FINAL REPORT

RESEARCH ON 'INTERNAL DISORDERS AND QUALITY OF ROCHA PEARS'.

September 1997 through June 1998

Confidential

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Summary

This report presents the results of the research, as conducted so far, for the cooperatives Frutus, Campotec and Agricola to improve the storage of Rocha pears to extent the storage period of these pears. The research is mainly focussed on the CA-storage. An important problem of CA-storage of Rocha pears is the development of internal browning and cavities in the fruits. The main objective is the determination of safe CA-conditions (without development of internal browning) where taste, texture, flavour and other quality aspects are maintained.

Rocha pears from two orchards were used for the storage experiment. They were picked in the beginning of August and the storage at ATO in CA-containers started early September. The pears from both orchards were stored at -0.5° C in combination with the following gas conditions: 0.5% CO_2 - 3% O_2, 3% CO_2 - 3% O_2, 0.5% CO_2 - 1% O_2, 3% CO_2 - 1%O_2 and normal cold storage. Every 7 weeks samples were taken from the containers to measure firmness, colour, vitamin C content and internal disorders. This took place immediately after storage and after 4 days of shelf life simulation (at 19°C). At three sampling times the respiration and diffusion characteristics of the pears of one orchard, stored at 0.5% CO_2 and 3% O_2, were determined.

After 15 weeks storage the first pears were found with internal damage in 3 out of 4 CA-conditons. The higher the CO_2-content and the lower the O_2-content, the more abnormalities were found. In the most extreme CA-conditions,100% of the fruits showed the disorder. In 0.5% CO_2 - 3% O_2 only a low amount of pears showed internal damage. After 22 weeks in normal cooling, also brown colouring developed at a higher level than in 0.5% CO_2 - 3% O_2. As yet for practical application this CA-condition (0.5% CO_2 - 3% O_2) is useful.

Furthermore, an important observation was that the green ground colour of the pears stored at a low oxygen content was maintained, whereas the CO_2 content was did not influence this aspect.

Pears in the selected CA condition showed a slight better colour retention than normal cooled pears. Continuously during the storage period, a higher firmness level was measured after shelflife, with pears from normal refrigerated storage.

The external disorder scald starts developing after 22 weeks in store and increased very rapidly during shelflife. The disorder was especially inhibited at a low oxygen content, whereas the CO_2 content was of less importance. This is in conflict with the influence of CA-conditions on internal browning. To find a CA-condition which gives a save protection for scald with no risk on internal disorders seems difficult. But as long as the use of DPA is allowed the scald problem is solved. Also the selected CA-condition (0.5% CO_2 - 3% O_2) gave more scald protection than normal storage.

Important was in this research to understand more about the background of the development of internal browning and cavities which should result in practical parameters. For this reason gas exchange and vitamin C was measured.

The vitamin C content was strongly influenced by the CA-conditions: the extreme CA-conditons showed a lower vitamin C content. This was related with an increased degree of browning.

It is possible to use vitamin C as a parameter during CA-storage. The initial levels before storage did not show a good relation with the later development of browncore as found with the pears from different orchards. But this observation should be proved with research on vitamin C levels at different harvesting times and orchards.

The respiration rates of Rocha pears were determined to i) relate respiration with the development of brown hearts and ii) to calculate the necessary scrubber-activity of CA-installations in practice.

It was found that the respiration rate of the fruits did not differ during the storage time and was fairly constant within the range of 21 and 4% O_2. At lower oxygen contents in CA the respiration rate is strongly reduced. The respiration rate of Rocha-pears is about equal to Conference-pears.

Measurements on gas diffusion showed large differences between individual pears, which may explain the different susceptibility for internal brown.

1.0 Introduction

In general Rocha pears can be stored and sold from August until February using normal cold storage. At these conditions a longer storage period is not possible, because the pears become tasteless and because of the development of storage diseases such as browncore and superficial scald. To extend the storage period some experience with CA-storage has been gained in research and in practice. The result was that the taste of the pears stored at CA was better, less scald occurred, but the pears developed internal abnormalities. The symptoms of this were browning around the core and the development of cavities.
These problems are also known for other pear cultivars (such as Conference) and occur at pears that are picked late and stored at too high CO_2 and too low O_2 conditions.
Together with the cooperatives Frutus, Campotec and Agricola dos Fruticoltures and the research centre at Alcobaca, an inventory was made last year to improve the storage conditions.
Since there is ample experience with CA-storage of pears at the ATO (experience with Conference pears), it has been decided to carry out storage research with Rocha pears at the ATO for the duration of one season.
The objective of this research is to select the CA-storage conditions for Rocha pears at which no internal and external abnormalities develop and the quality (taste, texture) of the pears will be maintained.
In this report the research activities will be described as carried out from August 1997 till June 1998.

1.1 Relationship with research on Conference-pears

The research on Rocha pears is facilitated because of 'Europear', a European Community research project at the supervision of ATO-DLO. The general objective of the Europear project is to prevent the development of tissue disorders in pears resulting in internal cavities and browning. The causes underlying the development of the disorder are unknown, although there is a lot of correlative knowledge on the development of internal disorders. The occurrence is influenced by weather factors, orchard factors (location, nutrition), picking date, post-harvest treatments and storage conditions. However, relationships found in one country cannot simply be applied for other countries or growing seasons.
The key element in Europear is the development of technology which enables a rapid measurement and decision about the post-harvest treatments and storage conditions. Part of the research is an extensive research on gas exchange rates, diffusion resistance and pear quality throughout the storage season. This will be further explained.

1.2 Theoretical background gas exchange characteristics

Because pear tissue has a very low porosity, small changes in metabolic rates or diffusion rates within the tissue can lead to very low internal oxygen levels. Increased fermentation can lead to an increase of toxic metabolites like acetaldehyde (Perata and Alpi, 1991). This may reduce cell viability and induce cell death, leading to the Brown Heart disorder. Another explanation is that the combination of oxidative and fermentative processes are not sufficient to maintain cell viability (Andreev *et al*, 1991; Zhang and Greenway, 1994). The reason for a difference in occurrence of Brown Heart between Northern and Southern European countries might be a the influence of different climate and/or agronomical factors on pear growth and development. This could result in different metabolic rates, resistance to gas diffusion, energy metabolism, energy needs for maintenance costs, etc. To check this hypothesis data is needed on climate and orchard conditions during the growing season and gas exchange rates, diffusion resistance and pear quality throughout the storage season, carried out in different European countries. Once a physiological explanation is found, storage conditions have to be adapted in order to prevent disorders. For this purpose predictive models are needed, which use information collected during the growth of the pear, or directly after harvest. Recently several models are developed describing gas exchange based on enzyme kinetics. The model of Peppelenbos *et al.* (1996) might function as a basis for the models that will be developed. The model uses ATP production rate as the main inhibitor of fermentative CO_2 production. This model enables to describe the relation between gas (O_2 and CO_2) concentrations and O_2 consumption and CO_2 and ATP production rates.

1.3 Background of vitamin C measurements.

The research with Conference Pears proved a strong relation between the occurrence of internal disorders (brown and cavities) and the vitamin C content. The background of this is that the vitamin C fulfils an important function in resisting browning. Initially the vitamin C prevents browning because it is a natural antioxidant. But in combination with other substances this substance can also catch so called radicals. These radicals affect the cell membranes leading to damage of the membranes. Therefore, enzymes (PPO) and polyphenols can react with each other and form a brown pigment (melanin) that finally forms the brown colour of the tissue.

The theory is, that at extreme CA-conditions less energy is available to produce vitamin C over and over again, causing a decrease of the content. When at a certain moment there is too little vitamin C available in a fruit, the radicals can no longer be caught, leading to browning. In case at specific storage conditions a significant decrease occurs during storage, this may be an indication for the development of brown hearts.

The relation with the respiration measurements is important because it gives an insight in the available energy.

2.0 Material and methods

2.1 Pears for the research

In the beginning of September the pears were transported from Portugal to the Netherlands. About the picking time (probably 2 August), the growing conditions of the pears and the period preceding the transport in Portugal no exact data are available. But after harvested product was cooled also during transport. The pears have not been subjected to any treatment before storage to prevent spoilage and scald. This was agreed beforehand.
The pears were transported in plastic crates and came from two different orchards, namely José Domingos dos Santos and Joáo Picarra.
At arrival at the ATO the pears were immediately stored at cooled conditions. Before the pears were put into the storage containers they were homogenized and randomized to obtain a fair distribution of pears over all the containers (storage conditions).

2.2 Storage facilities and storage conditions

After the pears were cooled (two days) they were placed in the CA-containers at the respective storage conditions. The containers with a content of about 600 litre were placed in a cold store room in which during the whole storage period a temperature of -0.5^0 C was maintained. Measurement and control of the CA-conditions (CO_2 and O_2) were almost continuously carried out by a process computer. Per hour an air sample of the container is measured and if necessary actions are undertaken to restore the set point of the conditions. This can be: scrubbing of CO_2 with potassium hydroxide, adding CO_2, ventilation or injection of nitrogen. The conditions were maintained during the whole storage period.
The pears of the two orchards were stored together in a container at the planned storage conditions.

Storage conditions:

CO_2 (carbon dioxide)	O_2 (oxygen)
0	21
0.5	3
0.5	1
3	3
3	1

2.3 Measurements of internal and external quality

During the storageperiod the pears were judged several times on quality. This took place at the start and after 8, 15, 22, 29 and 36 weeks in storage.
The measurements were done at a sample of pears directly from the storage container concerned and at a sample of pears that was kept for 4 days at atmospheric conditions at a temperature of 19^0 C (shelf life simulation). In addition, after 29 and 36 weeks, the pears were also judged after 6 days shelflife.
The quality measurements consisted of:
1) Colour measurement of the skin
Of 15 pears per storage condition and grower the colour of the skin was measured with the Minolta Chromameter. Using this method the colour path from green to yellow can be determined objectively.
2) Measurement of firmness
At the same 15 pears the firmness was measurement with the penetrometer. This was done with the 11 mm plunger.
3) Visual inspection of internal and external abnormalities
The same 15 pears were also cut into linear and cross direction. Eventual deviations were divided into 4 categories:

0 = no browning
1 = minor browning
2 = moderate browning
3 = strong browning

The same categories were used for cavity formation
4) After a storage period of 22, 29 and 36 weeks measurements were carried out on extractable juice. A small disk of the pear tissue with a dimension of 8 mm high and a diameter of 17 mm was used. The disk was compressed to 67% of its original hight between two flat plates by an Instron equipment. The content of released juice was measured with filter-papers.

2.4 Vitamin C determinations

The research at Conference-pears showed a strong relation between the occurrence of internal abnormalities (browning and cavities) and the vitamin C content. Vitamin C is known for its quality to combat browning (anti-oxidant effect) but can also prevent damage to the cell membranes. However, if, due to extreme storage conditions, vitamin C is no longer produced to a sufficient extent, the total vitamin C content in the pear will decrease. When at specific storage conditions a significant decrease occurs during storage, this could indicate a false storage condition.
At each judgement of all storage objects (storage conditions and origin) the vitamin C content was determined.
The measurement of vitamin C content was done to pears that came directly from the storage room. Of every 10 pears from each condition a sub-sample was taken of the tissue from the stalk to the core. This material was frozen in liquid nitrogen and subsequently stored under deep freeze conditions at respiration -80^0 C.
The vitamin C content in the fruit is the content of ascorbate plus de-hydro-ascorbate.
The determinations were executed with High-Performance Liquid Chromatography(HPLC).

2.5 Gas exchange characteristics of Rocha pears

The research on Rocha pears is partly comparable to a European concerted action on Conference pears called 'Europear' (FAIR3-CT96-1803). The general objective of the Europear project is to prevent the development of tissue disorders in Conference pears resulting in so called Brown Heart. Details of this project are described in the previous report. During the past year measurements were carried out on Rocha pears that were comparable to measurements within the Europear project (like gas exchange, diffusion resistance). Results on Rocha pears will be compared with Conference pears to facilitate a good interpretation of the results.

Rocha pears were stored at 0°C, 2% O_2 and 0% CO_2. Gas exchange characteristics were measured at 2°C. This part of the work focussed on the quantification of variation in gas exchange rates and diffusion resistance of Rocha pears. Pears were measured three times during the storage period:

a. Autumn 1997 (3 times: 13 and 15 October 1997);
b. Winter 1998 (3 times: 30 January and 2 February 1998);
c. Spring 1998 (2 times: 8 and 10 May 1998).

Gas exchange rates

Fresh weight and underwater weight (Bauman and Henze, 1983) were measured. Samples from pears were taken to measure the specific weight of pear juice (used in the calculation of the porosity). Outer surface area of the pears was estimated using the length and the maximum circumference of the pear. Two pears were put in 1500 ml cuvettes. The cuvettes were connected to a flow through system. Selected gas conditions were all combinations of 0, 0.5, 1, 2.5, 6 and 21% O_2 with 0 and 5% CO_2 (realised conditions are shown in the figures). Relative humidity was high (>95%) since the gas was led through water flasks. After 3 and 5 days of storage O_2 uptake and CO_2 production was determined. This was done by disconnecting the cuvettes from the flow through system, and sampling the headspace directly and after a period of 6 hours. The GC used was a Chrompack CP 2002. The measured O2, CO2 and N2 was corrected to 100% to account for possible pressure variations inside the GC. Then the concentration values (in %) were multiplied with the actual pressure inside the cuvette (in kPa). Because the volume of the cuvette and the volume of the pears is known, the gas exchange rates were calculated in nmoles/kg.s.

Diffusion resistance

The method of measuring diffusion resistance as described by Peppelenbos and Jeksrud (in press) was slightly adjusted. The inert gas neon was used as well, but instead of measuring the diffusion of neon into the fruit the diffusion of neon out of the fruit was measured. First the fuit was stored in a cuvette with a high concentration of neon (5000-6000 ppm) for one night, assuming that by that time an equilibrium between the concentration inside the fruit and in the cuvette was reached. The final concentration was measured. Then the fruit was transferred to another cuvette, and after specific time intervals (seconds) the neon concentration was measured. The time intervals were selected based on the paper of Banks (1985), using the so called 'linear method'.

Statistical analysis
The gas exchange rates were analysed using the nonlinear regression analysis of the statistical package Genstat. The models used within the package were derived from Peppelenbos and van 't Leven (1996) and Peppelenbos et al. (1996).

3.0 Results

The results are mainly presented in graph's, always the combination CO_2 - O_2 is used

3.1 Firmness measurements

During the whole storage period the firmness was measured with the penetrometer. This was executed directly after storage and after 4 days in shelflife. The firmness of the pears directly after storage showed no significant decrease during storage. The initial level for both orchards 11.5 kg, after 36 weeks in storage the firmness was still between 10 and 10.5 kg. Differences between storage conditions were not detectable.

Fig.1 Firmness of Rocha-pears from origin Picarra after shelflife

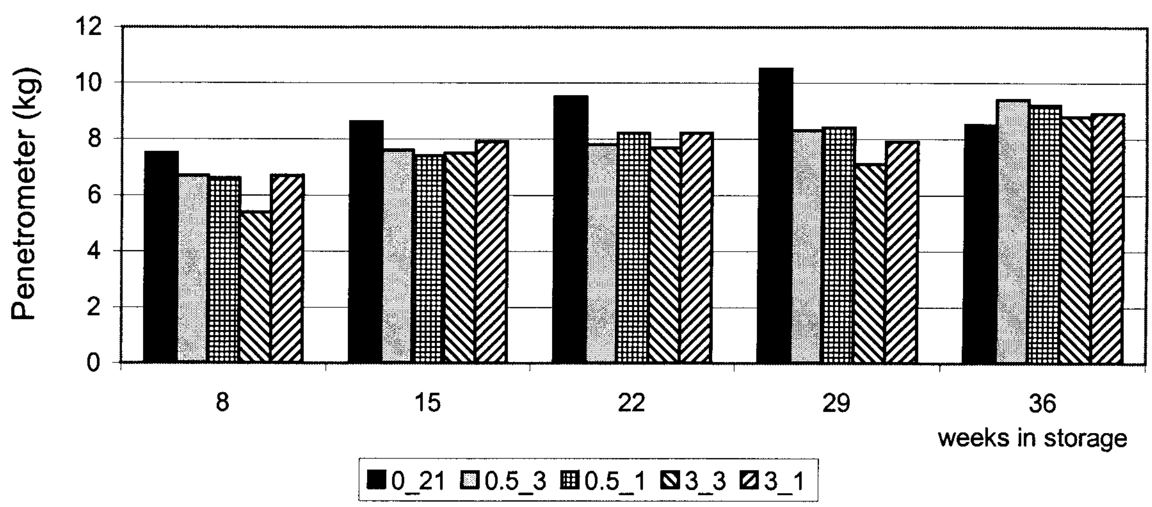
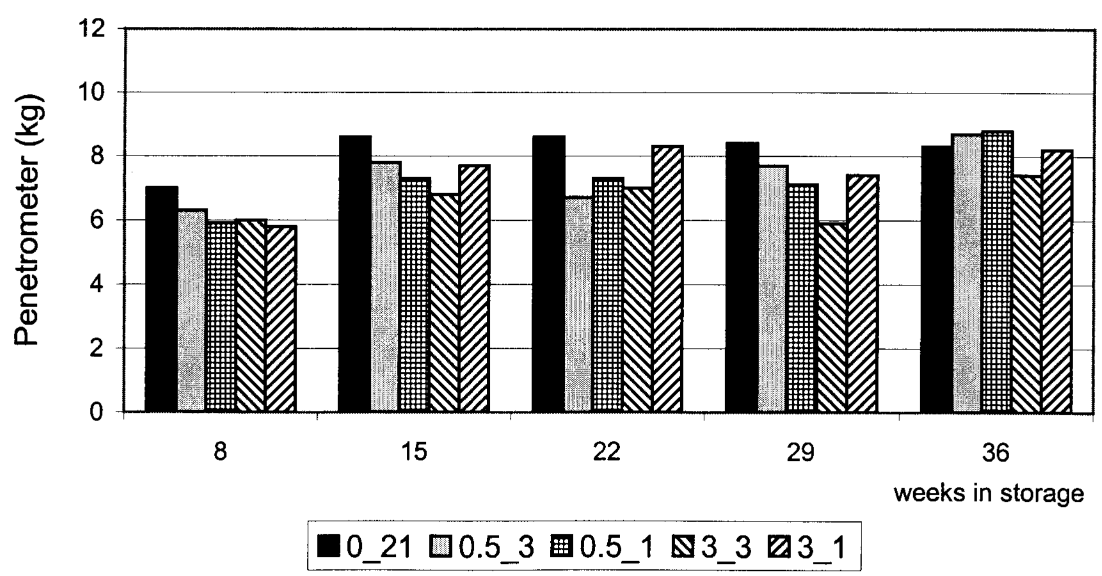


Fig. 2 Firmness of Rocha-pears from origin Santos after shelflife



However after 4 days shelflife in 19° C significant differences occurred, due to storage duration and storage-conditions. There is a tendency that pears from both orchards became firmer when the storage time prolonged. More or less this tendency was noticed for each storage condition. Remarkable is the higher firmness of the pears stored in normal cooling conditions (0% CO_2 and 21% O_2.). The reason for this phenomena is unknown, the lack of ripening capacity of this pears could be one of the reasons.

3.2 Colour measurements

The colour of the pears is measured with a Minolta Chromameter. This objective colour meter is used as an international standard for measuring colour of fruits. This meter allows measurements of several colour spectra. For the colour range from yellow to green the a-value usually satisfies. When the a-value is about zero, the pears are almost yellow. These are also shown in graphs 3 through 6.

Between pears of the two orchards a different pattern of colour development was noticable. Before storage a value of 16 was measured for both origins. The colour-reduction, measured directly after storage as well as measured after shelflife decreased, faster for the pears from origin Santos. Striking is the significant colour decrease in the first 8 weeks of storage.
So the initial colour value is not a guarantee for retention of the green colour.

Important is the influence of the storage conditions on colour-retention. For pears from both orchards a specific pattern is notified. The best colour retention is the storage in 1 % oxygen (0.5 and 3% CO_2 is equal). At second best is the result in 3 % O_2. Also in this circumstances the effect of CO_2 is minimal. Storage of the pears in normal refrigerated conditions resulted always in the most yellow colour.

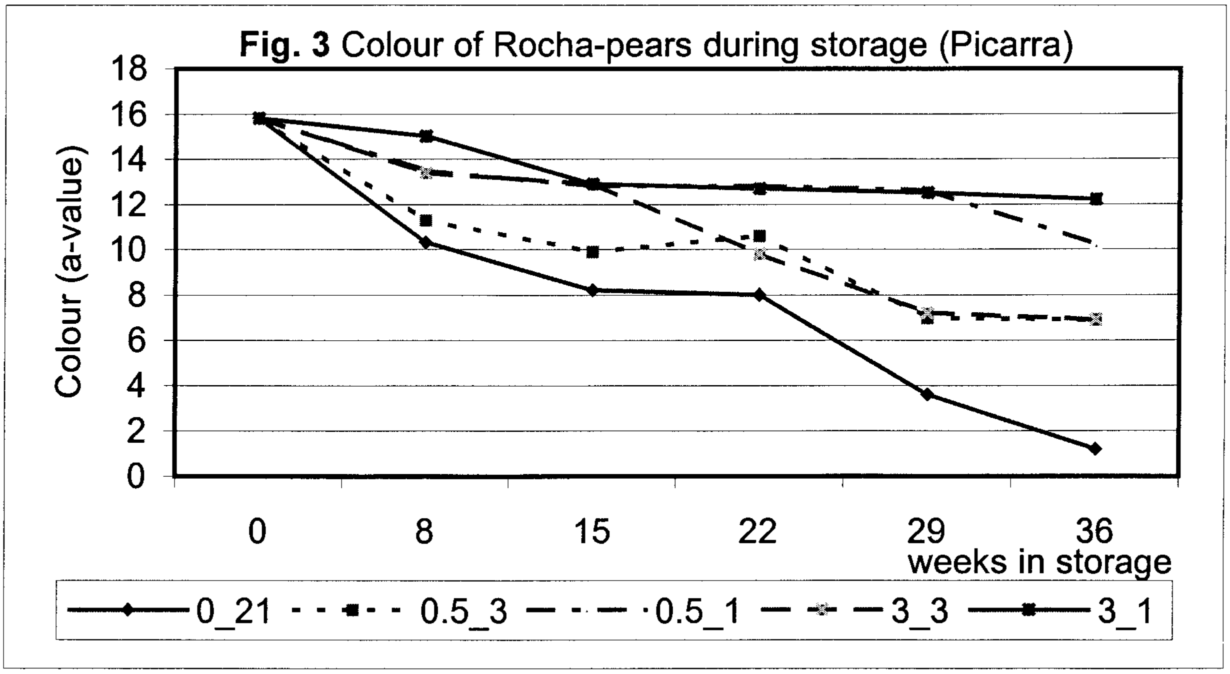


Fig. 3 Colour of Rocha-pears during storage (Picarra)
Colour (a-value)
18
16
14
12
10
8
6
4
2
0
0
8
15
22
29
36
weeks in storage
0_21
0.5_3
0.5_1
3_3
3_1

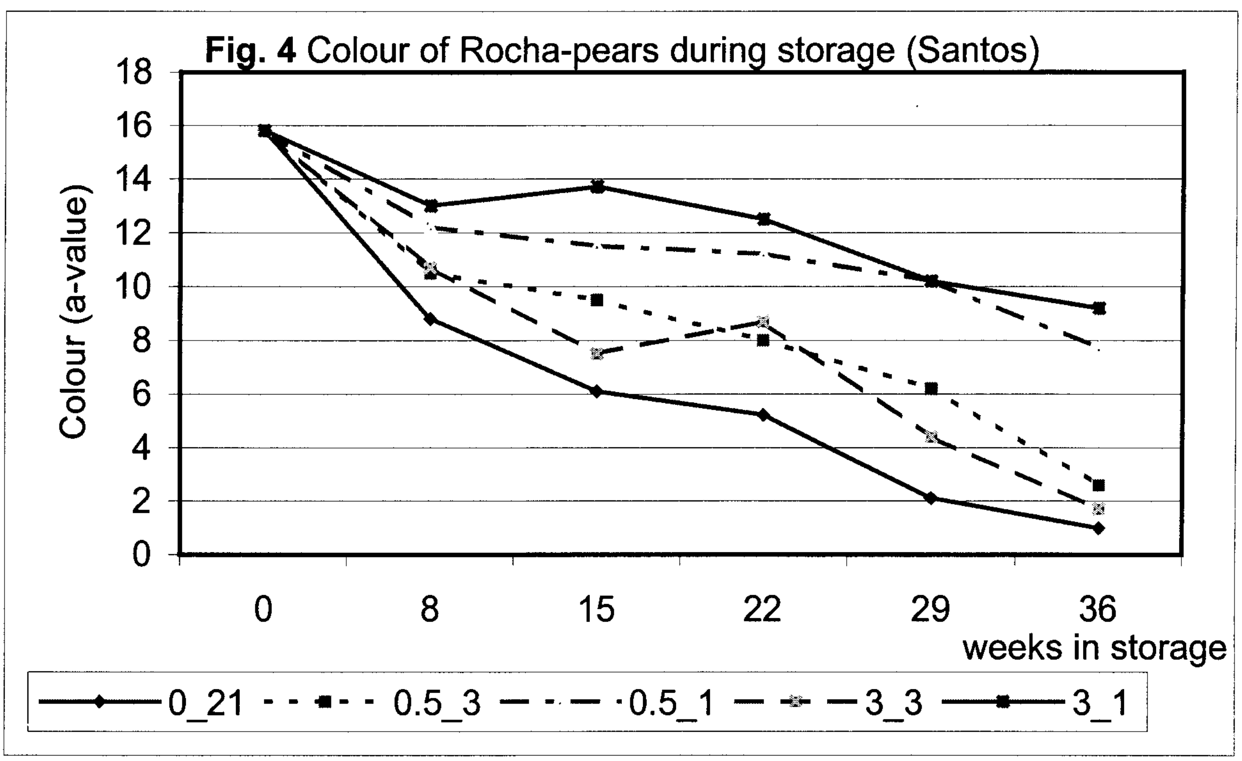


Fig. 4 Colour of Rocha-pears during storage (Santos)
Colour (a-value)
18
16
14
12
10
8
6
4
2
0
0
8
15
22
29
36
weeks in storage
0_21
0.5_3
0.5_1
3_3
3_1

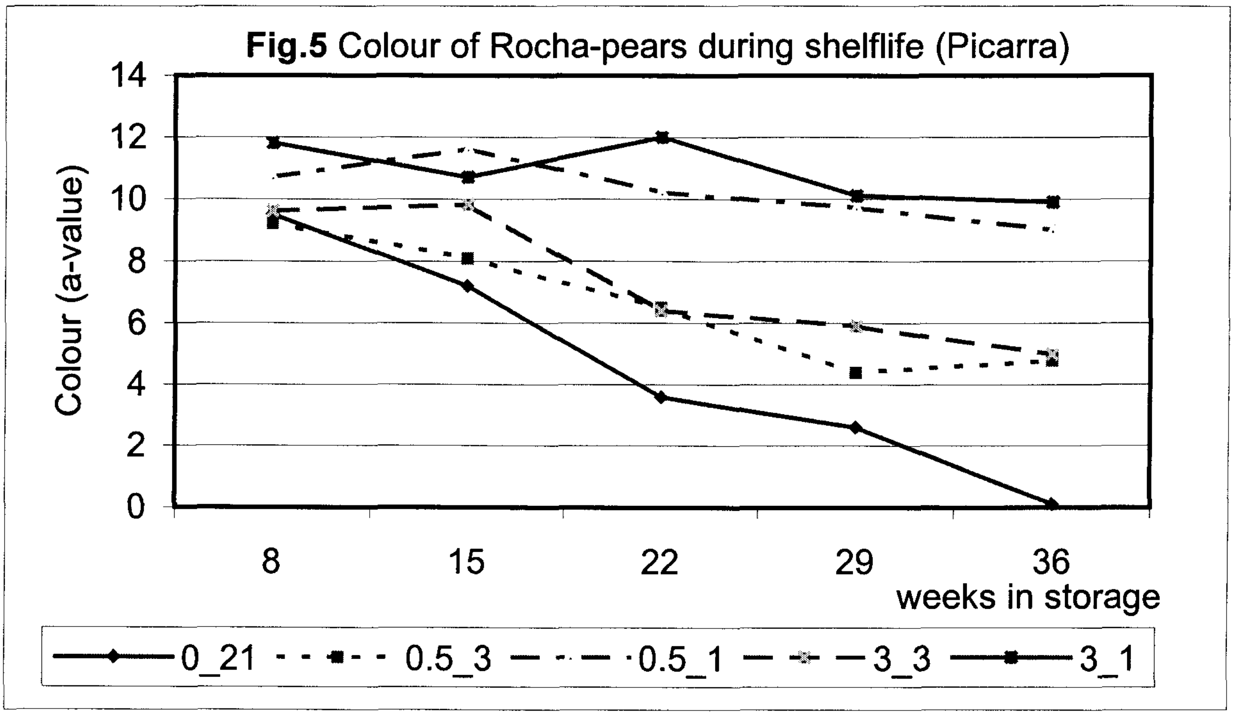


Fig.5 Colour of Rocha-pears during shelflife (Picarra)
Colour (a-value)
14
12
10
8
6
4
2
0
8
15
22
29
36
weeks in storage
0_21
0.5_3
0.5_1
3_3
3_1

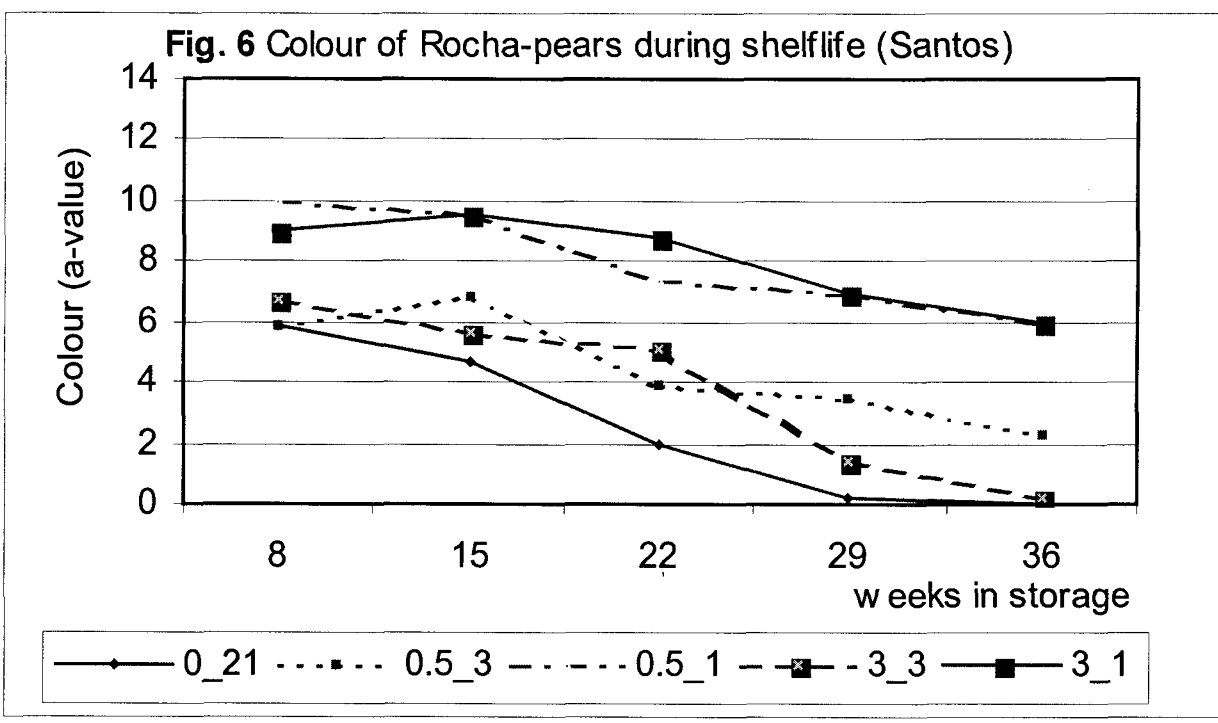


Fig. 6 Colour of Rocha-pears during shelflife (Santos)
Colour (a-value)
14
12
10
8
6
4
2
0
8
15
22
29
36
w eeks in storage
0_21
0.5_3
0.5_1
3_3
3_1

3.3 Browning and cavities in the pears

The judgement on internal browning and cavities is executed at 5 times during the whole storage season. This visual internal inspection was carried out directly after storage and after 4 day shelflife. During the first inspections after 8 and 15 weeks storage the amount of internal browning after storage was a little bit higher then after the shelflife period as described in the interim report. During the observations after 22, 29 and 36 weeks it could not be detected again. Therefore the given internal disorders in the fig. 7-10 is the average of storage- and shelflife observations.

Symptoms in Rocha-pears start with a small brown spot in the core especially at the calyx side. A further stage is enlarging towards the calyx and surrounding the core, and can further extend to the cortex tissue. Usually brown turns into cavities, but not always. Also small cavities without browning have been seen.

In the figures only the percentage of fruits attacked by the physiological disorder is calculated. For the market evaluation we can imagine the fruits should be only sound.

For scientific reasons it's interesting to have information on the intensity of the disorder. A relationship can be expected between the percentage of the disorder and the intensity and also the storage duration. In general the symptom (browning and cavities) starts with stage 1, and depended on the sensitivity of the pears, it increased to a maximum (stage 3). Gradually, until the storage period of February, the batches with maximum of 50% disorders showed only stage 1. Above 50% affected fruits, the intensity of the symptom increased. After February the area of the disorder in the pear increased when the amount of attacked fruits is higher.

The influences of storage-duration, -conditions and orchard on browning and cavities are equal, so it could be described in the same way as brownheart or disorder.

A small difference between the orchards for the disorder is noticeable. Pears from orchard Santos seems to be more sensitive especially the development of cavities.

After 15 weeks in "dangerous" storage-conditions the first symptoms have been detected, which increased very quick during the following judgements.

The most important question is the comparison of CA-conditions. A lower oxygen concentration and a higher CO_2 increased the sensitivity for the disorder but especially the combination of both factors.

It is very clear that the CO_2 - O_2 combinations 3-1, 0.5-1 and 3-3 are unsafe for Roch-pears. Only the combination *0.5% CO_2 and 3% O_2* gave a good result so without much brownheart. Surprising is that the results of this CA-regime are better than pears in the regular cold storage. The pears in cold storage showed after 22 weeks the first symptoms of brownheart. which increased at later judgements.

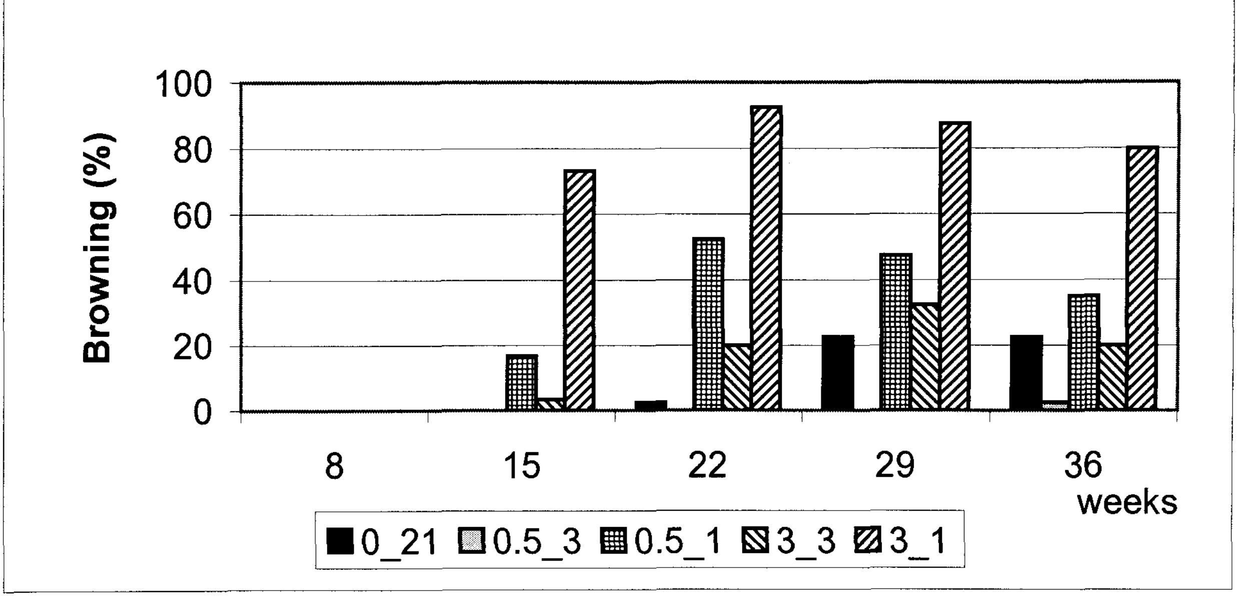


Fig 7 Browning in Rocha-pears during storage (Picarra)

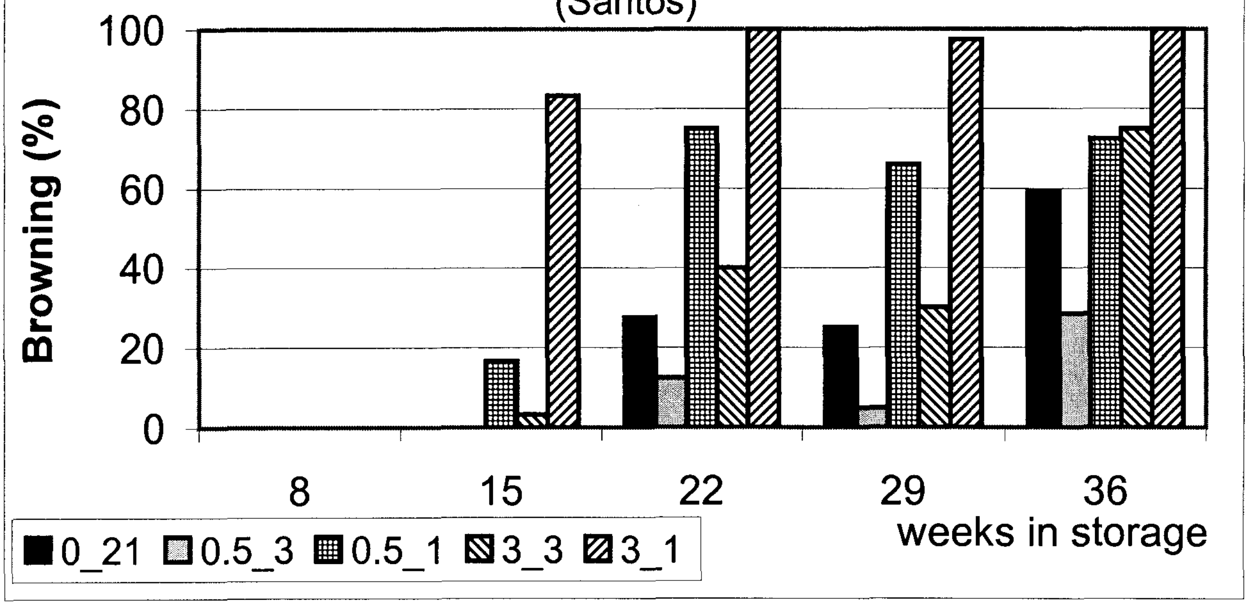


Fig 8 Browning in Rocha-pears during storage (Santos)

Fig. 9 Cavities in Rocha-pears during storage (Picarra)

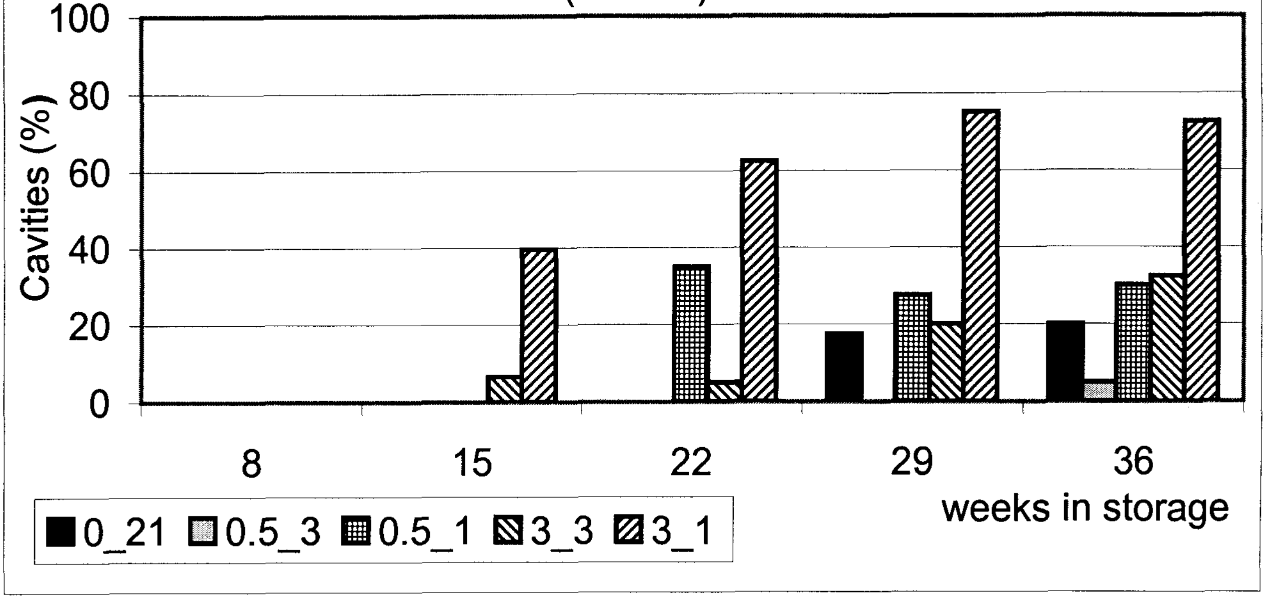
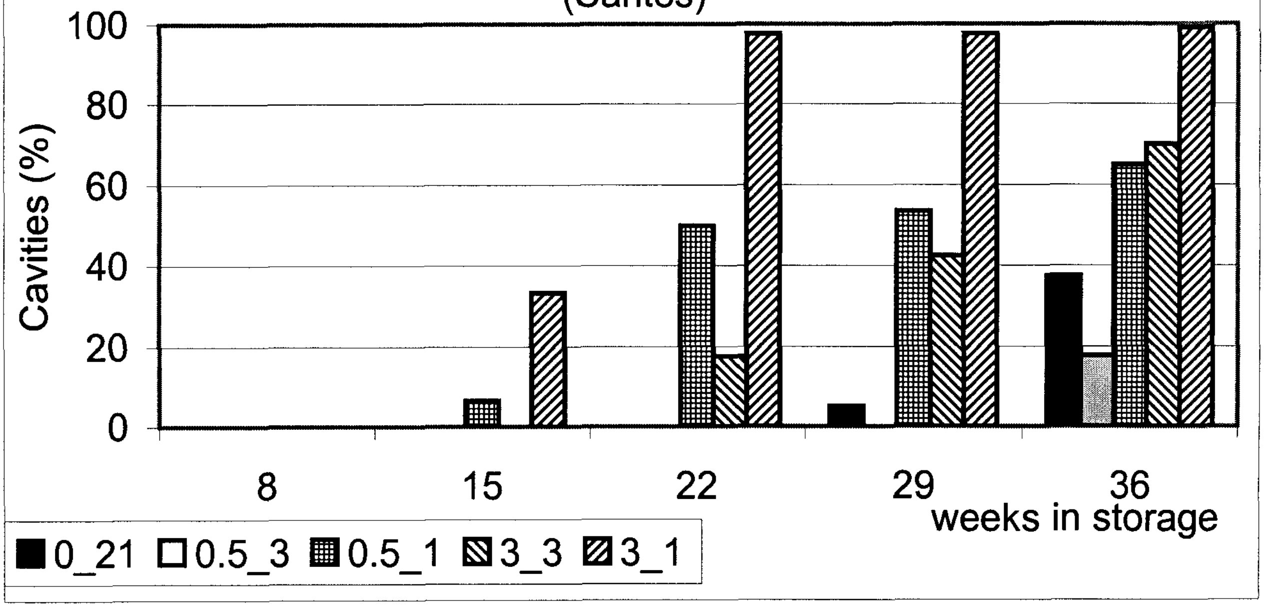


Fig. 10 Cavities in Rocha-pears during storage (Santos)



3.4 Juice content of the pears

In order to investigate the probable lack of taste of Rocha pears after prolonged storage in refrigerated storage is in each storage condition the extractable juice determined.
This was only with pears from the origin Santos after 29 and 36 weeks in storage.
The measurement was executed with pears stored 6 days in shelflife.

Table 2 Influence of storage conditions on extractable juice content

	Extractable juice (%)	
Storage condition (CO_2-O_2)	29 weeks	36 weeks
0-21	22.52	25.10
0.5-3	22.35	23.11
0.5-1	23.10	23.66
3-3	23.54	26.15
3-1	21.05	26.70

There are no significant differences in extractable juice % between pears stored at different conditions.

3.5 Scald development

The development of scald (percentage of fruits affected by the disorder) is given in the figures 11-14. This is the percentage of fruits attacked by the disorder. In the figures 11 and 12 the development of scald in the fruits judged directly from store, is visualized. In general fruits from the origin Picarra are much ore sensitive than fruits from origin Santos. After the shelflife period of 4 days in 19° C the external disorder increased significant, for some storage conditions up to 100%.
The first symptoms developed after 22 weeks in store and varied from some small area's on the skin towards great brown area's
For the influence of the storage conditions directly after storage and after shelflife the same tendency is found. Pears in normal refrigerated storage showed the highest sensitivity. Inhibition of scald is most effective in a lower oxygen concentration. Also a higher carbon dioxide level reduced scald. The only storage condition, which gave a good protection, is :
3% CO_2 -1 % O_2.
For untreated (DPA) fruits, the other conditions could not prevent to scald development. Otherwise it should be taken in account that only one CA-condition was more or less free from internal disorders: 0.5% CO_2 -3 % O_2. The sensitivity for scald in this condition, therefore is a relevant question. The percentage attacked fruits was in most cases also high, but lower than in normal refrigerated storage, which is an advantage.

For this most interesting condition for practical application, a comparison is made between the sensitivity for scald and internal browning (fig.15 and 16).
Pears from origin Santos are less susceptible for scald but more to internal damage, while pears from Picarra showed a bigger sensitivity for scald and almost no internal defects.

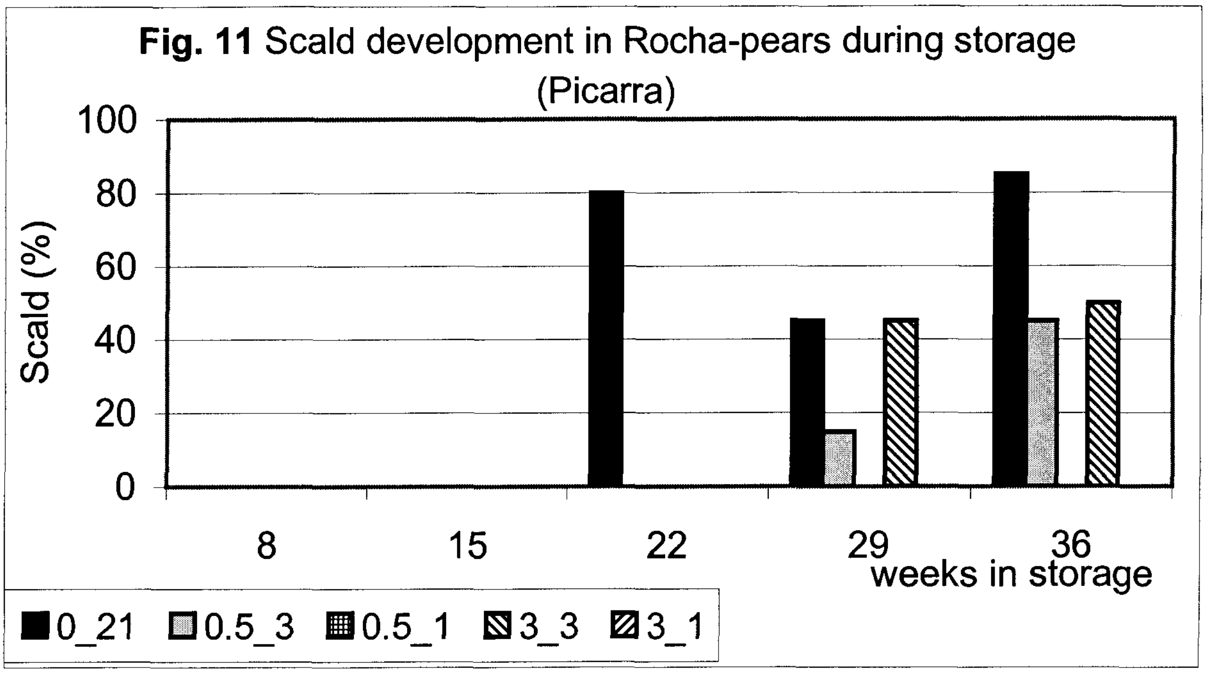


Fig. 11 Scald development in Rocha-pears during storage (Picarra)
Scald (%)
100
80
60
40
20
0
8
15
22
29
36
weeks in storage
0_21
0.5_3
0.5_1
3_3
3_1

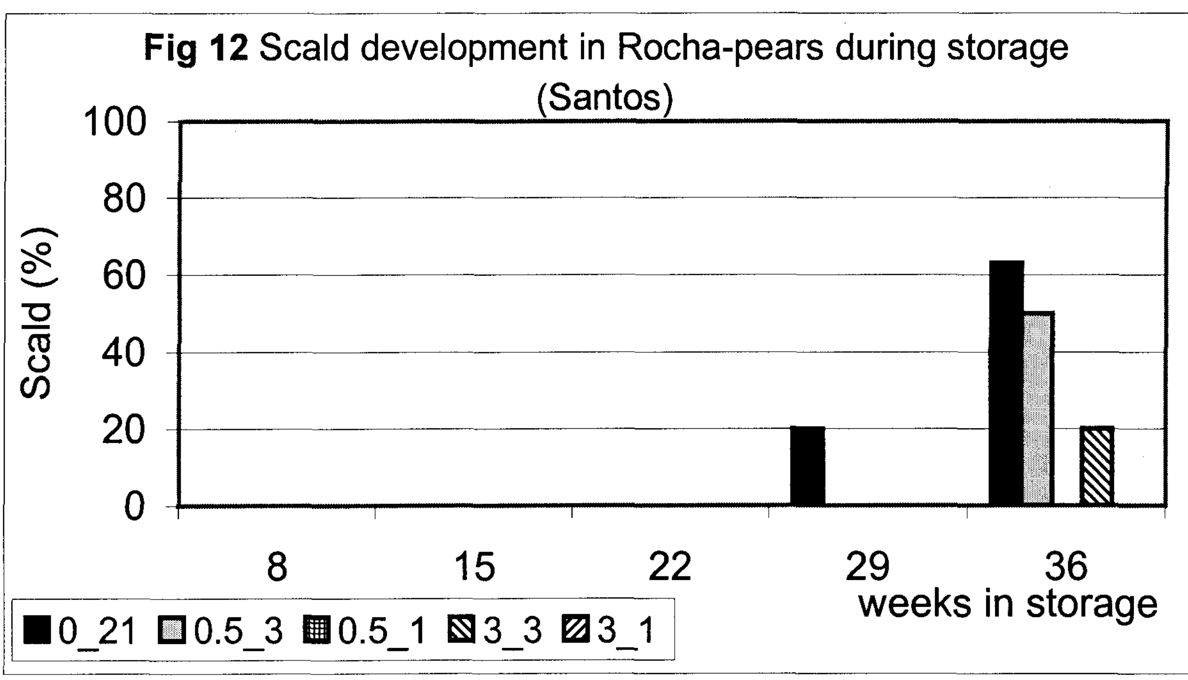


Fig 12 Scald development in Rocha-pears during storage (Santos)
Scald (%)
100
80
60
40
20
0
8
15
22
29
36
weeks in storage
0_21
0.5_3
0.5_1
3_3
3_1

Fig. 13 Scald development in Rocha-pears during shelflife (Picarra)

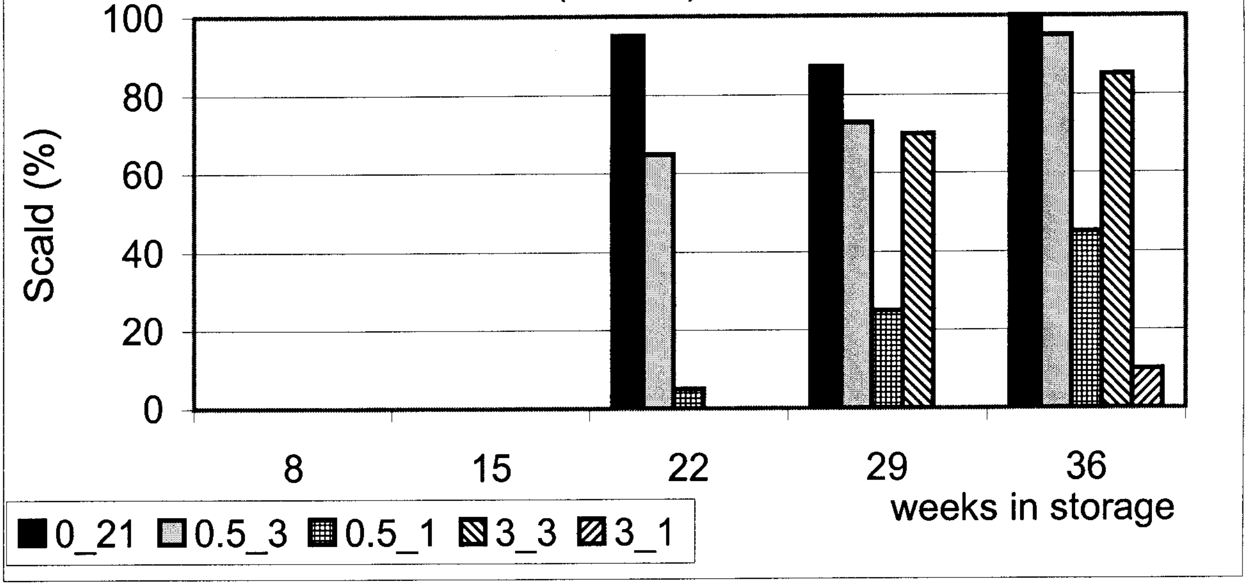


Fig. 14 Scald development in Rocha-pears during shelflife (Santos)

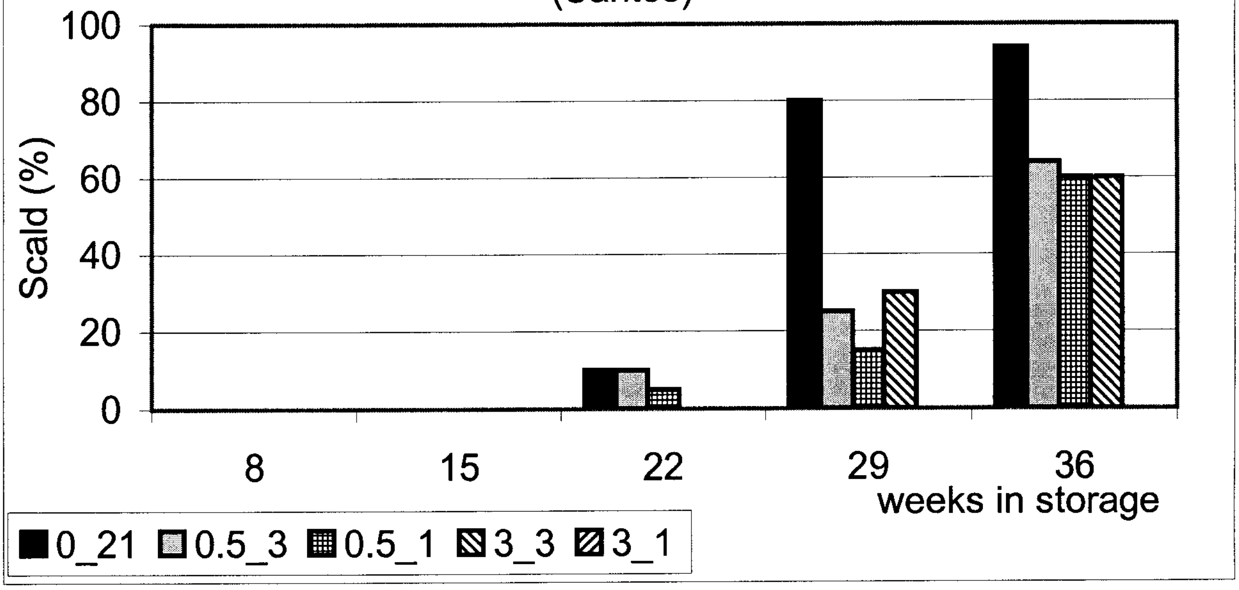


Fig. 15 Scald and brown development in Rocha-pears during storage in 0.5% CO2 and 3% O2 (Picarra)

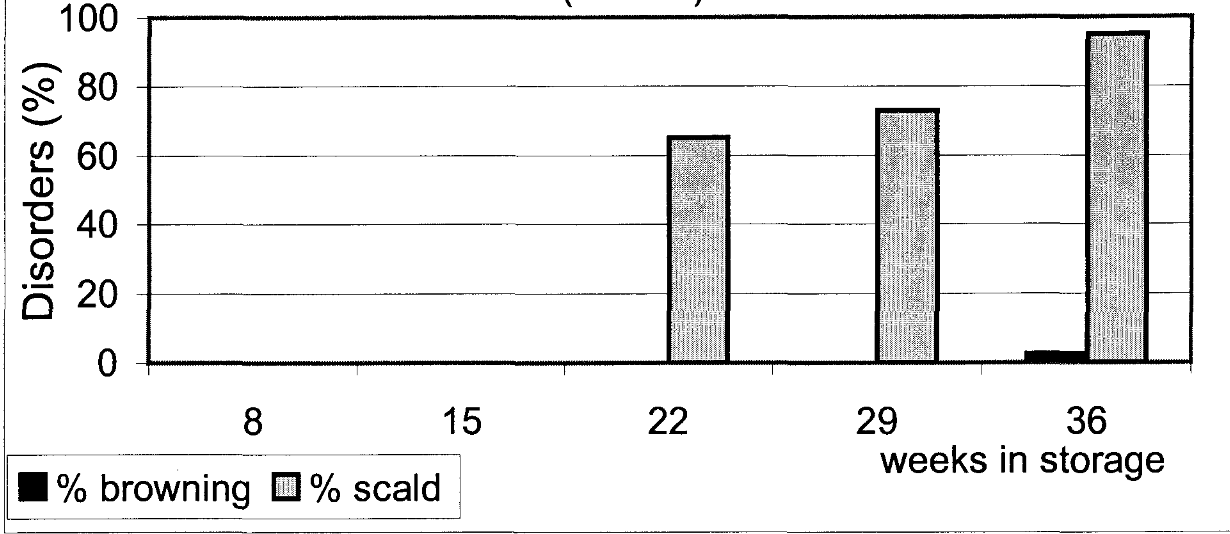
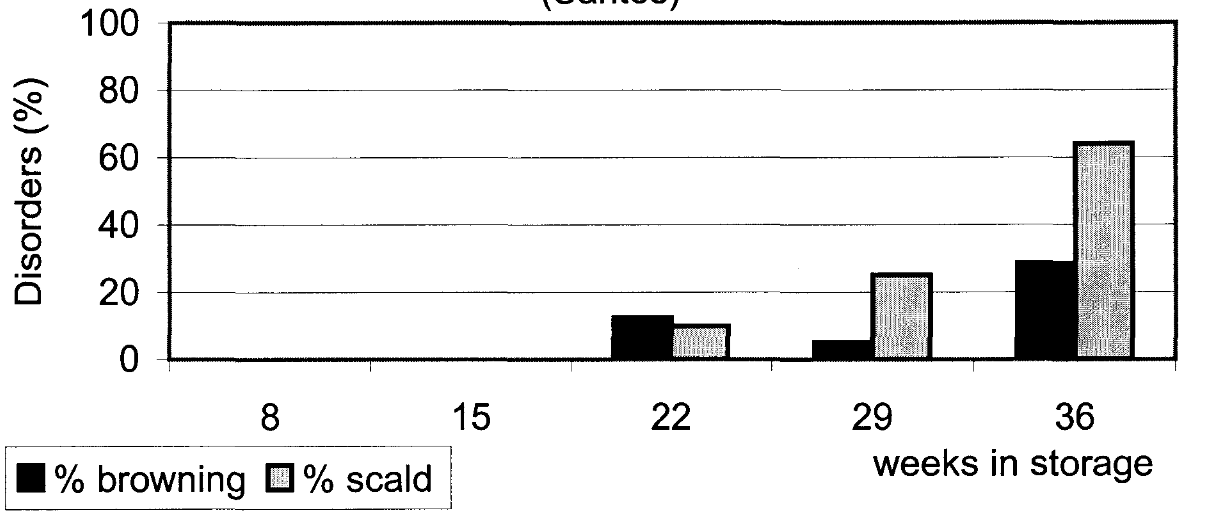


Fig. 16 Scald and brown development in Rocha-pears during storage in 0.5% CO2 and 3% O2 (Santos)



3.6 Vitamin C measurements

From the research with Conference-pears is known that especially during the period before browning and cavities are developed in the pears, the vitamin C content is a probable indicator for the disorders.
In this period only the gas conditions have an influence on vitamin C. When disorders occur in the pears, ultrastructural changes developed, resulting in different biochemical alterations. We can expect this process has a specific influence on natural antioxidants like vitamin C. So the initial measurements and after 8, 15 and 22 weeks should be taken in account. Therefore the period untill the first symptoms occur, are visualized (fig. 17-19). The initial values measured before storage in September were:

- Picarra 5.06
- Santos 7.84

It is interesting that pears stored in 0% CO_2 - 21 % O_2 and 0.5% CO_2 -3 % O_2 showed an increase in vitamin C content after 8 and 15 weeks. The other CA-conditions showed gradually a decline of vitamin C content, compared to the initial conditions.
Interesting is to note that pears stored in these conditions showed no internal browning during this first 22 weeks in storage. In Rocha-pears the same influence of the storage conditions on vitamin C content is found. According as the CO_2 concentration in store is higher and the O_2 level lower, the amount of ascorbate in the fruits lowered. Similar to Conference-pears, specific in this conditions browning and cavities developed first, as showed in the graphs. This lead to the idea that ascorbate can be seen as a parameter for browncore especially for Rocha-pears.
However also a contradiction is found. The initial value of vitamin C in pears from origin Picarra is significant higher than from origin Santos. But the pears from orchard Picarra showed a slight higher sensitivity for browncore. Therefore it seems to be impossible to give a limit for practice now, at which level of ascorbate browncore developed.

The measurements during the later judgement showed in generally for each storage condition, significant lower amounts of vitamin C. Difference between storage conditions disappeared, also the difference between the orchards.

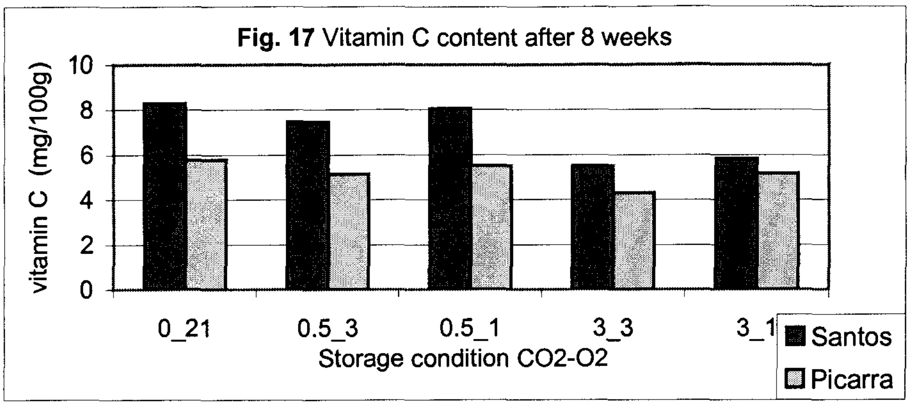


Fig. 17 Vitamin C content after 8 weeks
vitamin C (mg/100g)
10
8
6
4
2
0
0_21
0.5_3
0.5_1
3_3
3_1
Storage condition CO2-O2
Santos
Picarra

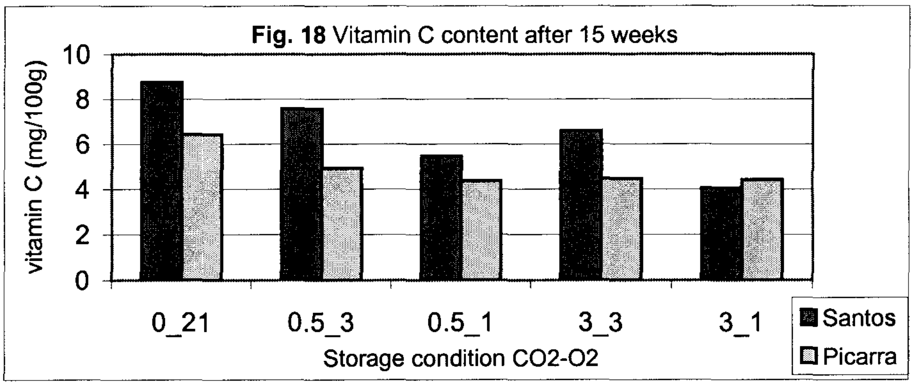


Fig. 18 Vitamin C content after 15 weeks
vitamin C (mg/100g)
10
8
6
4
2
0
0_21
0.5_3
0.5_1
3_3
3_1
Storage condition CO2-O2
Santos
Picarra

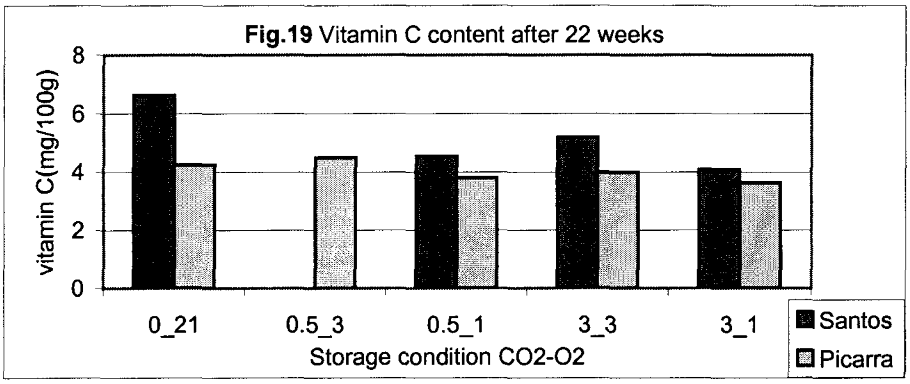


Fig.19 Vitamin C content after 22 weeks
vitamin C(mg/100g)
8
6
4
2
0
0_21
0.5_3
0.5_1
3_3
3_1
Storage condition CO2-O2
Santos
Picarra

3.7 Results gasexchange measurements

The experiment from May 1998 was limited to a few gas conditions only (table 2), since not enough good pears were available. When gas exchange rates from experiment 1 and 2 are compared, no big differences in gas exchange is observed. This is shown in both Figure 1 as in Table 1. Measurements on Conference pears, where gas exchange rates were measured 5 times between September 1997 and May 1998, also showed no changes throughout the storage season. Therefore the conclusion is drawn that the occurence of disorders cannot be related to changes in gas exchange rates during storage.

When modelling results of Rocha and Conference pears are compared (table 3), lower Km values are found for Rocha pears. This means that O_2 uptake of Rocha pears is reduced at lower O_2 concentrations than Conference pears, and also fermentative CO_2 production is increased at lower O_2 concentrations than Conference pears.

Diffusion resistance values are varying considerably between individual pears (table 4). Such a variation was not found for gas exchange rates. A difference in diffusion resistance will lead to different internal gas conditions. These internal gas conditions were calculated. Calculations were based on individual diffusion resistance values, an external gas composition of 1.84 kPa O_2 and 0.13 kPa CO_2 and gas exchange rates of 12.14 nmol/kg.s O_2 and 13.34 nmol/kg.s CO_2 (average values of measurements at a gas composition close to the storage conditions). It was found that internal O_2 pressure can vary between 1.1 and 1.7 kPa, where internal CO_2 pressure can vary between 0.3 and 1.2 kPa (Figure 2), at equal external gas conditions (1.84 kPa O_2 and 0.13 kPa CO_2). The variation in diffusion resistance is considerably higher than found for Conference pears. The main conclusion is that the first step in preventing disorders within a batch of pears is to reduce variation within the batch. This can be achieved by making a distinction between pears from different:

- harvest dates;
- orchards;
- location on the tree (for Conference it was found that pears from the top of the tree are more susceptible for the disorder);
- handling procedures between harvest and storage.

Because data are generated from one year, no conclusion can yet be drawn on the relation between the physiological parameters measured and the risk for the disorder. A repetition in another season is therefore necessary.

Table 1. Overview of the statistical results of fitting the gas exchange models to the data, using Genstat nonlinear regression analysis (Fit = fitted value, se = standard error, R^2 = percentage of explained variance)
Vm_{O2} = the maximum O_2 uptake rate (nmol/kg.s),Km_{O2} = is the Michaelis constant for the influence of O_2 on O_2 uptake (kPa), Km_{CO2} = is the Michaelis constant for the influence of CO_2 on the O_2 uptake (kPa), RQ_{ox} is the RQ value for oxidative processes, Vmf_{CO2} is the maximum fermentative CO_2 production rate (nmol.kg.s) and Kmf_{O2} the Michaelis constant for the inhibition of fermentative CO_2 production by O_2 (kPa).

	13 October 1997		2 February 1998	
	Fit	std	Fit	Std
R^2	86.7	2.0	89.0	1.7
Vm_{O2}	18.5	1.1	16.1	0.92
Km_{O2}	0.40	0.09	0.47	0.11
Km_{CO2}	19.0	6.6	9.11	2.16
R^2	74.5	1.6	75.8	1.4
Vmf_{CO2}	22.5	8.0	16.6	5.3
Kmf_{O2}	0.037	0.024	0.052	0.032
Rqox	0.965	0.030	0.954	0.034

Table 2. Results gas exchange measurements in May 1997.

O_2 (kPa)	CO_2 (kPa)	O2 uptake (nmol/kg.s)	CO2 production (nmol/kg.s)
0.03	0.06	0.35	9.52
20.73	5.23	14.42	11.12
0.03	0.06	0.41	7.98
20.73	5.23	12.65	6.33

Table 3. Comparison of modelling results of Rocha and Conference pears harvested in 1997.

	Rocha	Conference
Vm_{O2}	17.3	19.3
Km_{O2}	0.44	0.78
Km_{CO2}	14.1	12.3
Vmf_{CO2}	19.6	19.0
Kmf_{O2}	0.045	0.142
Rqox	0.961	0.87

Table 4. Overview of diffusion resistance values for O2, calculated from neon using Graham's law (Avg: average value, Std: standard deviation, Min: minimum value, Max: maximum value).

	Avg	Std	Min	Max
October 1997	1549	451	710	2610
February 1998	1438	295	951	2030
May 1998	2048	245	1798	2381

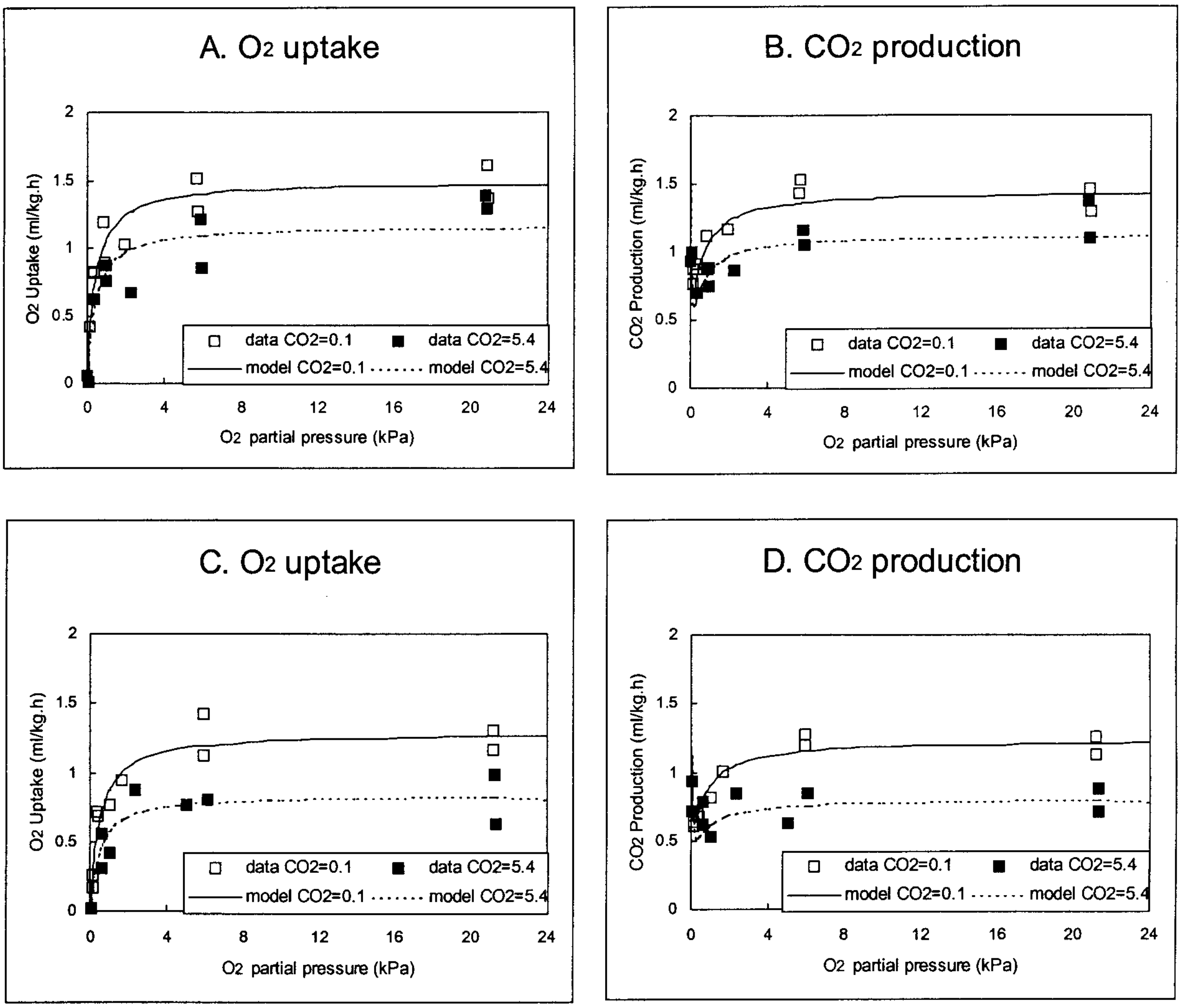


Figure 1. Gas exchange characteristics of Rocha pears. A: O_2 uptake in October 1997, B: CO_2 production in October 1997, C: O_2 uptake in February 1998, D: CO_2 production in February 1998.

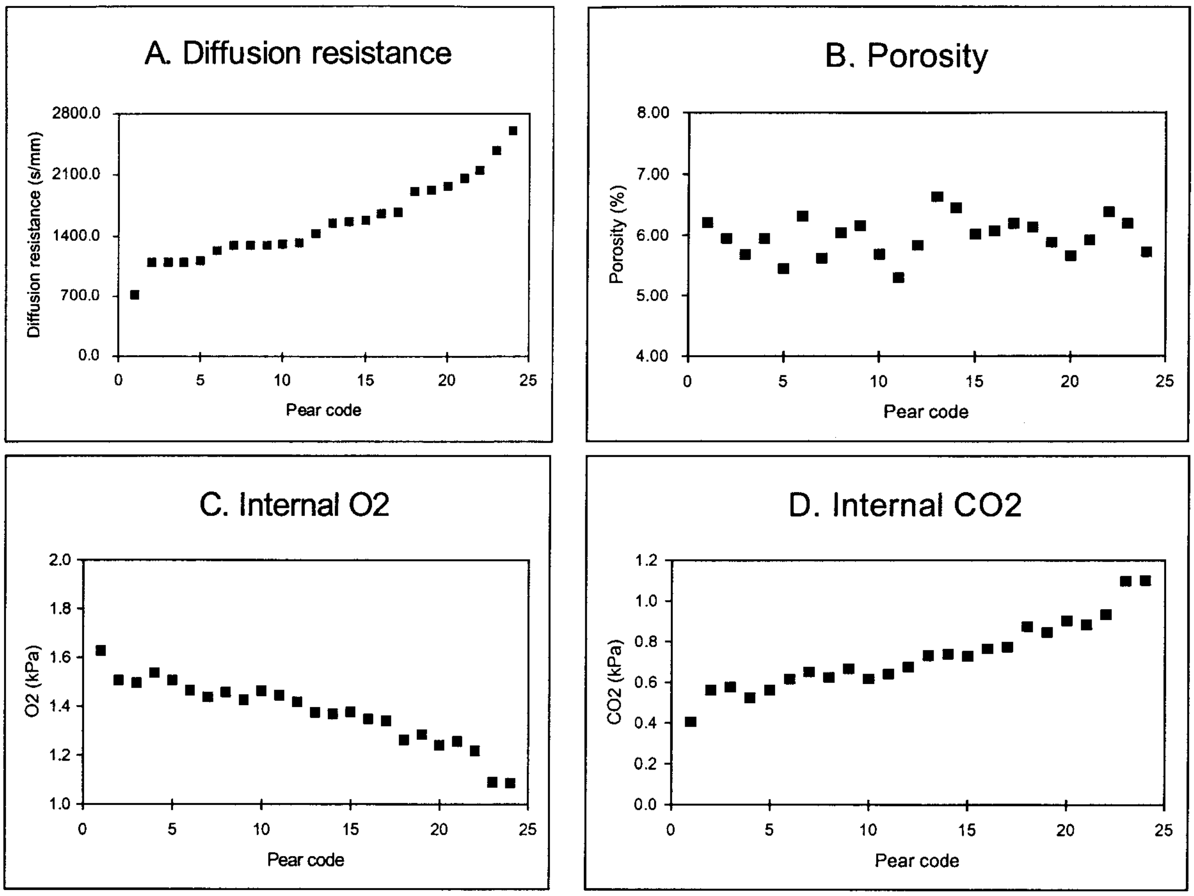


Figure 2. Gas diffusion characteristics of Rocha pears as measured in October 1997. Pears are sorted on diffusion resistance values. The pear codes in every graph are always related to the same pears. Graphs indicate variation between pears and the relation between diffusion resistance and internal gas conditions.
A: Diffusion resistance for O_2, B: Porosity, C: Calculated internal O_2, D: Calculated internal CO_2. Calculations in C and D are based on individual diffusion resistance values, an external gas composition of 1.84 kPa O_2 and 0.13 kPa CO_2 and gas exchange rates of 12.14 nmol/kg.s O_2 and 13.34 nmol/kg.s CO_2 (average values of measurements at a gas composition close to the storage conditions).

4.0 Discussion

The observations in the period of September until June on storage and quality research on Rocha pears give important indications on the behaviour of pears in various storage conditions. The aim of the research is to select a storage regime in which no internal abnormalities occur and the quality of the pears is maintained for a long period of time. With quality is meant the flavour quality, firmness, colour and the sensitivity to ex- and (especially) internal storage disorders.
With specific measurements it is the objective to understand more of the backgrounds of the processes that lead to internal abnormalities as well as the influence on quality and taste attributes. An important result was that during the 5 different judgments until June 1998, in the different storage conditions a range of internal abnormalities occurred in pears of both orchards. This concerned hollow pears as well as brown pears. Because in this research a so called square schedule was chosen, the influence of CO_2 as well as O_2 can be judged separately as well as in coherence.
It is clear that a higher CO_2 content and a lower O_2 content increase the development of internal browning and the formation of cavities. However, the combination of 3% CO_2 and 1% CO_2 leads to much more abnormalities as a result of the specific interaction between O_2 and CO_2. In the most mild CA-combination 0.5% CO_2 and 3% CO_2) only a slight development of cavities and browning occurred. Surprisingly pears stored in normal cooling showed after 22 weeks in storage a certain amount of browncore development which was clearly more than in the mildest CA-condition of 0.5% CO_2 and 3% CO_2. In the other CA-combinations with either 1% O_2 or 3% CO_2 the occurrence of browncore was unacceptable be seen from marketing viewpoint.
These results lead to the conclusion that the CO_2 level should not exceed 1% and the O_2 level should not be below 3%. Certainly when the results of the research in Alcobaca can be involved in this, where other conditions have been applied and also the influence of the harvesting time, better conclusions can be drawn about the optimum CA storage conditions.

Furthermore, it is possible to make the conclusion that in CA-storage the conditions that cause the most cavities and browning are the best guarantee for colour and firmness retention during storage and in particular the shelflife period as a simulation of the distributionchain. It is remarkable that the influence of the low oxygen content to colour preservation is very strong whereas a high CO_2-content hardly has an influence the yellow colouring. To maintain a green ground colour a low oxygen content seems very important.
With the selected CA-condition (0.5% CO_2 and 3% CO_2) still an advantage is noticable compared to normal cooling. Pears in this CA-condition are slight greener and showed a lower firmness than pears from normal cooling. Specific pears from this last condition showed continuously a higher firmness than CA-stored pears. If this is a result of the earlier mentioned lack of ripening capacity is not sure, it was not proved by the measurements on extractable juice.
Furthermore it is impossible to select a CA-storage condition that prevents the fruits from scald and showed no internal disorders, which is not easy. Therefore application of DPA seems to be necessary in future.

Important is the question to understand the process of internal disorders in Rocha-pears. Therefore the specific measurements like gasexchange and vitamin C content are introduced. This with the objective to find one or more parameters prediction for prediction of the disorder.
That the respiration rate below 4% O_2 showed a sharp decrease has the consequence that storage above 4-5% O_2 will not have much effect on other quality parameters. So a safe an effectual CA-storage regime can only be found in "a small O_2 window". A futher important result was the big variation in diffusion characteristics between individual pears in one batch. It can be expected that pears with a higher diffusion limitation are more sensitive to the disorder. This variation in a batch should have a reasons could related to picking time, orchards, within tree variation and handling procedures. Partly next research at Alcobaca and ATO can probably give indications for this.

At second the relation with vitamin C is hopeful as a possible parameter for the disorder. It is obvious that the more extreme the storage conditions the more the vitamin C content drops. Especially at the combination 1% O_2 and 3% CO_2 the vitamin C content is low. This might indicate that the vitamin C content may give an indication to the occurrence of browning. It is possible that this measurement has a predictive value. It is clear yet, that this is influenced by CO_2 and O_2.
At Conference pears vitamin C showed to be of great importance in resisting abnormalities. It appears that this also applies to Rocha pears. A follow-up conclusion could be that the processes of cavity formation and browning in Conference and Rocha pears have the same background.
To measure the initial vitamin C level as an indicator for brown susceptibility is discutable whereas the orchard with a higher initial level showed a little bit more brown occurrence. However more measurements should be carried out in relation with picking date and orchards.

5.0 Conclusions

- **In general for Rocha-pears the higher the CO_2 content and the lower the oxygen concentration in CA-storage the more internal disorders could expected.**
- **In contradiction these effects are useful to prevent scald and to maintain ground colour.**
- **The CA-condition *0.5% CO_2 and 3% O_2* seems to be a good alternative for practical application.**
- **This condition showed less disorders than normal cooling and maintained during storage the normal taste and quality attributes for Rocha pears.**
- **Vitamine C is a good parameter for internal browning but prediction based on initial levels should be studied in advance.**
- **Rocha pears within one batch showed a big difference in diffusion capacity with the consequence of different susceptibility per pear for internal damage.**
- **The respiration rate of Rocha-pears could be compared to Conference-pears, so within a range of 1-1.5 ml/kg/h.**
- **The respiration is equal during the storage season, below 4% O_2 the respiration showed a sharp decrease. The consequence is very small effective window for practical application.**

6.0 Literature cited

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