

A METHOD TO ASSESS CULTIVAR DIFFERENCES IN RATE OF PHYSIOLOGICAL AGEING OF SEED TUBERS

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Abstract

Inter-cultivar differences in rate of physiological ageing of potato tubers were assessed in a laboratory test and field tests.

The physiological status of the seed tubers was varied by storing them at 4°C and a range of higher temperatures for 28 weeks (= excluding curing and pre-sprouting periods), after which tubers were desprouted. In a laboratory test, growth vigor was assessed by determining number and weight of sprouts after 28 days of incubation. In a field test, percentage emergence, total plant height, haulm and tuber weights were determined and a stand score was allocated 4 weeks after emergence of the 4°C storage treatment. Ageing indices were calculated by dividing the values for the warm-stored treatments by values for the tubers stored at 4°C. Using the tests the cultivars were scored from 0-10 and ranked accordingly. The indices were more evenly distributed over the range of 0-10 in the field test than in the laboratory test. Fresh haulm yield proved very suitable for calculating ageing indices but it could be replaced by a visual score of the haulm development without a great loss in accuracy.

Compendio

Se determinaron las diferencias en las tasas de envejecimiento fisiológico de los tubérculos-semillas de diversos cultivares en una prueba de laboratorio y pruebas en el campo. Las condiciones fisiológicas de los tubérculos-semillas fueron modificadas almacenándolos durante 28 semanas a 4°C y un rango considerable de temperaturas altas (excluyendo los periodos de curado y rebrotamiento), después de lo cual los tubérculos fueron desbrotados. En una prueba de laboratorio, se determinó el vigor de las plantas calculando el número y peso de los brotes después de 28 días de incubación. En una prueba de campo se determinaron el porcentaje de emergencia, la altura total de la planta y el peso de la parte aérea y de los tubérculos y se estableció una escala para el número de plantas logradas cuatro semanas después de la emergencia del tratamiento almacenado a 4°C. Se calcularon los índices de envejecimiento dividiendo los valores

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para los tratamientos almacenados a altas temperaturas por los valores para los tubérculos almacenados a 4°C. Utilizando estas pruebas los cultivares fueron clasificados de 0 a 10 y puestos en orden de mérito de acuerdo con ésto. Dentro del rango de 0 a 10 los índices estuvieron distribuidos más uniformemente en la prueba de campo que en la prueba de laboratorio. El rendimiento fresco de la parte aérea probó ser muy aplicable para calcular los índices de envejecimiento, pero podría ser reemplazado por una clasificación visual de su desarrollo sin perder mucha exactitud.

Introduction

Toosey (5) defined the physiological age of a seed tuber of potato (*Solanum tuberosum* L.) as the physiological and biochemical status at any given moment between harvest and the end of real sprout formation. This status changes during storage. Van der Zaag and Van Loon (7) reviewed the effects of physiological age of the tuber on its growth vigor. Emergence, initial growth rate, number of stems, time of tuber initiation, haulm development and time of senescence may be affected by the physiological age of the seed.

The rate of physiological ageing varies among cultivars (2, 3, 4). This may be a very important characteristic in relation to the storage and pre-treatment of seed of the cultivar concerned. This article describes a method to measure inter-cultivar differences in the rate of physiological ageing.

Indexing Physiological Ageing of Cultivars

Krijthe (1) introduced the concept "sprouting capacity," which could possibly be considered to be a criterion of physiological age. Reust (3, 4) considered the incubation period as a measure in order to obtain tubers of the optimal physiological age. Van der Zaag and Van Loon (7) discussed the sprouting capacity, incubation period and many plant growth characteristics for measuring the physiological age of two cultivars (Jaerla and Désirée) stored at 4 or 12°C. After the rest period the growth vigor of seed tubers increases gradually until a maximum is reached. The length of the phase of maximum growth vigor depends on the cultivar, but is long at low storage temperatures (4°C). Finally, after the phase of maximum growth vigor, the growth vigor decreases again to zero (1, 2, 7).

The higher the storage temperature, the higher the rate of physiological ageing of seed tubers (6, 7). After a storage period of ca. 35 weeks seed tubers stored at 4°C are in the phase around maximum growth vigor whereas seed tubers stored at higher temperatures are in or beyond the phase of maximum growth vigor (7). At high storage temperatures, cultivars with a high rate of physiological ageing will attain the phase of decreasing growth vigor earlier than cultivars with a low rate of physiological ageing (7). Thus, the ratio in growth vigor between seed tubers stored at a high temperature

and those stored at a low temperature should give an indication of the rate of physiological ageing.

One experiment was carried out to elucidate the effect of different storage temperatures on growth vigor of 28 cultivars. The storage temperatures found to produce the most marked inter-cultivar differences in the first experiment were used in a second experiment to develop a method to measure inter-cultivar differences in the rate of physiological ageing.

Material and Methods

Experiment 1

Seed tubers of 28 cultivars were harvested in the first weeks of August 1981 and stored at ca. 15°C. On 21 September the seed was disinfected with ethyl mercury bromide (AARDISAN, Haren, The Netherlands, 4% a.i.; seed was immersed for 5 min in a 0.3% solution of the trade product). From 24 September 1981 until 22 March 1982 samples, each comprising 36 uniform tubers (mean tuber weight 65 g), were stored in the dark at 4, 10, 16, 22 or 28°C. At the end of the storage period the seed was carefully desprouted and then placed in the light at a temperature of 16°C until planting on 20 April 1982. Seed was planted by hand in the field in a fertile sandy soil. The rows were 75 cm apart and tuber spacing within the row was 30 cm. The usual cultural practices were carried out.

The experiment was laid out as a split-plot design with two replicates, cultivars as the main factor and storage temperature as the split factor. In each replicate 18 tubers were planted per treatment. Each plot consisted of two rows of 9 plants.

A number of observations were made to characterize early growth vigor of the seed. The total percentage of emerged plants, the rate of emergence (roughly equivalent to the percentage of emerged plants at the moment that 100% emergence of the 4°C treatment was achieved) and the plant height at 42 and 57 days after planting were assessed. After day 57 no further observations were made to estimate the early vigor of the seed, because of the increasing influences of the environmental conditions. Because the final yield at maturity was determined, no intermediate harvests were taken.

Experiment 2

Test Material—Ageing tests were carried out in 1986 with 18 cultivars, multiplied in the same field. After a curing period of 28 days at 15°C, the tubers were disinfected against *Rhizoctonia solani* with validamycine (Solacol, AAGrunol, Haren, The Netherlands, 30 g/l a.i.; tubers were immersed in a 3% solution of the trade product). On 26 September 1985 three samples of every cultivar, each comprising 25 uniform tubers (mean tuber weight 60 g) were placed in darkness at constant temperatures of 4, 12, 16 and 20°C

and a relative humidity of 80% for 28 weeks. After this storage period all sprouts were carefully removed. The tubers were pre-sprouted in light at 16°C for 12 days (laboratory test) or 23 days (field test) before they were planted. Four weeks curing period plus 28 weeks storage at different temperatures plus three weeks pre-sprouting means a total storage period of 35 weeks.

Laboratory Test—Twenty-four tubers from each of the 4 storage treatments were planted in a plastic tray (L x W x H = 45 x 30 x 10 cm) filled with quartz sand. Trays were placed in a dark growth chamber at 16°C and 80% RH. The sand was wetted frequently. After 28 days the number of sprouts, fresh and dry weights of sprouts and dry weight of roots were determined. Also, the fresh and dry weights of the newly formed tubers were determined. "Little potato" tubers formed directly on the mother tuber or on stems that had not emerged, were excluded.

Field Test—The field experiment was laid out as a split-plot design with two replicates and with cultivars as the main factor and storage temperature as the split factor. Within the main plot, temperature treatments were arranged from low to high in replicate one and from high to low in replicate two. This arrangement resulted in loss of randomization but its advantage was that it facilitated the visual observations during growth. Each plot consisted of two rows of 12 plants. On 26 April 1986 the tubers were planted by hand in a 25 x 75 cm² arrangement. The usual cultural practices were carried out.

The number of emerged plants was counted every 2-3 days. For all cultivars 80% emergence of the treatments stored at 4°C was reached within 16-22 days after planting. Plant height and light interception were measured 47 days after planting. Moreover, the relative haulm development (crop stand score) was estimated visually per plot, by relating the haulm mass of each of the warm-stored treatments of a cultivar to the haulm mass of the 4°C treatment (score 100). A score of 0 was given if no plants had emerged. In order to limit the interference of environmental conditions with the physiological age of the seed tubers, all treatments of a cultivar were harvested about five weeks (51-58 days after planting) after 80% emergence of the 4°C treatment of that cultivar. The fresh and dry weights of haulm and tubers and the numbers of stems and tubers per m² were determined.

Ageing Indices—Ageing indices were calculated by taking the results of a crop parameter obtained by one or more higher storage temperatures as the numerator and always using the value obtained for the 4°C temperature as the denominator. The main formulae used for calculating ageing indices were:

$F1 = 10 * T_{20} / T_4$, $F2 = 5 * (T_{16} + T_{20}) / T_4$ and $F3 = 3.33 * (T_{12} + T_{16} + T_{20}) / T_4$, in which F stands for formula number and T for storage temperature. The higher the score, the lower the rate of ageing. Ageing indices were calculated for many parameters.

Growth vigor is the potential of a tuber to produce sprouts and plants within a relatively short space of time. A fast development of fresh haulm indicates a potential for high yield. Therefore, the ageing index based on the fresh haulm weight per m², five weeks after 80% emergence of 4°C treatment in the field test, was used as the reference index (RI) against which the other indices were compared. Within each formula, correlation coefficients were calculated between the RI and the indices based on other parameters.

Results and Discussion

Experiment 1

The most appropriate parameter from Experiment 1 for assessing early growth vigor is the total plant height (cm/m²) 42 days after planting. When using this parameter allowance was made for number of emerged plants and the size of the plants without much interference from environmental conditions yet. The total plant height decreases (or does not increase) with increasing storage temperatures for 27 cultivars (Table 1). Only cv. Alpha has a higher total plant height after storage at 10 and 16°C than after storage at 4°C. We have no explanation for this different behavior of Alpha. An experimental error, such as exchanging the samples in the store of 4 and 16°C cannot be excluded, but we have no indications that this occurred.

The decrease of total plant height depends greatly on the cultivar. The rapidly ageing cultivar Jaerla (7) shows a very definite decrease in plant height, whereas cultivar Désirée, known as a slowly ageing cultivar (7), maintains a high total plant height even after storage at 22°C.

The mean plant height for 28 cultivars shows that 10°C is too low a storage temperature and 28°C is too high a storage temperature for distinguishing cultivars by rate of ageing. However, the intermediate temperature levels 16°C and 22°C result in very contrasting effects among cultivars in total plant height.

Experiment 2

A close correlation was observed between the ageing indices based on different crop parameters in the field test and the reference index RI (Table 2). Of course, a correlation is not very surprising because the fresh haulm weight is not independent of the other crop parameters. The advantage of basing ageing indices on crop parameters other than the fresh haulm weight has to be that they are easier and faster to determine. The total plant height and a visual estimation of the relative haulm mass by means of a crop stand score satisfy these requirements without loss of much reliability (Table 2).

The ageing indices based on certain growth parameters in the laboratory test showed a substantially lower correlation with the reference index

TABLE 1.—Total plant height (cm/m²) 42 days after planting for 28 cultivars after different storage temperatures of the seed. Experiment 1.

Cultivar	Storage temperature				
	4°C	10°C	16°C	22°C	28°C
Eersteling	124	102	89	60	32
Doré	109	93	84	51	11
Carina	82	76	30	27	8
Gloria	136	124	122	82	9
Primura	84	69	34	33	2
Alcmaria	78	64	45	11	0
Jaerla	107	93	29	7	0
Ostara	96	93	62	29	1
Sirco	100	93	73	27	14
Ehud	64	62	62	29	4
Resy	107	87	89	80	2
Estima	129	136	84	29	17
Spunta	140	104	61	26	4
Draga	91	87	87	36	4
Bintje	111	98	89	44	3
Emergo	89	73	82	20	1
Hertha	129	111	89	16	0
Kennebec	84	76	73	62	16
Désirée	89	82	84	64	26
Saturna	100	89	96	93	24
Eba	113	109	80	73	6
Pruceres	96	84	36	53	14
Prominent	82	87	89	35	1
Alpha	49	60	78	8	0
Exodus	75	60	51	21	0
Procura	104	89	84	73	18
Multa	109	96	98	60	15
Astarte	127	98	9	10	0
Mean	100	89	71	41	8

Least significant difference between treatments of one cultivar: 19.6 (according to the Studentized range test of Tukey; $P=0.05$).

RI than did ageing indices that were based on field parameters (Tables 2 and 3).

In Table 4 the cultivars have been ranked according to the index based on the fresh haulm weight (RI) and formula F2 in the field test. The field test gave a more differentiated view of the cultivar differences in rate of ageing than the laboratory test did. The laboratory test gave extreme differences; cultivars with a low rate of ageing scored very high, whereas those with a reasonably high rate of ageing scored very low, without any discrimination within groups. Bintje and Spunta scored quite well in the field test

TABLE 2.—*The linear correlation coefficients between ageing indices based on different parameters and the reference index RI (based on fresh haulm weight in the field test) by using the formulae: $F1=10 \cdot T20/T4$, $F2=5 \cdot (T16+T20)/T4$ or $F3=3.33 \cdot (T12+T16+T20)/T4$ for both calculations ($n=18$ cultivars). Experiment 2 - field test.*

Parameter	F1	F2	F3
% Emergence	0.67 ^{a,b}	0.57	0.51
% Light interception	0.91	0.88	0.85
Total plant height (cm m ⁻²)	0.94	0.94	0.92
Crop stand score	0.94	0.92	0.92
Dry weight of haulm (g m ⁻²)	1.00	1.00	0.99
Fresh weight of tubers (g m ⁻²)	0.94	0.92	0.85
Dry weight of tubers (g m ⁻²)	0.91	0.89	0.77
Number of stems (m ⁻²)	0.86	0.84	0.80
Number of tubers (m ⁻²)	0.86	0.80	0.70
Total dry matter weight (g m ⁻²)	0.98	0.98	0.96

^aIf $r \geq 0.71$, r differs significantly from 0 at $P \leq 0.001$.

^bCorrelation coefficient between index based on Formula F1 and fresh haulm weight (the RI for formula F1) and index based on Formula F1 and percentage emergence.

TABLE 3.—*The linear correlation coefficients between ageing indices based on different parameters and the reference index RI (field test) by using the formulae F1, F2 or F3^a. Experiment 2 - laboratory test.*

Parameter	F1	F2	F3
Number of sprouts per tray	0.76 ^b	0.72	0.73
Fresh weight of sprouts (g)	0.86	0.83	0.83
Dry weight of sprouts (g)	0.86	0.83	0.87
Dry weight of roots (g)	0.81	0.76	0.78
Dry weight of sprouts plus tubers (g)	0.77	0.54	0.48
Dry weight of sprouts plus roots (g)	0.85	0.84	0.87

^aFor explanation see Table 2.

^bIf $r \geq 0.71$, r differs significantly from 0 at $P \leq 0.001$.

^cCorrelation coefficient between index based on formula F1 and fresh haulm weight (field test) and formula F1 and number of sprouts per tray (laboratory test).

but in the laboratory test they scored extremely low. The physiologically old tubers of the warm-stored treatments that still emerged were able to produce quite well in the relatively warm spring of 1986. No such compensation is possible in a laboratory test.

It may be assumed that ageing indices based on formulae with two or more storage temperatures in the numerator give a more reliable view of the rate of ageing of seed tubers than ageing indices based on only one

TABLE 4.—*The classification of 18 cultivars based on ageing indices of the field test and the laboratory test, with the formulae F1 and F2^a. Experiment 2.*

Cultivar	Field Test				Laboratory Test	
	F2		F1		F2	F1
	Fresh haulm weight	Crop stand score	Fresh haulm weight	Crop stand score	Fresh sprout weight	Fresh sprout weight
Saturna	10	10	10	10	10	10
Multa	9	8	8	8	9	8
Désirée	8	9	8	10	9	9
Exodus	8	7	7	6	10	9
Bintje	7	8	6	7	2	2
Gloria	7	6	7	7	7	8
Kennebec	7	6	8	7	5	5
Sirtema	6	8	7	8	5	5
Spunta	6	5	5	4	1	0
Hertha	6	4	6	4	6	4
Procura	5	5	6	6	6	6
Doré	5	5	5	5	2	2
Primura	5	4	4	3	1	1
Astarte	5	4	4	3	1	1
Eersteling	4	3	3	3	1	1
Jaerla	4	3	3	1	1	1
Ostara	3	2	1	1	2	1
Alcmaria	2	2	2	2	2	2
LSD (P=0.05)	1.4	1.7	1.9	1.9		

^aFor explanation see Table 2.

temperature in the numerator. Nevertheless, there was a good relation between the ageing indices of the cultivars calculated with the formulae F2 and F3 ($r=0.99$, not shown in table) for the field test. Also, the classification of cultivars based on ageing indices calculated with formula F1 did not result in large deviations compared with the classification of cultivars based on formula F2 (Table 4), but the coefficient of variation of these indices is higher (ca. 17%) than those of indices based on two or three storage temperatures in the numerator (ca. 13%).

The test method based on only one temperature in the numerator (20°C) and a visual estimation of the haulm mass based on a crop stand score is attractive, because of its simplicity and lower labor requirement.

In Experiment 1 plants were not harvested at an early stage in the growing period. So, ageing indices based on fresh haulm weight could not be calculated for this experiment. However, Experiment 2 proved that the total plant height is also a good parameter for calculating ageing indices. Thus, ageing indices based on this parameter were calculated for cultivars

from Experiment 1. Seventeen cultivars were common to Experiments 1 and 2. To test the repeatability of the test method described the ageing indices of Experiments 1 and 2 were compared: the one from Experiment 1 based on total plant height 42 days after planting (cm/m²) and the formula $5*(T16+T22)/T4$ and the one from Experiment 2 based on fresh haulm weight and formula F2 ($5*(T16+T20)/T4$).

The ageing indices for the 17 common cultivars (Table 5) show that there is a good similarity between the two tests. Only the figures for the cultivars Exodus, Spunta, Procura and Astarte differ by more than two units. Cultivars could be classified into three groups, according to whether they had a low (7.1-10), moderate (4.1-7) or high (0-4) rate of ageing. Comparing the classification of the two years it can be seen that no cultivar changes from the first group to the third group, or vice versa. Désirée is classified as a cultivar with a low rate of ageing and Jaerla as a cultivar with a high rate of ageing. This is in full agreement with the findings of Van der Zaag and Van Loon (7).

Differences between years for one cultivar in values for an ageing index do not necessarily mean that the test is not reliable, but that the rate of physiological ageing differs from year to year as do other cultivar features. Therefore the test should be carried out for at least three years.

TABLE 5.—*The classification of 17 cultivars based on ageing indices of Experiments 1 and 2 (field test).*

Rate of physiological ageing	Cultivar	Experiment 1	Experiment 2	Mean
		$5*(T16+T22)/T4$ Total plant height	$5*(T16+T20)/T4$ Fresh haulm weight	
Low	Saturna	10	10	10
	Désirée	8	8	8
	Multa	7	9	8
	Kennebec	8	7	7.5
	Gloria	8	7	7.5
Moderate	Exodus	5	8	6.5
	Procura	8	5	6.5
	Bintje	6	7	6.5
	Doré	6	5	5.5
	Hertha	4	6	5
	Eersteling	6	4	5
	Spunta	3	6	4.5
	Primura	4	5	4.5
High	Ostara	5	3	4
	Alcmaria	4	2	3
	Jaerla	2	4	3
	Astarte	0	5	2.5

The test method discussed is clearly less labor-intensive than determining the sprouting capacity curves of each cultivar according to Krijthe (2). Moreover, the results of the laboratory test show that the similarity in results between a sprouting test and the performance in the field is not always high.

It is difficult to discuss the incubation period (3, 4) as a possible measure of the differences in rate of physiological ageing between cultivars because Reust did not determine the early growth vigor of warm- and cold-stored tubers. But in his tests the incubation period of Ostara, Bintje, Désirée and Saturna did not show much variation (2200-2400 day-degrees) whereas we found that their rate of physiological ageing differs greatly (Table 5).

Proposed Procedure

The following procedure is proposed for assessing the rate of physiological ageing of potato cultivars. Healthy seed tubers of different cultivars (grown in the same field) are harvested and stored for three weeks at 15°C for wound curing. Per cultivar, six samples of 25 uniform tubers each (mean tuber weight, e.g., 60 g) are selected. The samples are disinfected against silver scurf (*Helminthosporium solani*) and black scurf (*Rhizoctonia solani*) and stored in the dark at 4, 16 and 20°C; two samples at each temperature. After 28 weeks the tubers are carefully desprouted and pre-sprouted at 15°C in light for two or three weeks.

The tubers are planted in the field in plots of two rows with 12 tubers per row in a split-plot design with two replicates. Four weeks after the emergence of the 4°C treatments the haulm development is estimated visually by relating the haulm masses of the 16 and 20°C treatments to that of the 4°C treatment (4°C=100). If more labor is available or more accuracy is desired, the fresh haulm yield can be determined. The ageing index is calculated using the formula $F = 5 * (T_{16} + T_{20}) / T_4$. A somewhat rougher estimate of the rate of ageing may be obtained when only the 4°C and the 20°C storage temperatures are included (formula: $F = 10 * T_{20} / T_4$).

If it is wished to have a scale of 0-10, the ratios T_{16}/T_4 and T_{20}/T_4 are maximized to 1.

Because the rate of physiological ageing of one cultivar varies from year to year, the test should be carried out for at least three years.

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