STUDIES ON GERMINATION AND VIGOUR OF CABBAGE SEEDS

ASPECTEN VAN DE KIEMING VAN ZADEN VAN WITTE KOOL EN SAVOOIEKOOL

Publication 541, Department of Horticulture, Agricultural University, Wageningen, The Netherlands.

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STUDIES ON GERMINATION AND VIGOUR OF CABBAGE SEEDS

Proefschrift

ter verkrijging van de graad van
doctor in de landbouwwetenschappen,
op gezag van de rector magnificus,
Dr. C.C. Oosterlee,
in het openbaar te verdedigen
op dinsdag 22 september 1987
des namiddags te vier uur in de aula
van de Landbouwuniversiteit te Wageningen.

THEOREMS (STELLINGEN)

I

Mathematical models aiming to predict and optimalize crop establishment have to take into consideration the interaction between environmental conditions and the quality (vigour) of seed lots as determined in laboratory tests.

Bierhuizen, J.F. & Wagenvoort, W.A. 1974.
Some aspects of seed germination in vegetables. Scientia Hort. 2: 213-219.
This thesis.

II

Seed size of cabbage seeds is not a relevant parameter for the viability and vigour of a seed lot, but it is positively related to seedling vigour.

This thesis.

III

Osmotic pretreatment causes a number of different improvements in seed quality. It is questionable whether a common mechanism underlies these changes.

Weges, R. 1987. Physiological analysis of methods to relieve dormancy of lettuce seeds. Ph.D. Thesis. The Agricultural University. Wageningen. 121 pp.
This thesis.

IV

The nutritious garland chrysanthemum (Chrysanthemum coronarium, L.) which is an important edible leafy crop in China, deserves introduction in the Netherlands as a new vegetable and ornamental plant.

V

A successful seed-programme has to combine basic knowledge of plant physiology with practical experience in the technology of seed production and critical control of seed quality.

VI

The increase of germination rate and the invigoration of seeds by osmoti treatment reminds of the disappearance of the blockade of the human body ch' (energy) and the retardation of senescence due to Tai-Chi Ch'uan exercises.

VII

The danger of improper use of pesticides is comparable to that of arbitrarily discarded garbage. Both types of pollution can damage the ecologica system and diminish the quality of food production and recreation area. Thi threatening situation requires world-wide attention of scientists, politician and civilians.

VIII

Successful team work between universities and research institutes depend on the cooperation between heaven, earth and people.

The Natural Way of Lao Tzu. c. 2500 BC.

XI

The bicycle paths in the Netherlands are not only safe for the bicyclist but also offer a romantic luxurious exercise.

Tsung-dao Liou

Wageningen, 22 September 1987

dedicated to my parents
Mr. Chung-hou Liou
Mrs. Li-mien Liou

PREFACE

I like to express my gratitude to the Board of the Agricultural University, Wageningen, the Ministry of Agriculture and Fisheries, The Hague, The Netherlands and the Ministry of Education, the National Science Council of the Republic of China for offering me the opportunity to study in Wageningen and to perform this investigation.

I mention with great respect and thankful memory the late Prof.Dr.Ir. J.F. Bierhuizen who kindly organized my study before he untimely passed away.

I am greatly indebted to my adviser Prof.Dr. C.M. Karssen, Department of Plant Physiology and co-advisers Mrs. W.A. Wagenvoort, Department of Horticulture of the Agricultural University and Dr. H.L. Kraak, Government Seed Testing Station, for their supervision and critical comments throughout the course of this investigation and their help in the preparation of this thesis. I express my gratitude to the staff of the Department of Horticulture for their hospitality and their help in several occasions. Special thanks are due to Mr. R.J. Lamers and Mr. K. Steensma who prepared seed beds and took care of the plants during experiments in the field and in the phytotron.

Since various aspects of germination and vigour of cabbage seed were examined during the present study, some experiments and measurements had to be performed at other Departments and Institutes in Wageningen. I sincerely appreciate the cooperation I have received at all these occasions. Royal Sluis Seed Company, Enkhuizen, The Netherlands, kindly provided the seed material. I thank Prof.Dr. J. Bruinsma, Head of Department of Plant Physiology and Dr.Ir. G.P. Termohlen, Director of the Government Seed Testing Station for their permission to use facilities in their laboratories. Special thanks are due to Dr.Ir. J. Bekendam and Mr. A. van Geffen for advice on many theoretical and practical points, to Mr. H. van de Scheur for helping in seed size fractionation, to Mr. C. Witte for determination of seed moisture contents and to Mr. J.G. van Pijlen for assistance in my experiments. Miss H. Askamp is acknowledged especially for her kind and helpful cooperation. I express my gratitude to Mr. H.W.M. Hilhorst for his advice in the extraction of K^+ from cabbage seeds and to Mrs. E.A.M. Koot-Gronsveld and Dr. R. Weges for their technical assistance in the use of the

flame photometer and thermocouple psychrometer.

Several persons were involved in the technical preparation of the manuscript. In particular I am thankful to Mr. F. Dumoulin for his help in preparing the lay-out of the manuscript. Mr. H.H.W. van Lent was responsible for the drawing of the figures and Mr. R. Jansen photographed them.

I am very grateful to the present director of Fengshan Tropical Horticultural Experiment Station, Taiwan Agricultural Research Institute, Dr. T.F. Sheen and the former director of the Taiwan Agricultural Research Institute, Dr. H. Wan for offering me the opportunity to study in the Netherlands and to my colleagues Dr. M.S. Chen and Dr. C.C. Lin who guaranteed my studies.

I am very grateful to Mr. T.F. van Hell for offering convenient accommodation and social facilities as well as mental cooperation.

Finally, I thank my parents, my brothers and sisters, my wife and children, and my former adviser professor in National Taiwan University, Prof.Dr. L. Hung for their continuous encouragement.

CONTENTS

ABSTRACT

The effects of commercial storage of cabbage seeds on the germination of seeds and the emergence and growth of seedlings have been studied. Progressive ageing of seeds caused loss of seed vigour which resulted in poor emergence and growth of seedlings and the formation of abnormal seedlings. Loss of vigour was also observed indirectly in the controlled deterioration test (CD test) that speeds up the rate of ageing at controlled conditions. It was shown that the CD test gave the best indication of vigour loss. Low vigour seeds also differed from high vigour seeds in a more substantial loss of potassium ions during incubation in soil. Apart from seed vigour also soil conditions and other environmental factors influence the field performance that under certain atmospheric and soil conditions the best possible result is obtained.

Pre-incubation of cabbage seeds in osmotic solution clearly showed that several aspects of seed ageing are reversible. Pretreated seeds germinated earlier and faster and produced more normal seedlings than untreated seeds. The mechanism of osmotic pretreatment was studied. It is unlikely that such treatment favours the accumulation of osmotic constituents. The leakage of K^+ was inhibited during osmotic incubation. Loss of vigour might be related to membrane damage.

ABBREVIATIONS

A	percentage final emergence
CD	controlled deterioration
DW	dry weight
FW	fresh weight
h_m	the matric head of the soil
k	the time spread of emergence in the seed population
p	percentage emergence at time t
PEG	polyethylene glycol
r.h.	relative humidity
t_0	the time of first seedling emergence
t_{50}	the time of half-maximal seedling emergence
ψ	water potential
ψ_π	osmotic potential
$\psi_{\pi e}$	osmotic potential of the external PEG solution
ψ_p	pressure potential

CHAPTER 1

INTRODUCTION

Cabbage

This thesis studies several aspects of germination and vigour in seeds of *Brassica oleracea* L. Cabbage is a member of the Cruciferae family. The species forms a rosette of leaves surrounding a large bud or head in the first year and develops a flower-stalk in the second year. The shape of the head varies with the variety or cultivar, being either pointed, spherical or flattened. In general, the plant will be 20 to 30 cm tall in the first growing season. The leaves are moderately large, they sometimes reach a width of 30 cm. The shape is more or less ovate and is covered by wax at the surface. The colour of the outer leaves can be green, blue-green, red or white. The root system is extensive, fibrous, and finely branched. It reaches a maximum lateral spread of about 1 m before the head is formed. The taproot as well as many lateral roots penetrate into the soil to a depth of 1.5 m or more in the period of the maturation of the head. Cabbage plants that are used for seed production form an extremely dense root system that penetrates about 0.7 m into the soil.

Cabbage flowers are positioned in racemes. The flowers are yellow with four sepals, four petals, six stamens, and a two carpels ovary containing many ovules per carpel. Bees are the usual agents for pollination. The fruit is a silique. The siliques contain about 12 to 20 seeds. The seeds are rather small, globular and smooth and line up in rows in each silique. The seed contains mainly lipids as reserve food. The colour of the testa is light brown to darker brown depending on the maturity of the seeds or the variety. The seed consists only of an embryo and testa. The endosperm is absorbed by the cotyledons during seed formation. The flower and seed development and the cultivation methods have been described by Thompson (1933) and Ryder (1979), respectively.

Cabbage is one of the most important leafy vegetables for human consumption. The expanding use of cabbage is mainly due to its high nutritive value. In Far East Asian countries, it is commonly used either stir-fried or as pickles with other vegetables. In the western world, it is popularly used either boiled or as salad or sauerkraut.

It is believed that cabbage is indigenous to southern Europe. At the present time, the wild cabbage still occurs on the coasts of the North Sea, the English Channel and the northern Mediterranean Sea. The plants can also be found along the other coasts of the British Isles (Mitchell, 1976). Although it is a temperate crop, it grows well under a variety of temperature conditions due to its strong adaptation potentiality. At present, it is popular by the growers in both temperate and tropic areas.

In the tropics, poor emergence of cabbage seeds is often a serious problem. Emergence is hampered by too dry, too wet or too warm conditions, particularly in the season of the tropical monsoon. These extremely adverse conditions cause a gap in the cabbage production during the summer season in the humid tropics.

Germination tests

The essential first step in the growing of vegetables is the establishment of a stand in the field or in containers. Seed germination is the very first stage of that process. Therefore, it is of great importance that a high germination level is assured. In most countries sowing-seed is subjected to some form of germination test. Since germination tests need to be repeatable the seed analyst germinates seeds under optimal conditions. A minimum percentage of normal seedlings (Bekendam & Grob, 1979) is required, varying for different crops, before the seeds are regarded as acceptable for sowing. Nevertheless, there have been many reports of large differences in field emergence among seed lots of many crops that had all exhibited high and acceptable levels of germination in the standard laboratory germination test (Perry, 1972; Matthews, 1980). Most of the disappointments arise in less-than-optimum field conditions, for example, inadequate or excess moisture, low temperature, soil crusting or poorly prepared seed beds. Germination can fail if requirements for water, oxygen, and a suitable temperature are not provided. More often, however, poor emergence is not associated with failure to germinate, but with the failure to complete post-germination, pre-emergence stages of growth (Doneen & McGillivray, 1943). Obviously, there are clear differences in the capability of highly germinable seed lots to establish a seedling stand at suboptimal conditions. These differences are referred to as differences in seed vigour (Perry, 1972). Low vigour seed lots may also occur in commerce. A field emergence comparison at several sites of over 200 commercially available seed lots of small seeded vegetables including onion, carrot, lettuce and several Brassica crops, revealed considerable differences in the emergence ability of seed lots within the species tested (Matthews, 1980). The value of the standard laboratory germination test seems to be limited to the detection of those batches of seeds that have less than the acceptable levels of viable seeds provided that the seeds are not dormant (Mackay, 1972). Therefore, tests capable to detect low vigour seeds are required.

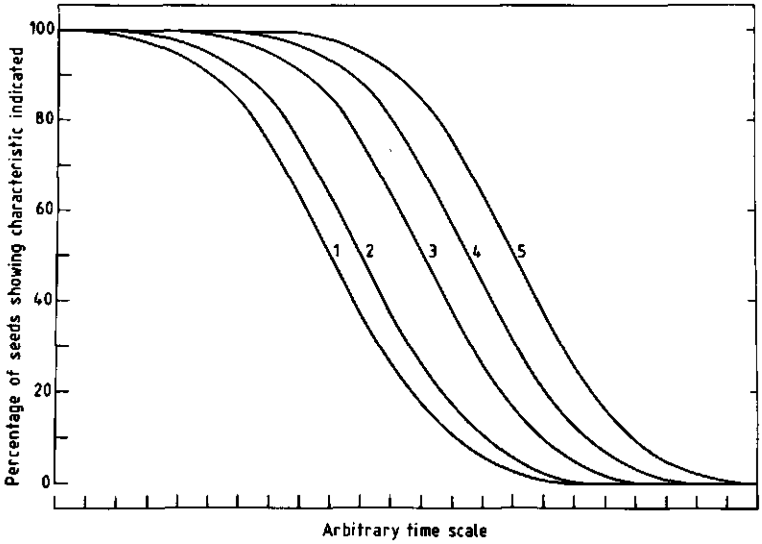

Fig. 1.1. Pattern of seed deterioration in time of an hypothetical seed lot: (1) all embryo tissue stained by the reduction of tetrazolium chloride; (2) electrical conductivity of imbibition water below minimum; (3) germination time below certain minimum; (4) normal seedlings and (5) radicle emergence (Redrawn after Ellis and Roberts, 1981).

Seed ageing

At present it is generally accepted that ageing both before and after harvest is a main cause of poor vigour in commercial seeds (Ellis & Roberts, 1980, 1981; Matthews & Powell, 1986). Ageing in all organisms is the sum total of the deteriorative processes that eventually lead to death.

For most practical purposes seeds are considered dead when in the absence of dormancy they fail to germinate under optimal conditions. Before the seeds become incapable of germination, several signs of the progressing ageing process appear (Fig. 1.1) (Heydecker, 1972; Roberts, 1972). The most obvious manifestation of ageing is a slowing down of the rate of germination (Ellis & Roberts, 1980). A second symptom of ageing is the production of smaller seedlings and an increase of the proportion of seeds producing abnormal seedlings (Parrish & Leopold, 1978). An additional feature of ageing seeds, seen especially in legumes with large cotyledons and in maize seeds, is a decline in the ability of seeds to retain solutes such as sugar and potassium when they are soaked in water (Powell & Matthews, 1977). Thus ageing in seeds involves a series of degenerative changes leading to loss of viability. It begins after physiological maturity when seeds are usually considered to be of optimum seed quality. The survival of a population of seeds follows the pattern typical of many organisms (Fig. 1.2) with an initial prolonged period when few seeds die, followed by a rapid decline in viability. All seeds with high laboratory germination will lie on the initial slow decline curve although their positions on the survival curve will differ depending on the degree of ageing of the lot. The rate of ageing is strongly influenced by the conditions of maturation before harvest and of storage after harvest. The time taken for half of the population of seeds to become incapable of germination is much shorter at a higher moisture content and/or temperature (Fig. 1.2 curve A) than at a lower moisture content and/or temperature (curve B).

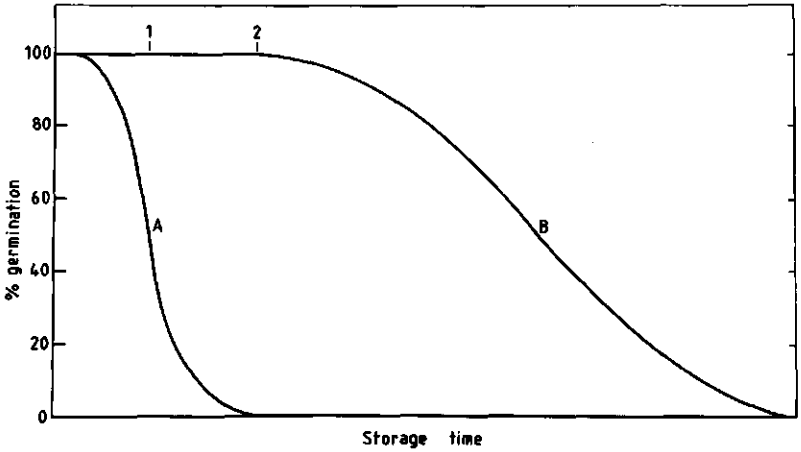

Fig. 1.2. Survival curves of an hypothetical seed lot held at relatively high moisture content and/or temperature (A) and relatively low moisture content and/or temperature (B). (Redrawn after Matthews, 1985).

Vigour tests

The acceptance of ageing as the main cause of poor vigour has encouraged Ellis and Roberts (1980, 1981) to propose laboratory germination as the most rational measure of vigour. They argue convincingly that it is the difference in position along the slow initial decline of the survival curve that gives rise to vigour differences (Fig. 1.2). Therefore, an accurate estimate of the actual germination of the population would suffice. This is not easy to do in practice, since it requires either germination tests with many thousands of seeds per sample or the production of a survival curve. To construct such a curve seeds are stored at conditions that accelerate ageing. When germination is converted to probits the survival curve can be extrapolated to zero time which gives an accurate estimation of the starting point of the seed lot, that is, of its position on the slow initial decline.

Both indirect and direct vigour tests have been developed and are in use. Two indirect tests that are closely related to the physiological causes of low vigour are the electrical conductivity test and the accelerated ageing or controlled deterioration test. The conductivity test detects seed lots in which low vigour is associated with ready leakage of ions into imbibition water. It is mainly used in grain legumes like peas (Matthews & Powell, 1987), field beans (Hegarty, 1977) and Phaseolus beans (Powell et al., 1984, 1986). Ageing or deterioration tests speed up the rate of ageing in a controlled manner by holding seeds at an elevated temperature and moisture either in a humid atmosphere or in moisture-proof containers following the raising of seed moisture by partial imbibition. Since seed lots of different vigour are at different points along the slow initial decline of the survival curve (Fig. 1.2 B, 1 and 2) they come to lie at different points on the steep decline following rapid ageing. And so, differences in germination percentage that are too close to be distinguishable in the germination test on unaged seeds, can be separated in the ageing test. Other indirect tests use slow germination and slow seedling growth to indicate low vigour (Smith et al., 1973; Gray & Steckel, 1983). The cold test is an example of a direct vigour test. Seeds are exposed to low temperature and soil-borne micro-organisms.

Subcellular changes with ageing

The observation that ageing in seeds is accompanied by a decline in the ability of seeds to retain solutes has given rise to the suggestion that an early event in the deterioration process is the degradation of cellular membranes. Peroxidation of unsaturated fatty acids in membrane phospholipids might be a significant early event in ageing. It is proposed that a variety of spontaneous and enzymatic oxidations, which is enhanced in the presence of oxygen, generates highly reactive free superoxide radicals (O_2^-) which attack both the unsaturated fatty acids of phospholipids and the membrane proteins, thereby disrupting membrane structure. However, the evidence that these events take place in seeds is conflicting (Osborne, 1982). In rapidly aged pea seeds the amount of unsaturated fatty acids decreased (Harman & Mattick, 1976). In soybeans such occurred in naturally but not in artificially aged seeds (Priestley & Leopold, 1983). In 600-years old lotus seeds unsaturated fatty acids were hardly changed compared to freshly cultivated seeds (Priestley & Posthumus, 1982).

Villiers and Edgecumbe (1975) observed that the longevity of lettuce seeds is much enhanced when they are held in the fully imbibed state. In imbibed seeds active metabolic processes repair molecular damage that had developed during dry storage. Repair has also been demonstrated in partially imbibed seeds (Ibrahim & Roberts, 1983; Ward & Powell, 1983). It also occurred during so-called priming or osmotic pretreatments (Burgess & Powell, 1984).

Outline of the thesis

It is the aim of this thesis to study the conditions that regulate in cabbage seed germination, seedling emergence and seedling growth in relation to seed vigour. Such studies are not only pertinent to improve summer cabbage production in tropical countries but also of basic importance to cabbage production in other areas.

The experiments are performed with a collection of cabbage seeds of different age. In Chapter 2 it is shown that these different seed lots vary widely in vigour. Different aspects of low vigour are studied in germination, emergence and seedling growth. In Chapter 3 it is demonstrated that in cabbage seed size is not relevant to seed vigour but is an essential factor in the growth and vigour of the seedlings.

Apart from seed quality, seedling emergence also depends on soil conditions. Chapter 4 describes a study on the influence of soil type and the matric head of the soil on emergence and seedling growth in relation to seed vigour.

The effects of osmotic pre-incubation are studied in the Chapters 5, 6 and 7. In Chapter 5 it is demonstrated that osmotic treatment considerably improves seedling quality, particularly of low vigour seeds. In Chapter 6 it is investigated whether the effects of osmotic pre-incubation are correlated to changes in the water relations of the seeds. Chapter 7 studies the differences in K^+ leakage in seed lots of varying vigour and the changes that occur due to osmotic treatment. Chapter 8 presents a general discussion of the results.

CHAPTER 2

ASPECTS OF LOW VIGOUR

Abstract

Seeds of Brassica oleracea L. Capitata group cv. Oscar that had been commercially stored during 1 to 6 years showed almost full germination in a standard laboratory germination test at diurnal temperatures of 20 ° - 30 °C. Controlled deterioration of the seeds during 24 hours at 45 °C and 24% seed moisture content revealed strong differences in seed vigour. Germination and emergence experiments at controlled conditions in laboratory or phytotron and in the field showed that low vigour was associated with reduced rate of germination and emergence, reduced seedling fresh weight and increased number of abnormal seedlings. Low vigour is one of the causes of low field emergence.

Introduction

It was argued in the first Chapter that ageing in seeds involves a series of degenerative changes leading eventually to loss of viability. As a consequence, several symptoms are associated with ageing which collectively are referred to as loss of vigour (Heydecker, 1972; Roberts, 1972). These symptoms are seen among others in rate of germination, seedling quality, resistance to stress conditions and performance of seeds and seedlings in the field.

In this Chapter several of these symptoms are studied in 5 seed lots of Brassica oleracea of different age and differing in vigour. Germination will be tested in standard laboratory germination test without and with preceding rapid ageing (controlled deterioration). Emergence will be tested in controlled phytotron conditions and in the field. Besides the percentage germination or emergence, in all tests also the quality of the seedlings will be taken into account. Finally, germination at a thermogradient plate will be tested at a range of temperatures.

Material and methods

Seed materials

Commercial F_1 hybrid seeds of Brassica oleracea L. Capitata group cv. Oscar obtained from Royal Sluis Seed Company harvested in three different years were used. Two seed lots of different origin were harvested in 1984, one seed lot in 1980 and two other lots were produced in 1979 on different locations (Table 2.1). The seeds were stored at commercial storage conditions till September 1984 and from then on at 5 °C and about 30% relative humidity until use. Experiments were performed in 1984 and 1985.

Table 2.1. The 1000 seed weight of cabbage seeds harvested in different years.

Year of harvest	1000-seed weight, g
1984-1	5.37
1984-2	3.51
1980	3.11
1979-1	3.15
1979-2	2.94

$LSD_{0.05} = 0.70$

Standard germination test

Standard germination tests were carried out according to ISTA rules (International Seed Testing Association, 1985). Four replicates of 100 seeds per lot were sown on filter paper saturated with tap water. The seeds were germinated on a Jacobsen germinator at alternating temperatures of 20 °C and 30 °C during 16 hours in darkness and 8 hours in white fluorescent light (Philips TL 40/33), respectively. Fluence rate was 8.5 Wm^{-2}. The seeds were considered to be germinated when the radicle became visible to the naked eye, seedlings were evaluated as normal or abnormal on the basis of ISTA rules (Bekendam & Grob, 1979).

Thermogradient plate test

On a thermogradient plate ranging from 10 ° to 34 °C a randomized complete block design with 3 replications was carried out. The thermogradient plate was covered with 9 rows each of 9 filter papers (Ø 9 cm). The temperature differed 3 °C between adjacent rows. On each water saturated filter paper 50 seeds were sown. Each paper was covered with a bell jar. During the first seven days of the test the germinated seeds were counted twice a day. The results of germination counts made on successive days were plotted as percentage germination against the temperature at the center point of the filter paper.

Controlled deterioration test

The vigour of different seed lots was determined by means of the CD test (Matthews, 1980). The moisture content of the seeds was determined in 3 replications on seed samples of 5 g by means of the oven method (103 °C for 17 hours). Of another 20 gram of each seed lot the seed moisture content was raised to 24% on a wet weight basis by adding an appropriate amount of water to the seeds within plastic bottles. Immediately afterwards the plastic bottles were closed and slowly rotated for at least 2 hours at 20 °C and then stored for 2 days at 10 °C to reach moisture equilibrium. At the end of the treatment the final seed moisture content was determined. In general the moisture content of the seeds differed less than 0.5% after equilibration. For the CD treatment proper, half of the equilibrated seeds was packed in aluminium foil and immersed in a water bath at 45 °C for 24 hours. The other half was kept overnight in an incubator at 5 °C. Following both treatments a standard germination test was carried out.

Phytotron test

Emergence and seedling vitality of the cabbage seed lots were tested in a phytotron at constant 9 °, 13 °, 17 °, 21 ° or 25 °C. A split block design with 4 replications was conducted. Fifty seeds were sown each in a container filled with peat in the lower part and a layer of 3 cm of river sand in the upper part. The seeds were covered with a sandy layer of

0.4 cm. During the germination period the soil was kept moist by means of a regular water spray. The seedlings were illuminated by high pressure sodium + mercury lamps (Philips, SON/T 250W E40, 400W E40; HPI/T 250W E40, 400W E40) during 8 hours per day, the light intensity at seedling level being approx. 35 Wm^{-2}. The seedlings were considered to be emerged when the cotyledons reached a horizontal position. At the 19th day after half-maximal seedling emergence the seedlings were classified according to fresh weight per plant as "strong" (>0.5 g), "medium" (0.25-0.5 g) or "weak" (<0.25 g).

Field test

Field experiments were conducted on the experimental field of the Department of Horticulture. The 5 seed lots were sown in the field on 4 sowing dates (7 June, 11 July, 16 August and 25 September, 1985) in a randomized complete block design with 4 replications of 200 seeds per seed lot. The seedlings were considered to be emerged when the cotyledons were raised above the soil. The final percentages of emerged seedlings were recorded. Nineteen days after half-maximal seedling emergence the seedlings were classified according to the same criteria as in the phytotron test.

Results

Standard germination test

Seeds of all 5 seed lots showed high germination percentages in the standard germination test (Fig. 2.1). The 1984-1, 1984-2 and 1980 seeds produced high percentages of normal seedlings, 81, 95 and 82%, respectively. A lower percentage of 49% was found in the two 1979 seed lots.

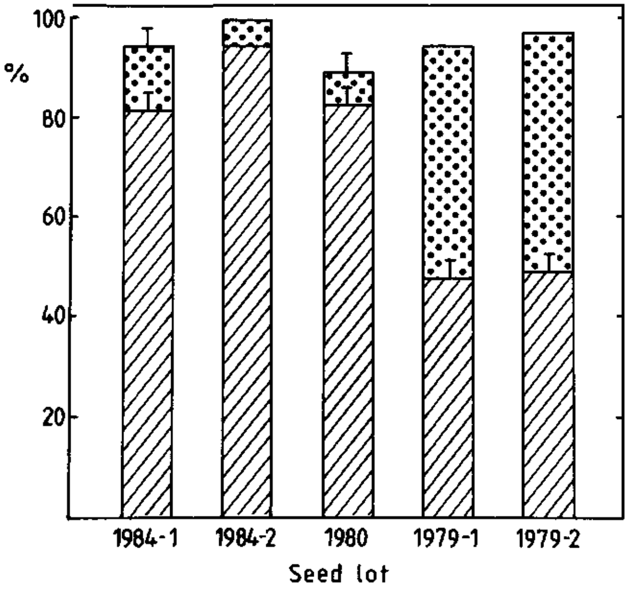

Fig. 2.1. Percentages germination (upper line of bars) of seeds of 5 lots in the standard laboratory germination test at alternating temperatures 20 °C - 30 °C. Seedlings were classified as normal (▨) or abnormal (⁙). Single vertical bars indicate standard deviations >3 %.

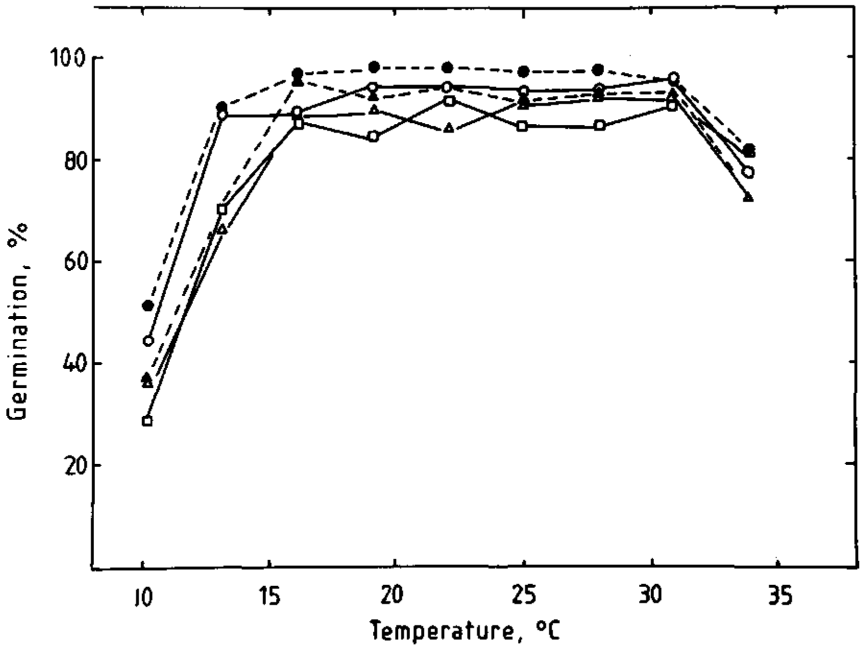

Fig. 2.2. Percentages germination of seed lots 1984-1 (○), 1984-2 (●), 1980 (□), 1979-1 (△) and 1979-2 (▲) at temperatures ranging from 10 ° to 35 °C.

Germination at the thermogradient plate

The experiment on the thermogradient bar showed that the final germination percentage of all seed lots was high between 16 °C and 31 °C (Fig. 2.2). At temperatures below 16 °C the germination of the 1984 seeds dropped slightly slower than of the 1979 seeds.

The germination rate, as seen in the reciprocal value of the time for half-maximal germination (Fig. 2.3) discriminated better between the seed lots. Both 1979 seed lots germinated slower than the younger seeds, the 1980 seeds were intermediate between the 1979 and 1984 seed lots.

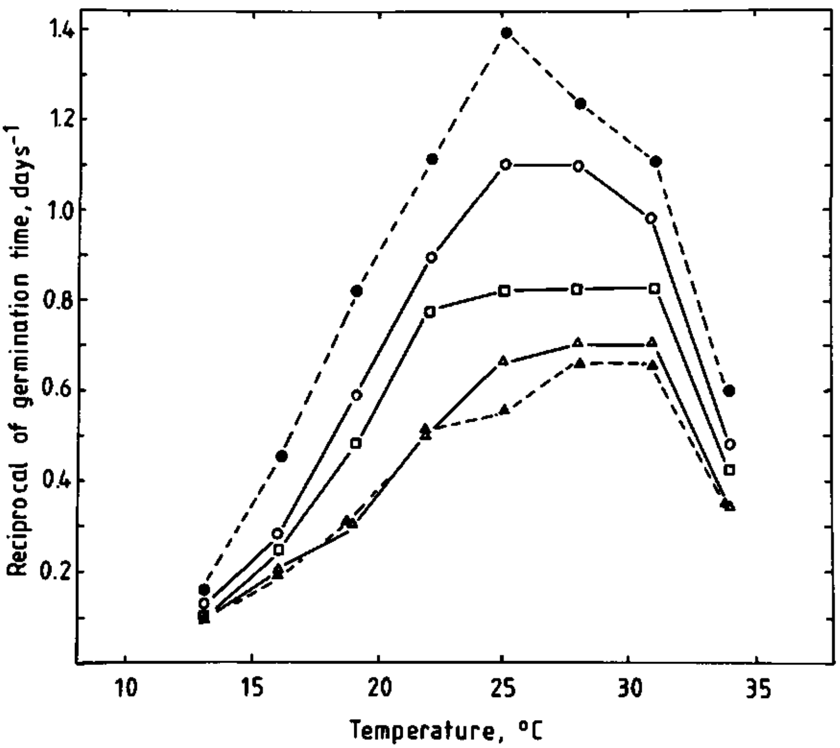

Fig. 2.3. The relation between the reciprocal value of the time for half-maximal germination and temperature for each lot 1984-1 (○), 1984-2 (●), 1980 (□), 1979-1 (△) and 1979-2 (▲).

CD test

The percentages germination in the standard test of control seeds that after moisture equilibration had been stored at 5 °C, were rather similar to those of seeds that were sown directly in that test (compare Fig. 2.4 with Fig. 2.1). However, the CD treatment (45 °C) strongly reduced germination. It discriminated clearly between the seed lots. Only the 1984 seed lots still showed high germination and a reasonable high percentage of normal seedlings, the 1984-2 seeds scored better than the 1984-1 seeds.

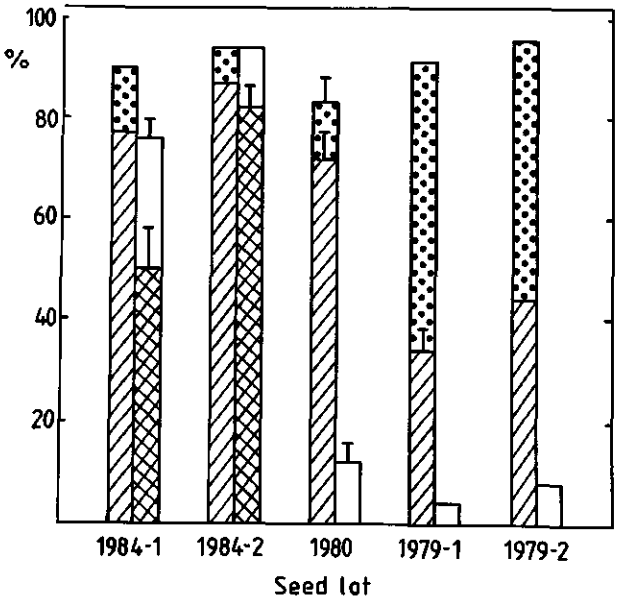

Fig. 2.4. Percentages germination (upper line of bars) of seeds of 5 lots at the standard laboratory germination test. Moisture content of the seeds was equilibrated at 24% on fresh weight basis. Seeds were stored for 24 hours at either 5 °C (left bar of each lot) or 45 °C (right bar). Seedlings were classified as normal (▨,▩) or abnormal (░,□). Single vertical bars indicate standard deviations >3%.

Emergence and seedling vitality at phytotron conditions

The influence of temperature was also tested on seedling emergence from soil at controlled conditions in the phytotron. In contrast to the experiment on the thermogradient plate, in the phytotron seeds of all 5 lots emerged for 80% or more in the range of 9 ° to 25 °C (data not shown). But again the reciprocal of the time for half-maximal seedling emergence revealed differences between the seed lots (Fig. 2.5). Clearly the germination rate of the oldest seed lots was lower than that of the younger ones. Nineteen days after half-maximal seedling emergence, only the seeds of the 1984 and 1980 lots had produced seedlings with a mean fresh weight above 0.25 g plant^{-1} and such only occurred at temperatures between 17 ° to 25 °C (Fig. 2.6).

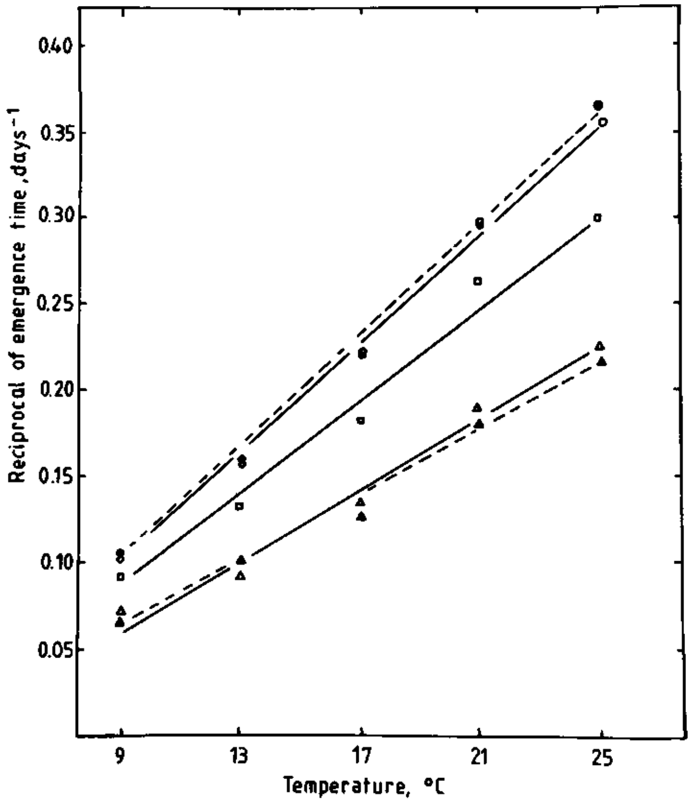

Fig. 2.5. The relation between the reciprocal value of the time for half-maximal emergence and temperature for seed lot 1984-1 (○), 1984-2 (●), 1980 (□), 1979-1 (△) and 1979-2 (▲) at 5 temperatures in the phytotron.

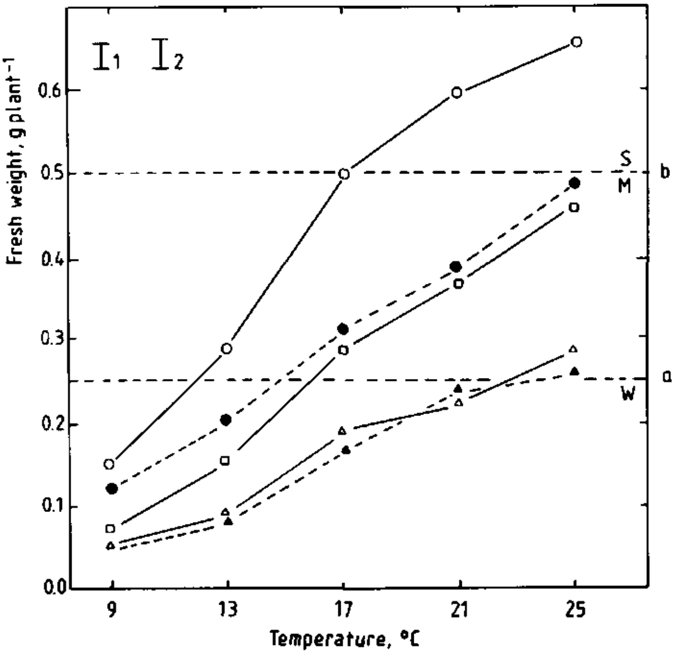

Fig. 2.6. The relation between the mean fresh-weight of seedlings and the temperature in the phytotron for seed lot 1984-1 (○), 1984-2 (●), 1980 (□), 1979-1 (△) and 1979-2 (▲) at the 19th day after half-maximal seedling emergence. Seedlings were classified as strong (S), medium (M) and weak (W) according to fresh weight. $LSD_{0.05}$ for the effect of temperature and seed lot is indicated by the vertical bars 1 and 2, respectively.

Emergence and seedling vitality at field conditions

Mean emergence of the 5 lots at all sowing dates in the field varied from 13 to 62% and it ranged from 6 to 73% at the 4 different sowing dates (Fig. 2.7). A low number of seedlings emerged in July, apparently due to the formation of a soil crust caused by water spraying during a dry weather period. The 1984 lots produced at all occasions the highest percentage emergence, whereas the 1980 and 1979 seed lots showed very low percentages of emergence.

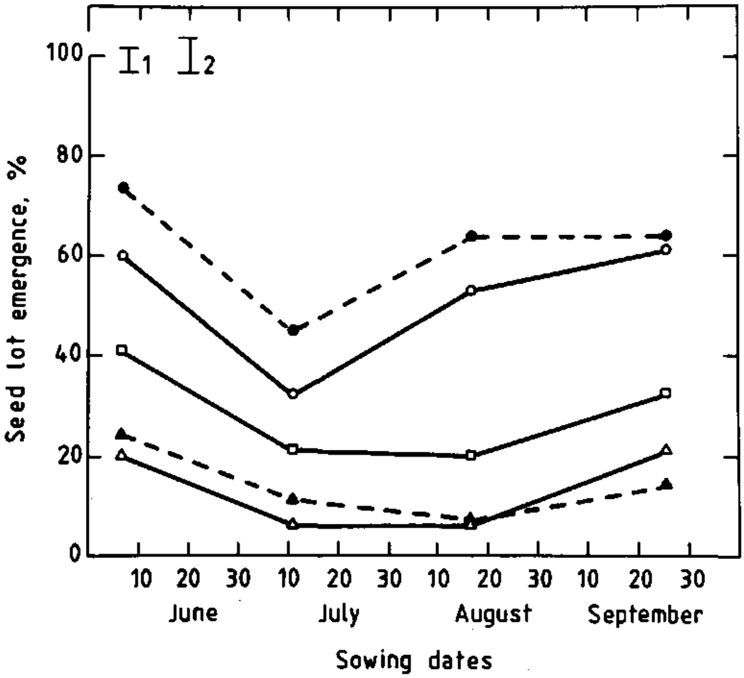

Fig. 2.7. The relation between seedling emergence and sowing date for seed lot 1984-1 (○), 1984-2 (●), 1980 (□), 1979-1 (△) and 1979-2 (▲) at field conditions. $LSD_{0.05}$ for sowing date and seed lot is indicated by the vertical bars 1 and 2, respectively.

The fresh weight of seedlings at the 19th day after half-maximal seedling emergence also showed a large variation among seed lots and sowing dates (Fig. 2.8). Average fresh weight per seedling of the 5 seed lots ranged from 0.09 g to 1.60 g at the 4 different sowing dates. The 1984 seed lots produced stronger seedlings than the older seed lots. When the sowing occurred in late September, small seedlings developed from all seed lots.

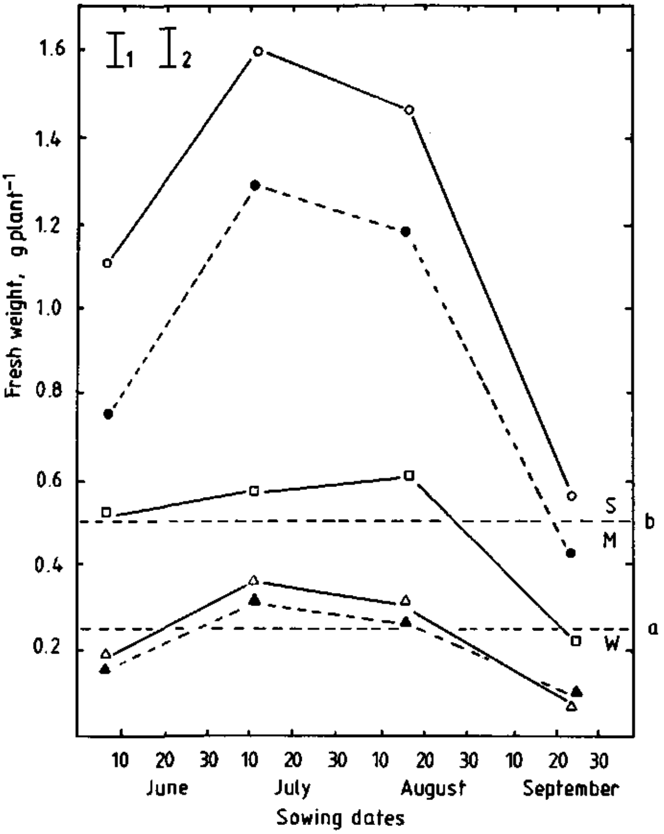

Fig. 2.8. The relation between mean fresh weight of seedlings and sowing date for seed lot 1984-1 (○), 1984-2 (●), 1980 (□), 1979-1 (△) and 1979-2 (▲) at the 19th day after half-maximal seedling emergence in the field. Seedlings were classified as strong (S), medium (M) or weak (W) according to fresh weight. $LSD_{0.05}$ for sowing date and seed lot is indicated by the vertical bars 1 and 2, respectively.

Discussion

The data presented in this Chapter demonstrate that also in cabbage seed lots that showed good germination, nevertheless may strongly differ in vigour. The first indication for the lower vigour of the 1979 seed lots was the low percentage of normal seedlings (Fig. 2.1).

The CD test (Fig. 2.4) also revealed strong differences in the physiological age of the seeds. Obviously the 1979 and 1980 seeds were much closer to the steep decline of the survival curve (Fig. 1.2) than the 1984 seed lots. Similar indications of a vigour difference between the 1984 and 1979 seed lots can be concluded from the rate of germination at different temperatures both at laboratory and phytotron conditions (Fig. 2.3 and 2.5), the percentage of seedling emergence in the field (Fig. 2.7) and the seedling quality after standard laboratory germination (Fig. 2.1), emergence in the phytotron (Fig. 2.6) and in the field (Fig. 2.8).

The 1980 seed lot reacted in the CD test in a similar way as the 1979 seed (Fig. 2.4), but in the other tests it took an intermediate position. The CD test seemed to underestimate the seed vigour slightly.

The difference in physiological age that is indicated by the different tests, roughly correlates with the real age of the seeds. However, the correlation is not linear. Thus, apart from the period of commercial storage that was 7, 6 and 1 year for the 1979, 1980 and 1984 seeds, respectively, also differences in preharvest conditions and production site influenced ageing of the cabbage seeds. The effect of site is also indicated by the differences within the two 1984 and the two 1979 seed lots that were each produced at different locations. The differences between the 1984 seed lots are not always similar, for instance the 1984-2 seed lot produced more seedlings but of a lower fresh weight than the 1984-1 seeds (Figs. 2.7, 2.8). The difference in fresh weight between the 1984-1 seedlings corresponds with the difference in seed weight (Table 2.1).

The present experiments also showed that apart from seed characteristics the actual ambient conditions strongly influenced emergence and seedling quality. Temperature is a prominent factor. Lowering of temperature reduced the rate of laboratory germination (Fig. 2.3) and the rate of emergence (Fig. 2.5) and the fresh weight of seedlings in the phytotron test. Lower temperatures were probably also the reason for reduced seedling growth in the field in early June (Fig. 2.8). In July field emergence was probably reduced by crust formation at the soil surface. Chapter 4 will deal with the influence of the matric head of the soil on emergence and seedling quality. In late September, both temperature and day length were unfavourable for seedling growth.

In conclusion, the present data show that viable and almost fully germinating seeds may strongly differ in vigour. Aspects of low vigour that have been shown in this Chapter are reduced rate of germination, lower seedling fresh-weight, and an increasing number of abnormal seedlings. Low vigour is one of the causes of poor field performance.

CHAPTER 3

EFFECT OF SEED SIZE ON GERMINATION AND VIGOUR

Abstract

The effects of seed size of cabbage seeds (Brassica oleracea L. Capitata group cv. Oscar) on germination, emergence, seedling growth and the sensitivity to rapid ageing were studied. Seeds of a 1984 lot showing high vigour were separated into 3 fractions, unsorted seeds served as control. Seed size had no effect on germination and percentage normal seedlings in the standard laboratory germination test. It did also not affect the final percentage emergence at temperatures ranging from 9 ° to 25 °C at phytotron conditions, nor the rate of emergence. However, seedling fresh-weight was closely related to seed size, the larger the seed, the better seedling growth. Thus, seed size influenced seedling growth irrespective of the physiological age of the seeds.

Introduction

Seed size is one of the most obvious physical parameters of seeds, and therefore is often used as a parameter of seed quality. Effects of seed size on various aspects of seed germination and seedling emergence and on growth and final yield of many vegetable crops have been documented. Germination of tomato and pimento seeds and seedling emergence of carrot seeds in the field were correlated positively with seed size (Cochran, 1974; Jacobsohn and Globerson, 1980). In onion, seed weight and emergence were not related, however (Bedford and Mackay, 1973). In vegetable crops such as, e.g., radish (Kubka et al., 1974), lettuce (Scaife and Jones, 1970), broccoli (Tompkins, 1966) and cucumber (Smittle and Williamson, 1978) seed size and seed weight correlated positively with seedling size and crop yield.

In this Chapter, the effects of seed size on germination, emergence, seedling growth and the sensitivity to rapid ageing of white cabbage seeds are investigated. Seeds of a seed lot that showed high vigour in the previous experiments (Chapter 2) were calibrated into different sizes and tested in the standard laboratory germination test as well as in the controlled deterioration test and were planted in the phytotron at temperatures ranging from 9 ° to 25 °C.

Material and methods

Two seed lots of hybrid cabbage seeds (Brassica oleracea L. Capitata group cv. Oscar) harvested in 1984 were used. The results of both seed lots were similar. Only the results of seed lot 1984-1 are presented. The seeds were separated at the Government Seed Testing Station, Wageningen by means of round sieves into different seed fractions: <2.0, 2.0 - 2.2 and >2.2 Ø mm. The unsorted seeds served as control. Experiments were performed in 1985. The conditions of the standard laboratory germination test, the controlled deterioration test, and the emergence and seedling growth in the phytotron were described in Chapter 2.

Table 3.1. Distribution according to size and weight of cabbage seeds from seed lot 1984-1.

Fractions (Ø mm)	distributions (%)	1000-seed weight (g)
Unsorted		5.37 ^{b}
<2.0	16	3.66 ^{d}
2.0 - 2.2	30	4.90 ^{c}
>2.2	54	6.26 ^{a}

Different letters demonstrate significant differences, $p = 0.05$ (Duncan's Multiple Range Test).

Results

The weight of the different seed-size fractions and their relative distribution in the seed lot are presented in Table 3.1.

In the standard laboratory germination test at diurnal temperatures of 20 ° - 30 °C seeds of all fractions and the unsorted controls germinated to a similar high percentage and produced a similar percentage of normal seedlings (Fig. 3.1A). Seed size also hardly influenced the reaction of the seeds to a controlled deterioration treatment of 24 hours at 45 °C at a seed moisture content of 24% (Fig. 3.1C). Both the percentages germination and normal seedlings were reduced in a nearly similar way. The control treatment at 5 ° instead of 45 °C did not differ from the untreated control (Fig. 3.1B).

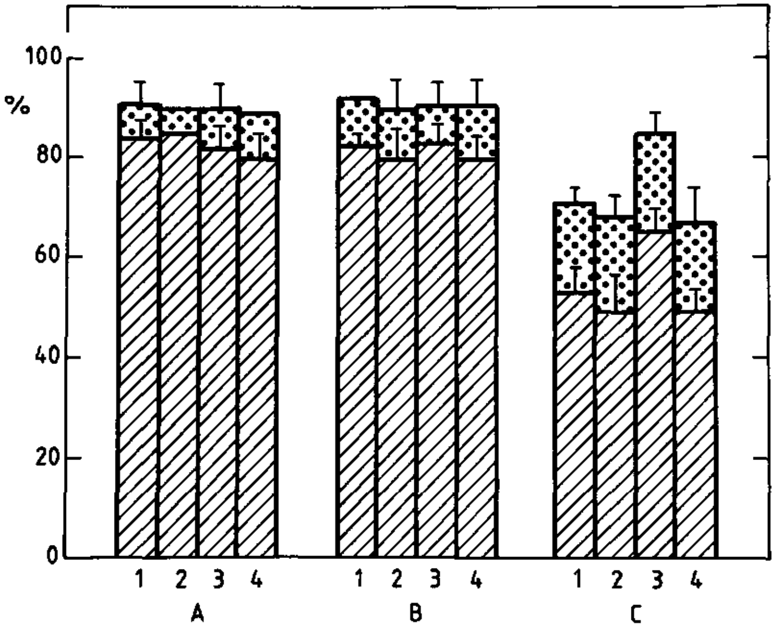

Fig. 3.1. Percentages germination (upper lines of bars) of seeds of the 1984-1 seed lot in the standard laboratory germination test at alternating diurnal temperatures of 20 ° - 30 °C. Seeds were either unsorted (1) or sorted in sizes with a diameter of <2.0 mm (2), 2.0 - 2.2 mm (3) and >2.2 mm (4). Seedlings were classified as *normal* (▨) *or abnormal* (⁘). *Seeds* were either sown directly (A) or after equilibration of the moisture content of the seeds to 24% on fresh weight basis. Those seeds were stored for 24 hours at either 5 °C (B) or 45 °C (C) before sowing. Single vertical bars indicate standard deviations >3%.

Seeds were also planted in the phytotron at temperatures ranging from 9 ° to 25 °C. In accordance with the results presented in Chapter 2 the temperature influenced the rate of emergence (Fig. 3.3) but did not affect the final percentage emergence (Fig. 3.2A). For both parameters the different seed size fractions and the unsorted control showed a similar response. An effect of seed size was seen, however, in the fresh weight of the seedlings at the 19th day after half-maximal seedling emergence (Fig. 3.2B). It is seen that the stimulating effect of rising temperature on seedling growth is strongly influenced by seed size, the larger the seeds, the heavier the fresh weight of the seedling. The unsorted seed lot had a position in accordance with the mean seed size and seed weight.

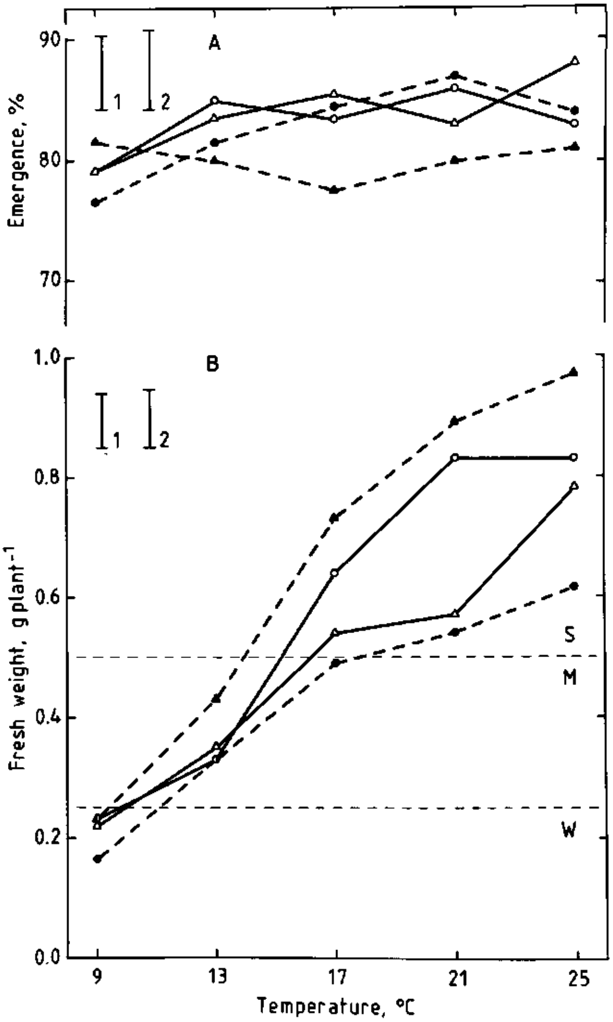

Fig. 3.2. The relation between temperature in the phytotron and the final percentage emergence (A) and seedling fresh weight at the 19th day after half-maximal seedling emergence (B) of seeds of the 1984-1 seed lot. Seeds were either unsorted (○) or sorted in sizes with a diameter of <2.0 mm (●), 2.0 - 2.2 mm (△) or >2.2 mm (▲). Seedlings were classified as strong (S), medium (M) or weak (W) according to fresh weight. LSD$_{0.05}$ for the effect of seed fraction and temperature is indicated by the vertical bars 1 and 2, respectively.

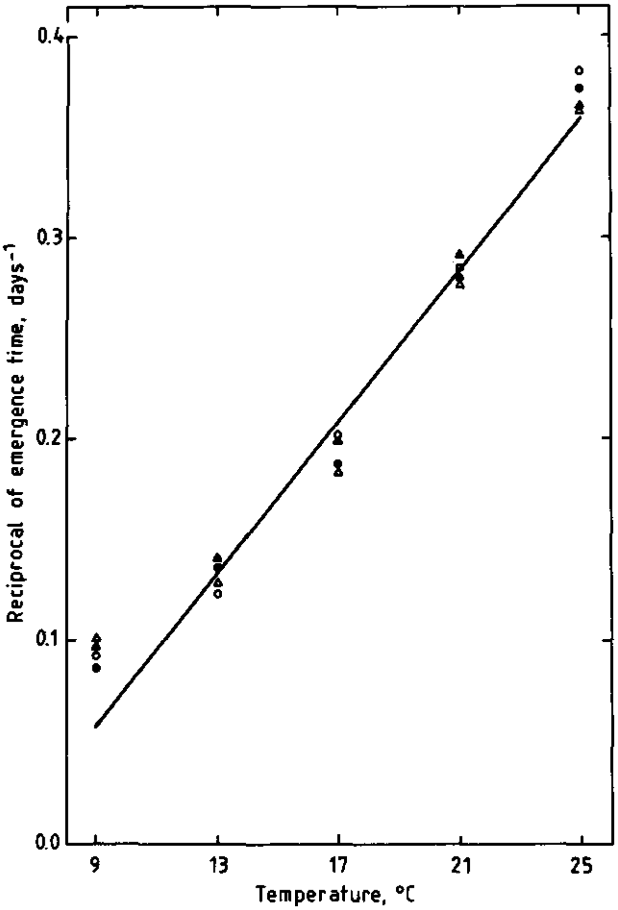

Fig. 3.3. The relation between the temperature in the phytotron and the reciprocal of the time for half-maximal emergence. Seeds were either unsorted (○) or sorted in sizes with a diameter of <2.0 mm (●), 2.0 - 2.2 mm (△) or >2.2 mm (▲).

Discussion

In cabbage, seed size and seed weight vary widely both within one seed lot (Table 3.1) and between seed lots harvested in different years and at different locations (See Chapter 2, Table 2.1).

The present data indicate that in commercial seed lot cabbage seed size is not a relevant factor in seed vigour. Seeds of the 1984-1 seed lot, separated into different fractions, reacted almost similarly to controlled deterioration (Fig. 3.1C). The same experiment was also performed with the 1984-2 seed lot. The data (not presented) fully supported this conclusion. The lack of correlation between seed size and seed vigour is also seen when 1984-1 and 1984-2 seeds are compared. Seeds of the 1984-2 lot have a lower seed weight (Table 2.1) and smaller seed size than the 1984-1 seeds. Nevertheless, the 1984-2 seeds germinated better after CD treatment than the 1984-1 seeds (Fig. 2.4). Thus, factors not related to seed size determine seed vigour in cabbage.

Seed size clearly affected seedling growth. Smaller seeds produced in the phytotron at all temperatures smaller seedlings than larger seeds (Fig. 3.2B). Again similar conclusions were reached with 1984-2 seeds (data not shown). In general, larger seedlings are more vigorous and have a better chance to survive in the field. Thus, seed size correlates positively with seedling vigour.

This conclusion is in good agreement with the studies of Smith et al. (1973) and Cochran (1974) on lettuce and pimento seeds.

It has to be remembered that seed vigour also influences seedling growth (Chapter 2). Seeds of nearly similar weight and size (1984-2, 1980, 1979-1) but of different vigour (Fig. 2.4) produced seedlings that strongly differed in fresh weight (Fig. 2.8).

That heavier seedlings grow from larger seeds is most obviously due to a larger embryo with more abundant reserve food and larger dimensions of the cotyledons. Such conditions favoured in soybean a more intensive process of photosynthesis and better growth (Burries et al. 1971; Carleton and Cooper, 1972).

The seed size and seed weight vary widely between the species of the Cruciferae, ranging from 3.6 mg seed^{-1} in Sinapsis alba to 0.06 mg seed^{-1}

in Diplotaxis harra (Takahashi and Suzuki, 1980). Small seeds disperse much easier than larger seeds. The present data clearly demonstrate that larger seeds produce heavier seedlings.

CHAPTER 4

EFFECT OF MATRIC HEAD AND SOIL TYPE ON EMERGENCE AND SEEDLING GROWTH IN RELATION TO VIGOUR

In collaboration with J.K. Wiemer and H. Koenraads

Abstract

The interaction of seed vigour and the matric head of the soil on emergence and seedling growth in river sand and sandy loam was studied in white cabbage seeds. The matric head of the soil was varied from 3 to 100 cm by means of a sand-box apparatus. Increasing matric head of both soils stimulated final percentage emergence and the rate of emergence. The reaction also depended on seed vigour. The influence of matric head on emergence is mainly due to the levels of oxygen, which depended on soil type. The fresh weight of the seedlings was different for high and low vigour seeds but was not influenced by matric head. It is concluded that emergence and seedling growth in soil are strongly affected by an interaction of seed and soil factors.

Introduction

Quick emergence of seedlings has long been the goal of agronomists and horticulturists in order to avoid the negative effects of soil crusting and pathogen attack on emergence and seedling growth. However, seedling emergence is affected by a complex interaction of seed and soil factors (Hegarty, 1976).

Seed vigour might be a crucial factor in seed emergence and seedling growth (Perry, 1982). It has been demonstrated in a number of species that the germination of seed lots after controlled deterioration (CD) is closely correlated with the field emergence (Matthews, 1980). It was shown in Chapter 2 that also in cabbage only those seeds that showed good germination and seedling growth after CD, performed well in the field.

Optimal soil temperature and soil moisture are required for a satisfactory start of crop growth in the field. However, the ideal combination of soil temperature and soil moisture seldom occurs in the field (Wagenvoort, 1981). An effect of soil factors such as soil type (Wagenvoort, 1981), soil temperature (Wagenvoort & Bierhuizen, 1977), soil capping (Sale & Harrison, 1964) and soil moisture (Gomaa, 1980; Wagenvoort, 1981) on emergence and seedling growth have been demonstrated in several crops. In vegetable crops such studies are very scarce, however. The present study investigates the effect of differing matric head of river sand and sandy loam on emergence and seedling growth of a series of cabbage seed lots with different seed vigour.

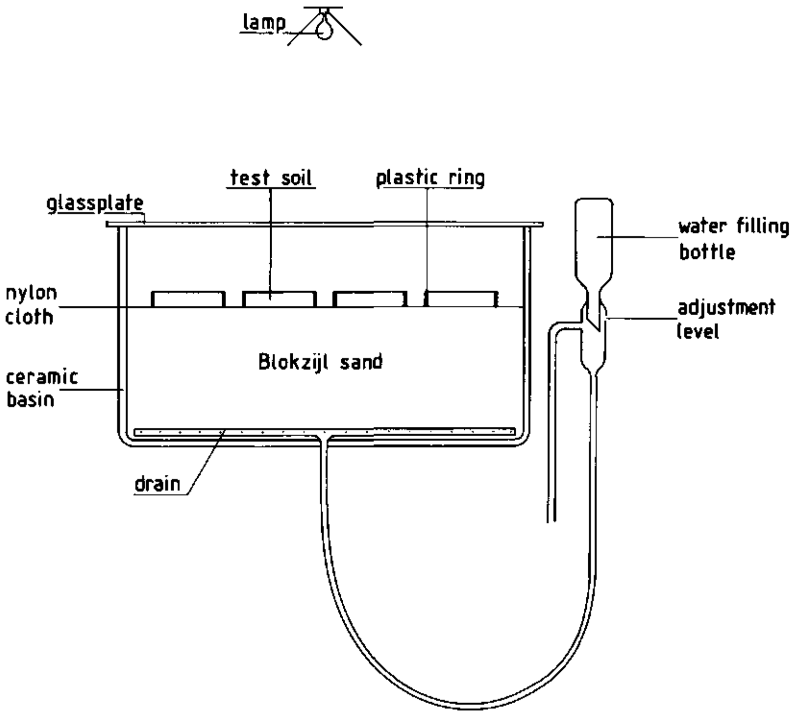

Fig. 4.1. Scheme of the sand-box apparatus.

Material and methods

Experiments were performed in 1985 and 1986 with the 5 seed lots of cabbage seeds (Brassica oleracea L. Capitata group cv. Oscar) that were described in Chapter 2.

Emergence and seedling growth were studied in a conditioned room at 18 ° ± 0.5 °C in soil media consisting of either sterilized river sand or sterilized sandy loam. Sterilization occurred at 60 °C during 8 - 12 hours before sowing. The effect of the matric head of the soil (indicated as h_m

and expressed as cm water column) within the range of 3.2 - 100 cm water column was measured in a sand-box apparatus (Stakman et al., 1969). The sand-box apparatus (Fig. 4.1) consists of a ceramic basin filled with Blokzijl sand. The sand was inundated with water to drain off the air. The moist surface was covered with a nylon cloth. Twelve samples of the test soil were placed on top of the cloth within plastic rings of 2 cm height, ø 10 cm. The bottom of the rings consisted of nylon cloth fixed with a plastic band. Equilibrium of sand and test soil to the appropriate h_m was achieved within four days. The basins were covered with glass to reduce evaporation. The soil-water retention curves were monitored both in undisturbed soil and in soil in which seeds were sown and seedlings had emerged. Soil water content was measured by weighing the soil in the rings before and after drying for 3 days at 60 °C in an oven with forced ventilation followed by 3 days at 110 °C. The germination of each seed lot was tested by means of standard laboratory germination test (Chapter 2) before the seeds were sown in the test soil. A split plot design with 3 replications was carried out. Fifty seeds per ring were sown at a depth of approximately 0.4 cm and covered with test soil. An 8 hours day-length was provided by a 160-watt blended light over each sand-box during the experimental period.

The seedlings were considered to be emerged when the hypocotyl disturbed the surface of the test soil. Counting of the emerged seedlings started 28 and 40 hours after sowing in the river sand and sandy loam, respectively. During the first 5 days the emerged seedlings were counted at 8 hours intervals and thereafter twice a day. Data of the emergence counts were fitted against time according to the following exponential equation, described by Milthorpe and Moorby (1979):

$$p = A\,(1 - e^{-k\,(t\,-\,t_0)})$$

where p = percentage emergence at time t; A = final percentage emergence; k = a value for time spread of emergence in the population (h^{-1}), k is the slope in the linear relationship between ln (p/A + 1) and (t -t_0); t_0 = the time of first seedling emergence (h). At 10 days after half-maximal seedling emergence (t_{50}) the fresh weight (FW) and dry weight (DW) of the seedlings were measured. Dry weight of the plants was measured after oven drying at 60 °C for one week. The experiments were statistically analysed with the VAX computer system of the Agricultural University, using a GENSTAT program. Analysis of variance was carried out to determine which effects significantly influenced the total variance in the experimental results.

Results

Soil-water retention curves

The effect of a variation of the matric head of soil on the retention of soil water is shown in Fig. 4.2. The air volume was calculated from the soil water data. The curves for river sand and sandy loam differed considerably. In both soils the soil-water content decreased and the air volume in the soil increased when the matric head increased. In river sand the main changes occurred at h_m values between 10 and 20 cm, in sandy loam the changes occurred more gradually. Evidently, sandy loam has a much higher water holding capacity than river sand, due to the considerable difference in pore diameter of both soil types.

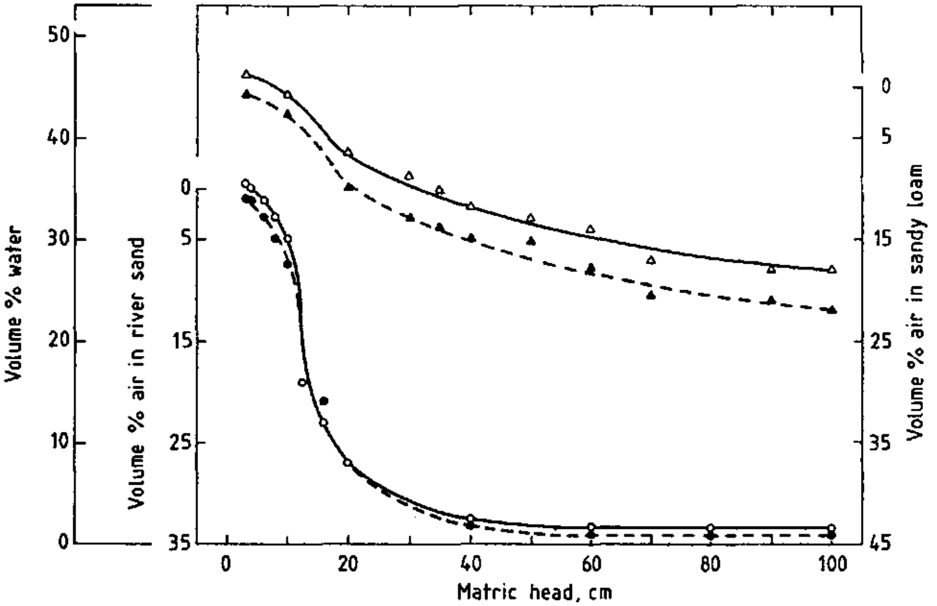

Fig. 4.2. Effect of matric head of the soil on the volume percentages H_2O and air for river sand (○●) and sandy loam (△▲) determined in undisturbed soils (—) and in soil in which seedlings had emerged (- - -).

Emergence and seedling growth

The effect of matric head on emergence and seedling growth was tested in river sand and sandy loam with seeds of 5 or 4 seed lots of different actual and physiological age (Chapter 2), respectively. The results of an analysis of variance on the combined data is shown in Table 4.1. The parameters A, t_0, t_{50}, k, FW and DW were both in river sand and sandy loam significantly affected by matric head and seed lot. Apart from the values of A, t_0, t_{50} and k in river sand the effects of matric head and seed lot showed a significant interaction. The effect of the matric head was particularly strong on the value of A and t_{50} in both test soils and of t_0 in sandy loam, while the seed age had its strongest effect on FW in both soils and on t_0 in river sand.

Table 4.1 The percentages total variance of soil matric head, seed lot, interaction and the residual value of 6 different parameters expressing different aspects of seedling emergence and development. The significance is indicated: *, significant at 5%; NS, no significant difference.

Soil type	Parameter	Percentage of total variance			
		Matric head	Seed lot	Interaction	Residual
River sand	A	89 *	2 *	3 NS	6
	t_0	10 *	64 *	7 NS	19
	t_{50}	52 *	10 *	18 NS	20
	k	20 *	16 *	19 NS	45
	FW	20 *	61 *	9 *	10
	DW	17 *	25 *	37 *	21
Sandy loam	A	84 *	8 *	3 *	5
	t_0	50 *	15 *	28 *	7
	t_{50}	73 *	10 *	9 *	8
	k	40 *	29 *	19 *	12
	FW	3 *	87 *	5 *	5
	DW	39 *	36 *	15 *	10

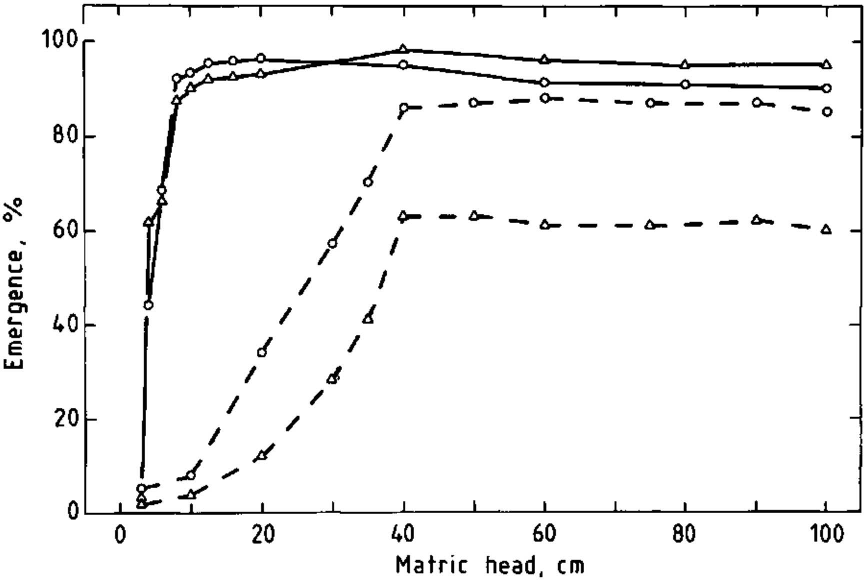

Fig. 4.3. The effect of matric head of the soils on the final percentage emergence of cabbage seed lots 1984-1 (○) and 1979-1 (△) in river sand (—) and sandy loam (---).

The results of the 1984-1 and 1979-1 seed lot are presented in detail in Figs. 4.3 - 4.8. The influence of the matric head on the final percentage emergence (A) is different in the 2 soil types (Fig. 4.3). In river sand emergence is optimal in both seed lots at h_m 10 - 100 cm whereas in sandy loam maximum emergence occurs at h_m 40 - 100 cm.

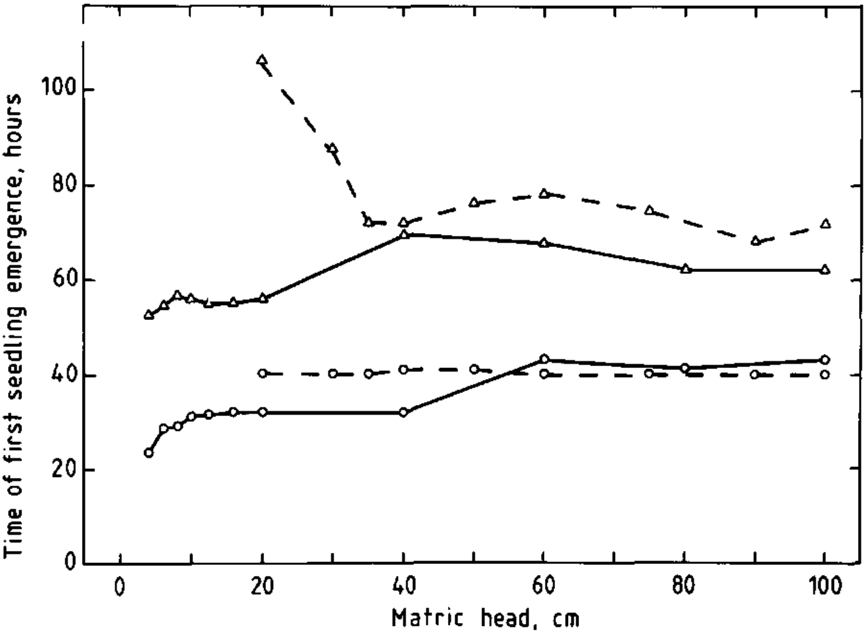

Fig. 4.4. The effect of matric head of the soils on the time of first seedling emergence (t_0) of cabbage seed lots 1984-1 (○) and 1979-1 (△) in river sand (—) and sandy loam (---).

The time of first seedling emergence (t_0) is hardly influenced by the range of matric head (Fig. 4.4). It mainly depended on the seed lot with the exception of 1979-1 seeds in sandy loam at h_m 40 cm. Similar effects of seed lot on t_0 are described in Chapters 5 and 6.

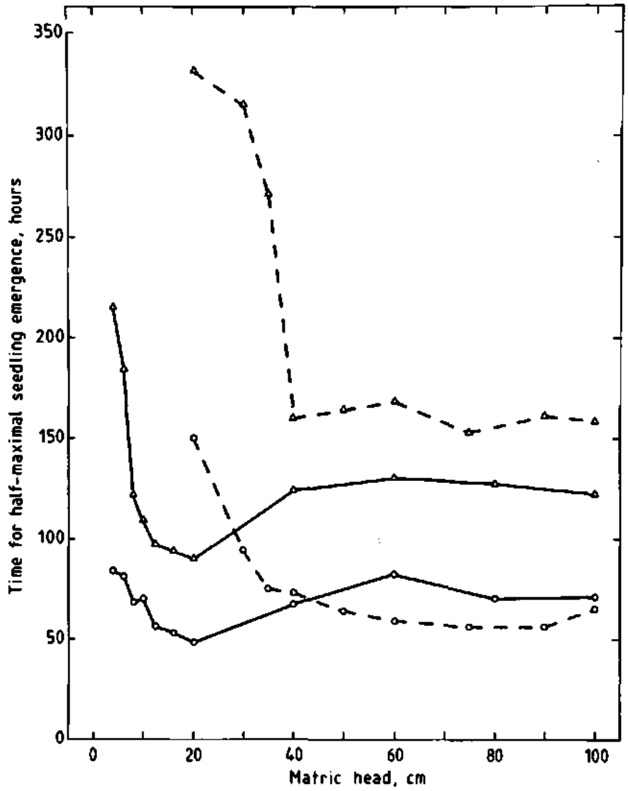

Fig. 4.5. The effect of matric head of the soils on the time for half-maximal seedling emergence (t_{50}) of cabbage seed lots 1984-1 (○) and 1979-1 (△) in river sand (—) and sandy loam (---).

The time for half-maximal seedling emergence (t_{50}) was strongly dependent on the matric head of the soil (Fig. 4.5). As in the case of the final percentage emergence (Fig. 4.3) the h_m values at which the value of t_{50} became more or less stable were 10 and 40 cm in river sand and sandy loam, respectively. At h_m values above 40 cm the t_{50} only depended on the seed lot.

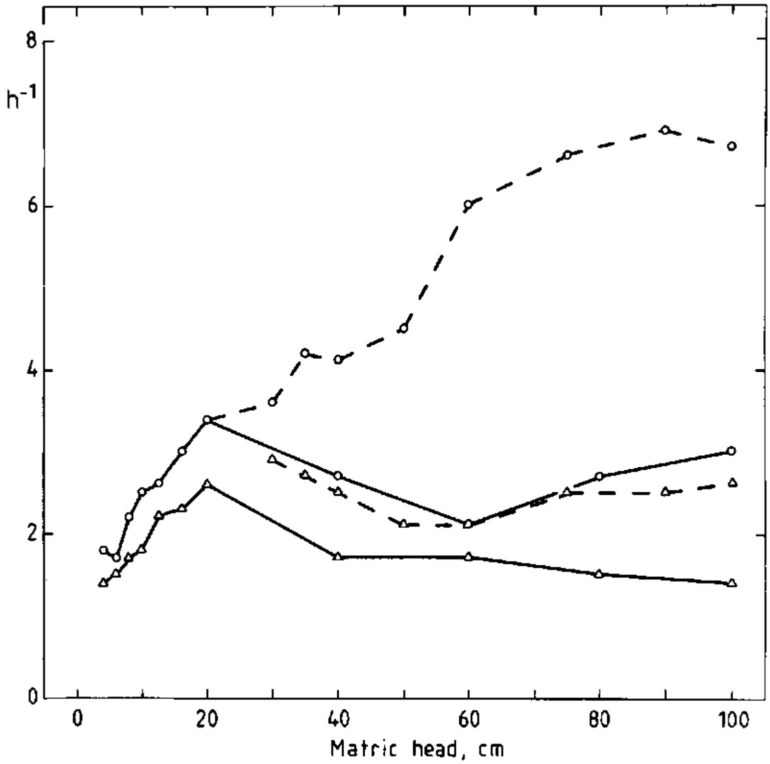

Fig. 4.6. The effect of matric head of the soils on the time spread of emergence (k) of cabbage seed lots 1984-1 (○) and 1979-1 (△) in river sand (—) and sandy loam (- - -).

The parameter k is an expression of the uniformity of emergence. The higher the k, the steeper the slope of the germination curve, the more uniform the seedlings stand. Its value in the 1979-1 seeds was not strongly influenced by the matric head in both soil types and in the 1984-1 seeds in sandy loam. However, k of the 1984-1 seeds was strongly affected in sandy loam. The higher the h_m the more uniform emergence.

The fresh and dry weight of the seedlings at 10 days after the moment of half-maximal seedling emergence was also hardly affected by the matric head of the soils (Figs. 4.7, 4.8), the only exceptions were the dry weights of 1984-1 seedlings in river sand. The FW values clearly depended on the seed lot.

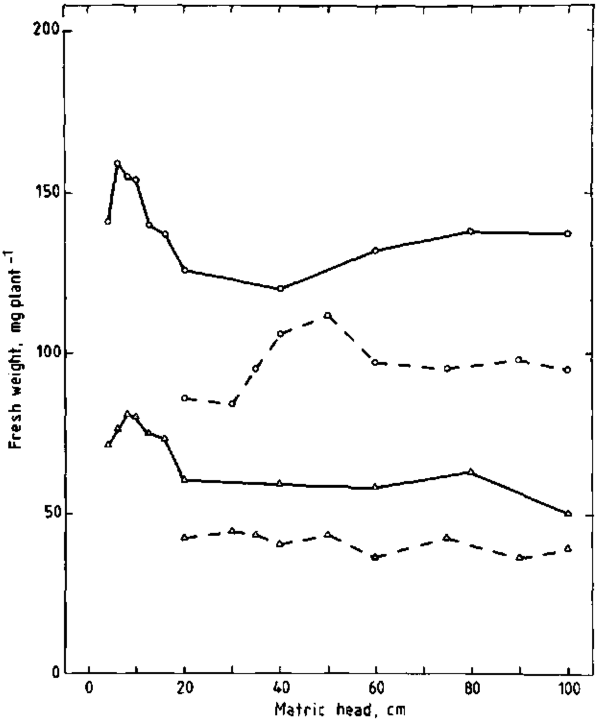

Fig. 4.7. The effect of matric head of the soils on the mean fresh weight of seedlings of cabbage seed lots 1984-1 (○) and 1979-1 (△) in river sand (—) and sandy loam (---).

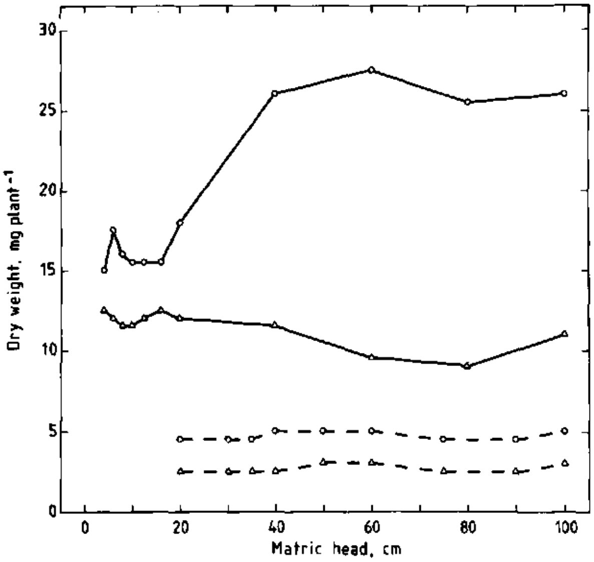

Fig. 4.8. The effect of matric head of the soils on the mean dry weight of seedlings of cabbage seed lots 1984-1 (○) and 1979-1 (△) in irver sand (—) and sandy loam (---).

Discussion

The data presented in this Chapter are a good illustration of the statement that emergence and development of seedlings in the field are strongly affected by a complex interaction of seed and soil factors (Hegarty, 1976).

The main seed factor that was studied in this Chapter was seed vigour. Several aspects of low vigour in white cabbage seeds have been described in Chapter 2. It was shown that the low-vigour 1979-seeds produced more abnormal and smaller seedlings than high-vigour 1984-seeds. Low vigour was also expressed as reduced resistance to accelerated ageing in the controlled deterioration test. The rate of germination and emergence of low vigour seeds was reduced. In the present experiments the seed lot factor had a strong effect on the t_0 value in river sand (Table. 4.1). In Figs. 4.4 and 4.5 it is shown that at h_m values above 40 cm in both soil types the values of t_0 and t_{50} of the 1979-1 seeds were always higher than of the 1984-1 seeds. The fresh weight of 1979-1 seedlings is lower than that of 1984-1 seedlings at all h_m values (Fig. 4.7). The seed lot effect on fresh weight may also partly depend on the difference in seed weight between the seed lots (Table 2.1). It was described in Chapter 3 that 1984-1 seeds of different size showed similar seed vigour, as judged from the controlled deterioration test, but differed in seedling fresh-weight (Fig. 3.2).

As soil factors the influence of matric head and soil type was studied. The matric head mainly influenced emergence. At increasing h_m values final emergence rose (Fig. 4.3) and t_0 and t_{50} decreased (Figs. 4.4 and 4.5). In river sand these changes occurred at h_m values between 3 and 10 cm, in sandy loam the curves reached final value around h_m 40 cm. These observations favour the conclusion that the influence of matric head on emergence is mainly due to the air volume of the soil. It is not likely that the decreasing water availability at increasing h_m values plays a major role, since nearly all parameters stayed at h_m values above 40 cm either stable or changed in a positive direction (Figs. 4.6, 4.8).

The critical h_m values for A, t_0 and t_{50} were different in river sand and sandy loam. In sandy loam the maximum emergence of 1979-1 seeds and the fresh and dry weight of seedlings of both seed lots were lower than in river sand (Figs. 4.7 and 4.8). It is proposed that the smaller air volume of sandy loam was the main factor that reduced emergence and seedling growth. An effect of soil nutrition is not likely since emergence and early seedling growth mainly depend on the food reserves of the seed.

In conclusion, the present data have shown that the emergence and seedling growth of white cabbage seeds depend as well on the vigour of the seeds as on soil type and matric head. Interactions between these factors have to be taken into account when data of laboratory germination tests are related to field performance.

CHAPTER 5

EFFECTS OF PRIMING ON GERMINATION AND VIGOUR

Abstract

The effects of priming or osmotic pre-incubation on germination, emergence and seedling growth of high and low vigour cabbage seeds were studied. Seeds of Brassica oleracea L. Capitata group cv. Oscar and B. oleracea L. Sabauda group cv. Samoa were primed in -1.5 MPa PEG. A temperature of 20 °C was optimal. Priming increased the number of normal seedlings that developed from low vigour seeds in the standard laboratory germination test. It also advanced the start of germination in all seed lots and enhanced the rate of germination in the low vigour seeds. Primed seeds also performed better than untreated seeds in the field. Seedling growth from primed seed was improved both at the controlled conditions of the phytotron and the natural conditions in the field.

Introduction

It was argued in Chapter 1 that ageing is the sum total of all deteriorative processes which eventually lead to death. For practical purposes, seeds are considered dead when in the absence of dormancy they fail to germinate at optimal conditions. Before seeds become incapable of germination several signs of the progressive ageing appear (Heydecker, 1972; Roberts, 1973). Obvious manifestations of ageing in seeds are reduction in the rate of germination and the production of smaller and often abnormal seedlings (Parrish & Leopold, 1978; Ellis & Roberts, 1980).

It has been suggested that some of the processes that precede seed death are reversible. Villiers and Edgecumbe (1975) explained the survival of seeds in soil by the assumption that molecular damage is repaired in fully imbibed seeds in contrast to seeds at low moisture contents. Seeds of aged, low vigour seeds of Brussels sprouts that were held for a certain time on the brink of germination during incubation in osmotic solutions, germinated at a higher rate and were more resistant to controlled deterioration than the control seeds (Burgass & Powell, 1984). Improvement of the quality of low vigour seeds adds another item to the advantages of priming treatments that are already known such as (1) enhancement of germination (Khan et al., 1980/81), (2) broadening of the temperature range of germination (Bodsworth & Bewley, 1981; Weges, 1987) and (3) synchronizing of germination

(Bodsworth & Bewley, 1981).

It is the aim of the experiments described in this Chapter to study invigoration of low-vigour cabbage seeds by osmotic priming.

Material and methods

Seed materials

Experiments were performed in 1985 and 1986 with 2 seed lots of white cabbage (Brassica oleracea L. Capitata group) cv. Oscar harvested in 1979 and 1984, respectively, and 1 lot of F_1 hybrid savoy cabbage (Brassica oleracea L. Sabauda group) cv. Samoa harvested in 1984. The seeds of cv. Oscar were stored dry under normal commercial conditions till September 1984 and from than on at 5 °C and 30% r.h. until use. Seeds of Samoa were changed to 5 °C in March 1986.

Priming of seeds

Osmotic potentials of PEG solutions (polyethylene glycol of molecular weight 6000) were calculated according to Michel (1983). Batches of 250 seeds were incubated in transparent plastic boxes (diameter 11 cm, height 7 cm) that contained 3 layers of filter paper (Ø 8 cm) and 1 layer of filter paper (Ø 10 cm) (Schut & Zonen, Heelsum, The Netherlands) saturated with 18 ml of -1.5 MPa PEG solution. Priming occurred at a range of temperatures between 5 ° and 35 °C, during a period varying from 1 to 21 days, or at 20 °C only during a fixed number of days. The pretreatment occurred in the dark. After the osmotic pre-incubation the seeds were thoroughly washed by means of tap water. In most experiments seeds were dried back in a stream of air at 20 °C during 16 hours to a moisture content that was very close to that of untreated seeds.

Germination test

The germination and seedling growth of primed seeds were tested in the standard laboratory germination test as described in Chapter 2. A randomized complete block design with four replications of 50 seeds of each treatment was carried out. Untreated seeds served as control. Any seed producing a radicle was counted as germinated. The germinated seeds were counted at 4 hours intervals during the first 64 hours and thereafter two times a day. The seedlings were evaluated as normal and abnormal in accordance with the ISTA rules (Chapter 2). The controlled deterioration test is also described in Chapter 2.

Phytotron test

Emergence and growth of seedlings developing from primed seeds were tested in the phytotron at 21 °C. A randomized complete block design with 4 replications was carried out. Fifty seeds per seed lot and per treatment were sown in a soil medium. The soil medium and other conditions in the phytotron, the criterion for emergence and the characterization of the seedlings were described in Chapter 2.

Field test

Seedling emergence and growth were also tested in the field. The experiment was carried out on the experimental field of the Department of Horticulture. On July 1, 1986, a randomized complete block design with four replications of 100 seeds of each treatment was sown. The row length was 300 cm and the distance between rows 50 cm. Untreated seeds served as control. The seeds were considered to be emerged when the cotyledons were in a horizontal position. Regular water sprays were carried out during the seedling growth period. In order to prevent bird damage, the field was covered by a net during the period of the experiment. The seedlings were evaluated and classified at the 19th day after half-maximal seedling emergence (Chapter 2).

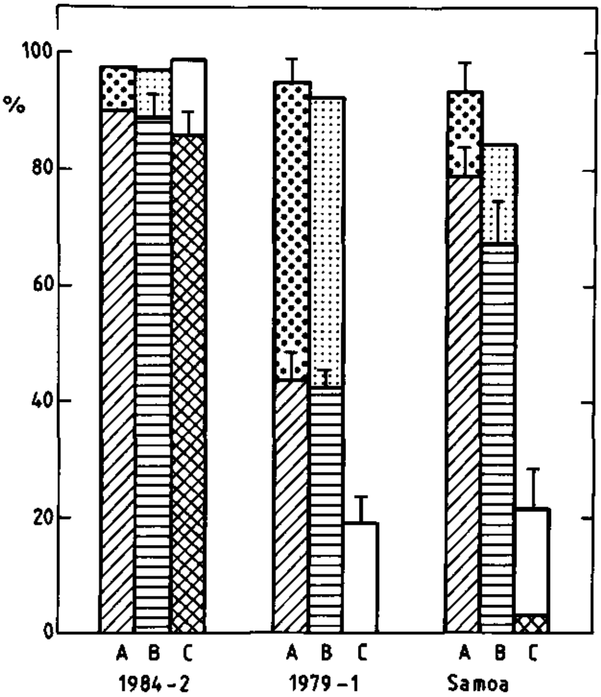

Fig. 5.1. Percentages germination (upper line of bars) of seeds of 3 lots in the standard laboratory germination test. Seeds were either sown directly (A) or moisture content of the seeds was equilibrated to 24% on fresh weight basis. Those seeds were stored for 24 hours at either 5 °C (B) or 45 °C (C). Seedlings were classified as normal (▨,▤,▩) and abnormal (⁘,⁙,☐). Single vertical bars indicate standard deviation >3%.

Results

Germination and vigour tests

At the start of the experiments the germination of the seeds of B. oleracea L. Sabauda group cv. Samoa F1-RS (to be indicated as Samoa seeds) were tested in the standard laboratory germination test without and with a preceding controlled deterioration treatment (Fig. 5.1). The Samoa data were compared to new standard laboratory test and CD test data on the 1984-2 and 1979-1 seed lots of B. oleracea L. Capitata group cv. Oscar (to be indicated as 1984-2 and 1979-1 seeds). It is shown that before treatment (A) or after 24 hours at 5 °C (B) nearly all seeds germinated. However, the differences in the number of normal seedlings pointed already to a variation in seed quality. This suggestion was supported by the reaction of the different seed lots to rapid ageing during 24 hours at 45 °C (C). Evidently, the 1979-1 and Samoa seeds had much lower vigour than the 1984-2 seeds.

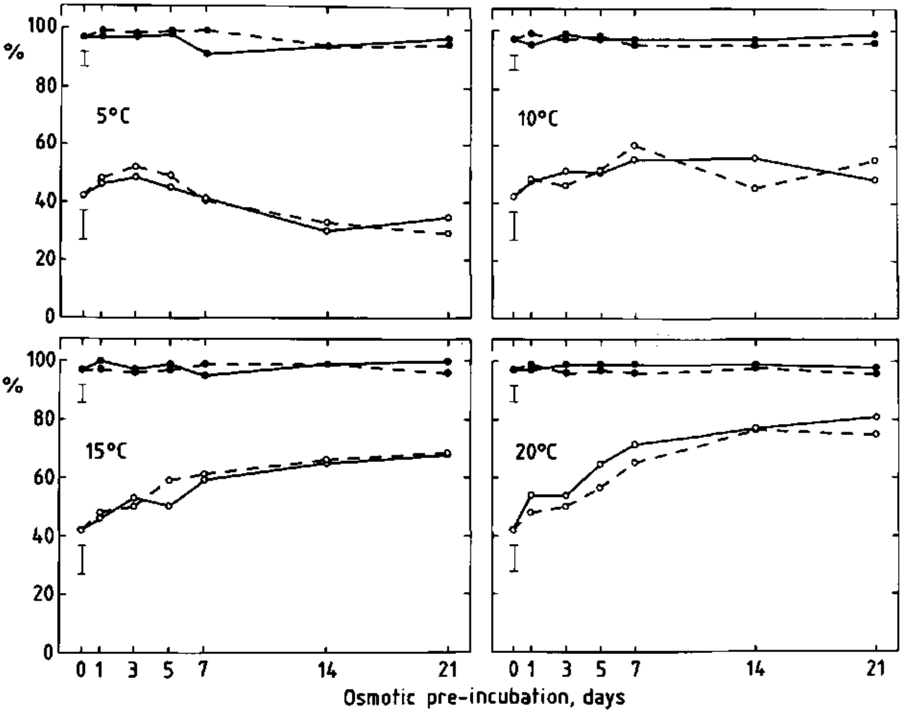

Fig. 5.2. Effect of osmotic pre-incubation at different temperatures on percentages germination (●) and normal seedlings (○) of the 1979-1 seed lot. Seeds were pre-incubated in -1.5 MPa PEG at 5 °, 10 °, 15 °, or 20 °C during the indicated period of time. One half of the seeds was dehydrated following osmotic pre-incubation (——), the other half was transferred directly to the conditions of the standard laboratory germination test (– –). Vertical bars indicate $LSD_{0.05}$.

Priming conditions

The optimal conditions of the priming treatment were tested in the low vigour 1979-1 seeds. Temperature was varied from 5 ° to 20 °C and the duration of the priming from 0 to 21 days (Fig. 5.2). Priming occurred in -1.5 MPa PEG. Preliminary experiments (not shown) indicated that incubation in higher osmotic potentials carried the risk of germination during the pretreatment. It is shown that in most of the conditions priming did not affect the final percentage germination. However, the number of normal seedlings clearly increased during priming at 20 °C and to a lesser extent

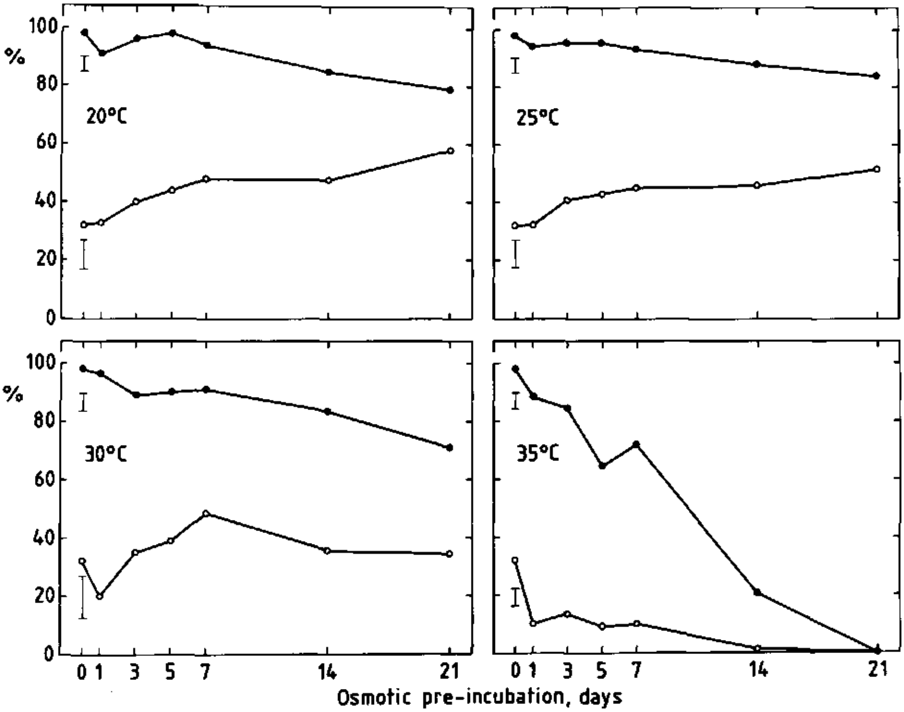

Fig. 5.3. As Fig. 5.2 but for pre-incubation at 20 °, 25 °, 30 ° and 35 °C. All seeds of 1979-1 seed lot were dehydrated following pre-incubation.

at 15 °C. Dehydration did not affect the result. The experiment was extended to higher temperatures (Fig. 5.3), dehydration was now a standard procedure. At 35 °C and to a lesser extent at the lower temperatures, priming decreased the percentage germination possibly due to induction of secondary dormancy. Nevertheless, at 20 ° and 25 °C the percentage normal seedlings increased. It is concluded that 20 °C is the optimal temperature for an osmotic pretreatment of cabbage seeds that is directed to the improvement of the seedling quality. In a second priming method, Erlenmeyer flasks (200 ml) containing 15 ml PEG solution and 250 seeds were constantly shaken. The results were similar or slightly inferior to the above results (data not shown). For practical reasons the pretreatment in plastic boxes was accepted as standard treatment.

Priming and germination

Another effect of priming was seen when the time course of germination was closely followed (Fig. 5.4). Priming in -1.5 MPa PEG at 20 °C evidently advanced the start of germination in all seed lots and, in addition, increased the rate of germination, particularly in the low vigour 1979-1 and Samoa seeds. The latter effect can be regarded as a synchronization of the germination process in the population. Priming during 7 days caused almost the maximal effect. Shorter priming treatments of 1979-1 seeds showed that also after 1 day of osmotic pre-incubation the start of germination was already advanced (Fig. 5.5). Priming not fully diminished the differences between high and low vigour seed lots, 1984-2 seeds germinated still earlier and faster than 1979-1 and Samoa seeds (Fig. 5.4).

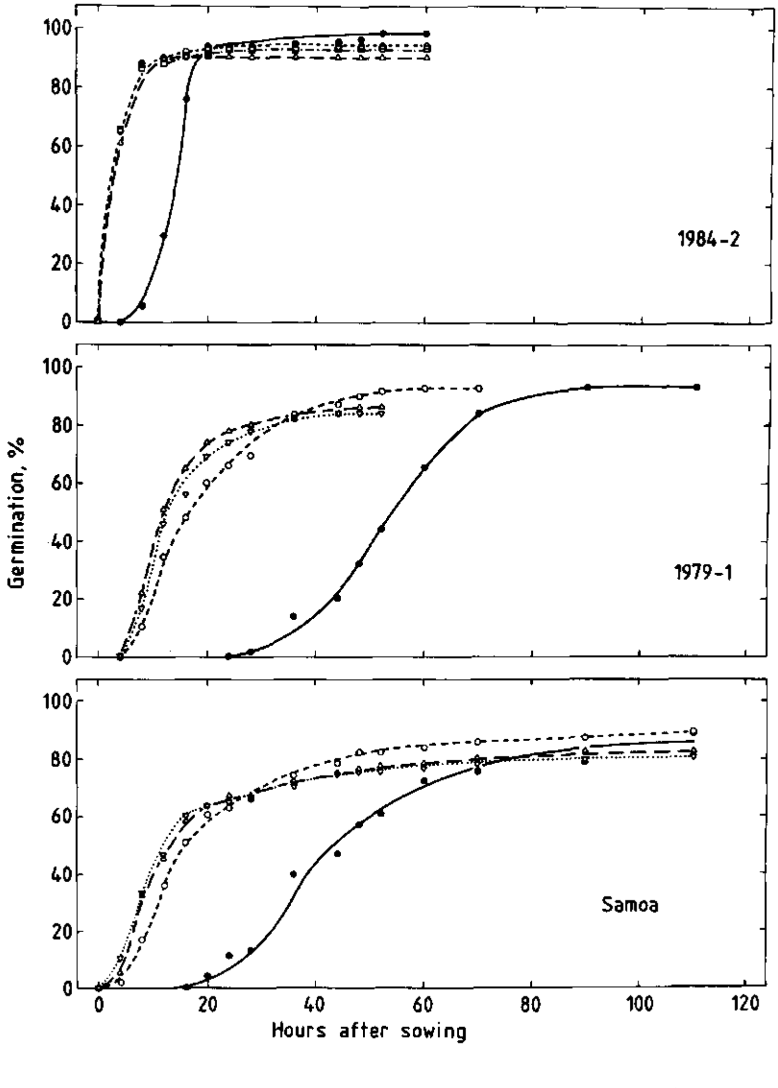

Fig. 5.4. Effect of osmotic pre-incubation on the time course of germination of the three seed lots. Seeds were pre-incubated in -1.5 MPa PEG at 20 °C during 7 (○), 10 (□), 14 (△) or 21 days (▽) or untreated (●). Germination occurred after dehydration at the conditions of the standard laboratory germination test.

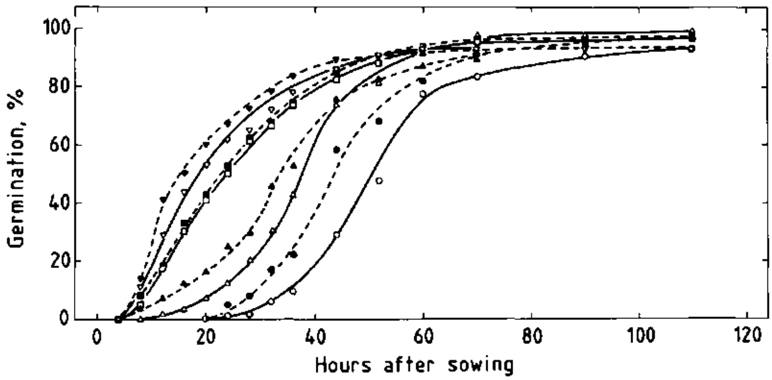

Fig. 5.5. Effect of osmotic pre-incubation on the time course of germination of the 1979-1 seeds. Seeds were pre-incubated in -1.5 MPa PEG at 20 °C during 1 (●), 2 (△), 3 (▲), 4 (□), 5 (■), 6 (▽) or 7 days (▼) or untreated (○). Germination occurred after dehydration at the conditions of the standard laboratory germination test.

Emergence and seedling growth

The effects of priming during 7, 14 or 21 days on seedling emergence and seedling growth were studied both in the controlled conditions of the phytotron at 21 °C and in the natural conditions of the field in July 1986. Seedlings were evaluated at the 19th day after half-maximal seedling emergence. This method separated effects on seedling growth from differences in the rate of emergence. In the standard laboratory test the effect of these prolonged priming times was also studied. It is shown in Fig. 5.6A that the germination of all seed lots in this test was slightly decreased. During the first 14 days the percentage normal seedlings increased in the 1979-1 seed lot from 30 to 60%, but was hardly affected in the other two seed lots (Fig. 5.6B).

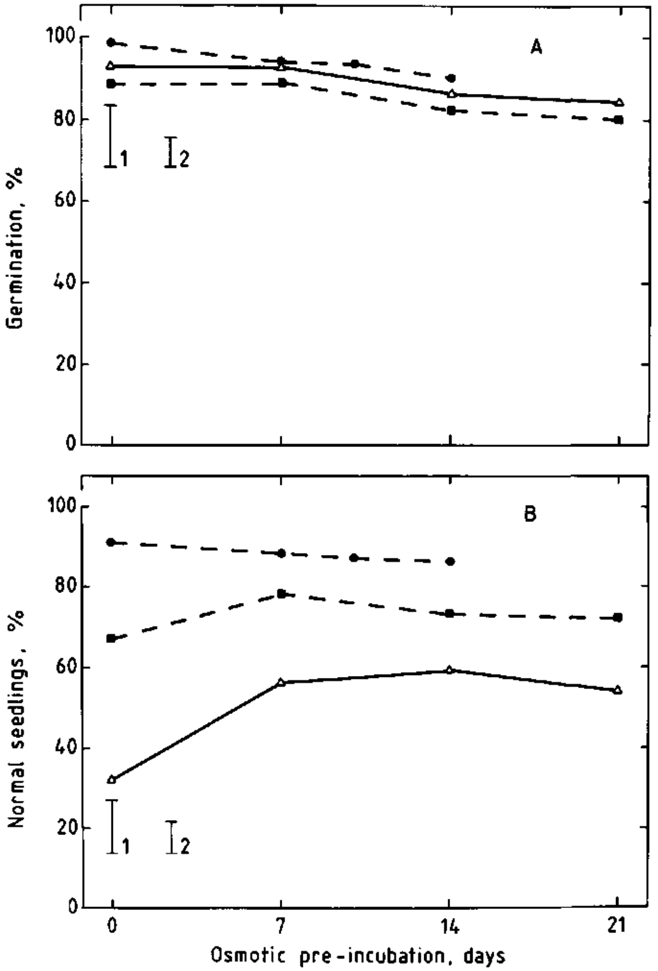

Fig. 5.6. Percentages germination (A) and normal seedlings (B) of 1984-2 (●), 1979-1 (△) and Samoa seeds (■) as a function of osmotic pre-incubation. Seeds were pre-incubated in -1.5 MPa PEG at 20 °C during the indicated periods of time. Germination occurred following dehydration at the conditions of the standard laboratory test. Vertical bar 1 indicates $LSD_{0.05}$ for 1984-2 and bar 2 for 1979-1 and Samoa seeds.

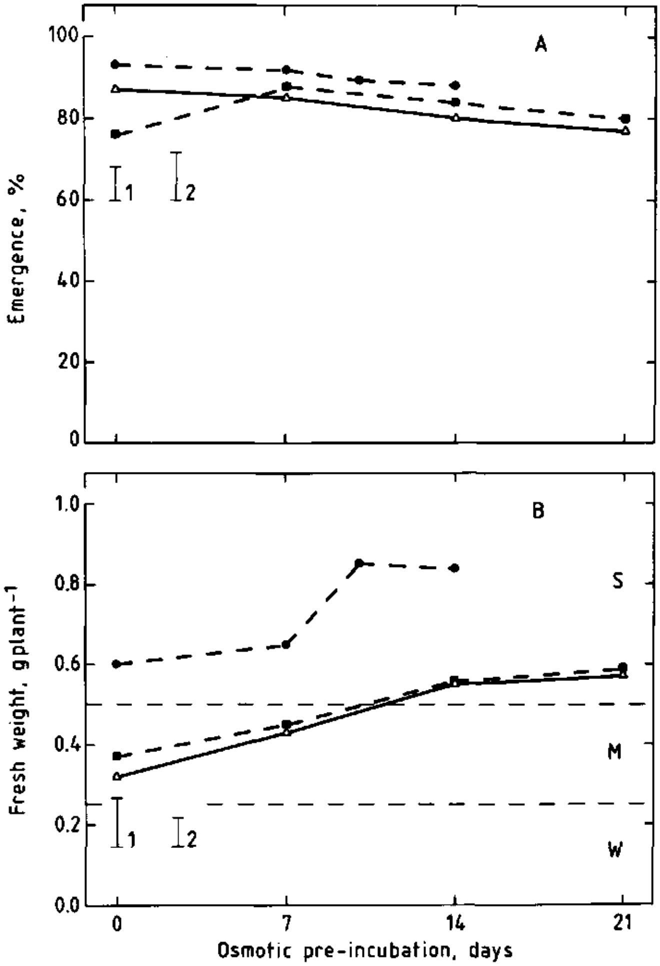

Fig. 5.7. Percentage of seedling emergence (A) and seedling fresh weight (B) in phytotron conditions at 21 °C of 1984-2 (●), 1979-1 (△) and *Samoa* (■) seeds as a function of osmotic pre-incubation. Seeds were pre-incubated as in Fig. 5.6. Vertical bars indicate $LSD_{0.05}$. Seedling fresh weight was determined at the 19th day after half-maximal seedling emergence. Seedlings were classified as strong (S), medium (M) and weak (W) according to fresh weight.

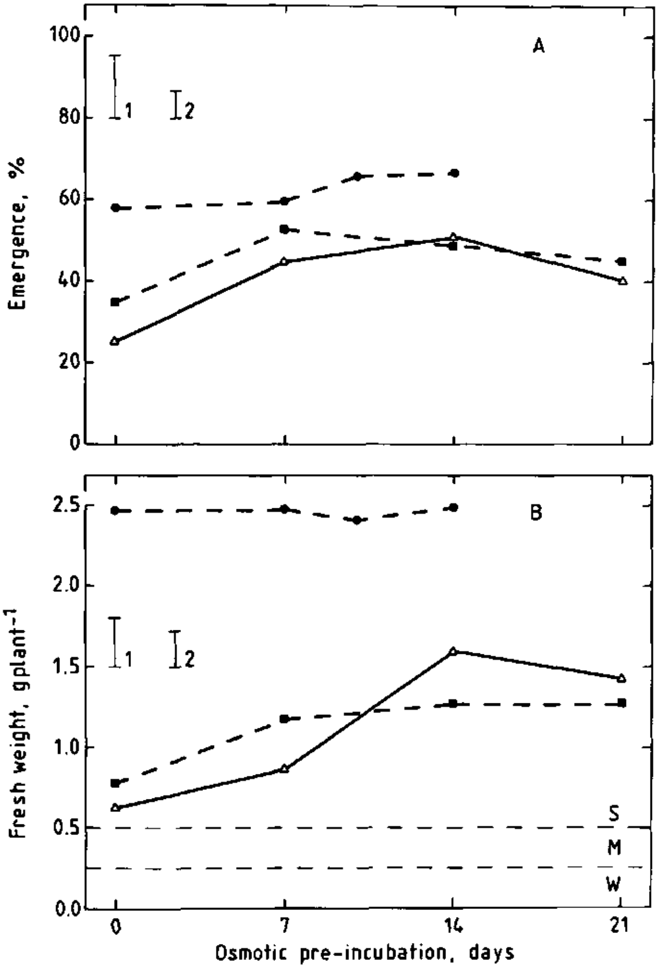

Fig. 5.8. Percentage of seedling emergence (A) and seedling fresh weight (B) for seeds sown in the field in July 1986 of 1984-2 (●), 1979-1 (△) and Samoa (■) seeds as a function of osmotic pre-incubation. Seeds were pre-incubated as in Fig. 5.6. Vertical bars indicate $LSD_{0.05}$. Seedling fresh weight was determined at the 19th day after half-maximal seedling emergence. Seedlings were classified as strong (S), medium (M) and weak (W) according to fresh weight.

Emergence of seedlings in the phytotron test (Fig. 5.7 A) resembled the results of the germination test. Again, prolonged priming tended to decrease total emergence slightly. In the field seedling emergence in July was rather low (Fig. 5.8 A). A similar result was obtained before (Fig. 2.7). Total percentage field emergence was positively influenced by priming, in the 1979-1 seed lot it rose from 25 to 50%.

Both in the phytotron and in the field the mean fresh weight of the seedlings was also clearly stimulated by seed priming (Figs. 5.7 B, 5.8 B). In the present experiment seedling growth in the field of the untreated 1984-2 and 1979-1 seed lots was more prosperous than in the study described in Chapter 2. Priming stimulated particularly the seedling growth of the two low-vigour seed lots.

Discussion

The present data clearly showed that osmotic pre-incubation of white cabbage and savoy cabbage seeds has two distinct effects: (1) germination is accelerated and (2) seedling growth is improved. One day of priming enhanced the start of germination of the 1979-1 seeds already with about 10 hours (Fig. 5.5), a 7 days treatment afforded almost an optimal effect (Figs. 5.4, 5.5). In most experiments 14 days of priming caused a larger improvement of seedling growth than 7 days (Figs. 5.6, 5.7, 5.8). Such long priming treatments have the disadvantage that the beneficial effects of the treatment started to be antagonized by inhibitory processes. It is shown in Figs. 5.6, 5.7 and 5.8 that 21 days priming resulted in somewhat lower percentages germination and emergence. Germination was strongly inhibited by osmotic pre-incubation at temperatures above 20 °C (Fig. 5.3). It was not examined whether the inhibition was caused by induction of secondary dormancy or loss of viability.

Acceleration and synchronization of germination have often been described as beneficial effects of priming in field crops such as barley, maize and soybean and vegetables such as celery, leek and lettuce. The effect is particularly dramatic in low vigour seed lots like the 1979-1 and Samoa seeds in the present experiments. Evidently, this effect of priming has great practical significance.

The invigoration effect of priming has been reported for a few species by Burgass and Powell (1984). Improvement of seedling performance in the field of primed seeds of carrot, celery and onion has been reported by Brocklehurst and Dearman (1983). The method certainly has no practical meaning for seed lots with extremely low vigour like the present 1979-1 and Samoa seeds. It is shown in Fig. 5.8 that the fresh weight of the seedlings of these seed lots in the field was still far below that of the high vigour 1984-2 seed lots. Nevertheless, the invigoration effect of a priming treatment might bear interesting perspectives.

The physiological mechanisms that mediate the action of a priming treatment are still for the greater part unknown. Therefore, it is not known whether the two priming effects that were distinguished in this study are the result of the same process or depend on different processes. In celery and onion the two priming effects correlated rather well (Brocklehurst & Dearman, 1983). The present data showed that the changes that occurred during priming were resistant to dehydration of the seeds.

In the next two Chapters the effect of priming on the water relations of the seeds and on the potassium leakage will be studied.

CHAPTER 6

THE EFFECT OF OSMOTIC PRIMING ON WATER RELATIONS DURING GERMINATION

Abstract

Comparison of osmotically primed and control seeds showed that the beneficial effects of priming on the rate of germination and on the vigour of white cabbage seeds is not mediated by changes in the water relations of the seeds. An osmotic pre-incubation during 14 days at -1.5 MPa PEG followed by redrying of the seeds did not cause a change in the critical osmotic potential that was inhibitory to germination. Direct psychrometric measurements of the water potential (ψ) and osmotic potential (ψ_π) of osmotically pretreated and untreated seeds of the low vigour 1979-1 seed lot showed that at the end of imbibition all seeds had the same ψ and ψ_π . The main effect of pretreatment was the disappearance of the transition phase in the water uptake curve. Pretreated seeds imbibed water at a higher rate than control seeds and started seedling growth immediately thereafter. Control seeds needed a transition phase of more than 1 day between imbibition and the start of seedling growth. Growth was accompanied by a further rise in ψ_π , a drop in turgor pressure and a decrease in the amount of osmotic constituents.

Introduction

The availability of water is of utmost importance in plant life. The uptake of water by a viable seed is the initial step of germination and therefore of seedling establishment and successful crop production. Initially, the water potential (ψ) of air-dry seeds is extremely negative due to strong matrix effects. Rapid water uptake occurs when seeds come into contact with a liquid (Koller & Hadas, 1982). The germination process of non-dormant seeds can be characterized as a triphasic pattern of water uptake. Phase 1 is the initial phase, the water-uptake during this phase is often called imbibition, it occurs in a similar way in seeds that are dormant or non-dormant, viable or non-viable. At the end of phase 1 the matrix potential and ψ approach zero. During phase 2, the lag or transition phase, ψ is in balance with the osmotic potential (ψ_π) and the pressure potential (ψ_p). During this phase unknown reactions prepare radicle emergence. The third or growth phase is characterized by renewed water uptake in concurrence with radicle elongation, it can be entered by germinating seeds only (Bewley & Black, 1985).

The use of a polyethylene glycol (PEG) '6000' solution, which is chemically inert but osmotically active, as germination medium, controls the water uptake of the seeds. The concentration of the PEG solution must be sufficiently low to have an almost normal phase 1 and 2 and sufficiently high to prevent the third phase of water uptake, thereby prolonging the transition phase (Heydecker et al., 1975). A pre-incubation in PEG, that is often also called priming or osmoconditioning, has many effects on seeds, which have been discussed in Chapter 5. The physiological processes involved in priming are for the greater part unknown. In some species priming stimulates the rate of germination and synchronizes germination within a population of seeds. It is obvious that in such seeds certain processes in phase 2 are limiting for germination. Therefore, prolongation of that phase by osmotic incubation causes a better preparation of germination and diminishes the differences in a population during subsequent germination in water. Since the favourable effects of priming often persist in redried seeds, the induced changes must be of a stable character.

It has been concluded that in lettuce seeds osmotic pre-incubation caused prior to germination an increase in the level of soluble amino-nitrogen compounds and of K^+ ions in the embryo, which resulted in a more negative osmotic potential (Carpita et al., 1979; Takeba, 1980 a, b). Due to a more accurate definition of the start of embryo elongation, Weges (1987) clearly showed that both processes are the result instead of the cause of better growth. The interpretation of priming data in lettuce is complicated because changes in dormancy are involved. Weges (1987) showed that pre-incubation at 15 °C both increased the maximum temperature at which 50% of the seeds germinated (T_{50}) and decreased the osmotic potential of the incubation medium at which 50% of the seeds was inhibited ($\psi_{\pi e\,50}$, for short ψ_{50}). These changes were not accompanied by a change in ψ and ψ_π of the seeds. Weges concluded that the changes in dormancy, i.e. in T_{50}, correlated with changes in the yield threshold Y of the turgor potential for cell expansion. This factor could be calculated because according to the general equation of extension growth, inhibition of germination can be regarded as a steady-state growth-rate of zero. Without growth ψ_{50} equals ψ_π + Y. And when ψ_π is stable, changes in ψ_{50} must be the result of similar changes in Y. It was proposed that Y is a function of the plasticity of the cell walls.

It has been shown in previous Chapters that in cabbage seeds priming accelerates the rate of germination, synchronizes the moment of germination in a population of seeds and improves the seedling quality (invigoration). It has been discussed in Chapters 1 and 5 that loss of vigour is often thought to be caused by damage to crucial processes. Therefore, invigoration might be associated with repair processes that get ample opportunity to proceed during prolonged incubation in PEG. It can not be excluded, however that priming acts through changes in the water relations of the seeds. It is the aim of the present paper to test the latter presumption.

Material and methods

Seed material

Experiments were performed with seeds of *Brassica oleracea* L. *Capitata* group cv. Oscar and *B. oleracea* L. *Sabauda* group cv. Samoa. From the Oscar seeds the 1984-2 and 1979-1 seed lots were used. Characteristics of the seed lots have been described in Chapters 2 and 5.

Effect of $\psi_{\pi e}$ on seed germination

To study the effect of water stress on germination batches of 250 seeds per seed lot were pre-incubated in -1.5 MPa PEG at 20 °C for 7, 10 and 14 days in darkness in plastic boxes as described in Chapter 5. After pretreatment seeds were dried back close to the original moisture content. Pretreated and untreated seeds were germinated at the conditions of the standard germination test (Chapter 2). Germination occurred in water or in various PEG concentrations. A seed with visible radicle protrusion was counted as germinated. The germinated seeds were counted after 4, 8 and 12 hours and thereafter three times per day. The viability of seeds that had not germinated in PEG was checked during a subsequent germination test during 21 days in water. The experiment was performed in 1986.

Germination and changes in the water relations

For the study of the time courses of germination, water uptake and the changes in ψ and ψ_π, 4 replicates of 20 seeds of both osmotically pretreated seeds and untreated control seeds were sown at 20 °C in darkness in 5 cm Petri dishes on one layer of filter paper moistened with 1.5 ml distilled water. For the determination of the time course of germination, the percentages of seeds with split testa and of seeds with radicles that protruded for more than 1 mm were determined at suitable intervals under green safelight.

Determination of moisture content

The changes in moisture content of the seeds which occurred during germination were determined with the low-constant-temperature oven method (ISTA, 1985). Seed were dried for 17 hours in an oven at a temperature of 103 °C.

Determination of ψ and ψ_π

The water potential (ψ) of cabbage seeds was determined with a thermocouple psychrometer. A C_{52} sample chamber (Wescor, Logan, USA) was adapted for connection to a NT-3 Nanovoltmeter (Decagon, Pullman, USA). The sample chamber was placed in a humid chamber (95 - 100% r.h., 25 °C) in order to prevent evaporation during manipulations with the seeds. Samples of 20 surface-dry seeds were introduced in the measuring chamber (5.07 mm deep, 9.15 mm in diameter). Water-vapour equilibration was reached after 60 minutes. Readings were taken 60, 80 and 100 minutes after closing the measuring chamber and the data were averaged. The highest value in µV of the instrument was read after a standardized cooling time of 20 seconds.

The osmotic potential (ψ_π) of the seeds was determined by the same procedure using samples of 20 seeds which were frozen in liquid nitrogen in small plastic vials and thawed for 5 minutes at 25 °C. Before the measurement the seeds were homogenized in the measuring chamber by means of crushing with a copper rod. Water vapour equilibration was reached after about 20 minutes. Readings were made 20, 30 and 40 minutes after closing the measuring chamber and the data were averaged.

The psychrometer was calibrated with PEG solutions, the ψ_π of the solutions was calculated according to Michel (1983).

The amount of solutes in the seeds can be calculated from the values of ψ_π and moisture content. If it was assumed that the cells only contained mannitol the amount of mannitol necessary to reach the measured value of ψ_π could be calculated according to Michel (1983).

Results

In the first experiment it was tested whether osmotic pre-incubation influenced the reaction of the seeds to a subsequent osmotic incubation during germination. Seeds were pre-incubated in -1.5 MPa PEG during 7, 10 or 14 days, redried and subsequently germinated in PEG solutions of different osmotic potential ($\psi_{\pi e}$). Untreated seeds were used as controls. Incubation in PEG decreased both the final percentage germination and the rate of germination. Osmotically pretreated seeds showed the same final percentages germination as control seeds (Fig. 6.1) and, moreover, the acceleration of germination due to pretreatment was similar at all PEG concentrations (Fig. 6.2). It is concluded that in cabbage seeds an osmotic pre-incubation does not increase the resistance of the germination process to osmotic stress. Therefore, it is unlikely that priming caused a dramatic change in the water relations of the seeds. This conclusion was further tested by direct measurements of ψ and ψ_π by means of psychrometric determinations.

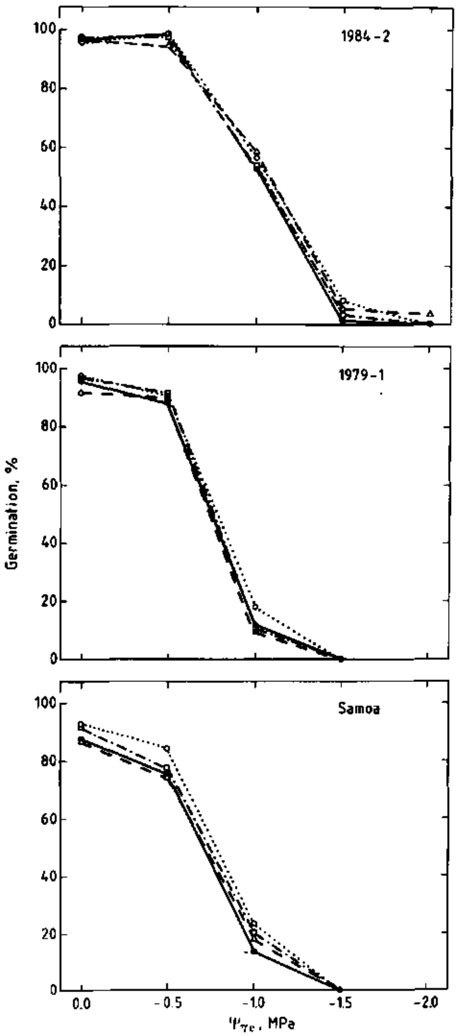

Fig. 6.1. Effect of osmotic priming on final percentage germination of cabbage seeds in osmotic stress conditions. Seeds of 1984-2 and 1979-1 seed lots of cv. Oscar and of the 1984 seed lot of cv. Samoa were pre-incubated in -1.5 MPa PEG at 20 °C during 7 (○), 10 (□) or 14 days (△) and subsequently redried. Germination of pretreated and control seeds (●) occurred at the conditions of the standard laboratory germination test in a range of external osmotic potentials arranged with different PEG concentrations. Germination was counted after 7 days.

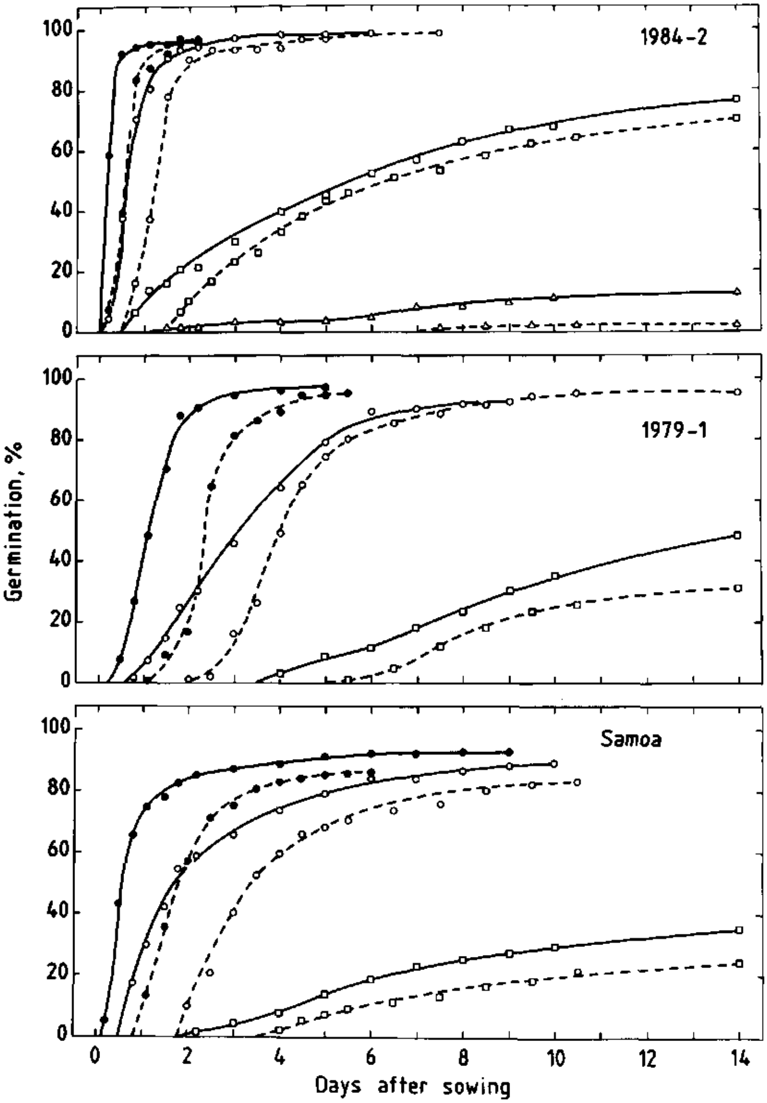

Fig. 6.2. Effect of osmotic priming on the rate of germination of cabbage seeds in osmotic stress conditions. Data are from the same experiment as Fig. 6.1. Seeds were without pretreatment (- - -) or were pre-incubated during 7 days in -1.5 MPa PEG at 20 °C (—). Germination test in water (●) or PEG at -0.5 (○), -1.0 (□), or -1.5 MPa (△).

The experiments were performed with the 1979-1 seed lot that was also used in the previous invigoration experiments (Chapter 5). Seeds were pre-incubated in -1.5 MPa PEG during 14 days at 20 °C and redried before incubation in water. Untreated seeds served as controls. To obtain the earliest possible information about the start of elongation growth the number of seeds with a split testa was recorded. In pretreated seeds the first split testas were observed 2 hours after start of imbibition, that is roughly 2 to 4 hours before visible rootlet growth (Fig. 6.3A). Two hours after the start of re-imbibition of pretreated seeds ψ was equal to zero (Fig. 6.3C). It is most likely that at that moment also the first phase of water uptake is completed. However, in pretreated seeds a distinct transition phase of water uptake is not observed. Apparently, phase 1 is immediately followed by phase 3, the water uptake related to extension growth (Fig. 6.3B).

After the moment that ψ approached zero in pretreated seeds the values of ψ_π continued to rise (Fig. 6.3C). Since $\psi = \psi_\pi + \psi_p$ as the matrix potential will be zero, the value of ψ_p further decreased (Fig. 6.3D). If it was assumed that all osmotic constituents were mannitol the amount of solutes per seed could be calculated from the data on water uptake and ψ_π. It is shown in Fig. 6.3E that from the second hour of imbibition the amount of solutes in pretreated seeds dropped considerably. Around the 12th hour the amount became stable.

Untreated control seeds differed in nearly all aspects from the pretreated seeds. Visible radicle protrusion started in the controls about 20 hours later (Fig. 6.3A). In about 20% of the untreated seeds the testa was split after about 24 hours of imbibition, further growth was retarded another 19 hours, however. The imbibition rate (Fig. 6.3B) and the rise of ψ (Fig. 6.3C) were slightly retarded in the untreated seeds as compared to the pretreated ones. Interestingly, at the moment ψ became zero, being after 2 and 6 hours in pretreated and untreated seeds, respectively, seeds of both groups contained the same moisture content of 43%. In contrast to pretreated seeds, in control seeds the second or transition phase of water uptake was clearly visible (Fig. 6.3C). It lasted from 6 till 34 hours after the start of water uptake. During this period the moisture content, ψ_π, ψ_p and the amount of solutes remained stable in the untreated seeds (Figs. 6.3B, C, D, E). At the moment visible growth started, all these parameters began to change in a similar way as occurred in the pretreated seeds about 32 hours earlier. The changes occurred slower in the untreated than in the pretreated seeds.

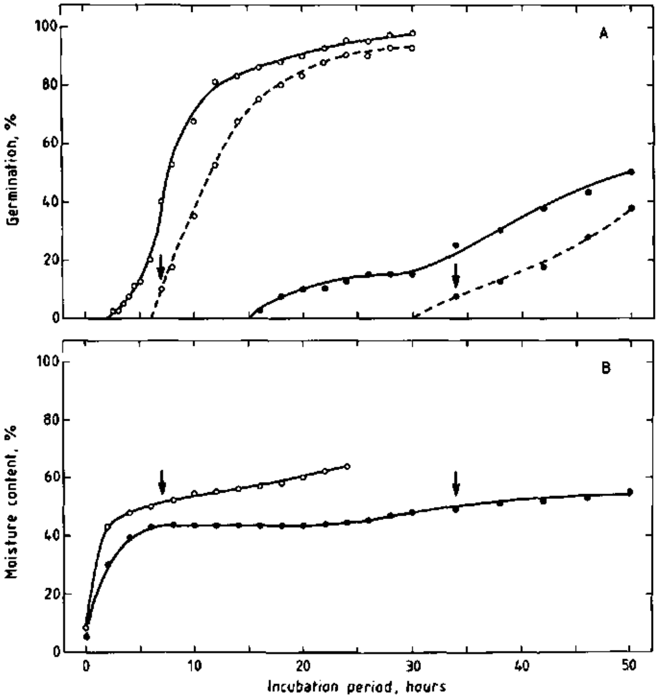

Fig. 6.3A, B. For legend, please see next page.

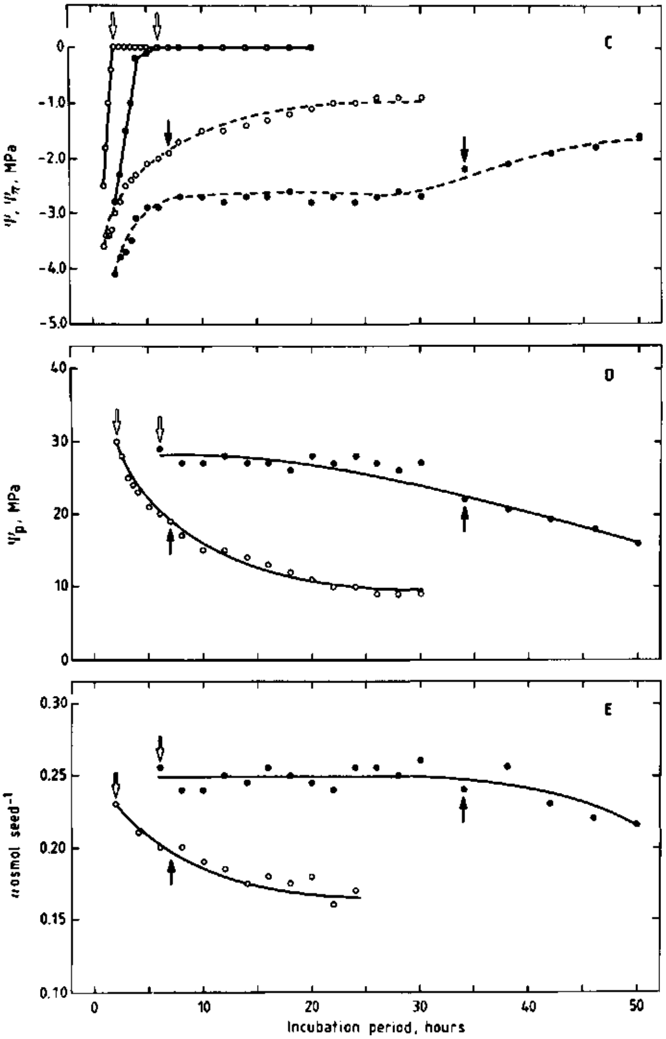

Fig. 6.3. Effect of osmotic priming on the time courses of germination (A), water uptake (B), water potential (ψ) and osmotic potential (ψ_π) (C), pressure potential (ψ_p) (D) and the amount of osmotic constituents, as calculated from the data on water uptake and ψ_π (E) of the 1979-1 seed lot of cabbage seeds cv. Oscar. Seeds were pre-incubated during 14 days at 20 °C in -1.5 MPa PEG and subsequently redried. Incubation of control seeds (●) and pretreated seeds (○) occurred in water at 20 °C in darkness. A. Germination was recorded both as percentage seeds with split testa (—) and seeds with visible radicle protrusion (---); C. ψ (—), ψ_π (---). Open arrows indicate the moment that ψ approaches zero, closed arrows indicate the first visible rootlet protrusion.

Discussion

An osmotic pretreatment in -1.5 MPa PEG strongly accelerated both the moment germination started and the rate of germination (Figs. 6.3 A and 5.5). The pretreatment also invigorated seeds (Chapter 5): pretreated seeds produced more vigorous seedlings than untreated controls. The present data clearly prove that the improved performance of pretreated seeds is not related to changes in the water relation of the seeds. This conclusion is based on 2 observations: (1) the critical $\psi_{\pi e}$ for imbibition of germination was not affected by a pretreatment (Fig. 6.1) and (2) at the moment ψ approached zero the ψ_{π} of both group of seeds amounted to -3.0 MPa (Fig. 6.3C).

The present data showed 2 distinct changes in pretreated seeds as compared to untreated seeds: (1) the rate of imbibition was increased (Fig. 6.3B) and (2) the transition phase (phase 2) in the water uptake of the seeds disappeared. The increased rate of imbibition caused a faster rise of ψ. It is shown in Fig. 6.3E that these changes are not due to a higher amount of osmotic constituents in the pretreated seeds. It is not clear yet which factors cause the accelerated imbibition in pretreated seeds. It is hypothesized that certain external or internal factors which impede the water uptake of the seeds become less dominant. In this respect changes in testa structure, involving reduced integrity of the testa or reduction of structures or layers that hinder direct seed-water contact, may play a role. Also internal changes in the swelling properties of cell constituents and ultrastructure may be involved.

The disappearance of the transition phase in the water uptake seems the most relevant change in pretreated seeds. Obviously, elongation growth in these seeds starts right from the completion of imbibition. In untreated seeds it took about 34 hours before growth started. The present data do not disclose the character of the retarded growth and of the improvement that occurred during osmotic pretreatment. However, the data show that a rise in osmotic constituents is certainly not involved.

It is clearly shown in Fig. 6.3 that the rise of ψ_{π} during early germination (Fig. 6.3C) is due to both extra water uptake (Fig. 6.3B) and a reduction in the amount of solutes (Fig. 6.3E). The latter fact might be caused by either leakage or active metabolism of substrates. Leakage will be dealt with in the next Chapter. A rise of the mean molecular weight of the cell constituents is not likely in a phase that is characterized by active hydrolysis of reserve food. Leakage or active metabolism may also be the cause of the slight reduction of the amount of osmotic constituents in pretreated seeds (Fig. 6.3E).

The present data support certain conclusions of the study on water relations in lettuce seeds by Weges (1987). Both in cabbage and lettuce seeds improved germination in pretreated seeds was not related to changes in ψ_{π}. However, in contrast to lettuce, in cabbage seeds osmotic pretreatment did also not affect the critical $\psi_{\pi e}$ for inhibition of germination (Fig. 6.1, 6.2). As was explained in the Introduction of this Chapter Weges (1987) concluded from the more negative values of $\psi_{\pi e}$ in pretreated lettuce seeds that the basic change that occurred during pretreatment was the decrease of the threshold Y of the turgor potential. It is very unlikely that the start of elongation growth in cabbage seeds is also blocked by such a mechanism. In the comparison of lettuce and cabbage seeds it has to be realized that in the former species pretreatment is meant to break dormancy, a condition that does not occur in cabbage. It was concluded before that in cabbage the advancement of germination in pretreated seeds is most probably the result of the repair of certain critical processes or ultrastructures. In untreated seeds repair took at least 34 hours, but the rate of germination was still reduced in comparison to pretreated seeds (Fig. 6.3A). Thus, osmotic pretreatments allows a much better repair than ever occurs in untreated seeds.

CHAPTER 7

LEAKAGE OF POTASSIUM DURING IMBIBITION IN RELATION TO VIGOUR AND OSMOTIC PRIMING

Abstract

The leakage of K^+ ions from high and low vigour cabbage seeds has been studied in relation to priming. Both groups of seeds contained similar amounts of total K^+. During a priming treatment of 7 days high vigour seeds maintained their K^+ level but low vigour seed lost about 20% of total K^+. During incubation of low vigour seeds in moist sand, about three times more K^+ leaked out than during a similar treatment of high vigour seeds.

Introduction

It has been argued in the Introduction of this thesis (Chapter 1) that ageing is a main cause of poor vigour in seeds. Ageing is the sum total of a series of deteriorative processes. Among these the general disorganization of cell ultrastructure and in particular, the disruption of cell membranes take a prominent position. As a consequence, aged seeds often loose more solutes to the imbibition medium. Mainly in grain legumes the association of low vigour with increased and prolonged leakage is applied in a vigour test (Hegarty, 1977; Powell & Matthews, 1981; Powell et al., 1984, 1986). Leakage is measured as electrical conductivity of the imbibition medium.

In fact all seeds loose solutes during the early hours of imbibition. In high vigour seeds membrane integrity is rapidly restored during imbibition. In low vigour seeds repair processes are required to restore damage to essential proteins (Osborne, 1982). Conditions during imbibition may interfere. In soybean and a few other grain legumes low temperatures during imbibition caused extra damage to membranes. When the cellular structures in a seed are in a weak condition the uninhibited inrush of water may cause extra damage. In soybean the rapid intake of water before repair of mitochondrial membranes could occur caused a stimulation of fermentation processes instead of normal respiration (Woodstock and Taylorson, 1981). The produced ethanol enhanced the deteriorative situation.

Imbibition in osmotic concentrations of PEG has been shown an effective means to reduce the rate of water uptake and decrease the leakage (Waggooner & Parlange, 1976). Thus, prolonged priming treatment might favour repair processes in seeds. Invigoration of low vigour seeds during priming treatment has indeed been demonstrated in seeds of Brussels sprouts (Burgass & Powell, 1984) and cabbage (Chapter 5).

The present Chapter describes a preliminary study on the leakage of K^+ ions from high and low vigour cabbage seeds in relation to priming treatments. Measurement of K^+ leakage is taken as a general criterion for membrane integrity.

Material and methods

The experiments were performed in 1987 with seeds of the 1984-2 and 1979-1 seed lots of white cabbage, cv. Oscar. Priming occurred during incubation in -1.5 MPa PEG solution at 20 °C for 7 days followed by rehydration almost to the initial moisture content. Untreated seeds served as control. To measure K^+ leakage triplicates of 20 seeds were sown in 5 cm Petri dishes to which 1.5 ml de-ionized water was added. Incubation occurred at 20 °C in darkness. K^+ leakage of 20 seeds was measured in 0.2 ml samples taken from the incubation medium in the Petri dish. The amount of K^+ ions was measured with a flame photometer (PEP 7, Jenway Ltd. England). To measure leakage of K^+ during a stay of the seeds in soil, samples of 100 seeds of the 1984-2 and 1979-1 seed lots were sown each in a plastic box containing moistened river sand. The seeds were placed in the phytotron at 13 ° or 17 °C. After certain periods of time the germinated and non--germinated seeds were sieved out and dried back to almost the initial moisture content. The K^+ content that remained in the seeds was measured by extraction.

To facilitate extraction of K^+ ions, seeds were punctured with a needle through the testa. Extraction occurred in a 5 ml mixture of 0.02 M HCl, 0.03 M CsCl and 0.14 M oxalic acid during 12 hours at 20 °C in darkness. Measurements occurred in 0.1 ml samples.

Table 7.1 K^+ content of intact white cabbage seeds cv. Oscar from seed lots 1984-2 and 1979-1 with and without pre-incubation in -1.5 MPa PEG at 20 °C for 7 days in darkness followed by redrying to almost the initial moisture content.

	K^+ content, µg.seed^{-1}	
	1984-2	1979-1
Control	20.7 ± 0.7	18.8 ± 0.8
Primed	21.1 ± 0.2	15.0 ± 0.2

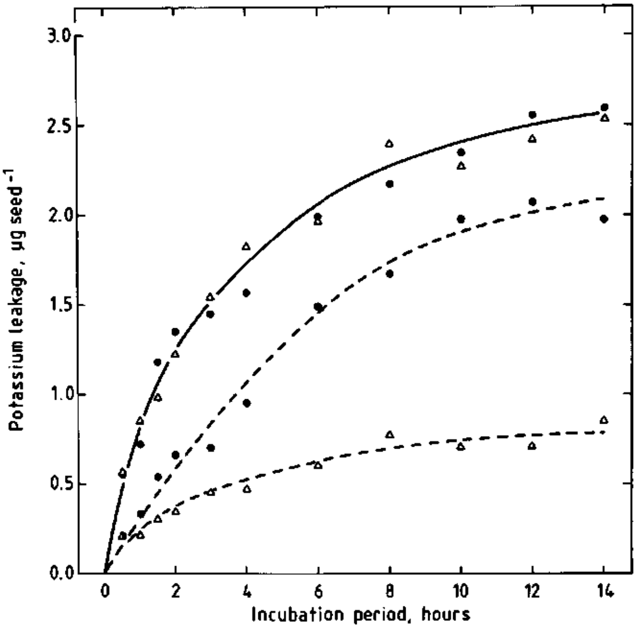

Fig. 7.1. The time course of K^+ leakage from seeds that were pre-incubated during 7 days at 20 °C in -1.5 MPa PEG and redried (- - -) and from untreated seeds (—) of the 1984-2 (●) and 1979-1 (△) seed lot in de-ionized water at 20 °C.

Results

The K^+ levels of dry seeds of the high vigour 1984-2 and of the low vigour 1979-1 lot are shown in Table 7.1. The time courses of K^+ leakage in de-ionized water from pretreated and untreated seeds of both lots are depicted in Fig. 7.1. Within the 14 hours of the test at 20 °C the K^+ leakage from untreated seeds of both age groups hardly differed. Both groups of seeds lost 2.6 $\mu g.seed^{-1}$ that is 12 to 14% of total K^+. After 7 days incubation in -1.5 MPa PEG the situation was different. The total K^+ content of the 1984-2 seeds had not changed during the pretreatment, but the 1979-1 seeds had lost 3.8 μg K^+ $seed^{-1}$, that is 20% of total K^+ (Table 7.1). Surprisingly, pre-incubated and redried 1984-2 seeds lost more K^+ during re-imbibition than similar 1979-1 seeds (Fig. 7.1). The germination of the two seed lots in the same conditions and during the same length of time as the K^+ leakage test is shown in Fig. 7.2.

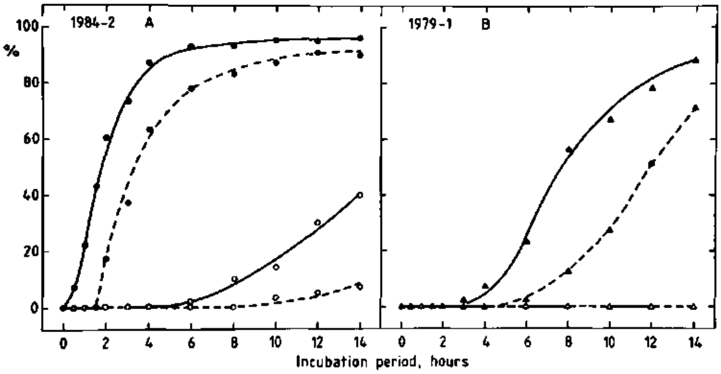

Fig. 7.2. The time course of germination at 20 °C, expressed as percentage seeds with a split testa (—) or with a protruded radicle (---). The seeds were pre-incubated in -1.5 MPa PEG during 7 days at 20 °C and redried (●▲) or untreated (○△). Data for the 1984-2 (A) and 1979-1 seed lot (B).

During their stay in moist sand in the phytotron, seeds of both seed lots lost different amounts of K$^+$ (Fig. 7.3). The low vigour 1979-1 seeds lost about 3 times more K$^+$ than the high vigour 1984-2 seeds. The end of K$^+$ leakage was not correlated with the start of visible germination.

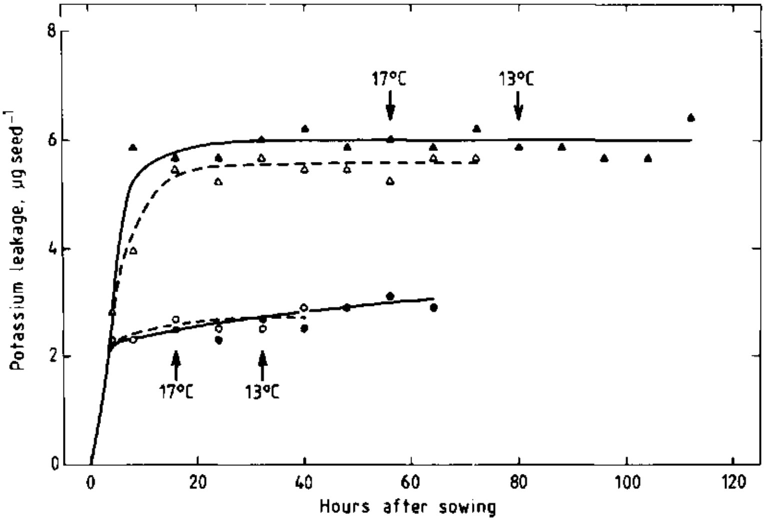

Fig. 7.3. The time courses of K$^+$ leakage (µg.seed^{-1}), as measured by the decrease of total K$^+$ content of the seeds, from seeds of the 1984-2 (●○) and 1979-1 (▲△) seed lots in river sand at 13 ° (—) and 17 °C (---) in the phytotron. The arrows indicate the start of seed germination.

Discussion

The K$^+$ leakage from untreated and osmotically pretreated seeds of white cabbage started immediately at the beginning of water uptake (Fig. 7.1). Comparison of the data on K$^+$ leakage (Figs. 7.1, 7.3) with the time course of germination (Fig. 7.2) clearly shows that the timing of germination, either expressed as the percentage seeds with a split testa or with a protruding radicle, was not related in any way to the time course of K$^+$ leakage.

The extent of the loss of K^+ from the seeds correlated to seed vigour. Low vigour 1979-1 seeds lost during incubation in sand about 3 times more K^+ than high vigour 1984-2 seeds (Fig. 7.3). K^+ is certainly not the only compound that is lost from the seeds. Amino acids, sugars, organic acids, phenolic compounds, phosphates and enzymes are among the substances that have been found in the imbibition medium of seeds (Abdel Samad & Pearce, 1970; Bewley, 1986). It is possible that the retarded seedling growth is partly due to diminishing levels of certain compounds in germinating seeds.

During osmotic pre-incubation in -1.5 MPa PEG no K^+ was lost from the 1984-2 seeds (Table 7.1), on the other hand, 1979-1 seeds lost considerable amounts of K^+ during PEG pretreatment. Thus, in low vigour seeds even a reduced rate of water-uptake did not prevent leakage. Nevertheless, during 7 days PEG the 1979-1 seeds lost less K^+ than during 2 days in sand, being 3.8 and 5.6 $\mu g.seed^{-1}$, respectively (Table 7.1, Fig. 7.3). K^+ loss also occurred when osmotically pretreated seeds were redried and reimbibed. Leakage from pretreated 1984-2 seeds occurred at about the same rate as from untreated seeds (Fig. 7.1). The pretreated 1979-1 seed had a much lower K^+ leakage than the controls, probably because the available K^+ became limiting. In lettuce seeds only 10% of total K^+ was available for leakage, the remaining 90% was never lost from intact seeds (Weges, 1987). The former fraction was most probably located on the outside of the K^+-impermeable endosperm cells.

The present experiments on K^+ leakage are in good agreement with the data presented in Chapter 6. It was shown in Fig. 6.3E that the amount of solutes in pretreated seeds was lower than in untreated seeds and that the solutes of pretreated seeds were further reduced during imbibition.

CHAPTER 8

GENERAL DISCUSSION

1. Germination, emergence and seedling growth in relation to vigour

Germination, emergence and early seedling growth are the first critical phases in crop establishment. The experiments described in this thesis have clearly shown that both emergence and seedling growth in the field depend on a complex interaction between seed and soil factors.

Aspects of seed quality

With respect to seed factors this study was mainly concentrated on seed vigour. Seeds of 5 lots of white cabbage (cv. Oscar) and 1 lot of savoy cabbage (cv. Samoa) of different age were compared. White cabbage seeds had been stored at commercial storage conditions during 1 (1984 seeds), 5 (1980 seeds) or 6 years (1979 seeds), savoy cabbage seeds during 1 year before they were stored at 5 °C and at 30% relative humidity. The seed lots differed in many aspects.

Germination and emergence

Germination of the seeds of all seed lots in the standard laboratory germination test was higher than 90%, but the percentages normal seedlings differed strongly with the age of the seeds (Figs. 2.1, 5.1). Particularly, the 1979 seeds produced less than 50% normal seedlings. The seed lots also differed in the lag time and the rate of germination. In general the rate of germination was retarded with increasing age of the seeds (e.g., Fig. 2.5).

The emergence of seedlings from soil was studied both at controlled conditions in the phytotron and at natural conditions in the field. Percentages emergence of 1979 and Samoa seeds were lower than of the 1984 seeds. In the field the total emergence of the 1979 seeds at certain sowing dates was reduced to about 10%, whereas the 1984 seeds still emerged for about 40 to 60% (Fig. 2.7).

The rate of germination and the rate of emergence were both strongly dependent on temperature (Figs. 2.3, 2.5) but at all temperatures the difference in rate between the 1984 and 1979 seeds was apparent.

Seedling growth

The seed lots strongly differed in the growth and development of the seedlings both in the phytotron (Fig. 2.6) and in the field (Fig. 2.8). The 1979 seeds always produced smaller seedlings than 1984 seeds.

Several differences observed between the seed lots correspond with the symptoms of progressive ageing as reported for various crop species (see Chapter 1). Ageing is generally accepted as one of the main causes of poor vigour in seeds.

Vigour

The conclusion that the differences in germination, emergence and seedling growth between the seed lots are indications of differences in vigour is favoured by the results of two vigour tests. The controlled deterioration (CD) test speeds up the rate of ageing in a controlled manner by holding seeds at an increased temperature and moisture content. Germination of CD-treated 1984 seeds germinated for 80 to 100% and produced considerable numbers of normal seedlings. In the CD test the 1980 seeds scored almost as bad as the 1979 seeds (Fig. 2.4) but with respect to other symptoms of low vigour, such as the reduction of the rate of germination (Fig. 2.3) and emergence in the phytotron (Fig. 2.5) and total emergence in the field (Fig. 2.7) they took an intermediate position between the seed lots of 1984 and 1979. Before CD treatment the 1980 seeds showed similar results as the 1984 seeds in the germination test (Fig. 2.1). Thus the CD test gave in the present experiments the most vigorous verdict on vigour. It is obvious that from a practical point of view an under-estimation of vigour is preferable above an over-estimation. However, a CD test at slightly less adverse conditions might give even more satisfactory results.

The preliminary studies on the K^+ leakage of the white cabbage seeds also support the conclusion that the seeds of different age differ in vigour (Chapter 7). Surprisingly, the K^+ leakage of 1984-2 and 1979-1 seeds did not differ during 14 hours of imbibition in de-ionized water in Petri dishes (Fig. 7.1), but during prolonged incubation in moist sand in the phytotron the 1979-1 seeds lost about 3 times more K^+ than the 1984-2 seeds (Fig. 7.3). These results certainly ask for more detailed experiments, but at this stage it seems evident that in 1979-1 seeds membrane damage had occurred during storage.

Seed size

The separation of the 1984-1 seeds into different size fractions learned that in white cabbage seed size is not a relevant factor to seed vigour (Chapter 3). However, seed size clearly affected seedling growth. Smaller seeds produced in the phytotron at all test temperatures smaller seedlings than larger seeds (Fig. 3.2). Thus, seed size correlated with seedling vigour.

Environmental factors

Field conditions

Several factors influence emergence of seed lots in the field. The effects of temperature, soil type and matric head on emergence and seedling growth of white cabbage were studied.

Temperature

Total germination or emergence of the seed lots at a range of constant temperatures on the gradient plate or in the phytotron were less affected by temperature than the rate of germination. In the field, also final emergence may be reduced at low temperatures because imbibed, non-germinated seeds are vulnerable to attack by soil-borne micro-organisms.

Soil type and matric head

The effects of the matric head (h_m) of soil was tested in two different soil types viz. *sandy loam and river sand, with seeds of a high and a low* vigour seed lot (Chapter 4). The matric head mainly influenced emergence. Within the range of h_m values in these experiments, at increasing matric head the rate of emergence and final emergence increased and finally stabilized (Figs. 4.3 and 4.5). It was concluded that the influence of h_m on emergence is mainly due to the air volume of soil. The smaller air volume of sandy loam as compared to river sand seemed the main reason for the different behaviour of the seeds in the two soil types during emergence and seedling growth.

Practical implications

The present data certainly do not allow accurate predictions of field performance of cabbage seed lots. It becomes evident, however, that information about seed vigour provides vital information in this respect. The differences in percentages normal seedlings in the standard germination test between the 1984 and 1979 seed lots corresponded well with the differences in field emergence between these lots. However, the standard germination test overestimated the 1980 seed lot with regard to field emergence. At field conditions, seeds with high vigour as indicated by the CD test always performed better than low vigour seeds (Figs. 2.7, 2.8, 5.8). But a high level of vigour does not guarantee a high percentage emergence. Soil and other environmental factors strongly influence field performance. E.g. soil crusting strongly inhibited seedling emergence in the field experiment performed in July 1985 (Fig. 2.7). In the field experiments seedling fresh weight appeared a function of temperature (Fig. 2.8).

In spite of interaction with abiotic factors (Table 4.1) selection of high vigour seeds guarantees that under certain atmospheric and soil conditions the best possible result is obtained.

2. Osmotic pretreatment

Invigoration

Osmotic pretreatment or priming of high and low vigour cabbage seeds has clearly shown that several aspects of ageing are reversible. Pretreated seeds germinated earlier and faster (Figs. 5.4, 5.5, 6.3, 7.2) than untreated seeds and produced a higher number of normal seedlings with a higher fresh weight both in the phytotron and in the field (Figs. 5.6, 5.7, 5.8). The beneficial effects of priming remained after drying back the seeds. In the present experiments seed lots with rather extreme differences in seed vigour were compared. Priming did not invigorate 1979 seeds to the level of the 1984 seeds. Nevertheless, an invigoration treatment may have practical implications for the improvement of seed lots that just fail to meet the higher quality requirements.

Mechanism

The present results add valuable information to the discussion about the mechanism of action of priming. It was clearly shown that priming did not afford better germination because of a drop of the osmotic potential (Chapter 6). Apparently, the prolonged period of incubation in low osmotic potentials is not utilized to accumulate osmotic constituents. In contrast to imbibition in de-ionized water (Fig. 7.1), during priming in -1.5 MPa PEG leakage of K^+ ions was not observed for 1984 seeds (Table 7.1). Low vigour 1979-1 seeds also lost K^+ during priming although to a lesser extent than during imbibition in moist sand (Fig. 7.3). Leakage of K^+ ions is an indication of membrane damage. In high vigour seeds membranes are rapidly reorganized during imbibition. In low vigour seeds repair of damaged ultrastructures is required which probably takes considerable periods of time. It is not possible to conclude from the present experiments whether inferior seedling growth from untreated 1979 seeds is caused by loss of K^+ and probably other cell constituents or is due to a general lack of integrity of the cellular system.

SAMENVATTING

In dit proefschrift zijn verschillende factoren bestudeerd, die de zaadkieming, opkomst en groei van kiemplanten van kool beïnvloeden. Het belang van zaadkwaliteit en omgevingsfactoren is onderzocht voor zaad van verschillende herkomst en oogstjaren. De experimenten zijn uitgevoerd met vijf partijen zaaizaad van witte kool (cv. Oscar) en een partij savooiekool (cv. Samoa).

Aspecten van zaadkwaliteit

In Hoofdstuk 2 blijkt, dat de onderzochte partijen witte kool onder optimale omstandigheden voor 90% of meer kiemen, maar in de diverse experimenten grote verschillen in kwaliteit vertonen. De verschillen komen ten dele tot uiting in de percentages normale kiemplanten in de kiemkrachttoets. De partijen van 1984 produceren meer dan 80% normale kiemplanten. Bij de partijen uit 1979 bedraagt dit percentage slechts ongeveer 50%. Bij een reeks van temperaturen op de gradiënttafel en in het fytotron is de snelheid van kieming en het opkomstpercentage voor de partijen van 1979 verreweg het laagst, evenals de opkomst in veldproeven. De groeisnelheid van de kiemplanten in het fytotron en in het veld (Hoofdstuk 2) en bij verschillende vochtspanningen van de grond (zand en zavel, Hoofdstuk 4) toont dezelfde kwaliteitsverschillen tussen de partijen uit 1984 en 1979. De partij uit 1980 heeft in de kiemkrachttoets een hoog percentage normale kiemplanten, maar het resultaat van deze partij in de overige uitgevoerde experimenten is matig. Bij deze partij werd de kwaliteit in de kiemkrachttoets overschat. In de "CD toets" vindt onder gecontroleerde omstandigheden wat betreft temperatuur en vochtgehalte van het zaad snelle veroudering plaats. Deze test resulteert in een zeer laag kiempercentage van de partijen van 1979 en 1980, terwijl de twee partijen van 1984 nog redelijk goed kiemen. De kwaliteit van de partij van 1980 wordt in de huidige CD toets, in tegenstelling tot de kiemkrachttoets, onderschat. Een CD toets onder gematigde verouderingscondities zal waarschijnlijk een goede voorspelling van de kwaliteit geven.

Veroudering van zaad lijkt een belangrijke oorzaak van de gevonden verschillen in kwaliteit te zijn. Uit de relatief kleine verschillen tussen partijen van één oogstjaar, maar van verschillende oorsprong, blijkt dat ook andere factoren een rol spelen.

Experimenten met drie zaadfracties van verschillende grootte van de partijen uit 1984 tonen aan, dat de zaadgrootte geen invloed heeft op de kieming en het percentage normale kiemplanten in de kiemkrachttoets, noch op de snelheid van opkomst en het opkomstpercentage bij temperaturen van 9 ° tot 25 °C in het fytotron. Er blijkt wel een verband te zijn tussen de zaadgrootte en de grootte van de kiemplanten (Hoofdstuk 3). Ook het verschil in versgewicht van de kiemplanten van de twee partijen uit 1984 onderling (Hoofdstuk 2) kan verklaard worden door het verschil in gemiddelde zaadgrootte. Enkele andere aspecten van zaadkwaliteit en verbetering daarvan komen later in deze samenvatting aan de orde.

Omgevingsfactoren

De zaadkieming in het veld is afhankelijk van verschillende factoren. Een aantal van deze factoren, de temperatuur, de grondsoort en de vochtspanning van de grond, zijn afzonderlijk bestudeerd.

Uit proeven op een gradiënttafel (10 ° - 34 °C) en in het fytotron (9 ° - 25 °C) blijkt, dat de temperatuur het kiempercentage en de uiteindelijke opkomst weinig beïvloedt. Op de kiemsnelheid heeft de temperatuur een duidelijke invloed. In het veld wordt het opkomstpercentage mogelijk gereduceerd door lage temperatuur, omdat geïmbibeerde, niet-kiemende zaden aangetast kunnen worden door micro-organismen in de bodem.

Tussen zaadkwaliteit, grondsoort en vochtspanning is een interactie aangetoond (Hoofdstuk 4). Binnen het onderzochte traject van vochtspanningen blijkt het volumepercentage lucht in de grond de belangrijkste factor te zijn, die de opkomst en beginstadia van de groei van de kiemplanten beïnvloedt.

Osmotische voorbehandeling

Verschillende aspecten van zaadveroudering blijken omkeerbaar door middel van osmotische voorbehandeling, hoewel voorbehandeling de kwaliteit van de onderzochte partij uit 1979 niet kan verhogen tot het niveau van de onderzochte partij uit 1984 (Hoofdstuk 5). Voorbehandeling resulteert in een hoger percentage normale kiemplanten in de kiemtkrachttoets bij de partij uit 1979. In het fytotron en in het veld kiemen voorbehandelde zaden eerder en sneller en produceren kiemplanten met een hoger versgewicht dan onbehandelde zaden. Bij terugdrogen na voorbehandeling van de zaden blijft het effect behouden.

Kieming van niet behandelde en voorbehandelde zaden wordt bij dezelfde osmotische potentiaal van het kiemmedium nog juist verhinderd (Hoofdstuk 6). Gedurende de verlengde incubatieperiode in PEG vindt geen accumulatie van osmotische stoffen plaats. Voorbehandelde zaden nemen iets sneller water op dan onbehandelde zaden. Bij voorbehandelde zaden groeit de kiemplant direct na imbibitie van het zaad, bij onbehandelde zaden pas na ruim een dag.

In tegenstelling tot zaden uit 1979, verliezen zaden uit 1984 geen K^+ ionen tijdens de voorbehandeling in -1.5 MPa PEG (Hoofdstuk 7). Tijdens imbibitie in vochtig zand lekt driemaal zoveel K^+ uit de zaden van 1979 dan uit de zaden van 1984. De relatief hoge uitlek van K^+ ionen uit de oudere zaden duidt op beschadiging van de membranen. De hoge uitlek van K^+ en vermoedelijk ook van andere stoffen tijdens de imbibitie van verouderd zaad is mogelijk een belangrijke oorzaak van de slechte kiemresultaten en slechte groei van de kiemplanten.

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