VARIETIES FOR RESISTANCE TO LATE BLIGHT,

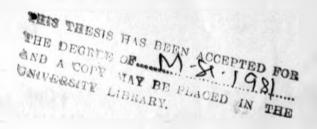
PHYTOPHTHORA INFESTANS (MONT.) DE BARY

BY

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of



MASTER OF SCIENCE

IN

- PLANT BREEDING

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FACULTY OF AGRICULTURE

DECLARATION

This thesis is my original work and has not been presented for a degree in any other University.

This thesis has been submitted for examination with our approval as University supervisors.

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ABSTRACT

Late blight caused by Phytophthora infestans

(Mont.) de Bary is one of the most destructive diseases which limits the production of potatoes in Kenya.

Most of the potatoes are produced by small scale farmers who cannot afford to spray their crops effectively due to the high costs involved. Therefore, screening and breeding of clones to develop resistant varieties is necessary to avoid the expensive chemical method of control.

Initial screening consisted of field and laboratory testing of 226 clones followed by yield evaluation of selected clones at Tigoni, N.A.L., Nairobi and Molo.

The laboratory screening included physical, anatomical and biochemical tests. The physical test consisted of inoculating whole plants and detached leaflets artificially and estimating the percentage leaf area infected, period of infection and sporulation, lesion size and number of sporangia produced. Results indicated that laboratory tests were more severe than field tests in separating the resistant lines from susceptible ones. The laboratory tests also indicated that some of the clones had a higher level of horizontal resistance than others while some appeared to have oligogenic resistance.

The anatomical test consisted of microtome sectioning of leaves and stems and measuring the thickness of the epidermal cells. There was no correlation between the degree of resistance and the thickness of these cells.

The biochemical test involved determination of total phenolic compounds in healthy leaves colorimetrically by the "Folin - Ciocalteu" reagent. Results showed a positive correlation between the level of resistance and the amount of phenolics determined.

Results of tuber inoculation indicated that in some clones there was a correlation between haulm resistance and tuber resistance while other clones did not show this correlation. Further tests involved nutritional studies where crude protein content, dry matter content, and cooking test revealed that potatoes could be a good source of proteins apart from supplying carbohydrates. The texture and flavour of the boiled potatoes depended on the dry matter content as well as soil conditions.

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CHAPTER 1

INTRODUCTION

Potatoes have been grown in Kenya for a long time but have received little attention from plant breeders. Potatoes were introduced into Kenya in the second half of the 19th Century by European settlers and were grown first in the Kenya Highlands between 1,600m to 3,000m, above sea level. Between 1917 and 1918 the crop was introduced into small scale farming especially among the African farmers who grew it for consumption and later on for export to neighbouring countries.

About 1940 the Government decided to bulk import potatoes for the British Army and it is believed that late blight caused by Phytophthora infestans (Mont.) de Bary was introduced then. Many varieties were imported from Scotland, Holland and West Germany for trial and most of them succumbed to diseases. Some of the early introductions were King George, White City, Rayners Park, Incomer, Kerr's Pink, Arran Comrade and B 53. The Plant Pathology section of the National Agricultural Laboratories, Nairobi, tested blight resistant introductions and recommendations of copper fungicides were given as a means to control blight.

About 1956, Black of the Scottish Plant Breeding Station

supplied selections for testing. In 1958, a large number of varieties were imported from Scotland, Holland and Ireland but most of them were wiped out by blight. The year 1961 marked the beginning of potato breeding in Kenya. Screening of seedlings from berry seed started in 1963. The main aim of the breeding project was to select wilt and blight resistant clones (Waithaka, 1976; Njoroge, 1978).

In Kenya the bulk of the potatoes are produced on small scale farms at altitudes ranging from 1,600 to 2,700m. These are the areas which are threatened by heavy blight damage during the growing season.

Despite the advances made in chemical methods of disease control, small scale farmers cannot depend on this method because of the high costs involved. Growing of resistant varieties is the most economical means of controlling the blight.

The current popular varieties grown in Kenya are Kenya Baraka, Roslin Tana, Roslin Gucha (these were selected in Kenya) Kerr's Pink, Roslin Eburu, (introduced from Scotland) Anett, Desiree, Maritta and Feldeslohn (introduced from West Germany between 1965 - 1966). Roslin Eburu and Anett are still resistant to late blight but Kenya Baraka, Roslin Tana, Roslin Gucha seem to be losing their resistance. Kerr's Pink,

Desiree, Feldeslohn and Maritta are very susceptible to late blight but they are popular because of other characteristics such as skin and flesh colour and their palatability. The other major defect of all these varieties is their susceptibility to Potato Leaf Roll virus. There is need for continued study to develop resistant varieties.

The objective of this study therefore is to screen and select clones with desirable level of resistance to late blight under field and laboratory conditions and with acceptable yield potential. The promising clones could be either released as varieties or their resistance incorporated into other clones and varieties in the breeding programmes.

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CHAPTER 2

REVIEW OF LITERATURE

2.1. Breeding of Potatoes

Potatoes introduced to Europe from Latin America were adapted to short day conditions. To improve productivity, new varieties adapted to long day conditions had to be developed by suitable breeding methods. Potato breeding in Europe started as early as the end of the 16th Century (Howard, 1970). Initially, breeding consisted of raising seedlings from naturally set berries but later, hybridization of chosen varieties was necessary to increase genetic variability. Breeding of new varieties in Europe was accelerated by the blight epidemic of 1845 and by the realization of the importance of immunity to wart disease caused by Synchytrium endobioticum (Toxopeus, 1956; Howard 1970). The breeding methods such as the pedigree method, mass selection and in some cases haploids of Solanum tuberosum have been used (Peloquin et al. 1966; Abdalla, 1970; Maine, 1978).

2.2. <u>Screening Techniques</u>

Screening techniques which have been used to test for resistance to late blight are:-

(i) Progeny testing of whole plants and detached leaflets

Progeny testing of whole plants can be done both in the laboratory and field. After hybridization botanical seeds are sown and four to six weeks old seedlings inoculated with known races under controlled conditions. Seven days after inoculation, plants are examined and rated on a 0-5 scale depending on the amount of damage incurred. The survivors are later tested under field conditions (Mastenbrock, 1953, Black and Gallegly, 1957, Black, 1972, Poehlman et al. 1972, Anonymous 1977, Malcolmson and Killick, 1980).

Field testing of the progeny is normally done over a period of years and at a number of locations. A known number of plants of each clone are planted and haulm defoliation is assessed by a quick overall visual estimate of the percentage of total haulm that has been killed by blight. No fungicides are applied (Lapwood, 1961; Thurston et al.,(1962).

Malcolmson and Stewart (1974) reported that
the old practice of expressing a clone foliage
reaction to blight as a single figure score has been
replaced by a more informative composite record of

reaction of both leaves and stems.

Detached leaflets and leaf discs are inoculated with either a sporangial or zoospore suspension of P. infestans by dipping small filter paper discs into the suspension and placing the discs on the leaflet and incubating in a moist chamber.

Resistance to infection, lesion size, rate and amount of sporulation are used as parameters for separating resistant and susceptible clones (Van der Zaag, 1959; Lapwood, 1961; Hodgson, 1962; Thurston et al., 1962; Knutson, 1962; Simmonds and Malcolmson, 1967; Black, 1972, Poehlman and Dhirendranath, 1972; Maine, 1978).

(ii) Anatomical and biochemical bases of resistance

In later years, the level of resistance was found to be associated with the anatomical differences and biochemical aspects of the plant.

Enzyme activity, for instance peroxidase and the accumulation of phytoalexins after wounding or infection with P. infestans have been used in determining the degree of resistance in potato plants (Umaerus, 1959; Sakai et al., 1964; Sakai and Tomiyama, 1964; Goodman et al., 1967; Clarke, 1969; Schöber, 1971; Currier and Kuc, 1975; Kuc

et al., 1976, Deverall, 1976, Albersheim, 1976).

Enzyme activity and the accumulation of phytoalexins have been found to be higher in resistant than in susceptible varieties.

(iii) Selection based on Tuber Index

Tubers have also been used as a criterion for selection. Plants with long stolons and long tubers at the seedling stage are discarded. Halved, whole and tuber slices are inoculated with P. infestans and the mycelial spread in the tissue assessed by measuring the rate of the discolouration of the tuber tissue. This and the zoospore production are used to differentiate between the resistant and the susceptible material (Hodgson, 1962; Lapwood, 1965; Howard, 1970; Woodward et al., 1975).

2.3. Genetics of blight resistance in potatoes

Resistance to late blight caused by Phytophthora
infestans (Mont.) de Bary occurs either in the foliage
or tubers (Howard, 1970; Abdalla, 1970; Black, 1970, 1972; Van der Plank, 1963, 1975; Poehlman and Dhirendranath, 1972). Tuber resistance is very important although it has been neglected by breeders. Resistance in the foliage is of two types, vertical and horizontal resistance (Van der Plank, 1963).

2.3.1. Vertical resistance

Vertical resistance is also known as hypersensitivity, differential or race specific resistance and is monogenically or oligogenically inherited. It is characterized by an interaction between the host and pathogen. This concept was introduced by Flor in 1942 when he was studying the genetics of flax and flax rust (Melampsora lini). He found that the host and the parasite possess complimentary genetic systems referred to as 'gene-for-gene hypothesis'. Resistance in the host shows only when there is a corresponding locus in the pathogen carrying an allele for avirulence. But if a corresponding locus in the pathogen carries a virulence allele, the resistance allele can not express itself (Black, 1953, Toxopeus, 1956; Flor, 1971; Robinson, 1973; Roane, 1973; Van der Plank, 1975; Parlevliet and Zadoks, 1977). Race 1 of P. infestans attacks potatoes with gene R₁ for resistance but not those with gene R₂ whereas Race 2 will attack genotypes with gene R₂ and not R₁. Similarly varieties with gene R₁, R₃, R₄ are susceptible to all races with 1, 3 and 4, respectively. This applies to haulm resistance but not tuber resistance.

Tubers can be attacked by races that cannot attack the foliage (Van der Plank, 1963; Talburt and

Smith, 1967, Abdalla, 1970). Black, as reported by Roane (1973) employed the gene for gene concept without reference to Flor's work to explain the relationship between R-genes of potato and races of P. infestans.

This relationship was referred to as vertical resistance and only major genes were assumed to operate within a gene-for-gene system (Black, 1953, Mastenbrock, 1953, Lapwood, 1961, Van der Plank, 1963, 1975, Abdalla, 1970, Roane, 1973, Favret et al., 1977). Parlevliet and Zadoks (1977) stated that there was no reason why minor genes in the host could not operate in a gene-for-gene way with minor genes in the pathogen.

Specific resistance to P. infestans was discovered in a wild Mexican hexaploid species Solanum demissum in the twenties (Toxopeus, 1956, Lapwood, 1961, Roane, 1973). This species was used to transfer R-genes governing major gene resistance to S. tuberosum by repeated backcrossing with commercial varieties. The transfer of at least one of these R-genes to hybrids with S. tuberosum was the basis for breeding hypersensitive varieties (Toxopeus, 1956, Lapwood, 1961).

The <u>S</u>. <u>demissum</u> derivatives appeared entirely immune from attack but this immunity did not last long.

The immune varieties succumbed to successively arising specialized races of P. infestans soon after their introduction (Black, 1953; Toxopeus, 1956; Umaerus, 1959; Lapwood, 1961; Van der Plank, 1963). Vertical resistance was therefore found to be unstable. P. infestans was found to be an easily mutable pathogen producing new races very quickly. According to Robinson (1973) vertical resistance involves mechanisms which are within the pathogens capacity to change. When this change occurs, the effectiveness of the resistance breaks down. According to Lapwood (1961) and Thurston (1971), Niederhauser et al. (1954) reported that no clones of S. demissum were free from the disease when they tested clones and varieties which were resistant to the fungus in test plots in Mexico. A sexual stage which had not been reported anywhere else in the world was reported in Mexico by Gallegly and Galindo (1957). The existence of the two sexually different strains rendered possible new combinations of existing genes. Elsewhere in the world, variation was thought to have arisen asexually (Toxopeus, 1956).

Analysing Flor's gene-for-gene hypothesis,

Van der Plank (1975) stated that the concept only

identified genes but did not reveal the gene qualities

whether useful to a breeder or not. This led to a second gene-for-gene hypothesis which deals with gene quality. This hypothesis explains the usefulness of a gene for resistance in the host in relationship with the matching virulence gene in the parasite. instance, the resistance genes R_1 and R_4 in potato differ in their usefulness. R_A has not successfully been used on its own except in combination with other R-genes whereas R₁ has been used successfully and has protected potato varieties against blight. difference between the two is that additional virulence on R₄ pre-existed abundantly in <u>P</u>. <u>infestans</u> populations whereas virulence on R₁ did not (Abdalla, 1970). Further examples are gene Sr_{ς} and $\mathrm{Sr}_{\widetilde{\mathsf{h}}}$ in wheat. former is ineffective on its own compared to the latter.

If resistance in plants can be divided into true resistance and pseudo or escape resistance, vertical resistance will fall under true resistance since this occurs only after intimate contact between the host tissue and the parasite. The resistance genes act in a gene-for-gene way with virulence genes in the parasite (Parlevliet, 1977). In the case of vertical resistance, the true resistance will be monogenic.

Nelson (1978) stated that Van der Plank's definition of vertical resistance is confusing. It should have been defined as a resistance that reduces the amount of effective initial inoculum.

Breeders realized that with the wide distribution of many of the unknown fungal races and the possibility of new races appearing, hypersensitivity based on single or multiple R-genes was inadequate protection against the disease. Emphasis in breeding for resistance to potato late blight began to shift from R-genes to polygenically inherited or field resistance to obtain a wider spectrum of resistance (Black, 1953; Umaerus, 1959; Lapwood, 1961; Driver, 1962; Thurston, 1971; Caten, 1974; Favret et al., 1977; Malcolmson and Killick, 1980).

2.3.2. Horizontal resistance

Horizontal resistance also known as uniform, race-nonspecific, partial, or field resistance is known to be controlled by a series of minor genes polygenically inherited. The minor genes involved supplement each other in the control of the disease. In individual plants, they determine:-

- (a) the degree of resistance to infection
- (b) the rate of spread after infection
- (c) the time required for sporulation to begin
- (d) the number of spores produced per unit leaf area.

In a resistant variety, the lesions formed will be fewer and smaller. more time will be required for sporulation to begin and few spores will be formed (Van der Zaag, 1959; Driver, 1962; Knutson, Thurston et al., 1962; Hodgson, 1962; Howard, Black, 1970, 1972; Poehlman et al., 1972, and Parlevliet, 1977). According to Thurston (1971) horizontal resistance was available for many years and fairly high levels were observed by Stuart (1905), Ito (1918), Reddick (1928) Bonde (1932) and Stevenson et al., (1937). These workers noted its occurrence in potatoes before the utilization of race specific resistance derived from S. demissum. The polygenic nature of inheritance was demonstrated by Black (1953). Toxopeus (1959 and 1961) stated that general resistance was governed by minor genes. Stevenson et al. (1937) as reported by Thurston (1971) studied the progeny of a cross between a susceptible and a resistant variety with general resistance and found all were more resistant than the susceptible check with 16.7% showing no infection and 7.3% more infected than either parent.

They concluded that resistance was controlled by a series of minor genes and it was quantitatively inherited.

Horizontal resistance is stable since it involves mechanisms which are beyond the pathogens capacity to change or because of its unspecific nature, the parasite finds it difficult to adopt to this type of resistance. To break horizontal resistance, the parasite would have to undergo considerable changes in its biology and this is not practicable (Robinson, Parlevliet, 1977). Thurston (1971) reported that, 1973: Davidson (1928) worked with the variety Champion which was introduced to Ireland in 1877 and found that it started losing its resistance by 1885 but it was still being grown by 1928. Muller and Haigh (1953) found this variety to have a very high level of general resistance both in the field and in the laboratory. Niederhauser and Cervantes (1956, 1962) reported that Mexican potato cultivars maintained the same level of general resistance under severe late blight conditions for many years. However, a slight decline was noted in certain clones over a period of 15 years of field testing in the Toluca valley, Mexico but no sudden

Thurston et al. (1962) reported that the general resistance levels of <u>S. tuberosum</u> subsp. <u>tuberosum</u>, <u>S. tuberosum</u> subsp. <u>andigena</u>, and <u>S. phureja</u> clones did not change from year to year when compared with standard cultivars.

Robinson (1973) stated that oligogenic horizontal resistance had been shown to occur and therefore not all horizontal resistance is polygenically inherited. Parlevliet (1977) stated that minor genes which control horizontal resistance could also operate in a gene-for-gene manner with minor genes in the pathogen. If polygenes are involved in this interaction or true resistance then we have horizontal resistance. Escape or pseudo resistance is of horizontal nature, and is often of morphological nature. Mechanisms in this type of resistance operate before contact between the host and the parasite is established. These mechanisms reduce the chances of contact between the host and the pathogen. Genes for escape resistance act independently from genes in the parasite, no gene-for-gene action. Morphological characters which prevent or affect entry of a pathogen are thick cuticle, waxy covering, small stomata, and

hairiness of leaves. Hairiness of leaves can offer resistance to several pests (Robinson, 1973; Parlevliet, 1977).

Parlevliet and Zadoks (1977) reported that in natural populations where separation of resistance into horizontal and vertical resistance is not practicable, the host-pathogen systems are characterized by great variability and the genetic system operates on a gene-for-gene basis to create a state of equilibrium. In this equilibrium, the diverse genes for resistance and their corresponding genes for virulence co-exist. The resistance of the host population is derived from cumulative effects of all the genes for resistance in the population. This equilibrium is disturbed by the change from heterogeneous to extremely homogenous population through agriculture. Through breeding, specific host genotypes were matched with specific pathogen genotypes and as a result, race specific resistance was selected at the expense of general resistance. explain the resistance in natural populations, Parlevliet and Zadoks (1977) put forward the intergrated concept, a concept including horizontal and vertical resistance.

The effectiveness or stability of resistance in a cultivar depends on genetic homeostasis or resistance to genetic changes. Stability is highest when many genes for resistance and pathogenicity are involved and recombination in the pathogen is reduced. This means that new races cannot be formed by recombination and the pathogen has difficulty in adapting to a large number of genes for resistance. The equilibrium in natural populations is due to the fact that all genes for resistance are effective even though their corresponding genes for virulence are present. Homeostasis operates very strongly. In our agro-ecosystems, some kind of dynamic diversity can be created by use of polygenes or multilines so that the pathogen does not adapt itself easily to the existing resistance.

2.4. Basis of resistance

Several workers have found an association between the thickness of the cuticle and the ability of the pathogen to penetrate. The association has been found in cases where penetration by the pathogen is by mechanical means. According to Royle (1976), Louis (1963) showed that cuticle thickness in bean and tomato were related to the ability of Botrytis cinerea to penetrate but Berry (1959) found no association between the

thickness of the outer epidermal wall in onion and resistance to <u>Peronospora destructor</u>. Some authors have stated that cuticle thickness in plant diseases provides no serious barrier to penetration and that its contribution whether physical or chemical to resistance cannot be great.

2.5. Phytoalexins and their role in disease resistance

The critical event which determines whether a plant will be resistant or susceptible occurs after penetration by the invading micro-organism. Many plants produce phytoalexins which inhibit the growth of the invading micro-organism whether or not the invading micro-organism is a pathogen. The ability to produce phytoalexins is in itself a defence mechanism.

In potato the primary event determining a compatible and incompatible interaction to P. infestans depends upon an interaction which occurs within minutes or hours after penetration. All the profound metabolic alterations including the accumulation of phytoalexins are the result of this initial interaction (Ingram, 1967, Robertson et al., 1969, Albersheim, 1976, Kuc et al., 1976).

Phytoalexins are antimicrobial compounds which are produced by the plant during the interaction between the plant and an infectious or invading agent. There are 2 classes of phytoalexins in potato (Deverall, 1976).

- (i) the phenolics and
- (ii) the terpenoids

2.5.1. Phenolic compounds

Phenolic compounds are associated with colour of raw potatoes and in part responsible for certain types of discolouration of processed products. The phenolics are oxidised to quinones and these are toxic to invading pathogens, for instance yellow and red pigmented onions, accumulate flavones, anthocyanins and simple phenolics, e.g. protocatechuic acid and catechol. Phenolics from the scales are water soluble and diffuse from the scales to infection sites and can inhibit germination and penetration of infective fungi. Potato plants resistant to verticillium wilt have been shown to contain high amounts of chlorogenic acid in roots than susceptible ones (Patil and Zucker, 1965; Goodman et al., 1967).

Types of phenolic compounds are:-

- (i) <u>Lignin</u> which is present in low amounts in the vascular tissue.
- (ii) <u>Coumarins</u> responsible for discolouration of potatoes. Coumarin derivatives are scopoletin and esculetin.
- (iii) Anthocyanins and flavones are responsbile for skin and flesh colour.
 - (iv) Tannins, e.g. chlorogenic acid constitutes

 0.025 0.150% of dry weight. It is

 concentrated in a thin layer in the

 periderm tissue next to the skin. The

 concentration of chlorogenic acid in the

 peel is 10 times that in the flesh and

 the concentration is more in the bud

 end than in the stem end.
 - (v) Monohydric phenols e.g. tyrosine which is present in the inner portion of the tuber and it constitutes 0.1 to 0.3% of dry weight. It crystallizes out from concentrates prepared from ethanolic extracts of potatoes. Tyrosine is exidised by tyrosinase into melanin.
 - (vi) Polyphenolics. These and chlorogenic acid accumulate at sites of infection.

sites of mechanical damage and when exposed to light (Talburt and Smith, 1967).

2.5.2. Terpenoids

The following terpenoids can be detected in potato tubers or leaf petioles after infection with incompatible races.

Rishitin which is detected 7-8 hours after (i) infection and reaches concentration of 100 µg/g fresh weight at sites of infection after 24 hours. In incompatible interaction, inhibition of fungal growth is apparent and is sufficient to stop growth of zoospore germ tubes in vitro (Sato and Tomiyama 1977). Varns et al. (1971) as reported by Kuc et al. (1976), demonstrated that incompatible interaction were associated with rapid necrosis of the host tissue and the accumulation of 16-18 terpenoids including rishitin and phytuberin. The optimum temperature for rishitin and phytuberin accumulation is about 19⁰C (Currier and Kuc, 1975). Accumulation of rishitin reduces at 25°C . The peak of rishitin accumulation in aged tissue

is 20 hours after treatment as opposed to 60-72 hours for unaged tissue (Kuc et al., 1976).

- (ii) Phytuberin accumulates at 19° C but is reduced at 30° C.
- (iii) Lubimin
- (iv) Chaconine and
- (v) <u>Solanine</u>

Solanine and chaconine are steroid glycoalkaloids which are associated with resistance and have been reported in tuber and foliage and appear localized around sites of injury in tubers. The steroid glycoalkaloids are largely restricted to the peel of whole tubers and are major antifungal compounds (Kuc et al., 1976).

Accumulation of solanine and chaconine at the surface of cut tissue slices is markedly reduced or suppressed by inoculation with P. infestans especially when incompatible races of the fungus are used, (Sakai et al., 1967, Kuc et al., 1976).

Aging tubers contain more steroid

glycoakaloids than unaged tissue. The Concentrations of about 139 μg and 72 μg solanine/g fresh weight in the top layers of the tuber have been reported (Kuc et al., 1976).

Previous workers have found that phenol and phenol derivatives are important in disease resistance. Sakai et al., 1964 and 1967 found that potato varieties resistant to P. infestans had high contents of total phenols and the metabolism of phenolic compounds was markedly increased by infection especially in cells surrounding the infection site.

CHAPTER 3

MATERIALS AND METHODS

3.1. Field tests

Clones and varieties were screened for three seasons at three locations and the following is a brief description of the experiments conducted.

Season	1000	Location
Short Rains, 1977-78		National Agricultural
		Laboratories, Nairobi.
A Committee of the same of	7	Potato Research Station,
		Tigoni.
Long Rains, 1978	700	National Agricultural
		Laboratories, Nairobi.
as a marga-	40	Tigoni
	· Lin	National Pyrethrum Research
4		Station, Molo.
Long Rains, 1979	-	Field Station, University of
		Nairobi, Kabete.
	-	National Pyrethrum Research
		Station. Molo.

3.1.1. Preliminary field screening of clones for resistance to late blight

A total of 226 clones were screened during the preliminary field screening and six out of these were established varieties. Two of these varieties namely Heiderot and Dextra are German varieties while Huancayo and Mariva are South American varieties. B 53 and Desiree are grown in Kenya and were used as the resistant and susceptible controls, respectively.

Out of the 226 clones tested, 221 were tested at National Agricultural Laboratories, Nairobi and 11 clones tested at Tigoni. Clones were planted in single rows of 10 plants in the field and the incidence of late blight following natural infection was recorded on all the clones. Weekly records were taken right from the time the first symptoms of late blight appeared on the leaves. No fungicides were applied. Other diseases appearing on the foliage were also recorded. At maturity, tubers were harvested and weighed. Records were also taken on skin and flesh colour, depth of eyes and shape of the tubers.

3.1.2. Second season testing

For the second season screening 15 clones were planted in single rows of five plants in the field at the National Agricultural Laboratories, Nairobi, and Tigoni. Late blight and yield records were taken as in the first season.

At the National Pyrethrum Research Station, Molo, 12 clones were tested. Single rows of 10 plants were planted in a randomized complete block design with three replicates and similar records taken.

3.1.3. Third season testing

6 clones were tested at the Field Station,
University of Nairobi, Kabete and at the National
Pyrethrum Research Station, Molo. Clones were planted
in two rows of 10 plants in a randomized complete
block design with four replicates and records taken as
in 3.1.1.

3.2. <u>Laboratory tests</u>

3.2.1. Screening of seedlings for resistance to late blight

Six week old seedlings grown from berry seed were inoculated with a sporangial suspension of P. infestans. The inoculum was prepared by washing the sporangia from infected incubated leaves with distilled water. The infected leaves were picked from the variety Roslin Eburu and any other diseased clone/variety from the field and incubated in sandwitch boxes lined with moistened cellulose tissue at about 18°C.

The concentration of the inoculum was 400 sporangia/ml and this was determined by sporangial counts using a haemocytometer. Seedlings were then inoculated by spraying the suspension on to the leaves using a hand-sprayer in a moist chamber. The ground under the chamber was watered before the inoculation and the chamber was covered for 48 hours to increase the humidity. Seven days later, the seedlings were examined and placed in one of the following resistance groups depending on the amount of damage incurred.

Group O

no blight

Ι

- 1-10% leaf area infected

II

- 10-25% area infected

III

 up to about 50% of the foliage effected and any sign of stem blight. 1.7

- plants severely defoliated
but stems still green

٧

- plants dead or dying.

3.2.2. Screening of whole plants for resistance to late blight

Tubers of 215 clones which were not tested at the seedling stage were planted in 15 cm pots and plants inoculated with a sporongial suspension of P. infestans by spraying the inoculum on to the plants using a hand sprayer. Inoculum concentration was 800 sporangia/ml. After 7 days, plants were graded as above (3.2.1.) and the grading was repeated for 3 weeks at weekly intervals.

3.2.3. Leaf inoculation

(a) Inoculation of whole leaves

Leaves of 16 clones were picked from similar positions on the plants, washed in distilled water and inoculated with a sporangial suspension of P. infestans 400 sporangia/ml and incubated at 17.5-18°C. The inoculum was prepared from infected incubated leaves. Test leaves were passed through the sporangial suspension and placed in plastic boxes lined with moist cellulose tissue and covered. The rate of infection, area infected and the period of sporulation were recorded.

(b) <u>Inoculation of distal and terminal leaflets</u>

Distal and terminal leaflets of six clones were inoculated by placing a drop of about 0.03 ml. of the suspension on the under side of each leaflet and incubated in plastic boxes lined with moist filter paper. Records taken included number of days for infection, size of the lesion, period of sporulation and number of sporangia per leaflet. Size of the lesions was recorded by measuring the length and width of the lesion, tracing the lesion on paper and determining the area by using an Allbrit planimeter. The number of sporangia per leaflet was counted by use of a haemocytometer. The concentration of the inoculum used was 400 sporangia/ml.

3.2.4. Anatomical studies

Leaves and stems of 8 clones/varieties were picked from one month and two month old potted plants and small sections of each were fixed in formaldehyde acetic acid and transverse sections made using a microtome at a thickness of 10-15 microns. The thickness of the epidermal cells was measured to see if there was any correlation between the cell thickness and the resistance of these clones to late blight.

3.2.5. Extraction of total Phenolics in the presence of a reducing agent

(a) Ethanol extraction of tissue

A 20g sample of each selection was ground in a waring blendor for 3 minutes with 96 ml of 95% ethanol. 10 ml of freshly neutralized 0.04M solution of cysteine were added to the sample in a flask and the ethanolic suspensions quickly taken to boiling and refluxed on a steam bath for 4 hours. The extracts were filtered at the pump and the respective residues re-extracted twice with 80% ethanol for a period of 3 hours. The extracts from each set were pooled and made up to a known volume.

(b) Ethanol extraction of homogenates

Simultaneously, a second 20g sample was ground with 80ml of 0.04M cysteine (freshly neutralized) to give 20% w/v homogenates. The homogenate was filtered and a 1.90ml aliquot of each was extracted five times in succession initially with 95% and subsequent 80% ethanol by heating in a bath of boiling water for 2 minutes each. The centrifuged supernatants were made up to a known volume. Cysteine was added to prevent oxidation of phenols.

(c) Acetone extraction in the cold

A sample of 50g tissue was extracted in the cold in a waring blendor with 300 ml anhydrous acetone. 10 ml of 0.04M solution cysteine were added. 50ml of anhydrous acetone were used for the next three extractions followed by 30 ml of 80% (v/v) acetone for 5 extractions. Finally, the residual solid was extracted twice with 30 ml anhydrous acetone (cysteine was not added).

3.2.6. Folin - Ciocalteu reagent

The reagent was prepared by dissolving 100g of sodium tungstate (Na₂WO₄2H₂O) plus 25g of sodium molybdate (Na₂MoO₄2H₂O) in 700 ml of distilled water in a 2 litre round bottomed pyrex flask. 50 ml of 85% H₃PO₄ and 100 ml conc. HCL were added and a reflux condenser attached and the solution was refluxed for 10 hours (not necessarily continuous). A few glass beads were added to suppress bumping. At the end of the refluxing period, heating was stopped and the flask allowed to cool. The condenser was removed, and 150g of lithium sulphate (Li₂SO₄H₂O) added. A small amount of 30% hydrogen peroxide was added. The hydrogen peroxide oxidises any traces of molybdenumtungsten blue, and the

final reagent is yellow. The solution was cooled made to one litre, filtered through sintered glass and stored in a glass-stoppered amber bottle.

3.2.7. Method of determination

I ml of the sample was mixed with 60 ml of water in a 100 ml volumetric flask. Folin-Clocalteu reagent, 5 ml were added and mixed, and after about 30 seconds and before 8 minutes 3.0 g of anhydrous Na₂Co₃ in aqueous solution (e.g. 15 ml of 20% solution) was added and mixed and the contents of the flask made to volume. The absorbance was determined calorimetrically versus a zero-absorbance reagent blank after 2 hours at 23°C in 1 cm cells at 765 mµ. The total phenolic content was calculated in gallic acid equivalents by comparison with a standard curve similarly prepared, (Fig. 1).

3.2.8. Tuber inoculation

Whole tubers of eight clones were inoculated with a sporangial suspension of P. infestans as described below. The skin and the top most few millimeters of the flesh were removed from each tuber and small disks of filter paper dipped into a sporangial suspension and placed on top of the small wounds. After 3 days, one tuber of each selection was cut into two and the extent of the brown discolouration due to infection was measured in millimeters.

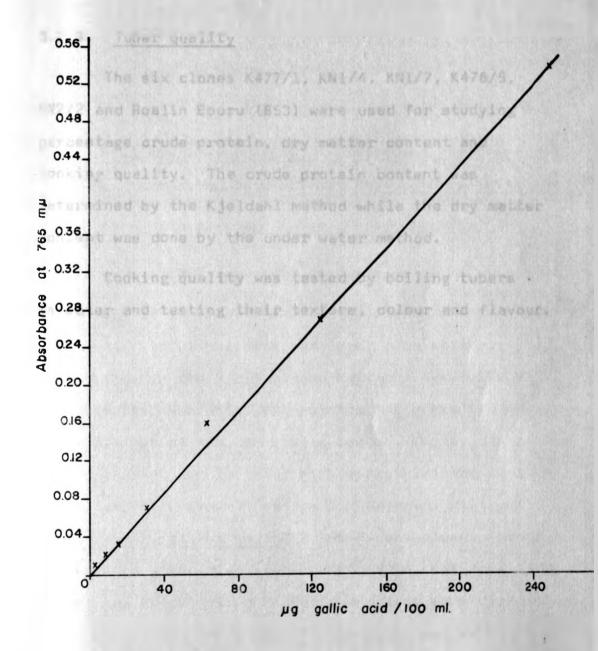


FIG. 1: THE STANDARD CURVE FOR THE DETERMINATION OF TOTAL PHENOLICS
IN POTATO LEAVES

3.2.9. Tuber quality

The six clones K477/1, KN1/4, KN1/7, K478/5, KN2/2 and Roslin Eburu (B53) were used for studying percentage crude protein, dry matter content and cooking quality. The crude protein content was determined by the Kjeldahl method while the dry matter content was done by the under water method.

Cooking quality was tested by boiling tubers in water and testing their texture, colour and flavour.

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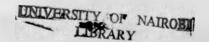
CHAPTER 4

RESULTS

4.1. Preliminary screening of clones and varieties for resistance to late blight, Phytophthora infestans other diseases and tuber characteristics

4.1.1. Late blight and other diseases

Out of the 226 clones tested during the preliminary screening, 215 clones were screened under natural infestation in the field at the National Agricultural Laboratories, Nairobi. The results are presented in Table 1. Out of the 215 clones tested, twenty one clones were discarded due to their high susceptibility to late blight and eighteen clones due to bacterial wilt (Pseudomonas solanacearum). Twenty one clones showed symptoms of virus diseases namely Potato Leaf Roll Virus (PLRV) and Potato Virus Y (PVY), and they were discarded. Early blight caused by Alternaria solani was recorded on eleven clones and these were discarded too. The remaining 176 clones showed low to moderate susceptibility of which eighteen clones were classified as group I, seventeen clones as group II and 141 as group III.



These 215 clones were tested in the laboratory under artificial infestation and only 78 clones passed the blight test. They were grouped between group I and III. These results therefore indicated that under field conditions only 10% were found to be susceptible (groups iv and v) as compared to 64% under laboratory conditions.

Table 1. Mean resistance grades to late blight and some tuber characteristics of potatoes screened at N.A.L., Nairobi.

S. No.	Clone	Pedigree	Resist	ance	Yield (gm/pl)	Skin colour	Flesh colour	Eyes depth	Tuber shape	Remarks
			Field	Lab.						
1.	K471/1	Roslin Athi x 66-658(3)	2.7	3.5	572	pink	white	medium	oval	potato leaf roll virus°
2.	K471/2	- do -	2.7	4.6	169	light purple	cream white	deep	round	purple ring in the flesh*
3.	K471/3	- do -	3.0	4.6	211	light pink	white	qseb	round	PLRV*
4.	K472/1	Roslin chania x 66-658(3)	1.0	2.3	538	white	white	medium	oval	purple specks in the skin
5.	K473/1	K023N/11 x	2.0	3.0	546	white	yellowish	shallow	oval	
6.	K473/2	K015N/12 I	2.3	3.3	447	white	cream	ve ry shallow	round	PLRV*
7.	K473/3	- do -	1.3	3.0	853	white	cream	shallow	round oval	tuber moth skin slightly rough
8.	K473/4	- do -	1.3	3.3	807	white	yellow	shallow	long	heavily russet and cracking*
9.	K473/5	- do -	1.3	3.3	580	white	white	shallow	round	slight cracking on tubers
10.	K473/6	- do -	1.7	4.0	702	white	cream	medium	pear shaped	cracked tubers, blighted, rough skin*
11.	K473/7	- do -	2.0	3.3	321	white	cream	shallow	round	slightly russet*
12.	K473/8	- do -	1.0	3.0	383	white	white	shallow	oval	
	W 470 /0			2.0	F42			doop	mumd*	THE PARTY OF THE P

Table 1 (Contd..) Pedigree Resistance Yield Skin. S. No. Clone (0-5)(gm/p1) colour Field Lab. 14. K474/3 KO23N/2 III 2.5 2.7 674 white x KOIN(1) I K79a(1) Ix 15. K475/1 1.0 2.3 808 white Anett 16. K475/2 - do -3.0 3.0 483 white K475(3) 466 white 17. - do -3.0 3.66 18. K475(4) - do -3.0 3.3 472 white K475(5) 3.0 4.3 478 white 19. - do -20. K475(6) - do -2.7 3.3 544 white K475(7) 2.3 3.0 645 white 21. --- do -(8) 407 white 22. - do -3.0 3.0 23. (9) do -162 white 2.3 3.7 - do -24. (10) 2.3 4.5 482 white 25. (11) - do -2.3 4.5 279 white 26. (12) - do -2.3 4.0 389 white 27. (13)- do -2.66 3.3 324 white 28. (14)- do -2.7 3.0 white 444

2.7

3.0

378

white

29.

(15)

do -

Flesh colour	Eyes depth	Tuber shape	Remarks
white	deep	round	heavily russet, misha-pen tubers*
white	shallow	oval	russet, hollow heart*
white	shallow	round	slight cracking, hollow heart*
cream	shallow	round	tuber moth*
white	shallow	round	PLRV*
cream	shallow	round	tuber moth, slightly rough skin*
cream	very shallow	round	PLRV, russet and cracking*
cream	medium	round	russet, cracking*
white	shallow	oval	
white	shallow	long	PLRV (heavy)*
white	medium	round	tuber moth and soft rot*
cream	madium	round*	
cream	very shallow	round*	
white	shallow	long*	
cream	shallow	round	cracking, heavily russet*
cream	medium	round*	
	45.000	DouBA .	and the state of t

unmer mounts akin

eracking.

Table 1 (Contd...)

S. No.	Clone	Pedigree	Resist	ance	Yield (gm/pl)	Skin colour
			Field	Lab.	(8.11)	
31.	K475(17)	- do -	1.3	2.7	339	white
32.	(18)	- do -	2.7	2.3	362	white
33.	(19)	- do -	2.0	2.3	790	white
34.	(20)	- do -	2.3	2.7	257	white
35.	(21)	- do -	2.7	3.0	284	white
36.	(22)	- do -	2.0	3.0	153	white
37.	(23)	- do -	3.0	3.3	340	white
38.	(24)	- do -	2.0	3.0	510	white
39.	(25)	- do -	2.7	3.3	524	white
40.	K476 (1)	Feldeslohn x K79a (1) I	1.7	3.3	485	white
41.	(2)	- do -	2.3	3.3	457	white
42.	(3)	- do -	2.7	3.3	483	white
43.	(4)	- do -	2.0	3.0	685	white
44.	(5)	- do -	3.0	3.3	907	white
45.	(6)	- do -	3.0	5.0	247	white
46.	(7)	- do -	2.0	3.0	235	white
47.	(8)	- do -	2.3	4.0	731	white
48.	(9)	- do -	2.3	3.3	581	white
49.	(10)	- do -	3 . D	4.0	358	white

Flesh colour	Eyes depth	Tuber shape	Remarks
white	shallow	round	hollow heart*
cream	medium	round	heavily russet, cracking*
cream	medium	round	
yellow	shallow	round	heavily russet*
yellow	shallow	round	russet and tuber moth*
yellowish	shallow	round	cracking, russet*
white	very shallow	oval- long	hollow heart, russet*
white	shallow	long & flat*	
white	shallow	oval	slightly russet*
white	medium	round* oval	
white	medium	round	slightly russet*
cream	medium	round*	
yellowish	medium	round	slightly rough skin
yellowish	medium	oval	cracking*
yellowish	shallow	round*	
yellowish	medium	round	russet*
yellowish	shallow	round	good yielder*
cream	medium	round	cracking*
cream	very	oval*	

shallow

Table 1 (Contd..)

S. No.	Clone	Pedig	iree	Resist	ance	Yield (gm/pl)	Skin colour
			· · · · ·	Field	Lab.		
50.	K476(11)	- do	-	2.7	3.0	315	white
51.	(12)	- do	-	2.3	3.7	380	white
52.	(13)	- do	_	2.0	2.3	606	white
53.	(14)	- do	-	2.7	4.0	240	white
54.	(15)	- do	-	2.7	3.0	484	white
55.	(16)	- do		2.7	4.0	410	white
56.	(17)	- do	-	2.3	4.0	241	white
57.	(18)	- do	-	2.3	3.3	359	white
58.	(19)	- do	-	1.7	3.0	360	white
59.	(20)	- do	-	2.3	3.3	519	white
60.	(21)	- do	91.11	2.0	4.0	500	white
61.	(22)	- do	-	2.7	3.0	368	white
62.	(23)	- do	-	2.7	4.0	276	white
63.	(24)	- do	leg h	2.0	3.0	424	white
64.	(25)	- do	-	2.3	4.0	734	white
65.	(26)	- do	-	3.0	5.0	301	white
66.	(27)	- do	-	2.7	2.3	257	white
67.	(28)	- do	-	2.3	3.0	357	white

Flesh colour	Eyes depth	Tuber shape	Remarks
yellowish	shallow	round	cracking, russet*
cream	medium	round	slightly rough skin*
cream	medium	round	cracking, heavily russet*
yellowish	medium	round*	
cream	medium	oval round	a few cracked tubers*
yellowish	shallow	round	heavily russet*
cream	shallow	oval	russet*
yellowish	deep	round	cracking, slight russet*
white	qeeb	round oval	heavily russet, slightly cracking*
cream	medium	round	blighted tubers*
cream	deep	round	a few cracked tubers*
cream	medium	round	tuber moth*
white	shallow	round	russet, hollow heart*
cream	shallow	round oval	russet*
cream	medium	oval*	
white	shallow	round	slightly cracking*
white	medium	round	
yellow	shallow	round	slightly russet

Table 1 (Contd...)

S. No.	Clone	Pedigree	Resist	ance	Yield (gm/pl	Skin colour
			Field	Lab.	Cam pr	001001
68.	K476(29)	- do -	2.7	3.5	324	white
69.	(30)	- do -	3.0	3.0	376	white
70.	(31)	- do -	3.0	3.0	371	white
-						
71.	(32)	- do -	2.3	2.7	414	white
72.	(33)	- do -	2.3	3.0	447	white
73.	(34)	- do -	3.0	4.0	603	white
74.	(35)	- do -	2.7	3.0	331	white
75.	(36)	- do -	2.7	3.0	462	white
76.	(37)	- do -	3.7	4.0	147	white
77.	(38)	- do -	3.0	3.3	531	white
78.	K477(1)	Atzimba x W88	1.3	2.0	747	white
79.	(2)	- do -	1.7	4.0	301	white
80.	(3)	- do -	2.0	3.3	683	white
81.	K478(2)	Atzimbax W 25	1.0	4.0	584	white
82.	(3)	- do -	2.0	2.3	547	white
83.	(4)	- do -	1.0	2.7	515	white
84.	(5)	- do -	1.0	1.6	885	white
85.	(6)	- do -	1.0	1.3	686	white
86.	(7)	- do -	1.0	1.0	782	white

		Eyes depth	Tuber shape	Remarks
CL	otodi	gehen	Suahe	
CI	ream	medium	round*	
CI	ream	qeeb	round*	
уe		very shallow	round	cracking and heavily russet*
wh	nite	medium	round	cracking*
wh	nite	shallow	round	heavily russet, cracking*
ye	ellow	shallow	round	heavily russet, cracking*
C	ream	shallow	oval	cracking*
C	ream	medium	round	
wl	nite	shallow	oval*	
W	hite	medium	round*	
ye	ellow	shallow	oval long	
ye	ellowish	shallow	round	cracking*
C	ream	shallow	round*	
C	ream	shallow	round	soft rot and tuber moth*
W	hite	shallow	oval	
C	ream	medium	oval	
C	ream	shallow	long oval	good yielder
С	ream	shallow	long	nematodes
W	hite	shallow	oval	

Table 1 (Contd..)

S.No.	Clone	Pedigree	Resist	ance	Yield gm/pl	Skin colour	Flesh colour	Eyes de pth	Tuber shape	Remarks
			Field	Lab.						
87.	K478(8)	Atzimbax W 25	1.0	3.0	741	white	yellow	shallow	round	nematodes
88.	(9)	- do -	1.3	3.5	641	white	white	shallow	long	tuber moth*
89.	(10)	- do -	1.0	1.3	581	white	cream	shallow	oval long	slightly cracking
90.	(11)	- do -	1.5	3.0	1007	white	cream	medium	round	
91.	(12)	- do -	1.7	2.3	100	white	white	medium	round	PVY*
92.	(13)	- do -	2.0	3.0	419	white	cream	shallow	long	
93.	(14)	- do -	1.0	2.0	1030	white	cream	medium	oval long	
94.	(15)	- do -	1.3	3.7	454	white	cream	shallow	long*	
95.	(16)	- do -	1.0	1.0	560	white	white	shallow	round	
96.	(17)	- do -	1.7	4.0	365	white	yellow	medium	round*	
97.	(18)	- do -	2.0	3.3	511	white	yellow	shallow	long*	
98.	(19)	- do -	1.0	2.3	758	white	yellow	medium	round*	
99.	(20)	- do -	1.0	2.3	214	white	cream	shallow	round*	
100.	K479(1)	Kenya AkibaxW88	1.0	1.0	746	white	cream	shallow	long and	
101	(2)	- do -	2.0	2.3	687	purple	white	medium	round* oval	
102.	(3)	- do -	1.0	4.0	586	purple	white	medium	round	sprouts in field*

Table 1 (Contd...)

S. No.	Clone	e Pedigree	Resistance (O-5)		Yield gm/pl	Skin colour	Flesh colour	Eyes depth	Tuber shape	Remarks
			Field	Lab.						
103.	K480(1)	K79a(1) I x P 95	1.7	2.3	505	white	white	medium	pear shaped	
104.	(2)	- do -	2.3	2.7	325	white	white	shallow	long curved	
105.	(3)	- do -	1.0	3.0	802	white	white	shallow	oval	tuber moth
106.	(4)	- do -	2.3	3.3	756	white	cream	shallow	oval	tuber moth*
107.	(5)	- do -	2.0	3.7	306	white	white	shallow	long*	
108.	(6)	- do -	2.7	4.0	518	white	white	medium	oval*	
109.	(7)	- do -	2.7	3.3	447	white	white	shallow	oval	tuber moth*
110.	(8)	- do -	2.3	4.0	113	white	white	medium	oval*	
111.	(9)	- do -	2.3	3.3	325	white	yellow	medium	oval*	
112.	(10)	- do -	1.3	3.3	462	white	white	shallow	oval* flat	
113.	(11)	- do -	3.0	3.3	274	white	white	shallow	oval*	
114.	(12)	- do -	1.0	4.0	450	white	yellow	very shallow	long*	
115.	(13)	- do -	2.0	2.3	338	white	white	shallow	oval	soft rot*
116.	(14)	- do -	2.0	2.3	506	white	white	very shallow	round oval	cracking*
117.	K482(1)	K79a (1) I x Roslin Karura	1.3	1.5	550	white	white	shallow	oval	blighted tubers and hollow heart*

Table 1 (Contd...)

S.No.	Clone	Pedigree	Resistance (0-5)		Yield gm/pl	Skin colour	Flesh colour	Eyes depth	Tuber shape	Remarks
			Field	Lab.						
118.	K482(2)	K79a(l) I x Roslin Karura	2.3	2.7	859	white	white	shallow	round	hollow heart*
119.	(3)	- do -	1.3	3.0	383	white	white	shallow	round	
120.	K483(1)	66-700(1) IIIxK80a (19)I	2.7	3.3	605	white	white	medium	round*	
121.	(2)	- do -	2.3	4.0	1520	white	cream	medium	round*	
122.	(3)	- do -	3.0	3.0	350	white	white	shallow	round	
123.	(4)	- do -	2.0	3.0	845	, white	white	shallow	round	
124.	(5)	- do -	2.0	3.0	480	white	white	deep	oval	
125.	(7)	- do -	2.7	3.5	328	white	white	shallow	round* oval	
126.	(8)	- do -	3.0	3.5	855	white	cream	shallow	long*	
127.	(9)	- do -	4.0	4.0	-	white	white	shallow	round	no yield*
128.	(10)	- do -	3.0	4.0	1345	white	white	shallow	round*	
129.	(11)	- do -	3.7	4.0	150	white	white	shallow	round*	
130.	(12)	- do -	3.0	3.5	270	white	yellowish	medium	round*	
131.	(13)	- do -	2.0	3.5	55	white	white	shallow	oval	PLRV stunted plants with browning leaves. •
132.	(14)	- do -	3.0	3.3	240	white	white	shallow	round	leaves rolling, PLRV*
133.	(15)	- do -	3.0	3.0	1155	white	white	shallow	oval	many strong lateral branch

Table 1 (Contd...)

144.

145.

146.

147.

148.

149.

(56)

(12)

(15)

(16)

(17)

K484(2)

- do -

66-700(1) III

x Roslin Tana

S. No.	Clone	Pedigree	Resista (O-5)	ance	Yield (gm/pl)
			Field	Lab.	
134.	K483(16)	66-700(1)III × K80a(19)I	2.5	3.3	83
135.	(17)	- do -	3.0	4.0	634
	(20)				
136.	(18)	- do -	3.0	4.0	**
137.	(19)	- do -	-	4.0	-
138.	(20)	- do -	-	4.0	-
139.	(21)	- do -	-	4.3	-
140.	(22)	- do -	-	4.0	-
141.	K483(50)	66-700 (1) IIIx	2.0	3.3	438
142.	(54)	K80a (19) I	3.3	5.0	387
143.	(55)	- do -	2.3	4.0	1925

2.3

3.0

3.0

3.0

4.0

2.3

5.0

3.0

3.0

3.0

4.0

3.5

115

503

1065

715

875

950

	0			
Skin colour	Flesh colour	Eyes depth	Tuber shape	Remarks
white pink with white stripes	white white	shallow	oval* round*	
- stithes	-	-	-	wilt* bacterial wilt*
-	-	-	-	do* do*
pinkish white	white	shallow medium	oval*	00
white white	cream	medium	round	very strong haulms* PLRV*
white white	white white	shallow	round round	
white white	white white cream	medium shallow shallow	round oval*	tiny leaflets and thin

Table 1 (Contd...)

S.No.	Clone	Pedigree	Resist	ance	Yield (gm/pl)
			Field	Lab.	
168.	(9)	- do -	4.3	3.5	165
169.	(11)	- do -	2.7	3.5	467
170.	(12)	- do -	3.0	3.0	355
171.	(13)	- do -	2.7	3.0	783
172.	(15)	- do -	2.7	2.7	678
173.	(17)	- do -	4.0	4.0	-
174.	K486a(2)	66-631(1) x K8Oa(19) I	3.0	4.0	565
175.	(3)	- do -	3.0	3.3	1040
176.	(4)	- do -	3.7	2.0	110
177.	(6)	- do -	3.0	1.0	160
178.	(9)	- do -	3.7	4.0	-
179.	(10)	· - do -	4.3	4.0	35
180.	(13)	- do -	-	4.0	un.
181.	(15)	- do -	-	2.0	400
182.	(17)	- do -	-	4.0	
183.	K486a(18)	- do -	3.0	4.0	685
184.	(19)	- do -	4.3	4.0	-
200	(20)	4-	A 2	2 0	-

Skin colour	Flesh	Eyes depth	Tuber shape	Remarks
white	white	shallow	round*	
white	cream	medium	oval*	
purple	white	medium	round	
purplish	white	medium	oval*	
white	white	shallow	round	
-		-	-	no yield*
white	white	very shallow	round* oval	
white	white	deep	round* oval	
white	white	medium	round*	
white	white	deep	round*	
-	-	-	-	no yield*
white	white	shallow	oval*	
-	-	-	-	wilt*
-	-	-	-	wilt*
-	-	-	-	wilt*
white	cream	medium	round*	
7	-	-	-	no yield*
-	-	-	+	no vield*

Table 1 (Contd...)

S.No.	Clone	Pedigree	Resista (O-5)	nce	Yield (gm/pl)	Skin colour	Flesh colour	Eyes depth	Tuber shape	Remarks
			Field	Lab.						
186.	(22)	- do -	3.7	5.0	230	white	cream	medium	pear shaped*	
187.	(23)	- do -	3.5		-	-	-	-		wilt*
188.	(24)	- do -	2.7	4.0	115	white	cream	shallow	round*	
189.	(25)	- do -	3.0	5.0	530	white	cream	shallow	round*	
190.	(26)	- do -	3.0	4.0	1013	white with pink eyes	cream	deep	long*	
191.	(17)	- do -	3.0	4.0	750	white	white	shallow	round*	
192.	K486b(1)	- do -	3.0	4.0	140	white	white	shallow	oval*	
193.	(2)	- do -	2.3	1.3	582	white	cream	shallow	round oval	hollow heart*
194.	(3)	- do -	3.0	5.0	417	white with purplish eyes	white	shallow	round	russet*
195.	(4)	- do -	3.0	4.0	647	white	white	shallow	oval*	
196.	(5)	- do -	2.7	4.0	444	white	cream	medium	round	hollow heart*
197.	(6)	- do -	9	3.0	-	-	-		-	wilt*
198.	(7)	- do -	2.7	3.0	370	white	white	shallow	round	
199.	(8)	- do -	3.0	4.0	505	white	white	medium	long* oval	
200-	(10)	- do -	3.0	3.0	59	white	white	medium	round	

Table 1 (Contd....)

S.No.	Clone	Pedigree	Resista (O-5)	Resistance (O-5)		Skin colour	Flesh	Eyes depth	Tuber shape	Remarks
			Field	Lab.						
201	(11)	- do -	3.0	5.0	433	white	white	shallow	round oval	black rot*
202.	K486c(1)	- do -	-	4.0	-	-	-	-	-	wilt*
203.	(2)	- do -	-	3.5	-	-	-	-	-	wilt*
204.	(3)	- do -	-	3.5	-	-	-	-	-	wilt*
205.	K486c(4)	- do -	3.0	3.3	470	white	white	medium	oval	leaves wrinkled at top.*
206.	(5)	- do -	3.0	3.0	110	white	white	medium	round	
207.	(7)	- do -	2.7	3.0	380	white	white	shallow	round	
208.	K486c(8)	- do -	2.7	3.0	465	white	white	medium	oval round	
209	(10)	- do -	3.0	3.0	1180	bluish	white	qeeb	oval*	
210.	(11)	- do -	2.7	3.5	1090	white	white	medium	round*	
211.	(12)	- do -	2.3	4.0	376	white	cream	shallow	round	russet*
212.	(16)	- do -	3.0	4.0	585	white	cream	shallow	round*	

Table 1 (Contd...)

S.No.	Clone	Pedigree	Resistance (O-5)		Yield (gm/pl)		Flesh colour	Eyes depth	Tuber shape	Remarks
			Field	Lab.						
213.	K486c (17)	- do -	3.0	3.0	1618	white with	cream	shallow	round	
						eyes				
214.	(18)	- do -	3.0	4.0	1020	purplish	yellow	medium	round*	
215.	(19)	- do -	2.7	3.0	1398	white	white	medium	round oval	

^{*} discarded due to high infestation by bacterial wilt, virus diseases and late blight.

PLRV - Potato leaf roll virus

PVY - Potato Virus Y.

The distribution of the progenies into different resistance groups revealed that most of the crosses I made yielded progenies which were moderately resistant (group III) to susceptible (group iv) or highly susceptible (group v) (Table 2). Cross numbers K478 and K486 however had few resistant clones (group I and II).

4.1.2. Tuber characteristics

At harvest, clones which had blighted tubers or whose tubers had deep eyes, hollow hearts or cracked were discarded. Hollow heart characteristic was found on eight clones. Cracking of tubers was recorded on twenty seven clones. Thirty three clones had slight to very rough skin. The cracking and rough skin was more in certain crosses, like K475 and K476 as compared to smooth skin of others like K478 (Fig. 2). The two characteristics in most cases occurred together.

Variation was also found in other characteristics such as tuber shape, skin colour, flesh colour and eye depth. 106 clones had round tubers, 43 clones had oval tubers, 15 had long tubers and three clones had pear shaped tubers. Eight clones had long oval tubers while 15 had round oval tubers. Round tubers were more common than other tuber shapes. Similarly white skin colour was more common than the pink and light purple colours of tubers. 178 clones had white tubers, four

clones had pink tubers and seven clones had purple skin colour.

Creamy to white flesh colour was more common than yellow flesh colour. One hundred and sixty three clones had cream and white flesh colour and only twenty six clones had yellow flesh.

Shallow eyes were more common than medium or deep eyes. A total of 113 clones had either shallow or very shallow eyes, 63 had medium eyes and fourteen clones had deep eyes.

At the moment, local consumers generally prefer the red skin types to the white skin varieties. Potatoes with shallow eyes are easier to peel than those with deep eyes.

Table 2. Distribution of clones from different crosses into resistance groups under field and laboratory screening

Cross	Number of	Field resistance groups					Lab. resistance groups				
number	plants	I	II	III	ΙV	V	I	ΙΙ	III	IV	V
K471 K472	3 1	1	-	3 -	-	_	-	1	-	1 -	2
K473	9	1	5	3	-	-	-	-	4	-	5
K474	1		-	1	-		-	-	1	-	-
K475	25	1	2	22	-	-	-	-	12	8	5
K476	38	_	1	35	2	-	-	-	14	19	5
K477	3	-	1	2	-	-	-	1	-	2	_
K478	19	11	4	4	-	-	2	3	7	6	1
K479	3	2	-	1	-	-	1	-	-	2	-
K480	14	2	2	7	3	-	-	-	5	9	-
K482	3	_	2	1	-	-	-	1	2	-	-
K483	25	-	-	17	2	1	-	-	4	19	2
K484	5	-	-	4	-	1	-	-	3	2	-
K485	24	-	-	13	4	2	-	-	2	19	3
K486 (a) (b) (c)	18 10 14	- - -	-	8 9 11	3 -	3 - -	1 -	2 1 -	2 3 6	10 4 8	3 2 -
Total	215	18	17	141	14	7	4	9	65	109	28





Fig. 2. Comparison of rough skin and cracking (K475/20 and K476/31) and smooth skin (K478/14) tubers.

4.1.3. Evaluation of introduced varieties at Tigoni

Four introduced varieties namely Heiderot, Mariva, Huancayo and Dextra were tested together with seven local clones under natural infestation in the field at Tigoni and Kabete. The results are given in Tables 3a, 3b, 3c, 3d, and 4a, 4b, 4c and 4d. The analysis of variance for the blight infestation at Tigoni is given in Table 3b. There were no significant differences in the infection rate as the interaction between the varieties and sampling dates was not significant. However the infection generally increased with the age of the plants. There were significant differences between the varieties in their reaction to late blight. The first sampling date (D₁) Heiderot, Mariva, Huancayo and Desiree (susceptible control) had mean resistance grades of 3.0, 3.0, 4.0 and 3.0 respectively. At D_A , all the four varieties recorded 5.0. The resistant control variety B53 and KN1/7 had mean resistance grades of 1.0 at D1 and 2.0 at D_A . The other clones ranged between 1.50-2.50 at D_1 and 2.5 at D_A , (Table 3a).

The Duncan's multiple range test indicated that the clone KN1/7 and B53 were not significantly different from each other in their susceptibility to late blight. Similarly KN1/14, Desiree, Dextra, Mariva and Heiderot were not significantly different from each other, but were

Table 3a. Resistance grades of four introduced varieties and seven local clones evaluated at Tigoni

Clone		Resist	tance grad	de (O - 5)	
Number	D ₁	D ₂	^D 3	D ₄	Mean
B53	1.00	1.50	2.00	2.00	1.63ab
Heiderot	3.00	3.00	4.00	5.00	3.75d
KN1/4	1.50	2.00	2.00	2.50	2.00b
Mariva	3.00	3.50	4.00	5.00	3.88d
KN2/2	2.50	2.50	2.50	3.00	2.63c
KN1/12	2.50	2.50	2.50	4.00	2.88c
Dextra	2.50	2.50	4.00	4.50	3.38d
KN1/7	1.00	1.00	1.00	2.00	1.25a
Desiree	3.00	3.00	4.00	5.00	3.75d
Huancayo	4.00	4.00	5.00	5.00	4.50e
KN1/14	2.50	3.50	4.00	4.00	3.50d
Mean	2.50	2.77	3.45	4.00	3.18

Means followed by the same letter are not significantly different from each other, e.g., clones KN2/2 and KN1/12 are followed by the letter c and are therefore not different from each other in their mean resistance grades but different from means followed by the letters a, b, d, and e.

 \mathbf{D}_1 , \mathbf{D}_2 , \mathbf{D}_3 and \mathbf{D}_4 represent first, second, third and fourth sampling dates.

Table 3b. Analysis of variance of infestation grades for the blight at Tigoni.

Source	df	SS	MS	F	
	0.7	320.0000			
Total	87	128.9886			
Reps	4	1.1364	0.2841	1.35875	
Varieties	10	85.11364	8.51136	40.7067*	
Dates	3	26.03409	8.67803	41.5038*	
Varieties					
× Dates	30	8.34086	0.27803	1.3297	U
Error	40	8.3636	0.20909		
					_

S.E. of a variety mean = 0.1617

^{*}Significant at 5% level

significantly different from B53, KN1/7, KN1/4, KN2/2, KN1/12 and Huancayo (Table 3a).

From the overall means, the introduced varieties and local clones can be arranged in the order of resistance as follows: KN1/7, B53, KN1/4, KN2/2, KN1/12, Dextra, KN1/14, Heiderot, Desiree, Mariva and Huancayo.

Yield

The analysis of variance (Table 3d) showed that there were significant differences between the varieties. Using the Duncan's Multiple Range Test it was found that the yield differences between Mariva and Huancayo were not significant. Similarly, yield from Desiree, KN1/12, Heiderot and KN1/14 were not significantly different. B53, KN1/4 and KN2/2 were not different but were significantly different from KN1/7, Desiree, KN1/12, Heiderot, KN1/14, Dextra, Mariva and Huancayo. The clone KN1/7 was significantly different from all the ten clones and varieties (Table 3c). It was the best yielder with a mean of 801 gm/plant compared to 368 gm/plant given by the resistant control variety B53 and 112 gm/plant of the susceptible control variety Desiree. Huancayo which was the most susceptible yielded 12 gm/ plant. The mean yield of the other clones and varieties were as follows KN1/4 (399 gm) KN2/2 (349 gm) Heiderot (144 gm) KN1/12 (136 gm) KN1/14 (104 gm) Dextra (96 gm), and Mariva (32 gm/plant).

Table 3c. The yield of the four introduced varieties and sevel local clones evaluated at Tigoni.

Clone	Yie	eld (gm/plant)	,
number	REP 1	REP 2	Mean
B53	371	365	368 d
Heiderot	126	161	144 c
KN1/4	366	431	399 d
Mariva	026	038	032 ab
KN2/2	320	378	349 d
KN1/12	163	109	136 с
Dextra	071	121	096 bc
KN1/7	822	780	801 e
Desiree	131	093	112 с
Huancayo	012	011	012 a
KN1/14	107	100	104 с
Mean	229	235	

Table 3d. Analysis of variance for the yield of clones tested at Tigoni.

				-
Source	df	SS	MS F	
Total	21	1086323.818		
Varieties	10	1077489.818	107748.9818 125.313	34
Blocks	1	235.6364	235.6364 0.274	10
Error	10	8598.3636	859.8364	

S.E. of a variety mean = 20.73

^{*}Significant at 5% level

C.V. = 12.64

4.1.4. Evaluation of introduced varieties at N.A.L., Nairobi

The four varieties tested at N.A.L., Nairobi under natural infestation in the field were less blighted than at Tigoni. This can be seen from the overall means which ranged between 0.67 to 3.3 (Table 4a). The resistant control variety 853 had an average resistance grade of 0.67 at N.A.L and 1.63 at Tigoni. Desiree, the susceptible control variety had a resistance grade of 3.33 at N.A.L and 3.75 at Tigoni. Mariva had an average grade of 3.0 and 3.88 at N.A.L. and Tigoni, respectively. Huancayo which had a mean resistance grade of 3.0 at N.A.L had 4.50 at Tigoni while Heiderot had 2.83 at N.A.L and 3.75 at Tigoni. Dextra had a mean resistance grade of 1.50 at N.A.L and 3.88 at Tigoni.

The interaction of the varieties and dates of sampling was significant (Table 4b), Desiree was infected earlier than the rest of the clones and varieties. It was group 1 at the first sampling date (D_1) but by D_2 it was a group 3. Clones KN1/7, Dextra and K478/14 were not infected even by D_2 . The clone K477/3 was group 1 at D_2 and D_3 but a group 3 at D_4 (Table 4a).

There were significant differences between the varieties in their susceptibility to blight (Table 4b).

The clone K478/14 and the resistant control variety B53

Table 4a. Comparison of blight resistance of four introduced varieties and seven local clones at N.A.L., Nairobi

Clone	S	ampling d	ates and re	sistance g	rades (0-5)
number	D ₁	02	D ₃	D ₄	Mean
B53	0.00	0.00	1.00	1.00	0.67 a
Heiderot	0.00	2.00	3.00	3.50	2.83 d
Mariva	0.00	3.00	3.00	3.00	3.00 d
Huancayo	0.00	2.00	3.00	4.00	3.00 d
Dextra	0.00	0.00	1.50	3.00	1.50 b
Desiree	1.00	3.00	3.00	4.00	3.33 d
K482/2	0.00	1.50	1.50	3.00	2.00 bc
K478/14	0.00	0.00	0.50	1.00	0.50 a
K479/2	0.00	0.50	1.50	3.00	1.67 bc
K476/21	0.00	1.00	2.50	3.00	2.17 c
K477/3	0.00	1.00	1.00	3.00	1.67 bc
Mean	0.09	1.3	2.0	2.9	

Means followed by the same letter, e.g. those followed by the letters b or bc are not significantly different but are significantly different from those followed by the letters a or d.

Table 4b. The analysis of variance of infection grades for the blight of clones tested at N.A.L., Nairobi

Source	df	SS	MS	F
Total	87	99.9394		
Reps	4	1.3637	0.34093	2.0549
Varieties	10	31.9394	3.19394	14.387*
Dates	3	28.0303	9.34343	56.31626*
Varieties x Dates	30	31.9696	1.06565	6.42306*
Error	40	6.6364	0.16591	

S.E. (Var. Mean) = 0.1228

^{*}Significant at 5% level

were not significantly different in their degree of resistance. Dextra, K477/3, K479/2 and K482/2 were not significantly different from each other but were significantly different from B53, K478/14, K476/21, Heiderot, Mariva, Huancayo and Desiree. Three of the introduced varieties namely Huancayo, Mariva and Heiderot were therefore not significantly different from the susceptible control variety Desiree (Table 4a).

There were significant differences between the mean tuber yield of the clones K479/2, K482/2, K477/3, K478/14 and the yield of the varieties B53, Mariva, Dextra, Desiree, Heiderot and Huancayo. The yield of clone K476/21 differed significantly from the yield of all other clones and varieties except clone K478/14. There were also significant differences between the yield of Huancayo and that of Dextra, Mariva and B53 (Table 4c).

Table 4c. The yield of four introduced varieties and seven local clones tested at N.A.L., Nairobi

Clone	Y	ield (gm/plant)	
number	REP I	REP 2	Mean
DES	407	400	453 c
B53	407	499	184 ab
Heiderot Mariva	145 478	223 386	432 bc
	057	032	045 a
Huancayo - Dextra	312	339	326 bc
Desiree	243	321	282 abc
K482/2	666	859	763 d
K478/14	1030	824	927 de
K479/2	687	901	794 d
K476/21	1145	1000	1073 e
K477/3	885	. 683	784 d
Mean	550	552	

Means followed by different letters are significantly different.

Table 4d. The analysis of variance of the yield of clones tested at N.A.L., Nairobi

Source	df	SS	MS	F
Total	21	2328112.000		
Reps	1	6.5455	6.5455	J
Varieties	10	2219232.000	221923.200	0.0006
Error	10	108873.4545	10887.3455	20.3836

S.E. variety means = 73.78

^{*}Significant at 5% level.

C.V. = 18.94%

4.1.5. Second season screening-Molo

During the second season (Long Rains, 1978) clones tested at Molo showed minor infection (visual observation) throughout the growing season. All the twelve clones with the exception of clones KNI/4 and K476/4 behaved as if they were very resistant (Table 5a).

From the analysis of variance, there were significant differences in the degree of resistance of the clones and sampling dates but the interaction between the clones and dates of sampling was not significant (Table 5 b). At the first sampling date (D $_{\parallel}$) clones KNI/4 and K476/4 were graded as 2.33 and 3.00 respectively. The rest of the clones had mean resistance grades of 1.0 except the clone (K476/15 which was graded as 1.33. At the second sampling date (D_2) the clone KNI/4 was graded as 3.0 while K476/14 was 3.33. The clones K473/5, K473/8, K478/16 and K482/2 were graded as I.O. The clones KNI/7, KN2/2, K478/8, and K479/I and K476/I5 were graded as 1.33. The clone K478/I3 had a mean resistance grade of 1.67. From the overall means, the clone K476/4 was the most susceptible followed by clone KNI/4 (Table 5 a).

The Duncans Multiple Range Test indicate that the clones KNI/7, KN2/2, K473/5, K473/8, K478/8, K478/13, K478/16, K479/1, K482/2 and K476/15 did not show significant differences in their disease scores. These

Table 5a. Mean resistance grades per sampling date of clones tested at Molo.

Clone		Resistance grades (0-5)				
Number	01	D ₂	Mean			
(N1/4	2.33	3.00	2.67 b			
KN1/7	1.00	1.33	1.17 a			
KN2/2	1.00	1.33	1.17 a			
K473/5	1.00	1.00	1.00 a			
K473/8	1.00	1.00	1.00 a			
K476/4	3.00	3.33	3.17 c			
K478/8	1.00	1.33	1.17 a			
K478/13	1.00	1.67	1.34 a			
K478/16	1.00	1.00	1.00 a			
K479/1	1.00	1.33	1.17 a			
K482/2	1.00	1.00	1.00 a			
K476/15	1.33	1.33	1.33 a			
Mean	1.31	1.55				

Table 5b. The analysis of variance for the blight infection grades at Molo

Source	df	SS	MS	F
	7.1	44.5278		
Total	71		0.9028	7.5468*
Reps	4	3.6113		
Varieties	11	33.48608	3.04419	25.4467*
Dates	1	1.1250	1.1250	9.4039*
Varieties x Dates	11	1.0417	0.0947	0.7916
Error	44	5.2637	0.11963	

S.E of a variety mean = 0.1412

^{*}Significant at 5% level

clones were however significantly different from the clones KN1/4 and K476/4. These latter two clones were also significantly different from each other in their blight reaction.

The analysis of variance for yield (Table 5d) showed that there were significant differences between the tuber yield of clones. The Duncans Multiple Range Test indicated no significant differences between, KN1/4, K473/5, K478/8 and K478/13, but they were significantly different from the yield of clones KN1/7, KN2/2, K473/8, K476/4, K478/16, K479/1, K482/2 and K476/15. However, the yield of clone K476/15 and KN2/2 were not significantly different. The yield of clone KN1/7 was significantly different from the yield of all the eleven clones. It out yielded all the clones with a mean yield of 927 gm/plant. This was followed by clone KN2/2 (579gm), K476/15 (515 gm), K482/2 (423gm), K479/1 (367gm), K476/4 (338gm), K478/16 (320gm), K473/8 (303gm), KN1/4 (238gm), K473/5 (221gm), K478/13 (174gm) and K478/8 (164gm/plant), Table 5c).

4.1.6. Second season screening at Kabete and Tigoni

In the second season (Long Rains 1978) 15 clones were further tested for resistance to late blight at Kabete and Tigoni. At Kabete, the means of the resistance grades showed that the infection increased for all the

Table 5c. The yield of clones tested at Molo in the second season

Clone		,	Yield (gm/pla	ent)
number	R ₁	R ₂	R ₃	Mean
KN1/4	207	249	259	238 ab
KN1/7	933	954	894	927 f
KN2/2	553	668	515	579 e
K473/5	275	255	234	221 a
K473/8	294	291	324	303 bc
K476/4	284	351	379	338 с
K478/8	183	138	172	164 a
K478/13	169	177	176	174 a
K478/16	320	294	345	320 c
K479/1	411	321	368	367 cd
K482/2	493	391	386	423 d
K476/15	544	548	452	515 e
Means	389	386	375	

Table 5d. The analysis of variance for the mean yield of clones tested at Molo

Source	df	SS	MS	F
Total	35	1526358.972		
Reps	2	1243.7222	621.8611	0.3355
Varieties	11	1484333.639	134939.4217	72.7943*
Error	22	40781.6108	1853.7096	

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S.E of variety mean = 24.86

^{*}Significant at 5% level

C.V = 11.23%

varieties with advance in the age of the plants. The mean of the first sampling date (D_1) was 1.77 while that of the fourth sampling date (D_4) was 2.90 (Table 6a).

From the analysis of variance of the resistance grades, it was found that there were significant differences between the clones (Table 6b). Using the Duncans Multiple Range Test, clones K478/7, KN1/7, K472/1 and K478/14 were found not to be different from each other. Clones K478/6, B53, K472/1 and K478/14 did not differ significantly in their reaction to blight.

Clones KN2/2, K476/4, K480/1 and K473/1 were not significantly different from each other but they differed from clones KN1/14, K478/6, B53, K472/1, K478/14, K478/7, KN1/7, K481/25 and K480/3. Clones K476/4, K480/1 and K473/1 did not differ from K475/19 and K476/28. These last two clones did not differ from K481/25 and K480/3. The clone KN1/4 differed significantly from all the 14 clones in its reaction to blight.

The same clones when tested at Tigoni showed an increase in the infection with time. The mean of the first sampling date (\mathbb{O}_1) was 0.87 while that of the 3rd sampling date (\mathbb{O}_3) was 2.43 (Table 7a).

Table 6a. Mean resistance grades at different sampling dates and tuber yield of clones tested at Kabete

Clone			Resista	nce grades	(0-5)	
number	D ₁	02	D ₃	D ₄	Mean	Yield (gm/plant)
K478/7	0.00	0.00	1.00	1.00	0.50 a	553 ab
K481/25 ·	3.00	4.00	4.00	4.50	3.88 f	242 a
K478/6	1.50	1.50	1.50	1.50	1.50 b	810 ab
B53	1.00	1.50	1.50	2.00	1.50 Ь	564 ab
K476/4	3.00	3.00	3.50	3.50	3.25 de	560 ab
KN1/4	1.00	2.00	3.00	3.00	2.25 c	816 ab
KN1/7	0.00	0.00	0.50	1.50	0.50 a	1812 c
K480/1	3.00	3.00	3.00	3.00	3.00 de	506 ab
K480/3	3.00	3.00	5.00	5.00	4.00 f	646 ab
K475/19	3.00	3.00	4.00	4.00	3.50 ef	479 ab
K472/1	0.50	1.00	1.00	1.50	1.00 ab	398 ab
K476/28	3.00	3.00	4.00	4.00	3.50 ef	450 ab
K478/14	0.00	1.00	1.50	1.50	1.00 ab	867 b
KN2/2	1.50	3.00	3.50	3.50	2.88 d	421 ab
<u>K</u> 473/1	3.00	3.00	3.00	4.00	3.25 de	298 ab
	1 44	0.10	2 67	ים כ	2.37	0.628

Table 6b. The combined analysis of variance for the blight infection grades at Kabete

Source	df	SS	MS	F
Total	119	227.78		
Reps	4	0.60	0.15	0.58
Varieties	14	175.62	12.54	48.79*
Dates	3	23.67	7.89	30.70*
Varieties × Dates	42	13.58	0.323	1.26
Error	56	14.40	0.257	

S.E of a variety mean = 0.179

^{*}Significant at 5% level.

Table 7a. Mean resistance grades at different sampling dates and tuber yield of clones tested at Tigoni

Clone	Sampling dates and resistance	
	groups O-5 scale	

number	D ₁	D ₂	D ₃	Mean :	Yield(gm/plant)
K478/7	0.50	1.50	1.50	1.17 abc	392 ab
K481/25	2.50	3.50	3.50	3.17 g	206 a
K478/6	0.00	1.00	1.00	0.67 a	528 bc
B53	1.50	2.00	2.00	1.83 de	231 a
K476/4	2.00	3.00	3.50	2.83 f	269 a
KN1/4	0.00	2.00	3.00	1.67 de	579 c
KN1/7	1.00	2.50	2.00	1.83 de	678 c
K480/1	0.50	2.00	3.00	1.83 de	329 a
K480/3	1.00	2.00	3.00	2.00 de	382 ab
K475/19	0.00	2.00	3.00	1.67 cde	226 a
K472/1	1.00	1.00	1.50	1.17 abc	307 a
K476/28	0.50	2.00	2.50	1.67 cde	290 a
K478/14	1.00	1.00	1.00	1.00 ab	347 a
K N12 /2	0.00	1.50	3.00	1.50 bcd	295 a
				2 17 2	383 ab

The analysis of variance for the resistance grades showed that the interaction between the clones and the sampling dates was significant and therefore the infection rate of all the varieties was not the same. There were also significant differences between the clones (Table 7b). There were no significant differences between clones K478/6, K478/14, K478/7 and K472/1 but they differed significantly from clones K481/25, K476/4, K473/1, KN1/4, KN1/7, B53, K480/1 and K480/3. Clones K476/4 and K481/25 differed significantly from all the 14 clones. Clones KN1/4, KN1/7, K480/1, K480/3, B53, K473/1, K476/28 and K475/19 were not significantly different from each other. The clone KN2/2 did not differ from clones K478/7, K472/1, K478/14, K475/19, K476/28, B53, KN1/4, KN1/7, K480/1 and K480/3 but differed from K481/25, K476/4, K473/1 and K478/6.

The tuber yield of the 15 clones was generally higher at Kabete than Tigoni (Tables 6a and 7a). The clone KN1/7 recorded the highest yields at both sites, and the clone K481/25 the lowest. The clone KN1/7 had a mean yield of 1812gm at Kabete and 678gm/plant at Tigoni, while clone K481/25 had a mean yield of 242 gm at Kabete and 206gm/plant at Tigoni. The mean yields at Kabete showed that clones K481/25, K478/7, K478/6, B53, K476/4, KN1/4, K480/1, K480/3, K475/19,

Table 7b. The combined analysis of variance for the blight
Infection at Tigoni

Source	df	SS	MS	F
			7.1. 1.0267.2	GENERAL SERVICE
Total	89	99.12		
Reps	3	0.30	0.100	0.457
Varieties	14	35.62	2.544	11.616
Dates	2	38.42	19.210	87.717*
Varieties				
x Dates	28	15.58	0.556	2.539
Error	42	9.20	0.219	

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S.E of a variety mean = 0.19

^{*}Significant at 5% level

K472/1, K478/28, KN2/2 and K473/1 were not significantly different from each other but were significantly poorer than clone KN1/7. The tuber yield of clone K478/14 was significantly different from that of clones KN1/7 and K481/25 (Table 6a).

At Tigoni, the tuber yield of clones K481/25, B53, K476/4, K480/1, K475/19, K472/1, K476/28, KN2/2, K478/7, K480/3, K478/14 and K473/1 were not different but they were different from clones KN1/7 and KN1/4 in their tuber yield. Similarly clone K478/6 was not significantly different from clones KN1/4, KN1/7, K478/7, K480/3, and K473/1 but was significantly different from clones K481/25, B53, K476/4, K480/1, K475/19, K472/1, K476/28 and KN2/2 (Table 7a).

From the combined analysis of variance (Table 8)
the interaction between the clones and site was
significant.

4.1.7. Third season screening (Long Rains 1979)

Six clones were tested at Kabete and Molo. The mean infection grades per sampling date are given in Tables 9a and 10a. From the analysis of variance for the blight infestation at Kabete, the interaction between the treatments and dates of sampling was significant at 5% level (Table 9b). This means that the infection rate was not the same for all clones. For instance the

Table 8. Combined analysis of variance for the yield of clones tested at Kabete and Tigoni

Total 59 6474157.600 Reps 2 56346.1333 2817 Varieties 14 3365728.600 24040 Sites 1 1054965.600 10549	The series in the series
Reps 2 56346.1333 2817 Varieties 14 3365728.600 24040 Sites 1 1054965.600 10549 Varieties ×	MS F
Varieties ×	73.0666 0.8613 409.1857 7.3499*
1001267 400 777	4965.600 32.2531
Error 28 915849.8667 32	7233.3857 2.3612* 2708.9238

S.E of a variety mean = 90.43

^{*}Significant at 5% level.

Table 9a. Mean resistance grades per sampling date of clones tested at Kabete

Clone			Sampling	dates and re	esistance gra	des (0-5 sca	ale)
	01	D ₂	D ₃	D ₄	^D 5	D ₆	Mean
KN1/4	0.75	1.25	1.25	3.00	3.00	3.00	2.04 d
KN1/7	0.00	0.00	0.00	2.00	2.00	2.00	1.00 b
KN2/2	. 0.00	0.00	0.00	1.00	1.00	1.00	0.50 a
K477/1	0.25	0.50	1.00	2.50	2.75	2.75	1.62 c
K478/5	0.00	0.25	0.50	1.50	2.00	2.25	1.12 ь
B53	0.75	1.75	1.75	2.75	3.50	3.50	2.33 е
Mean	0.29	0.62	0.75	2.12	2.38	2.46	1.44

Table 9b. The analysis of variance for the blight infection at Kabete

Source	df	SS	MS	F
Total	143	197.4400		
Reps	18	3.8166	0.2120	1.6695
Varieties	5	56.8983	11.3797	89.6036*
Dates	5	116.1483	23.2297	182.9107*
Varieties × Dates	25	9.1450	0.3658	2.8803*
Error	90	11.4318	0.1270	

S.E of a variety mean = 0.0727

^{*}Significant at 5% level

clones KN2/2 and KN1/7 had no infection till the fourth sampling date ($\mathbf{D_4}$), while clones KN1/4, B53, K477/1 got infected at $\mathbf{D_1}$. The clone K478/5 showed symptoms of infection at $\mathbf{D_2}$ and the infection increased gradually to 2.25 at $\mathbf{D_6}$. B53 and clone KN1/4 had the same mean resistance grade of 0.75 at $\mathbf{D_1}$ but by $\mathbf{D_4}$ clone KN1/4 had a mean resistance grade of 3.00 while B53 had 2.75. At $\mathbf{D_6}$, B53 and KN1/4 had mean resistance grades of 3.50 and 3.00 respectively. B53 and the clone KN1/4 were the most heavily infected followed by the clones K477/1, K478/5, KN1/7 and KN2/2.

From the analysis of variance, the treatments were significantly different at 5% level (Table 9b).

The overall means of the resistance grades when compared using the Duncans Multiple Range Test showed that clones KN1/7 and K478/5 were significantly different from clones KN1/4, KN2/2, K477/1 and B53.

The tuber yield of the six clones tested at Kabete is given in Table 9c. The analysis of variance (Table 9d) showed that there were significant differences between the varieties as well as the replicates at 5% level. The Duncans Multiple Range Test indicated that the tuber yield of B53 and clone KN1/4 were not significantly different. Clones KN1/7, KN2/2, K477/1 and K478/5 were significantly different from B53. The tuber yield of

Table 9c. The yield (gm/plant) of the six clones tested at Kabete.

Clone		Rep	lication		
number	I	II	III	IV	Mean
KN1/4	539	689	765	505	575 ab
KN1/7	665	555	789	578	649 bc
KN2/2	925	674	668	685	738 bc
K477/1	735	694	700	500	657 bc
K478/5	885	653	990	663	798 c
· B53	500	341	500	517	465 a
Mean	708	601	702	576	

Table 9d. Analysis of variance for the tuber yield of clones tested at Kabete.

S	1.6	00	MS	F
Source	df	SS	115	
T-4-3	2.2	F15475 223		
Total	23	515475.333	27799.222	2.7189
Reps	3	83397.667		
Treatments	5	278711.833	55742.367	5.4519*
Error	15	153365.833	10224.389	

S.E of a variety mean = 50.56

C.V = 15.63%

^{*}Significant at 5% level.

clone KN1/4 was not significantly different from that of clones KN1/7, KN2/2 and K477/1 but significantly different from clone K478/5.

In Molo, the infection was generally low (Table 10a). The rate of infection however, increased slightly with the sampling dates (Table 10a). The analysis of variance for the blight infestation is given in Table 10b. At 5% level, the interaction between the clones and the sampling dates was significant indicating infection rate was not the same for all the clones. For instance at D2 clone KN2/2 had a lower resistance grade (2.50) than clone KN1/7 (0.50). Similarly clone K477/1 had a higher grade (0.75) than B53 (1.25) and clone K478/5 (1.25). B53 which had a mean resistance grade of 1.25 at D2 recorded an resistance grade of 2.00 at D₃, but clone KN1/7 which had an resistance grade of 0.50 at D_2 increased to 2.0 at D_3 . This shows that the infection rate of clone KN1/7 was higher between the two sampling dates than that of B53. The treatments were not significantly different (Table 10a). The overall means ranged between 1.25 to 1.75.

The tuber yield of the six clones planted at

Molo is given in Table 1Oc. The analysis of variance

(Table 1Od) showed that there were significant differences

between the varieties. Applying the Duncans Multiple

Table 10a. Mean resistance grades per sampling date of clones tested at Molo.

Clone	Sa	mpling dat	es and resist	
number	D ₁	D ₂	D ₃	Mean
KN1/4	0.25	1.00	2.25	1.62 a
KN1/7	0.00	0.50	2.00	1.25 a
KN2/2	0.00	2.50	1.00	1.75 a
K477/1	0.75	0.75	2.00	1.38 a
K478/5	0.00	1.25	2.00	1.62 a
B53	0.00	1.25	2.00	1.62 a
Mean		1.21	1.88	

Table 10b.. The analysis of variance for the blight infection of clones tested at Molo.

Source	df	SS	MS	F
Total	47	31.9167		
Reps	6	2.2499	0.3750	1.0466
Varieties	5	1.4167	0.2833	0.7908
Dates	1	5.3334	5.3334	14.8853*
Varieties x Dates	5	12.1666	2.4333	6.7913*
Error	30	10.7501	0.3583	

S.E. (Variety mean) = 0.2116

^{*}Significant at 5% level.

Table 10c. The tuber yield (gm/plant) of the six clones tested at Molo.

Clone		Replication				
number	I	II	II	IV	Mean	
K478/5	775	700	813	763	763 b	
K477/1	1057	1061	844	875	959 c	
KN2/2	1074	957	1058	935	1006 c	
KN1/4	658	633 ,	600	675	642 b	
KN1/7	694	536	867	733	708 Ь	
B53	371	526	539	425	465 a	
	771	736	787	734		

Table 10d. The analysis of variance for the yield of clones tested at Molo

Source	df	SS	MS	['] F
Total	23	955568.9583		
Reps	3	12457.7917	4152.5972	0.4877
Varieties	5	815390.7083	163078.1417	19.1525*
Error	15	127720.4583	8514.6972	

[.]S.E (variety mean) = 46.14

C.V = 12.19%

Range Test, the yield of B53 was found to be significantly different from that of clones KN1/4, KN1/7, K478/5, K477/1 and KN2/2. Clones KN1/4, KN1/7 and K478/5 were not significantly different from each other though their tuber yield differed significantly from the yield of clones K477/1 and KN2/2 (Table 10c).

4.2.1. Screening of seedlings under laboratory conditions for resistance to late blight

Seedlings obtained from nine different crosses were screened under artificial infestation for resistance to late blight and results are given in Table 11. The highest frequency of resistant progenies were recovered from crosses whose parents were group III and I and group I and II. The combination of group III and I parents yielded between 18-35% resistant progenies. Cross number K920 had 35% resistant progenies followed by K916 with 30%, K914 with 25%, K482 with 24%, K913 with 20%, and K915 with 18%. Cross numbers K917, K918 and K919 had only susceptible (group IV) and very susceptible (group V) progenies.

4.2.2. Leaf inoculation

Leaves of selected clones were inoculated and the percentage leaf area infected was estimated (Table 12a). Period and degree of sporulation were also recorded.

Table 11. Distribution of seedlings in different reaction groups (I-V)

cross		Percentage	Reaction of parents	No. of seedlings tested	No. of seedlings (%) in reaction groups				
					I	II	III	IV	V
(482	K015N/12	× KO1N/13	III × I	1087	1	3	20	49	27
(913	P95	x R96 (6)	I × II	340	4	4	12	67	13
<914	K013N/12	x K80a(19)	III × I	313	2	3	20	65	10
K915	S45	x R88 (1)	III × I	91	0	3	15	26	55
K916	K013N/5	x K79a (1)	III × I	335	2	6	22	48	23
K917	7653 a(5)	× 66-646 (3)	II × -	32	0	0	0	31	69
K918	R96 (1)	× S42	- × -	16	0	0	0	94	6
K919	Atzimba	× K113/36	I × III	15	0	0	0	67	33
K920	Roslin Ruaka	a x Montserrate	III × I	17	6	0	29	12	53

⁻ blight reaction not known

4.2.2.1. Incubation period

The incubation period for most of the clones ranged between three to four days except K478/7 and K478/15 whose incubation period was two days. Kerrs pink (the susceptible control) and clone K478/16 had an incubation period of 3 days. The incubation period for clones K472/1, K478/5, K473/8, K476/4, K476/9, K480/1 and K482/2 was 4 days while B53 (the resistant control) and Dextra had an incubation period of 5 days. Clones K476/13 and K476/19's incubation period was 7 days while that of K478/14 was 6 days.

4.2.2.2. Sporulation period

Clones K472/1, K478/7, K478/15, K478/16, Dextra and Kerrs pink sporulated by the 5th day after inoculation. Clone K473/8 sporulated on the 6th day while K476/13 sporulated on the 9th day. Clones K473/5, K476/4, K476/9, K476/19, K480/1, K482/2 and B53 did not sporulate. These clones showed tiny specks on their leaves followed by rapid disintegration of the cell walls of the leaves. The process took one or two days for the entire leaf to rot completely. Clone K473/5 started rotting on the 5th day after inoculation while K476/4 and B53 started rotting on the 7th day after inoculation. K476/9 and K482/2 started rotting on the 8th day and K476/19, and K480/1 on the 9th day after inoculation (Table 12a).

Table 12a. Rate of infection expressed as percentage leaf area infected and the period of sporulation in potato leaves inoculated with Phytophthora infestans.

Clone number		Percentage laaf area infected after inoculation							Period of sporulation (days)	Degree of sporulation	
1	1	2	3	4	5	6	7	8	9		
K472/1	_	-		1	25	50	80	97	100	5	Profuse
K473/5	_	-	_	ī	75*	100				_	None
K473/8	_	_	_	1	1	25	60*	100		6	Sparse
K476/4	_	-	_	<1	10	25	60*	100		_	None
K476/9	-	-	_	1	2	5	10	95*	100	_	None
K476/13	_	_	_	_	-	-	<1	3	25	9	Sparse
K476/19	_	-	-	-	-	-	<1	2	50*	_	None
K478/7	-	:<1	10	30	50	75	80	100*		5	Sparse
K478/15	-	2	15	45	75	80	90	100*		5 5	Profuse
K478/16	-	-	5	30	75	80	100	100	5		Sparse
K480/1	-	-	-	1	2	15	50	50	100*	-	None
K482/2	-	-	-	<1	1	1	1	60*	100		None
Dextra	-	-	-	-	1	25	50	75	100*	5	Profuse
B53 Kerrs	-	-	-	-	1	1	7*	45	60	-	None
Pink	_	_	<1	4	2	5	55*	60	100	5	Profuse
K478/14	-	7	-	1	1	1	3	10	15		None

^{*}leaf started rotting

The degree of sporulation varied between sparse and profuse. Clones K472/1, K478/15, Dextra and Kerrs pink sporulated profusely while clones K478/16, K478/7, K476/13 and K473/8 had sparse sporulation.

4.2.2.3. Degree of resistance and the amount of sporulation

Distal and terminal leaflets of some selected clones were inoculated and the lesion size was measured in $\rm cm^2$. The amount of sporulation was expressed as sporangia/ml (Table 12b). Clone KN1/4 had the smallest lesion size of 2.34cm² followed by KN1/7 (3.04cm²), B53 (3.10cm²), KN2/2 (4.02 cm²) and Kerrs pink (4.18 cm²). The clone K478/14 recorded the largest lesion size of 6.28 cm².

Despite the larger lesion size exhibited by clone K478/14, the amount of sporulation recorded was only 11 sporangia/ml. as compared to 38 sporangia/ml recorded for Kerrs pink. Clones KN1/4 recorded 29 sporangia/ml, KN2/2 had 20 sporangia/ml, B53 10 sporangia/ml and KN1/7 had 9 sporangia/ml.

4.2.3. Anatomical studies

The length and thickness of the epidermal cells of some selected clones were measured in microns to find out if there was any relationship between their thickness and the degree of resistance to late blight.

Table 12b. The area of lesions in cm² and the number of sporangia per ml. on inoculated leaflets of some potato varieties.

number	Days after inoculation and lesion size cm ²			Mean lesion size	Period of sporulation (days)	No. of sporangia/	Resistance level
	4th	5th	6th	(cm ²)			
K478/14	4.79	6.49	7.56	6.28	4	11	R
Kerrs Pink	2.28	3.59	6.66	4.18	3-4	38	S
KN1/7	2.10	2.55	4.48	3.04	4	9	R
KN1/4	1.50	1.70	3.82	2.34	4	29	MR
B53	1.95	2.80	4.54	3.10	3-4	10	R
KN2/2	3.45	3.48	5.15	4.02	4	20	MR

The thickness of the epidermal cells of the leaf of one month old plants ranged between 14 and 19 microns (Table 13). B53 the resistant controlhad an epidermal thickness of 14 microns compared to Kerrs pink's 17 μ thick. The clones K479/1 measured 19 μ , K472 measured (17.4 μ), K478/14 and KN1/7 had epidermal cells measuring 17 μ in thickness. KN1/4 measured 15 μ while KN2/2 measured 14 μ . In the two month old samples, B53 had a thicker epidermis (24.4 μ) than KN2/2 (22.5 μ), Kerrs pink (22 μ), K479/1 (21.6 μ), K478/14 (20.3 μ), K472/1 (18.6 μ), KN1/7 (18.1 μ), and KN1/4 (17.1 μ).

The epidermal cells of the stem of B53 measured 21.8 μ and 28 μ thick in one and two month old samples, respectively. The thickness of the epidermal cells for the other clones were as follows: K472/1 (23.8 μ), K479/1 (22 μ), KN1/4 (21.9 μ), KN2/2 (21.3 μ), KN1/7 (20.7 μ), Kerrs pink (20.4 μ) and K478/14 (16 μ) in one month old samples. The results of the two month old samples were as follows: Kerrs pink (28 μ), K472/1 (27.3 μ), K479/1 (25.2 μ), KN1/4 (24.4 μ), KN2/2 (23.8 μ), KN1/7 (21.8 μ) and K478/14 (20.0 μ).

9 1

Table 13. Length and thickness of epidermal cells given in microns

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160 E a

Clone Leaf Stem 1 month old 2 months old 1 month old 2 months old Resistance Length Width Length Width Length Width Length Width grade B53 2 24.0 14.0 38.4 24.4 29.6 21.8 34.0 28.0 Kerrs Pink 5 33.0 17.0 33.0 22.0 30.4 32.0 28.0 20.4 KN1/4 3 26.5 15.0 28.3 17.1 27.7 34.4 24.4 21.9 KN1/7 2 26.0 17.0 28.5 18.1 31.2 20.7 32.4 21.8 K478/14 2 26.0 28.0 20.3 17.0 26.0 16.0 40.2 20.0 KN2/2 3 '20.9 14.0 23.8 22.5 25.6 38.6 23.8 21.3 K479/1 1 25.0 19.0 27.6 21.6 32.5 22.0 34.3 25.2 K472/1 2 27.0 17.4 361. 18.6 30.7 23.8 33.0 27.3

Fig. 3. Comparison of the thickness of epidermal cells in one month old leaves of K479/1, B53, and Kerrs pink.

Top: K479/1 (x 1394.2)

Middle: B53 (x 2245.1)

Bottom: Kerrs pink (x 2247.7)

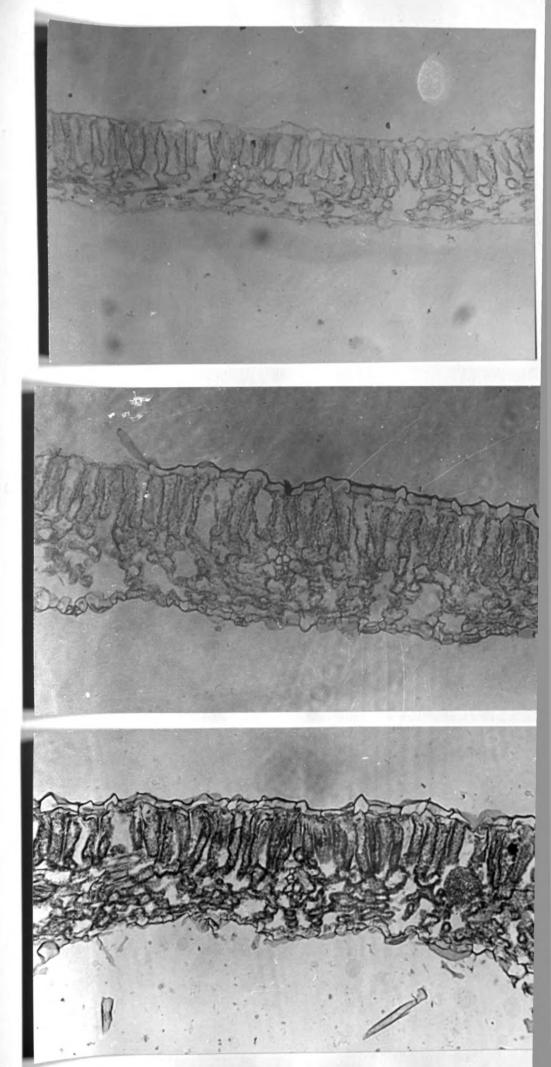
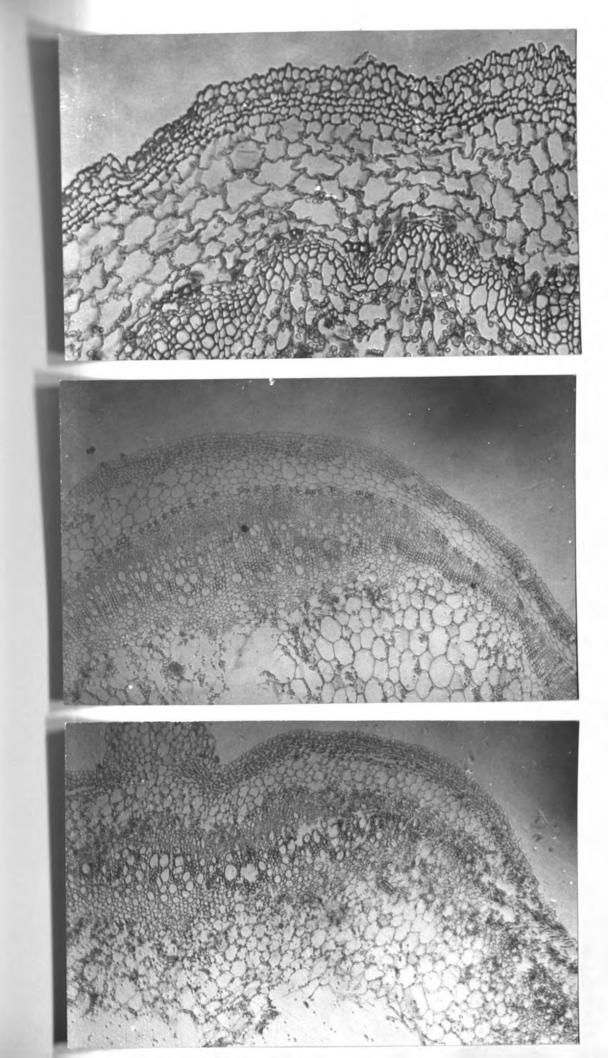


Fig. 4. Comparison of the thickness of epidermal cells in one month old stems of clone K479/1, B53 and Kerrs pink.

Top: K479/1 (x 1411.2)

Middle: B53 (x 438.5)

Bottom: Kerrs pink (437.7)



4.2.4. Total Phenolics in healthy leaves

Results of the ethanol extraction (a) indicated that the clone KN1/7 had the highest amount of total phenolics (1225 μ g) followed by clones KN2/2 with 894.375 μ g, clone K478/5 (866.45 μ g), KN1/4 (847.3 μ g), K477/1 (830.5 μ g), B53 (769.8 μ g), and Kerrs pink (335.34 μ g/g tissue).

Results of the ethanol extraction of the homogenates (b) are as follows: Kerrs pink (437.5 μ g), KN1/7 (275 μ g), K477/1 (252.5 μ g), K478/5 (162.4 μ g), B53 (160.125 μ g), KN2/2 (126.25 μ g), and KN1/4 (102.63 μ g/g tissue).

Acetone extraction (c) gave the highest amounts of phenolics in the leaves. Results were as follows: Clone KN1/7 (2053.35 μ g), K478/5 (1628 μ g), KN1/4 (1375 μ g), KN2/2 (1167.25 μ g), K477/1 (1134 μ g), B53 (814 μ g) and Kerrs pink (290.95 μ g /g tissue).

The average results, (Table 14) indicated that clone KN1/7 had the highest amount of phenolics (1184.45 μ g/g tissue), followed by K478/5 (885.617 μ g), KN1/4 (774.977 μ g), K477/1 (739 μ g), KN2/2 (729.292 μ g), B53 (581.308 μ g) and Kerrs pink (354.596 μ g /g tissue).

Table 14. Phenolic compounds in healthy potato leaves (µg/g tissue)

Clone		Extraction	n methods	
number	(a)	(b)	(a)	Average
KN1/7	1225	275	2053.35	1184.45
KN1/4	847.3	102.63	1375	774.977
KN2/2	894.375	126.25	1167.25	729.292
.K477/1	830.5	252.5	1134	739.0
K478/5	866.45	162.4	1628	885.617
B53	769.8	160.125	814	581.308
Kerrs pink	335.34	437.5	290.95	354.596

⁽a) Ethanol extraction of tissue

⁽b) Ethanol extraction of homogenates

⁽c) Acetone extraction in the cold

4.2.5. Resistance grouping of clones

Based on field and laboratory tests clones

KN1/7, K478/14 and B53 could be classified as resistant.

The field rating was 2.0, 1.0 and 2.0, respectively

while laboratory grades were 1.0, 2.0 and 1.8,

respectively. Area infected 7 days after inoculation

was 5% for KN1/7, 7% for B53 and <1% for clone K478/14.

These three clones sporulated on the 4th day after

inoculation except B53 in one experiment sporulated on

the 3rd day after inoculation. Sporulation was sparse

for all the three clones as follows: KN1/7 (9 sporangia/

m1), K478/14 (11 sporangia/m1) and B53 (10 sporangia/m1)

(Table 5). Despite the small number of sporangia

produced in the case of clone K478/14, this clone had

the largest lesion size (6.28 cm²).

The clones KN1/4 and KN2/2 were graded as 3.0 in the field as well as in the laboratory. The two clones could be classified as moderately resistant. The susceptible control Kerrs pink was group IV in the field and group V in the laboratory screening.

The area infected 7 days after inoculation was as follows: KN1/4 (50%), KN2/2 (60%), Kerrs pink (55%).

Period of sporulation was 4 days after inoculation except

Kerrs pink which sporulated 3 days after inoculation

in one experiment.

The three clones sporulated profusely and the average number of sporangia/ml was as follows: KN1/4 (29).

KN2/2 (20) and Kerrs pink (38).

KN1/4 had the least lesion size $(2.34 \, \text{cm}^2)$, KN2/2 had a lesion size of $4.02 \, \text{cm}^2$ and Kerrs pink $(4.18 \, \text{cm}^2)$ (Table 15).

4.2.6. Tuber resistance

Tubers of some selected clones inoculated with a sporangial suspension of Phytophthora infestans
showed some differences in their degree of resistance
(Table 16). Clone K477/1 was the most susceptible
(grade 4) as the discolouration was more than 5mm deep.
Kerrs pink was less susceptible than K477/1 with a resistance grade of 3.0. The other clones were fairly resistant. Clone K478/5 had a mean resistance grade
of 2.2, the clones KN1/4, KN1/7, KN2/2 and B53 had the same resistance grade (1.6).

4.2.7. Dry matter and protein contents in tubers

Estimates of dry matter content and protein

content of some selected clones are given in Table 17.

In general the protein content and dry matter content

was higher for Molo samples as compared to Kabete except

clone K478/5 which showed a decrease in the protein content.

However, at a particular site the increase in the dry

matter content did not follow a corresponding increase in

Table 15. Classification of clones into resistant, moderately resistant and susceptible grades based on field and laboratory scores.

Clone number	Field rating (1-5)	Lab. rating (1-5)	Percentage of area infected 7 days inoculation	Period of sporulation (days)	Degree of sporulation	Spores/ ml	Lesion size	Ratira
KN1/7	2.0	1.0	5	4	Sparse	9	3.04	R
B53	2.0	1.8	7	3 - 4	non-sparse	10	3.10	R
K478/14	1.0	2.0	<1	4	Sparse	11	6.28	R
KN1/4	3.0	3.0	50	4	Profuse	29	2.34	MR
KN2/2	3.0	3.0	60	4	Profuse	20	4.02	MR
Kerrs Pink	4.0	5.0	55	3 - 4	Profuse	38	4.18	S

R = Resistant, MR = Moderately resistant, S = Susceptible

Table 16. Resistance grades of inoculated tubers

Clone number	Mean resistance grades
K477/1	4.0
B53	1.6
K478/5	2.2
KN1/4	1.6
KN1/7	1.6
KN2/2	1.6
Kerrs Pink	3.0
	10 2 2 2 3 7 1

Grade 1 - No response to infection

2 - Penetration less than 2 mm.

3 - Penetration less than 5 mm.

4 - Penetration more than 5 mm.

Table 17. Dry matter content and protein content in potato tubers

Clone	Dry	matter cont	% (Crude) protein content				
number	Kabete	Molo	Mean	Kabete	Molo	Mean	
B53	21.7	23.3	22.5	8.94	9.44	9,19	
K477/1	19.5	22.4	21.0	8.25	9.13	8.69	
K478/5	22.8	23.7	23.3	8.06	7.00	7.53	
KN1/4	21.5	21.7	21.6	9.44	10.69	10.08	
KN1/7	22.0	22.2	22.1	9.44	10.13	, 9.79	
KN2/2	21.2	24.1	22.7	8.75	11.13	9.94	

the protein content for each clone. The dry matter for Molo samples were as follows: Clones KN2/2 (24.1%), K478/5 (23.7%), B53 (23.3%), K477/1 (22.4%), KN1/7 (22.2%) and KN1/4 (21.7%). For Kabete samples, the dry matter content was slightly lower as the following figures show: K478/5 (22.8%), KN1/7 (22.0%) B53 (21.7%), KN1/4 (21.5%), KN2/2 (21.2%) and K477/1 (19.5%). The dry matter content ranged between 19.5% to 22.8% for Kabete samples and 21.7 - 24.1% for Molo samples. The clone K477/1 had the lowest and the clone K478/5 had the highest dry matter content for Kabete samples while the clone KN1/4 and KN2/2 had the lowest and highest dry matter content for Molo, respectively.

The protein content ranged between 7.0% to 11.13% for Molo. The clone KN2/2 had the highest protein content (11.13%) while the clone K478/5 had the lowest (7%). Clones KN1/4 had 10.69%, KN1/7 (10.13%), B53 (9.44%) and K477/1 (9.13%). The protein content of samples from Kabete were as follows: the clones KN1/4 and KN1/7 had 9.44% each, B53 had 8.94%, KN2/2 had 8.75%, K477/1 had 8.25% and K478/5 had 8.06%. For both sites clone K478/5 recorded the lowest protein content (7% and 8.06%).

4.2.8. Texture, colour and flavour

The texture of the cooked tubers did not follow the amount of dry matter recorded. A potato with a high dry matter content is expected to be floury but this was not the case. The clones K478/5, KN2/2 and B53 had the highest dry matter content (23.3, 22.7 and 22.5% respectively) on average but when cooked they were waxy (Table 18). The clones KN1/7 and K477/1 were rather floury and their dry matter content was 22.1 and 21.0%, respectively. The clone KN1/4 with a dry matter content of 21.6 comparable to that of clone K477/1 was medium waxy.

The tubers of clones KN1/4, KN1/7, KN2/2 and B53 had white flesh while those of K478/5 had whitish-yellow and K477/l had yellow flesh. The flavour of the cooked tubers of clones KN1/7 and K477/l were good as compared to those of B53, K478/5, KN1/4 and KN2/2 which were fair (Table 18).

Table 18. Quality aspect of cooked tubers

Clone	Texture	Colour	Flavour
KN1/4	medium waxy	white	fair
KN2/2	rather waxy	white	fair
KN1/7	rather floury	white	good
K477/1	rather floury	yellow	good
K478/5	medium waxy	whitish-yellow	fair
B53	medium waxy	white	fair

CHAPTER 5

DISCUSSION

5.1. Reaction of clones to late blight in the field and laboratory screening

Results of the preliminary screening (Table 1) indicated that the laboratory testing of clones was more severe than the field screening for separating susceptible lines from resistant ones. Under field conditions, 64% of the clones tested were grouped between group I to III as compared to only 10% under laboratory conditions. This difference could be attributed to uniform inoculum and the ideal conditions, i.e., lower temperatures and higher relative humidity in the laboratory screening.

The introduced varieties were found highly susceptible to late blight. Although the blight was less severe at Kabete, the Tigoni results showed that in certain seasons, due to the high blight incidence, the yield of these varieties could be drastically reduced. The difference in the severity of the disease at Kabete and Tigoni could have been due to differences in the weather conditions. For instance, Tigoni received an average of 188 mm of rain as compared to 160 mm at

Kabete during the sampling period. The mean temperatures for Tigoni were lower (15.9 - 16.5° C) than those of Kabete (18.3 - 18.4° C). At both sites, Huancayo was the most susceptible followed by Mariva, Heiderot and Dextra.

The incubation period of B53, K478/14 and Dextra was much longer than that of the other clones at Kabete. In case of Dextra, however, the defoliation was rapid after infection. There was no blight for the first and second sampling dates (D, and D,) but at D, Dextra had a mean resistance grade of 1.5 and reached 3.0 at D. This could have been due to reduced initial inoculum or because of little or no horizontal resistance to slow down the attack of blight. Clone K478/14 and B53 showed no symptoms of blight at D_1 and D2 but got infected before D3 and they had a resistance grade of 1.0 at D4. Hence the rate of infection was slow suggesting the presence of horizontal resistance. The clones K482/2, K479/2 and K477/3 were not as susceptible as the introduced varieties and Desiree, the susceptible control. Their resistance score was 3.0. These three clones appear to have some level of horizontal resistance.

At Tigoni, B53 and KN1/7 showed a gradual rate of infection. Infection grades of B53 and KN1/7 indicated that they were not significantly different.Clones

KN1/4 and KN2/2 were less blighted than the introduced varieties and clones KN1/12 and KN1/14. Clones KN1/4 and KN2/2 could have had some level of horizontal resistance while KN1/7 and 853 had higher levels of horizontal resistance. These results agree with the findings of Lapwood (1961) and Van der Plank (1963). They reported that field resistant varieties were destroyed more slowly during an epidemic or disease caused less damage than in susceptible varieties.

At Tigoni, the best tuber yielder was the clone KN1/7 (801 gm/plant) followed by clones KN1/4 (399 gm/ plant) and B53 (368 gm/plant). The tuber yield of the clone KN1/4 was slightly more than that of B53 although it was more susceptible (grade = 2.0) than B53 (grade = 1.6). At Kabete, the outstanding clone in yield was the clone K476/21 (1073 gm/plant) and its blight resistance grade was 2.7. B53, the resistant control, had a mean resistance grade of 0.67 and its yield was 453 gm/plant. Clones K482/2, K479/2 and K477/3 also gave higher tuber yields than B53 although they had more blight infestation than B53. This indicated that these clones had better yielding capacity than B53 and were fairly tolerant to late blight. The results also indicated that the yield was affected by other factors other than late blight. Huancayo had the lowest tuber yield of 45 gm and 12 gm/plant, respectively at Kabete

and Tigoni (Tables 3c and 4c). In this case the yield reduction could have been due to its higher susceptibility to late blight. At Kabete, it was almost bare of leaves while at Tigoni the haulms were completely dead by the 3rd sampling date and hence the least yield.

During the second season, clones tested at Molo had very little blight compared to those tested at Kabete and Tigoni (Tables 5a, 6a and 7a). The outstanding resistant clones to late blight at Molo were K482/2, K478/16, K473/5 and K473/8 all with a mean of 1.0. most susceptible were clones K476/4 and KN1/4 with mean resistance grades of 3.17 and 2.67, respectively. Clone K476/4 at Kabete had a mean resistance grade of 3.25 and 2.83 at Tigoni. This clone can be grouped as tolerant (group III) for all sites. Clone KN1/7 was classified as group I in Molo and Kabete and was almost within group II in Tigoni. Thus KN1/7 showed resistance at all the three sites. The clone K481/25 was susceptible both at Kabete and Tigoni. The other resistant clones at Kabete and Tigoni were K478/6, K478/14, K478/7 and K472/1. There was not much difference in the mean resistance grades at Kabete and Tigoni. Clone KN1/4 was more susceptible in Molo (2.67) than in Kabete (2.25) and Tigoni (1.67).

The poorest yielder was the clone K481/25 both at Tigoni (206 gm/plant) and Kabete (242 gm/plant). The best yielder at Tigoni and Kabete was the clone KN1/7 yielding 678 gm and 1812 gm/plant), respectively. It is interesting to note that the yields of clone KN1/7 and K478/14 at Kabete were twice as much as those at Tigoni. This may be due to the fact that Tigoni had very little rain compared to Kabete. Again at Molo, the clone KN1/7 had the highest tuber yield (894 gm/plant).

During the third season, clone KN2/2 tested at Molo and Kabete had very little blight on the foliage. In both cases, it behaved as if it was immune. At Kabete, it had a mean resistance grade of 0.5 and at Molo 1.75. The resistance grade was higher at Molo because of the stem blight recorded during the second sampling date (O2). At D3, no stem blight was obvious therefore the clone was given a group I (Table 10a). The clone KN2/2 therefore appears to have a major gene for resistance. The clone KN1/7 was the second best for resistance to late blight (resistance grade 1.0) at Kabete. At Molo, it was the most resistant clone to late blight (mean resistance grade 1.25) followed by the clone K477/1 (1.38).

At Kabete B53 and clone KN1/4 were the worst hit by late blight with resistance grades of 3.5 and 3.0, respectively (Table 9c). This breakdown of resistance of 853 could be attributed to the occurrence of a new virulent race since B53 had remained resistant for all the seasons and at all sites in previous experiments in Kenya.

The highest yielder in Molo was the clone KN2/2 (1006 gm/plant) although its yield was less at Kabete (738 gm/plant). The second highest yielder was clone K477/1 (959 gm/plant). The highest yielder at Kabete was clone K478/5 (798 gm/plant) followed by clone KN2/2 (738 gm/plant). The clone KN1/7 did not do very well because of the high incidence of Potato Leaf Roll Virus (PLRV). In both cases, B53 had the least tuber yield (464 gm at Kabete and 465 gm/plant at Molo).

Progeny testing

Inoculation of seedlings (Table 11) indicated
that the highest percentage of retainable seedlings
was given by cross number K920 whose parents were
classified into groups I and III. The seedlings in
group I could have arisen due to the good breeding value
of the group I parent which passed on its resistance
to the progeny. This could have been the case with cross
numbers K916, K914, K482 and K915. Cross number K919

involving group I and III parents gave only susceptible progeny. This was a cross between the variety Atzimba and K113/36 - a S. tuberosum clone. The reportedly high level of partial resistance (Anonymous, 1972) was apparently not reflected in its progenies. Similarly cross number K918 between R96 (1) - a S. andigenum clone and S42- a S. tuberosum clone gave susceptible (group IV) or very susceptible seedlings (group V). These results do not reflect what Thurston et al. (1962) reported that S. andigenum clones exhibited a higher level of field resistance than S. tuberosum clones. Since none of the seedlings showed field resistance, the R96 clone did not pass on its resistance to the progenies.

.3. Detached leaf inoculations

Results of detached leaf inoculations (Table 12a) showed that clones K476/13 and K476/19 were highly resistant. These clones did not show any infection till the 7th day after inoculation and the leaf area infected on the 9th day was 25% and 50%, respectively. Clone K476/19 did not sporulate while K476/13 showed sparse sporulation. Results of the laboratory and field screening (Table 1) showed that clone K476/19 had mean resistance grades of 2.0 (field) and 2.3 (laboratory). Clone K476/19 had mean resistance grade of 1.7 (field) and 3.0 (laboratory). These two clones might have had

a higher degree of field resistance. The results agree with what was reported by Hodgson (1962) and Van der Plank (1963) that partial resistance may be expressed as resistance to penetration, mycelial growth or spore production.

Clone K478/7 was highly resistant both in the field (1.0) and in the laboratory (1.0) but the detached leaf test indicated that it was susceptible, (Table 1 and 12a). Clone K478/16 was group I both in the field and laboratory but the detached leaf test indicated that it was susceptible. On the third day after inoculation (Table 12a) 5% of the leaf area was infected, by the 7th day the infection had increased to 100%. The amount of sporulation was, however, sparse. The sparse sporulation and the high resistance grades both in the field and laboratory indicated the presence of partial resistance. The uniform infection and spread of the fungus in the detached leaf test could have been due to ideal temperatures and humidity at which the leaves were incubated. Black and Gallegly (1957) and Knutson (1962) reported that environmental conditions under which the host is growing prior to and during infection could greatly influence the expression of field resistance. Clone K478/15 was susceptible under laboratory conditions and the results of the detached leaf test also showed that it was susceptible.

However, in the field, it was fairly resistant (1.3). This clone may have escaped infestation in the field.

Clone K478/14 and B53 showed a slow rate of mycelial growth and no sporulation was observed. They were highly resistant both in the laboratory and field and could have had a high level of partial resistance. Kerrs pink and Dextra showed 100% infection by the 9th day with profuse sporulation. Dextra however, had a longer incubation period than Kerrs pink suggesting that Dextra had vertical resistance. Clones K482/2, K480/1, K476/9 and K473/5 had their leaves rotten and did not sporulate. Clone K472/l showed a high rate of infection and profuse sporulation and the results showed it was susceptible. The clones that sporulated profusely had at least 50% of their leaf area infected by the 7th day and they could be classified as susceptible. Van der Plank (1963) reported that if lesions form more sporangia, the result is a rapid spread of the disease and an indication of less horizontal resistance.

5.4. Lesion size and amount of sporulation

Results of lesion size and the number of sporangia produced (Table 12b) indicated that lesion size did not follow the degree of resistance. Clone K478/14 which was resistant had the highest lesion size (6.28 cm) as compared to Kerrs pink the susceptible control (4.18cm²).

B53 and KN1/7 were resistant and their lesion size was 3.10cm² and 3.04cm², respectively. Clones KN1/4 and KN2/2 were moderately resistant and their lesion size was 2.34 and 4.02cm², respectively. These results do not agree with what was reported by Van der Zaag (1959), Thurston et al. (1962), Black (1970, 72) and Dowley et al., (1975). Results of this study agree with Kuntson's report (1962) who found that the area of the lesion in square cm was 44 for the resistant variety, 54 for the moderately resistant variety, 77 for the moderately susceptible and 54 for the susceptible varieties. The susceptible and the moderately resistant varieties had the same lesion size, therefore he concluded that lesion size was not a good indicator of resistance.

The number of sporangia produced had a relationship with the degree of resistance. Clones KN1/7, B53 and K478/14 were resistant and had 9, 10 and 11 sporangia/ml, respectively. Clones KN2/2 and KN1/4 were moderately resistant and they had 20 and 29 sporangia/ml, respectively. Kerrs pink the susceptible control had the most (38 sporangia/ml). Knutson (1962) found that the resistant variety had the least sporangia/leaf followed by the moderately resistant variety. The moderately susceptible variety had the most sporangia/



leaf. Results of this study indicated a correlation between the degree of resistance and the number of sporangia produced but there was no correlation between lesion size and the amount of sporulation. Lapwood (1961), Poehlman and Dhirendranath (1972) also reported that fewer spores were produced on resistant than on susceptible varieties.

5.5. Basis of resistance

The thickness of the cuticle of both resistant and susceptible clones varied considerably, some giving the same thickness (Table 10). The trend in the one month old plants was more or less similar to that of the two months old plants. Royle (1976) reported that Louis (1963) found that cuticle thickness in beans, tomato and other hosts was related to the ability of Botrytis cinerea to penetrate but Berry in 1959 found no relationship between the thickness of the outer epidermal wall in onion and resistance to Peronospora destructor. Royle (1976) also reported that Venturia inaequalis penetrates cuticles of all thickness encountered on apple fruits. Results obtained in this study suggest that there is no correlation between the epidermal thickness and level of resistance to late blight though clone K479/1 had a thicker epidermis than B53 and Kerrs pink in both cases, (Figs. 3 and 4).

Phenolic compounds in healthy leaves showed a positive correlation with the level of resistance (Table 14). The higher the level of resistance, the more the amount of phenolic compounds. For instance, clones KN1/7 and K478/5 were placed in group II when the blight incidence was high and they had the highest amounts of phenolic compounds (1184.45µg and 885.617 μg/g tissue, respectively). The clones KN2/2, KN1/4 and K477/1 were classified as group III under favourable conditions and had 729.292µg, 774.977 µg and 739.0 µg/g tissue, respectively. Kerrs pink, the susceptible control had the lowest amount of phenols 354µg/g tissue. Similar correlations between levels of phenols and the degree of resistance have been reported by Sakai et al. (1964), Goodman et al. (1967) and Sakuma et al. (1967). B53, the resistant control was graded as group II in the first and second season of the field testing but in the third season, it showed greater susceptibility to late blight and got a low resistance grade 3.5. It had $581.38\mu g/g$ tissue. The low level of phenolic compounds in B53 could have been due to environmental factors. Goodman et al., (1967) reported that environmental factors such as large amounts of nitrogen fertilisers may increase susceptibility of cereals to rust disease and decrease the level of total phenolics in leaf tissue.

Although a correlation exists between the phenols and the level of resistance, their role in disease resistance is still questionable (Tomiyama, 1980, Henfling, 1981; personal communication). Sakuma et al. (1967), Sato and Tomiyama (1977) reported that the phenols or phytoalexins e.g. rishitin found in potatoes are inhibitory rather than toxic to invading microorganisms.

5.6. Tuber Resistance

Results of tuber resistance (Table 16) showed that Kerrs pink which is susceptible under Kenya conditions had fairly resistant tubers (group 3) while clone K477/1 had susceptible tubers and yet the latter was more resistant (haulm resistance) than Kerrs pink. Clone K478/5's tubers were more susceptible than those of clone KN1/4 and yet the former was more resistant in the field than the latter. Talburt and Smith (1967) reported that tuber resistance did not follow haulm resistance though in some cases it followed but it was weaker and less consistent. The present results, (Table 16) agree with their findings. B53 which is a resistant variety in Kenya had resistant tubers (1.6). Clone KN1/7 which was outstanding in its resistance to late blight throughout the three seasons had also resistant tubers (1.6). Here, the tuber resistance followe haulm resistance. On the other hand, clones KN1/4 and KN2/2 were group III in the field when the blight was severe but their tubers were quite resistant (1.6) suggesting no relationship between tuber resistance and haulm resistance. Similarly tuber resistance of clone K477/1 did not follow the haulm resistance since the tubers had a resistance grade of 4.0 and the haulms were classified as group III.

5.7. Nutritional tests

Nutritional tests indicated that potatoes could be a good source of proteins. The crude protein content ranged between 7.00 - 11.13% and the dry matter content ranged between 19.5 - 24.1%. In the CIP Annual Report (1977) the crude protein content reported ranged between 6.13 - 11.69% and the dry matter content was 1.27 -27.98%. The protein content agrees well with the results of this study. Miedema et al. (1976) reported a dry matter content of 17.2 - 29.3% and crude protein content of 4.78 - 10.05%. The upper limit of the dry matter content reported in both cases was slightly higher than the 24.1% reported in this study. There was no correlation between the dry matter content and the protein content. This agrees with CIP Annual Report of 1977. Li and Sayre (1975) also found no significant correlation between the dry matter content and the crude

protein content. Miedema <u>et al</u>. (1976) however, reported a negative correlation between the crude protein and the dry matter content.

The variation in the texture and flavour of cooked tubers could have been due to the dry matter content as well as soil conditions. Pushkarnath (1964) reported that flavour and taste of cooked flesh was influenced greatly by soil and manuring conditions.

*

CONCLUSION

Field and laboratory screening of potato clones is necessary to identify the resistant and the susceptible clones and those that may be having partial resistance. The laboratory tests were more severe than the field tests because of the ideal conditions under which the laboratory tests were conducted. Based on the high level of resistance exhibited in the field and laboratory, and the sparse sporulation, clones K478/14, KN1/7 and B53 could be having high levels of horizontal resistance. Clone KN2/2 was characterized by frequent stem blight but little or no infection on the leaves. The detached leaf test gave profuse sporulation. This clone seems to have major gene resistance.

There was no relationship between the thickness of the cuticular cells and the level of resistance but there was a correlation between the level of phenols and the degree of resistance. The phenols appear to play a role in the resistance of potato clones to late blight.

The tuber resistance of B53, KN1/7 and K478/5 was found to follow the haulm resistance but the tuber

resistance of clones KN1/4, KN2/2 and K477/1 did not.

The results of this study suggest that resistance or susceptibility could be a response to several factors and all the factors play a role in determining the resistance or susceptibility of each clone.

The dry matter content and the protein content were higher at Molo than at Kabete except clone K478/5 which had higher protein content at Kabete than at Molo. This variation with site indicates that environmental conditions influence or affect the dry matter content and the protein content of the potato clones.

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