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The influence of harvest maturity and basic macroelement content in fruit on the incidence of diseases and disorders after storage of the 'Ligol' apple cultivar

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ABSTRACT

Research was carried out during the 1999-2007 growing and storage season using 'Ligol' apples from trees grafted on M.26, and fertilised in accordance with recommendations for commercial orchards. The approximate optimum harvest date was determined mainly on the basis of starch index measurements and Streif index calculations, evaluated 7-8 times every 4-5 days each year. In addition to samples collected to determine OHD each year, there were four harvests of fruit intended for storage. The apples were stored in a cold storage room at 1-2°C and RH of around 90% for about five months. Following a storage period lasting the same number of days for each harvest, the quality of fruits was examined and the correlation between the concentration of minerals in apples and all fungal diseases and physiological disorders was calculated. In years with high precipitation in the period preceding the harvest, the share of fruits affected by fungal diseases and physiological disorders after storage depended more on weather conditions prevailing in the growing season than on the harvest date. The incidence of bitter pit increased with the number of days preceding OHD. Other recorded losses were caused by internal breakdown and superficial scald, but they were small and did not seem significant for 'Ligol' apples. Each characteristic mineral concentration in the fruit at harvest (N, P, K, Ca and Mg) was correlated with the incidence of physiological disorders and/or fungal diseases. The incidence of bitter pit and lenticel blotch pit and the sum of physiological disorders increased along with the increase in nitrogen concentration. The feature that best predicted the storability of 'Ligol' was the K/Ca ratio.

Key words: bitter pit, fungal diseases, mineral content, physiological disorders, quality, storability

Abbreviations:

OHD – optimum harvest date, TSS – total soluble solids, TA – titrable acidity

INTRODUCTION

The storability of apples depends on many factors. Although the genetic factor is the most important (Ahmadi-Afzadi 2012), environmental and weather conditions (Failla et al. 1990), agrotechnical measures regulating mineral nutrition (Sams 1999, Andziak et al. 2004) and the harvest date (Łysiak 2011) also have a strong influence on storability. Dry weather during the vegetation season disrupts Ca uptake, whereas abundant rains in the pre-harvest period are conducive to the incidence of

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fungal diseases (Ferguson and Watkins 1989). Crop load strongly influences Ca uptake. Regardless of fruit size, apples from light-cropping trees had lower Ca and higher K concentrations and are more susceptible to bitter pit than did fruit from trees with heavy crop loads (Ferguson and Watkins 1992).

Low Ca concentrations in apples at harvest have often been associated with fruit susceptibility to disorders that may develop during cold storage (Saks et al. 1990). Calcium-deficient apples are sensitive to bitter pit, cork spot, water core, scald, core browning, flesh breakdowns, and low-temperature disorders (Ferguson and Watkins 1989, Tomala 1997). However, fruit with low Ca concentrations does not always show breakdown (Perring and Preston 1974), and breakdown has sometimes been reported in fruit with high Ca levels (Perring and Plocharski 1975). In such instances, low P appeared to be involved in the development of the disorder (Perring and Plocharski 1975). High K levels and a high K/Ca ratio (Dilmaghani et al. 2004) strongly affect the incidence of physiological disorders, and sometimes also fungal diseases (Fallahi et al. 1997). Losses caused by Ca deficiency in fruit result not only from the incidence of physiological disorders but also from decay (Conway and Sams 1983).

The proper harvest date is crucial for the storability of autumn and winter apple cultivars intended for long storage (Łysiak 2011). Fruits harvested at an unripe stage are more prone to shrivelling (Łysiak and Kurlus 2000, Nguyena et al. 2004), physiological disorders and diseases (Valero and Serrano 2010). Over mature fruits are likely to become soft and mealy (Kader 1997) and have an insipid flavour after a short period of storage (Kays 1991).

Because mineral nutrition and OHD are modified by the genetic factor, the correlation between these three factors needs to be identified for important cultivars to minimize losses during their storage. In Poland and in other Central and Eastern European countries, 'Ligol' has belonged to the more economically important apple cultivars since the mid-1990s (Czynczyk et al. 2009). Therefore, this research was conducted to measure the relationship between mineral nutrition, OHD and the genetic features of this cultivar to help optimise its storability.

MATERIAL AND METHODS

The research was conducted in the cold storage facility and laboratory of the Department of Pomology of the Poznan University of Life Sciences between 1999 and 2007 and was designed to determine and evaluate the storability of the 'Ligol' cultivar depending on the harvest date and mineral concentration. Trees, grafted on M.26, were planted in an experimental orchard in 1993 in a distance of 3.5 × 1.2 m. The trees were fertilised every year with nitrogen (soil application, 60 kg N per ha) and calcium (foliar application five times 5 kg per ha of $CaCl_2$); other minerals were applied depending on the result of the leaf and soil analysis, in accordance with standard recommendations for commercial orchards (PIORIN 2003). Calcium was applied when the soil analysis showed a decrease in the pH below 5.5. During the research, two tons of CaO per ha were applied in the autumn of 2001 and 2004, respectively. Magnesium was applied three times: in 2000, 2003 and 2005 (80 kg MgO per ha each time). Potassium was added in the spring of 2002 and 2005 (100 kg K_2O per ha, each time).

Sampling

Fruits were collected every four to five days starting some weeks before the estimated OHD (Tab. 1). The approximate OHD was determined on the basis of starch index measurements and Streif index calculations. Besides samples intended for OHD determination each year, there were four harvests of fruit intended for storage.

The sample used to determine OHD consisted of 20 fruits picked from a minimum of 10 trees, from 120 to 160 cm high, outside part of the tree, from the same east-southern side. Fruits over or under size, infected by pests or diseases were rejected. However, the uniformity of the crop was quite high because the share of rejected fruits in each sample in the respective years ranged between about 2% and 10%. The fruits should represent the same stage of maturity (as well as have the same size and colour) as that of the fruits to be harvested for storage. The samples intended for storage were picked according to the rules applicable to the samples picked to determine OHD, but the size of a single sample intended for storage was considerably larger and amounted to four boxes of 10 kg each box (50-60 apples per box).

Measurement

The fruit maturity at harvest was evaluated according to well known standard methods:

– firmness: penetrometer EFFEGI FT011 (manufactured by Facchini srl, via REale 63, 48011 ALFONSINE (Ra), Italy). The maximum penetration force to the depth of the probe – 8 mm and 11 mm in diameter, two

Table 1. Schedule of experiments

Dates	1999	2000	2001	2002	2003	2004	2005	2006
Full bloom	26 IV	25 IV	4 V	27 IV	4 V	30 IV	3 V	8 V
Harvest	1 IX	27 VIII	5 IX	28 VIII	23 VIII	28 VIII	5 IX	1 IX
	6 IX[1]	1 IX	10 IX	31 VIII	28 VIII	2 IX	10 IX	6 IX
	11 IX[2]	6 IX	14 IX	5 IX	2 IX	6 IX	16 IX	11 IX
	16 IX[3]	11 IX	19 IX	**9 IX**	6 IX	11 IX	21 IX	21 IX
	21 IX	**16 IX**	24 IX	15 IX	11 IX	16 IX	26 IX	26 IX
	27 IX	21 IX	29 IX	19 IX	**15 IX**	**20 IX**	**30 IX**	2 X
	1 X	26 IX	**4 X**	24 IX	20 IX	24 IX	5 X	**6 X**
		30 IX	9 X		24 IX	29 IX		
End of storage	10 II 2000	12 II 2001	28 II 2002	02 II 2003	3 II 2004	6 II 2005	10 II 2006	12 II 2007
Length of storage period (days)	147	149	147	145	149	147	141	143

[1]Not underlined dates present dates of measurements carried out for the determination of OHD
[2]Underlined dates present dates of sample harvesting for storage
[3]Bold underlined dates present OHD based on judgement and Streif Index

opposite sides of the fruit with a small area of the skin removed – was registered in kgf;
- refractometer value (total soluble solids) in % was measured with an Atago PR-32a digital refractometer;
- starch disintegration according to a 10-point scale, where one means “no conversion” and 10 means “totally converted”; the reference value for the starch index was 4-5 (Ctifl 1995);
- titratable acidity was determined by titrating a 1 ml sample with 0.1 N NaOH to an end point of pH 8.2 using an Accumet pH meter manufactured by Fisher Laboratory Equipment Division, Pittsburgh, PA, and an automatic burette.

Storage conditions and evaluation of storability

Fruits were stored in a cold storage room at 1-2°C and RH of around 90% for about five months. The schedule of all measurements is shown in Table 1. The storability of apples was evaluated after the same number of days of storage respectively to their harvest date. The evaluation of storage efficiency was based on judgments and on measurements. The judgments was made using sensory tests. In the case of subjective evaluations, a panel of five independent judges experienced in postharvest evaluation conducted all of the tests. The results of individual evaluations were averaged. Incidences of diseases and disorders, fruit mass loss and internal quality criteria (firmness, TSS, titrable acidity) were assessed by two judges experienced in postharvest evaluation.

Each criterion was scored separately for each date of harvest. The results were processed statistically using an analysis of variance. The incidence of physiological disorders and/or fungal diseases was summed and averaged for each harvest date. The results obtained as percentage variables were subject to the Bliss transformation to meet the assumption of normal distribution. Mean comparisons were performed using the Duncan Multiple Range Test to examine the differences among harvest dates for statistical significance ($p < 0.05$).

Fruits from the respective harvests were examined in the laboratory for the presence of all fungal diseases and physiological disorders after the same number of days of storage. For each stored box, the percentage of fruits affected with each individual disease or disorder was determined.

Mineral analyses and correlation

For the mineral analysis, a sample was collected at the second harvest date, following the procedure applied to determine the fruit maturity and ripeness at harvest. The chemical analysis was conducted only once because the research by Saks et al. (1990) had shown that the total contents of Ca, Mg, and K in the pulp tissue of apples were constant between early and late harvest dates and during storage and subsequent shelf-life. The contents of N and K were determined with a wet ash procedure and the content of Ca with a dry ash procedure (Cunniff 1995). Total N in fruit samples was determined using the Kjeldahl method (Chapman and Pratt 1961). Ca, Mg and K concentrations were determined using atomic absorption spectrophotometry after lanthanum was added, and the content of P was measured using the colorimetric method. Fruit mineral content was expressed as mg 100 g on a fresh weight (FW) basis. Correlations between fruit mineral concentration and disorders and diseases were calculated. The correlation significance was tested for p = 0.05 and p = 0.01.

RESULTS AND DISCUSSION

The accumulation of nutrients in fruits depends on several factors, including the relative strengths of vegetative and reproductive links (Zavalloni et al. 2001); however, the weather conditions, primarily the accessibility of water, are of basic importance (Failla et al. 1990). The eight-year research showed that the accumulation of individual nutrients might vary considerably each year regardless of a similar level of nutrients in the soil maintained by means of fertilisation (Tab. 2). The nitrogen content in 'Ligol' apples ranged between 0.17 and 32 mg per 100 g d.m., which represents a high variability between years. Assuming that the highest nitrogen content found during the research in 2002, stands for 100%, the variability in per cent reached even 47%. Similarly, the variability in percentage between experiment years amounted to 29% for phosphorus, 39% for potassium and 24% for magnesium. The percentage variability of the calcium content was found to amount to no less than 45%, which inevitably had a significant influence on the ripening processes and the development of physiological disorders.

The number of fruits affected by fungal diseases varied depending on the harvest year and date. The greatest losses were reported in 2003 and 2006 (Fig. 1). Often, the later the harvest date, the greater incidence of storage diseases causing fruit decay (Błaszczyk 2006); however, such a pattern was observed only in two years of the research (1999 and 2002). In two years (2001 and 2006), the infection incidence even declined with the harvest date, which allows for the conclusion that the vulnerability of 'Ligol' apples to bull's eye rot (*Neofabraea* sp.), blue mould (*Penicillium expansum*) and brown rot (*Monilinia* sp.) does not depend on the stage of maturity. It should be noted, however, that in the years 2001 and 2006 the total precipitation in the period preceding the harvest (August-September) was very high (159.6 and 184.6 mm, respectively) as compared with the total average precipitation for these two months in all of the years of the research (108 mm) (Tab. 4). Such high precipitation levels create good conditions for the development of diseases and in the years concerned they most certainly disturbed the aforementioned pattern according to which the incidence of storage diseases increases with consecutive harvest dates.

Bitter pit is the most frequent physiological disorder in countries with high apple production (Ferguson and Watkins 1989). In all years of the research, at least half of the fruit losses caused by post-cold-storage physiological disorders were attributable to bitter pit. Bitter pit is a well-studied disorder, and the pre-harvest factors have been relatively well defined (Ferguson and Watkins 1989). Those factors include, in particular, low Ca and high K concentration and premature harvest.

Table 2. The concentration of macroelements (% d.m.) in 'Ligol' fruits during the second harvest of fruits intended for storage

Year	N	P	K	Mg	Ca	K/Ca	N/Ca
1999	0.30 ± 0.02*	0.058 ± 0.002	0.58 ± 0.04	0.031 ± 0.001	0.034 ± 0.008	18.2 ± 5.0	9.5 ± 2.5
2000	0.26 ± 0.03	0.059 ± 0.008	0.59 ± 0.04	0.031 ± 0.002	0.051 ± 0.010	11.7 ± 2.0	5.3 ± 1.2
2001	0.24 ± 0.03	0.054 ± 0.003	0.87 ± 0.04	0.038 ± 0.003	0.042 ± 0.010	22.3 ± 7.9	6.3 ± 2.7
2002	0.32 ± 0.02	0.058 ± 0.002	0.86 ± 0.03	0.033 ± 0.002	0.062 ± 0.007	14.1 ± 1.6	5.2 ± 0.7
2003	0.17 ± 0.02	0.042 ± 0.005	0.95 ± 0.03	0.038 ± 0.001	0.059 ± 0.013	16.6 ± 3.3	2.9 ± 0.4
2004	0.19 ± 0.02	0.048 ± 0.004	0.91 ± 0.09	0.038 ± 0.004	0.042 ± 0.004	22.1 ± 4.0	4.6 ± 0.5
2005	0.27 ± 0.03	0.045 ± 0.005	0.66 ± 0.04	0.029 ± 0.002	0.049 ± 0.003	13.6 ± 1.4	5.5 ± 0.7
2006	0.20 ± 0.02	0.042 ± 0.002	0.87 ± 0.09	0.037 ± 0.007	0.035 ± 0.004	25.3 ± 5.0	5.9 ± 1.0
Mean	0.24 ± 0.05	0.051 ± 0.007	0.79 ± 0.15	0.034 ± 0.004	0.047 ± 0.010	18.0 ± 4.8	5.6 ± 1.9

*Standard deviation

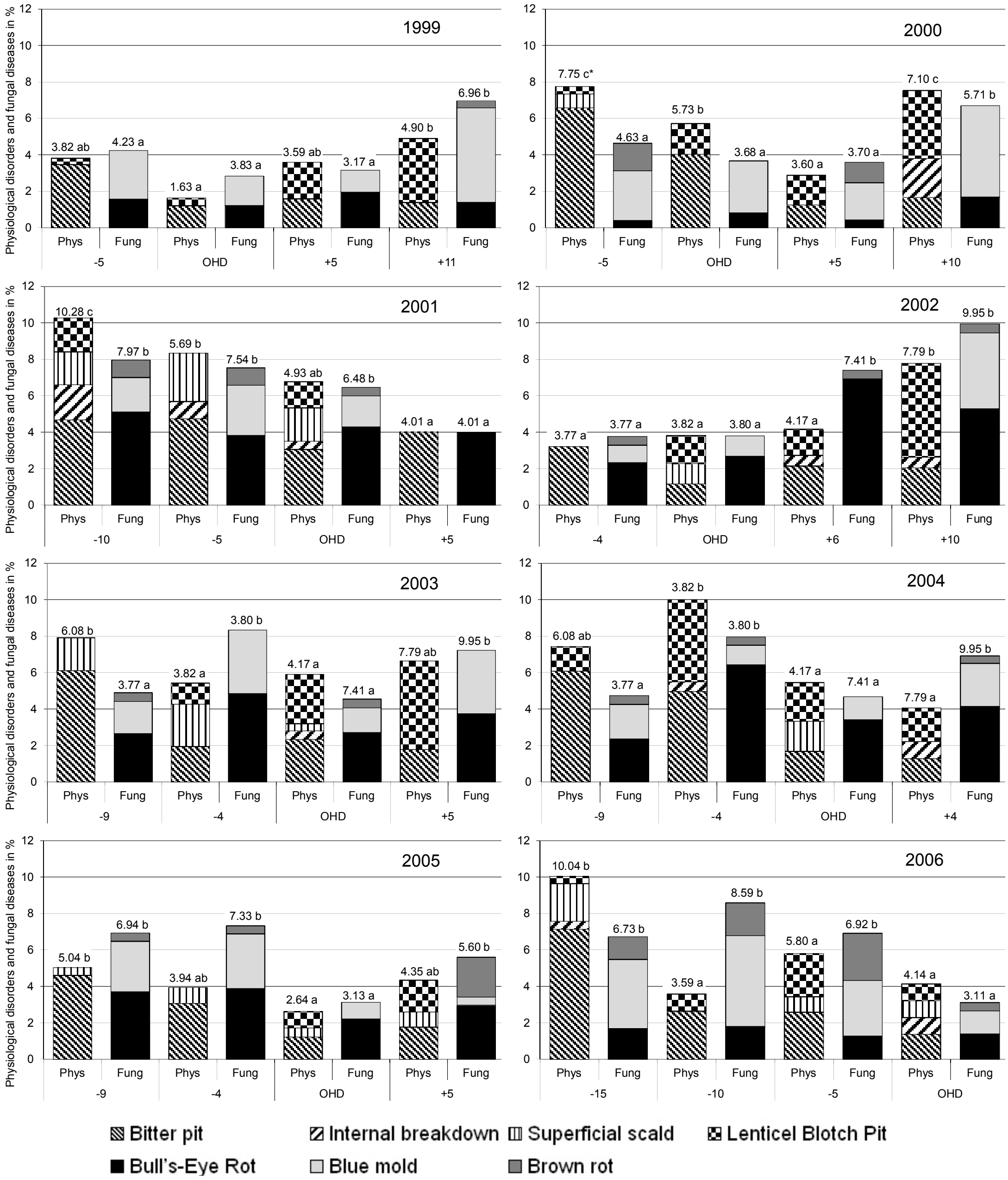

Figure 1. Incidence of fungal diseases and physiological disorders (%) on 'Ligol' fruits after five months of storage depending on year and harvest date

*Second row of X axis – numbers of days before or after the optimum harvest date (OHD)

Although the harvest date is believed to influence the incidence of bitter pit (Ferguson and Watkins 1989), this has not always been confirmed (Volz et al. 2006). In this research, the incidence of bitter pit increased with the number of days preceding OHD. However, this could be clearly observed only in 2000, 2004 and 2006.

In other apple disorders, pre-harvest factors are largely still speculative (Ferguson et al. 1999). The second most frequent physiological disorder observed in 'Ligol' apples was lenticel blotch pit. Its causes are similar to those of bitter pit (Perring 1984). Lenticel blotch pit only occurs after cold storage – Casero et al. (2010) detected this disease in fruit only after four months of cold storage under a controlled atmosphere of ultra low oxygen (CA-ULO) conditions. Other recorded losses were caused by internal breakdown and superficial scald, but they were small and seem not to be important for 'Ligol'.

A correlation between the losses caused by both fungal diseases and physiological disorders and the chemical composition of fruits at harvest was measured for all eight years of the research

(Tab. 3). The results show that the concentration of each basic mineral can affect the losses in 'Ligol' apples during storage. The weakest correlation was observed between the phosphorus concentration and the loss factors. The phosphorus concentration influenced only the incidence of blue mould and other fungal losses since phosphorus improves the cell structure and reduces susceptibility to fruit rot (CODEX 2003, Quaglia et al. 2011).

A very strong correlation was observed between nitrogen concentration and losses caused by both fungal diseases and physiological disorders. There was no strong correlation between the incidence of individual fungal diseases and nitrogen concentration, but there was a strong correlation between the accumulated presence of those diseases and the increase in nitrogen concentration. The incidence of bitter pit and lenticel blotch pit and the sum of physiological disorders increased along with the increase in nitrogen concentration. This phenomenon has been frequently observed and explained as resulting from an increase in the fruit mass, loosening of fruit tissue, rapid loss of firmness during storage and the disproportion between nitrogen concentration and the concentration of other minerals in fruits (Fallahi et al. 1997, Ferguson

Table 3. Correlation coefficients between concentration of macroelements and incidence of fungal diseases and physiological disorders after five-month storage of 'Ligol' fruits

Loss type	N	P	K	Mg	Ca	K/Ca	N/Ca
Bull's-Eye Rot	0.140	-0.185	0.680**	0.448	-0.178	0.381	-0.228
Blue mould	0.439	-0.513*	-0.063	0.135	-0.742**	0.400	0.195
Brown rot	0.339	-0.325	0.214	0.330	-0.718**	0.592**	0.308
Total diseases	0.569**	-0.656**	0.725**	0.672**	-0.607**	0.888**	0.028
Bitter pit	0.738**	-0.393	0.389	0.612**	-0.782**	0.706**	-0.074
Internal breakdown	0.105	0.110	0.436	0.547*	-0.268	0.513*	0.291
Superficial scald	0.463*	-0.332	0.538*	0.661**	-0.319	0.591*	-0.063
Lenticel Blotch Pit	0.570*	-0.096	0.473	0.643*	-0.336*	0.493*	-0.329
Total physiological	0.733**	-0.313	0.623**	0.853**	-0.654**	0.814**	-0.116
Total loses	0.721**	-0.502*	0.726**	0.843**	-0.690**	0.921**	-0.059

*Significant at p = 0.05
**Significant at p = 0.01

Table 4. Temperature and precipitation during vegetation periods in 1999-2006

Parameter	Year	April	May	June	July	August	September	Mean
Temperature (°C)	1999	10.6	14.7	17.3	21.6	19.5	18.4	17.0
	2000	13.5	17.1	18.7	17.1	19.5	13.4	16.6
	2001	8.2	15.8	15.8	20.8	20.4	12.1	15.5
	2002	9.4	17.4	18.6	21.0	22.0	14.4	17.1
	2003	8.9	16.2	20.0	20.8	20.9	15.2	17.0
	2004	10.0	13.2	16.7	18.7	21.0	14.9	15.8
	2005	9.6	14.3	17.5	20.4	17.6	16.7	16.0
	2006	9.2	14.4	19.5	22.9	16.7	16.1	16.5
	Mean	9.9	15.4	18.0	20.4	19.7	15.2	16.4
Precipitation (mm)	1999	34.3	49.5	63.0	75.7	59.7	44.3	54.4
	2000	11.7	50.2	41.6	70.8	86.4	34.3	49.2
	2001	31.6	7.2	77.7	41.7	65.4	94.2	53.0
	2002	33.6	59.9	26.2	25.0	53.4	28.6	37.8
	2003	24.5	14.6	24.6	85.7	14.5	19.8	30.6
	2004	14.9	46.9	63.8	41.7	41.8	33.3	40.4
	2005	14.2	68.0	11.5	96.6	52.8	52.7	49.3
	2006	39.8	33.3	17.4	23.8	162.0	22.6	49.8
	Mean	25.6	41.2	40.7	57.6	67.0	41.2	45.6

et al. 1999, Drake et al. 2002, Fallahi et al. 2006, Tahir et al. 2007, Kühna et al. 2011).

A similar relationship as that between nitrogen concentration and the incidence of diseases was found for potassium and magnesium. Whereas a high K concentration is treated on equal terms with a high N concentration due to ion antagonism between K and Ca (Ferguson et al. 1999, Casero et al. 2010), much less attention is paid to Mg concentration (Casero et al. 2004). In this research, the incidence of losses was even more strongly correlated with Mg concentration than with K concentration, which was especially visible in the losses caused by physiological disorders.

Ca concentration showed a high correlation with the incidence of bitter pit and total physiological losses, but with other physiological disorders the relationship was very weak. As early as in 1974, Perring and Preston observed that apples with low Ca concentration did not always fall to internal breakdown. In this research, the above observations applied not only to internal breakdown, but also to superficial scald and lenticel blotch pit. However, a high correlation was found out between Ca concentration and two of the three fungal diseases (blue mould, brown rot).

The highest correlation was found between losses and the K/Ca ratio. The incidence of both fungal diseases and physiological disorders strongly increased along with the increase in the ratio. The K/Ca ratio may prove very helpful in determining fruit storability in a given year because its determination during harvest helps estimate the losses after storage. In this research, we found out that a K/Ca ratio above 20 during harvest led to an increase in post-storage losses caused by diseases and disorders. Unlike some other studies, which reported the correlation between N/Ca ratio and losses during storage (Amiri et al. 2008, Casero et al. 2010), this study showed no such correlation for 'Ligol'. Therefore, N/Ca ratio seems to be useless for the prediction of storage losses in this cultivar.

CONCLUSIONS

1. In years with high precipitation in the period preceding the harvest, the share of fruits affected by fungal diseases and physiological disorders after storage depended more on weather conditions prevailing in the growing season than on the harvest date.
2. Mineral concentration in fruit at harvest (N, P, K, Ca and Mg) is correlated with the incidence of physiological disorders and/or fungal diseases.
3. K/Ca ratio can be a good indicator of the five-month-long storability of 'Ligol'.

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WPŁYW DOJRZAŁOŚCI ZBIORCZEJ I ZAWARTOŚCI PODSTAWOWYCH SKŁADNIKÓW MINERALNYCH NA WYSTĘPOWANIE CHORÓB PO PRZECHOWYWANIU JABŁEK ODMIANY 'LIGOL'

Streszczenie: Badania przeprowadzono w okresie wegetacyjnym i przechowalniczym w latach 1999-2007, na jabłkach odmiany 'Ligol', zebranych z drzew zaokulizowanych na podkładce M.26 i nawożonych zgodnie z zaleceniami dla sadów produkcyjnych. Przybliżoną optymalną datę zbioru określono głównie na podstawie pomiarów indeksu skrobiowego i wyliczeń indeksu Streifa, ocenianych corocznie 7-8 razy, co 4-5 dni. Oprócz pobrania próbek celu określenia optymalnej daty zbioru w każdym roku, przeprowadzono cztery zbiory owoców przeznaczonych do przechowywania. Jabłka przechowywano przez około 5 miesięcy

w komorach chłodniczych w temperaturze 1-2°C i wilgotności względnej wynoszącej około 90%. Po okresie przechowywania trwającym tyle samo dni podczas każdego zbioru sprawdzono, zarówno jakość owoców, jak również obliczono korelację pomiędzy zawartością składników mineralnych w owocach a występowaniem wszystkich chorób grzybowych i fizjologicznych. W latach, w których występowały wysokie opady w okresie poprzedzającym zbiór, udział owoców porażonych przez choroby grzybowe i fizjologiczne po przechowywaniu był zależny w większym stopniu od warunków pogodowych panujących w okresie wegetacyjnym niż od daty zbioru. Występowanie gorzkiej plamistości podskórnej było tym większe, im więcej było dni poprzedzających optymalną datę zbioru. Inne zanotowane straty były wynikiem rozpadu wewnętrznego i oparzeliny powierzchniowej, lecz były one nieznaczne i zdawały się nie odgrywać większej roli w przypadku odmiany 'Ligol'. Zawartość każdego z podstawowych składników mineralnych w owocach podczas zbioru (N, P, K, Ca i Mg) była skorelowana z występowaniem chorób fizjologicznych i/lub grzybowych. Występowanie gorzkiej plamistości podskórnej i plamistości przetchlinkowej oraz suma występowania chorób fizjologicznych wzrastały wraz z wzrostem zawartości azotu. Stosunek K/Ca był najlepszym wskaźnikiem pięciomiesięcznej zdolności przechowalniczej jabłek odmiany 'Ligol'.

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