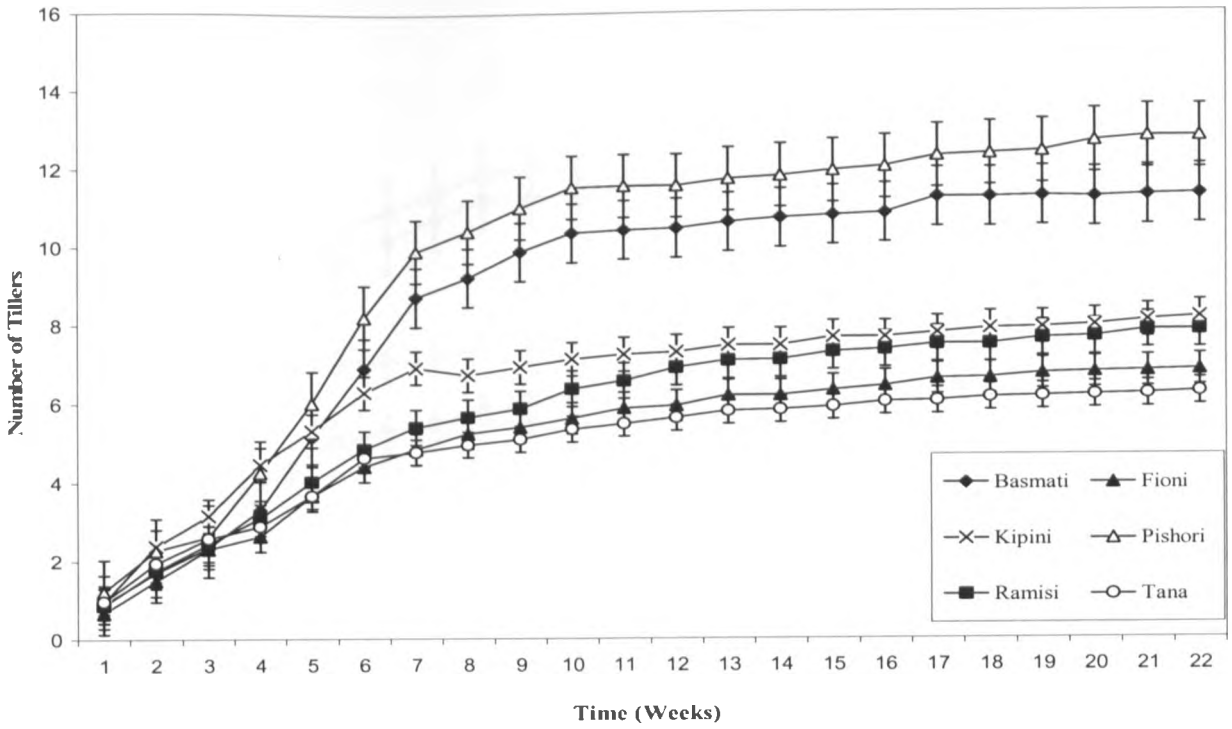


Figure 5: Trend in tillering ability for four ecotypes of *O. longistaminata* and the two cultivars of *O. sativa* at different time (Weeks) in mixed planting. Vertical bars indicate standard error of means

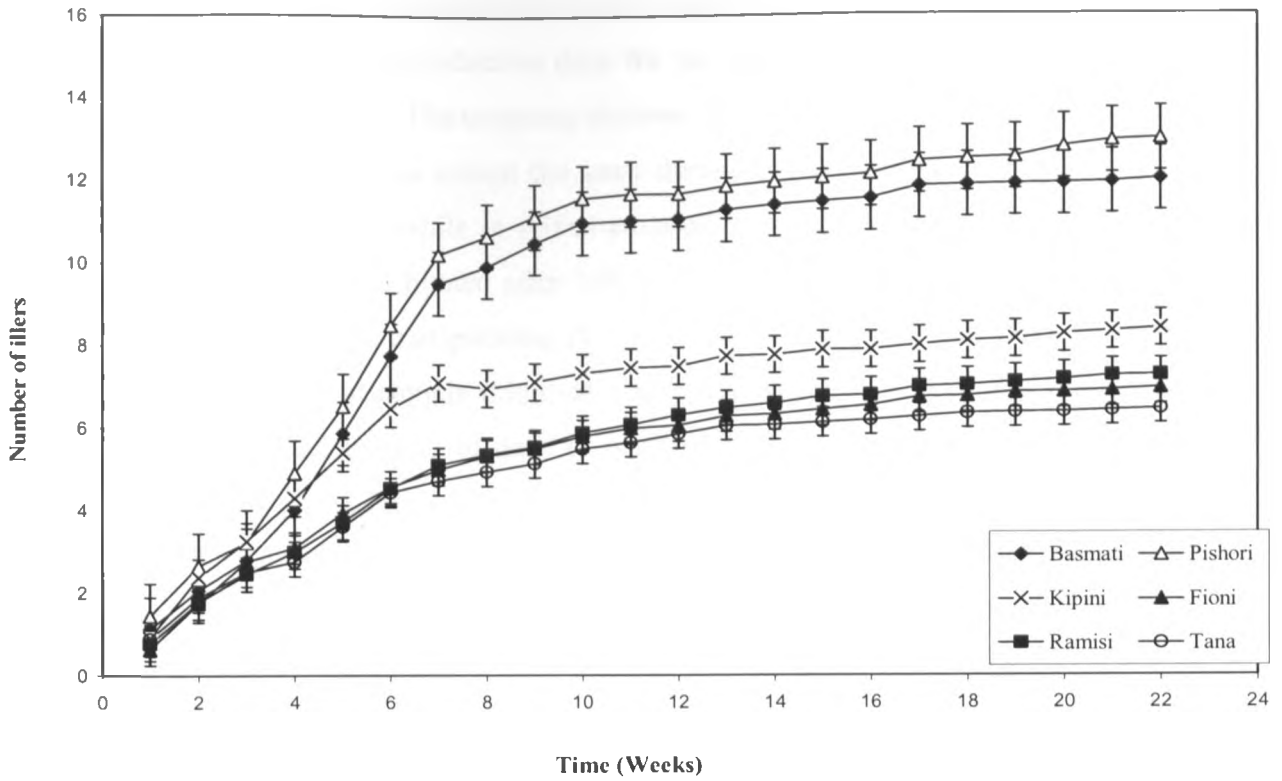


Figure 6: Trend in tillering ability for four ecotypes of *O. longistaminata* and the two cultivars of *O. sativa* at different time (Weeks) in pure planting. Vertical bars indicate standard error of means

4.1.3 Reproductive stages

The comparisons during reproduction were made between Basmati and Pishori alone as shown in table 4. The plants reproductive days for the two cultivars were not significantly ($P>0.05$) different in all stages. The cropping systems did not differ significantly ($P>0.05$) in all stages. The two cultivars took almost the same duration (103 days after transplanting) to booting stage in pure planting while in mixed planting, Basmati took more time (105 days after transplanting) and Pishori booted after 103 days (Table 4). At panicle initiation stage, Basmati took 103 days after transplanting (DAT) while Pishori took 102 DAT in mixed planting. In pure planting, at panicle initiation stage Basmati took 99 DAT and Pishori 102 DAT. Pishori took equal number of days to panicle initiation and booting stages in both plantings. At heading stage, Pishori took 109 DAT in mixed planting and 118 days in pure planting while Basmati took 111 DAT in mixed planting and 109 DAT in pure planting. At 50% flowering, Pishori and Basmati took almost the same time (115 and 116 DAT respectively) in mixed planting and in pure planting Basmati and Pishori took 117 and 124 days respectively.

Table4: Days after transplanting to end of panicle initiation, booting, heading and 50% flowering in mixed and pure planting populations

		Plant Growth Stage			
Type of Planting	Plant Population	Panicle Initiation	Booting	Heading	50% Flowering
		Number of Days after Transplanting			
Mixed Planting	Basmati	103	105	111	115
	Phisori	102	103	109	116
Pure Planting	Basmati	99	103	109	117
	Phisori	102	103	118	124

Rice takes about 10 to 20 days from panicle initiation to 50% flowering depending on the cultivar and the environment or climate in which it is growing (Fukai 1999). In mixed planting, Basmati and Pishori rice plants took 12 and 14 days respectively while in pure planting Basmati took 18 days and Pishori 22 days from panicle initiation to 50% flowering which is in close agreement with the findings of Fukai (1999), who found that most of the cultivars took 10 to 28 days from panicle initiation to flowering depending on the air and water temperature, drought tolerance and photo period they were exposed to. Basmati matured earlier than Pishori. This would be the desirable cultivar since farmers would like having a fast maturing variety.

The interaction with the wild/weedy populations did not affect all the reproductive stages. This may have happened because the research was done for only one season and it may not have been sufficient to detect the change in reproduction cycle of the cultivated and/or the wild/weedy *Oryza* species when they interact over a length of time. In addition, the wild/weedy populations (Ramisi, Fioni Tana and Kipini) did not attain reproductive stage of their growth and as such, there was no synchronization of the reproductive stage by the wild/weedy populations and the cultivars.

4.1.4 Flag Leaf Area

The planting system affected flag leaf areas of the two cultivars significantly ($P < 0.05$). Basmati had significantly ($P < 0.05$) higher mean flag leaf area (54.03cm^2) in pure planting and lower flag leaf area (51.07cm^2) in mixed planting while Pishori had a higher mean flag leaf area (55.35cm^2) in mixed planting and lower area (45.62cm^2) in pure planting (Table 5). Flag leaf area affects the production of above ground biomass and grain yields in rice. The higher the flag leaf area, the higher the grain yields (Li *et al.*, 2003). Since Basmati is the improved cultivar, it is expected that it would have a higher leaf area than Pishori, which is a landrace. Pishori has no improvement made on it and as such it is a better performer on mixed planting than Basmati. It may be closer to the wild type than to the cultivars, since it had been growing close to them or with them for a long time.

Table 5: Flag leaf mean area (cm^2) in Basmati and Phisori rice in mixed and pure planting populations

	Mixed Planting		Pure Planting	
	Basmati	Phisori	Basmati	Phisori
Mean Area (cm^2)	51.1	55.4	54.0	56.0
SD	12.02	16.6	11.4	9.9

4.1.5 Other morphological characteristics

There was no formation of grain and so no grain characteristics were recorded. There was a drastic change in climatic conditions and it turned too cold in the month of July when the cultivated rice was at reproductive stage making it hard for grain formation. In rice production, there is a comparatively cool optimum temperature as rice progress towards panicle initiation (only $24 - 26^\circ\text{C}$) in many cultivars, which was not obtained during this stage of development in the month of July when the mean temperatures in the screen house was averaged 22°C . Low temperatures are a major restriction on rice cultivation, and thus temperature is a major concern in areas at high latitudes or high altitudes (Shimono *et al.*, 2007). This is a major constraint in rice production at Mwea Irrigation Scheme, because of this, there is no rice in production at Mwea Irrigation Scheme fields during the months of July and August. Low temperatures cause damage to the male organ (pollen) rather than to the female organ (stigma) causing sterility. A shortage of sound pollen number at flowering stage limits pollination and this result in sterility (Plate 1).



Plate 1: Panicles of rice cultivars with empty spikes due to the effect of low temperatures

Rice panicles with empty spikes

During panicle development, the susceptibility of the male organ to low temperatures is extremely high during the booting stage of microspore formation, which occurs approximately 10 days before heading; low temperatures interfere with sound division of pollen mother cells (Shimono *et al.*, 2007 and Fukai, 1999).

Another problem encountered was attack by blast rice disease (Plate 2). The attack was very severe since this was during off-season and being the only crop around the fields, it was very difficult to control the disease. The disease was controlled by use of fungicide goldazim.



Plate 2: Brown parts of rice plants showing infected plants with blast disease in the screen house

this rice blast is caused by a fungus (*Pyricularia oryzae*). Its symptoms include, spots or lesions on leaf node and panicles, spots are elongated and spindle shaped, no grain fill and in severe cases the plant dies. The development of blast is favoured by high fertilizer level, cloudy sky and high humidity (Wanjogu *et al.*, 1995). This was perfect time for it since the weather at that time was cloudy with very high humidity inside the screen house.

4.2 Above Ground Biomass

Cropping system affected biomass production significantly at $P < 0.05$. Ramisi population differed ($P < 0.050$) from all other populations and cultivars. Kipini, Fioni and Tana Delta populations were not significantly ($P > 0.05$) different from each other as shown in Table 6. The biomass of the two cultivars was not significantly ($P > 0.05$) different. Cultivars had higher biomass than the wild and weedy populations (Figure 7). The cultivars had higher biomass than the wild/weedy populations since they had more tillers as observed in Figures 3 and 4. The more tillers a rice plant has, the higher the biomass (Awan *et al.*, 2007) and the larger the rice hill. This is in agreement with a study carried out by Li *et al.* (2003) where they controlled tillering in rice and found that the rice plants with the least number of tillers had the least biomass but the largest grain and greatest leaf area.

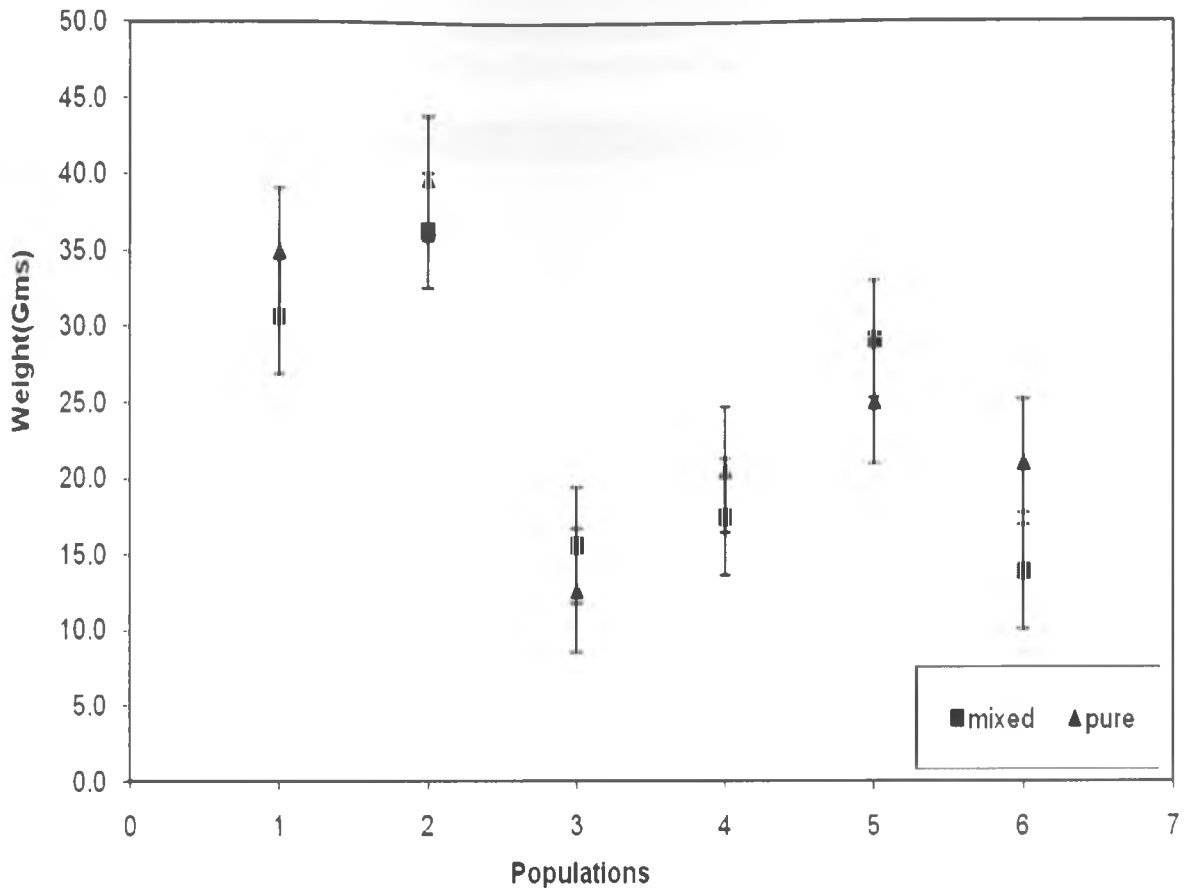


Figure 7: Difference in weight (gm) for sample biomass for all populations (1-Basmati, 2-Pishori, 3-Kipini, 4-Fioni, 5-Ramisi, 6-Tana - Delta) in mixed and pure planting. The vertical bars are standard error of means

Prolonged vegetative growth duration and higher plant heights in the wild populations was expected to result in greater biomass production as was observed by Shao-Bo *et al.* (2005), but this was out done by the number of tillers. Rice plants in pure planting had a significantly ($P < 0.05$) higher biomass than the ones in mixed planting. This may have been caused by competition for resources and also the change in climatic conditions especially for the ecotypes which were collected from the coastal region of Kenya.

Table 6: Mean separation for above-ground biomass (gm) for wild/weedy populations and the rice crop cultivars. Means followed by the same letter are not significantly different

Population	Mean Plant Biomass (gm)
Kipini	14.08a
Tana Delta	17.41a
Fioni	18.94a
Ramisi	27.05b
Basmati	32.11c
Phisori	37.45c

CHAPTER FIVE

5.0 CONCLUSIONS AND RECOMMENDATIONS

5.1 conclusions

Rice morphological characteristics showed no difference between the landrace cultivar (Pishori) and the improved cultivar (Basmati) in both mixed and pure planting apart from the flag leaf area. This likely happened because most improvement in rice is made to increase yield and disease resistance and the flag leaf area is a major determinant of yields in rice. The wild/weedy populations differed significantly in their morphological characteristics with the Kipini population which had been collected in abandoned rice fields showing great difference from all other populations. The Kipini population showed some similarities with the cultivars. Ramisi population showed characteristics which were between wild/weedy populations and the cultivars. Since Ramisi population was collected near the landrace rice fields, it can be concluded that it was in the process of mimicking some of the characteristics from the cultivars, which were not expressed wholly by morphological features. During the growth stage in rice the main difference was during physiological maturity where none of the wild/weedy populations attained this stage. The planting system, affected the morphological characteristics, where plants in mixed planting had lower plant height, less tillers and lower above ground biomass.

Above ground biomass of cultivars is higher than the one for wild/weedy populations. Generally, there were differences in morphology, phenology and growth of wild/weedy populations of rice from that of the cultivars, whether grown in pure or mixed plantings. The above ground biomass between wild/weedy populations and cultivars too were different.

5.2 RECOMMENDATIONS

1. Planting in pure stands is recommended for the plants to be able to express their morphological characteristics fully. This will make it easier to distinguish a character from, an example, nutrient deficiency or disease attack.
2. Morphology and phenology alone are not enough to study the mimicking characteristics and other methods should be used, for example, genetic markers, AFLP and SSR, which can detect a slight resemblance in genetic make up of the plant.
3. Experiment should be carried out in the areas where the wild ones grow naturally. This will reduce the time for the plant to acclimatize.

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APPENDICES

Appendix 1: Treatment combinations of Kipini, Ramisi, Fioni and Tana-Delta populations with Basmati and Pishori (cultivars)

Wild/Weed ⇒	Kipini (K)	Ramisi (R)	Fioni (F)	Tana-Delta (T)
Crop Cultivar ↓				
Basmati (B)	Kipini-Basmati (KB)	Ramisi-Basmati (RB)	Fioni-Basmati (FB)	Tana-Delta-Basmati (TB)
Pishori (P)	Kipini- Pishori (KP)	Ramisi-Pishori (RP)	Fioni- Pishori (FP)	Tana-Delta-Pishori (TP)

Appendix 2: Mean height (Cm) of all populations at different growth time (Weeks) in mixed planting

Population/Height(cm)						
week	Basmati	Fioni	Kipini	Pishori	Ramisi	Tana
1	31.1	45.6	51.1	36.2	53.9	43.4
2	39.3	42.8	37.5	45.9	41.0	44.6
3	52.9	57.9	46.6	56.5	55.3	62.4
4	63.4	68.9	56.6	64.0	66.8	78.4
5	92.1	93.9	80.8	91.3	95.6	100.4
6	107.6	112.9	95.9	106.3	114.1	116.5
7	120.2	127.5	110.8	122.4	131.3	132.3
8	129.9	135.8	117.2	129.8	142.7	141.6
9	134.1	139.4	119.4	133.8	146.6	145.7
10	136.5	143.2	122.0	136.3	149.0	148.0
11	136.8	144.3	124.7	138.6	151.6	150.6
12	139.2	147.0	127.2	141.0	154.0	153.2
13	140.1	150.2	133.1	141.7	157.8	156.3
14	130.2	152.3	137.0	124.6	163.2	160.2
15	130.7	154.5	140.7	124.8	169.4	164.5
16	130.9	157.0	145.1	124.8	175.5	169.1
17	129.9	162.2	141.8	125.8	184.0	174.7
18	129.9	168.4	146.7	125.9	193.4	181.5
19	130.2	172.1	149.9	126.0	199.3	186.4
20	130.4	173.8	152.0	126.2	200.1	185.7
21	130.5	172.6	149.4	126.3	196.1	182.1
22	130.5	173.1	149.5	126.3	194.9	181.4

Appendix 3: Mean height (Cm) of all populations at different growth time (Weeks) in pure planting

Population/Height(cm)						
Week	Basmati	Fioni	Kipini	Pishori	Ramisi	Tana
1	26.5	57.0	43.1	34.9	59.9	46.1
2	40.8	41.2	32.9	46.3	43.9	47.5
3	59.7	50.3	40.5	58.2	59.3	61.6
4	75.2	57.9	46.4	67.1	69.7	73.9
5	97.1	81.8	72.7	91.0	104.8	98.2
6	114.8	105.1	90.0	104.2	125.8	117.3
7	128.9	120.5	103.9	117.3	141.2	132.2
8	136.3	129.0	112.5	125.1	149.8	145.3
9	143.4	132.9	116.5	131.9	153.9	146.1
10	145.8	135.9	119.1	134.3	156.9	148.7
11	146.9	138.8	122.0	135.8	159.6	154.5
12	135.6	142.0	124.5	126.2	162.3	154.6
13	135.9	145.6	126.9	126.9	166.0	160.8
14	129.4	149.6	128.2	121.3	168.9	162.6
15	132.1	154.5	129.6	124.1	172.2	163.4
16	132.1	160.9	131.3	124.1	176.3	164.6
17	132.1	168.3	135.1	124.1	180.5	167.2
18	132.1	178.1	138.9	124.1	186.3	170.4
19	132.2	184.1	142.1	124.1	191.0	171.9
20	132.2	186.3	136.9	124.1	192.1	172.5
21	132.3	185.9	133.4	124.1	190.3	171.0
22	132.3	187.6	134.0	124.1	191.3	170.9

Appendix 4: Mean tillers (Number) for all populations at different time of growth (Weeks) in mixed planting

Population/Number of tillers						
week	Basmati	Fioni	Kipini	Pishori	Ramisi	Tana
1	1	1	1	1	1	1
2	2	2	2	2	2	2
3	2	2	3	3	2	3
4	3	3	4	4	3	3
5	5	4	5	6	4	4
6	7	4	6	8	5	5
7	9	5	7	10	5	5
8	9	5	7	10	6	5
9	10	5	7	11	6	5
10	10	6	7	12	6	5
11	10	6	7	12	7	6
12	10	6	7	12	7	6
13	11	6	7	12	7	6
14	11	6	7	12	7	6
15	11	6	8	12	7	6
16	11	6	8	12	7	6
17	11	7	8	12	8	6
18	11	7	8	12	8	6
19	11	7	8	12	8	6
20	11	7	8	13	8	6
21	11	7	8	13	8	6
22	11	7	8	13	8	6

Appendix 5: Mean tillers (Number) for all populations at different time of growth (Weeks) in pure planting

Population/Number of tillers						
week	Basmati	Fioni	Kipini	Pishori	Ramisi	Tana
1	2	1	1	2	1	1
2	3	2	2	3	2	2
3	4	3	3	4	2	2
4	5	4	4	6	3	3
5	7	4	5	8	3	4
6	9	5	7	9	4	4
7	11	5	7	11	5	5
8	11	5	7	11	5	5
9	12	6	7	11	5	5
10	12	6	8	12	5	6
11	12	6	8	12	6	6
12	12	6	8	12	6	6
13	13	6	8	12	6	6
14	13	6	8	12	6	6
15	13	7	8	12	6	6
16	13	7	8	12	6	6
17	13	7	8	13	6	6
18	13	7	8	13	7	7
19	13	7	8	13	7	7
20	13	7	9	13	7	7
21	13	7	9	13	7	7
22	13	7	9	13	7	7

Appendix 6: Mean monthly weather measurements for MIAD from January, 2007 to August, 2007

Month	Rainfall(mm)	Temperature(°C)	RH (%)
Jan-07	30.00	21.33	80.12
Feb-07	0.00	22.57	66.90
Mar-07	85.30	24.16	65.16
Apr-07	249.50	23.66	75.97
May-07	121.10	23.05	76.45
Jun-07	6.50	21.72	72.17
Jul-07	20.20	21.74	72.58
Aug-07	9.20	21.60	69.07