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LONG TERM STORAGE OF 'ROCHA' PEARS AT LOW TEMPERATURE

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ABSTRACT

Changes in chlorophyll concentration, hardness and internal atmosphere concentration of 'Rocha' pears stored under three temperatures were followed in store. All these parameters showed a differential response to temperature.

Rocha pears were successfully stored for 2 ½ months at 4.4 ± 0.5° C; 6 months at 0.4 ± 0.4° C and 8 months at − 0.7 ± ± 0.4° C.

INTRODUCTION

ETHYLENE biosynthesis plays a very important role in postharvest physiology of fruits because it occurs at temperatures used for storage. Consequently the transition to senescence can begin in store after prolonged exposures to these temperatures.

Previous work with 'Rocha' pears shows that there is an interval between the climacteric in store and the end of storage life (Torres & Rhodes, 1973). During this interval the fruits are still commercially acceptable.

In this work we followed the chlorophyll loss of 'Rocha' pears stored at three different temperatures in an attempt to correlate the readily apparent phenomena associated with ripening with the physiological age of the fruit.

MATERIAL

On the 6th September 1971 'Rocha' pears from one orchard in Alcobaça (Portugal) were picked, immediately transported to Estação Agronómica Nacional, Departamento de Fisiologia Vegetal, Oeiras, and stored in three different cold rooms. The

temperatures of the cold rooms (1) were regularly recorded and in Table I are the results of these observations. However due to a refrigeration failure which occurred in December (40th to 46th day in store) the fruit experienced temperatures up to 15.5° C. in C2; 15.3° C. in C3 and 14.1° C. in C4.

TABLE I

Temperatures (C. degrees) of the cold rooms
Temperaturas (em graus centígrados) das câmaras frigoríficas em que os frutos foram armazenados

Cold room *Câmara frigorífica*	Mean temperature *Temperatura média*	Standard deviation *Desvio-padrão*	Range *Amplitude de variação*
C2	4.4	0.5	3.7 to 6.5
C3	0.4	0.4	- 0.3 to 1.2
C4	- 0.7	0.4	- 1.6 to 1.0

METHODS

The analysis of chlorophyll was based on the Laval-Martin (1969) method. At regular intervals five fruits were taken from the cold rooms, peeled and two samples of 12 discs (7.76 mm diameter) were cut from each fruit, and the samples transferred to ice cold mortars. The discs were homogenized in the dark with 80 % acetone (v/v) at 4° C. in the presence of sand and calcium carbonate. The homogenate was centrifuged at 15,000 × g for 10 minutes at 0° C. The supernatant volume was measured and the optical density values at 665 and 645 nm were determined in a Unicam SP. 800 spectrophotometer. The quantities of chlorophyll *a* and *b* were calculated by the method of Vernon (1960) and the results expressed as μg chlorophyll/100 cm^2.

Pressure tests were made in the equatorial region of peeled fruits (3 measurements) using a penetrometer.

Measurements of CO_2 in the internal atmosphere of the fruit were made in a Grubb-Parsons SB2 infra-red gas analyser using methods previously described (Reid, Rhodes & Hulme, 1973).

(1) In this paper we shall refer to these rooms as C2, C3 and C4.

The curves in the graphs were fitted by the programs INTPLT and LOGINT written by D. H. Fremlin for the University of Essex Computing Centre, England.

RESULTS

1. *Chlorophyll loss*

Fruit stored under the highest temperature conditions, in C2, ripened very quickly and by the end of the second month in store had lost all its chlorophyll. Pears in other rooms took much longer to develop signs of ripening. Figure 1 shows that total chlorophyll loss occurred more rapidly in C3 than in C4, and the lowest level was reached 75 days earlier. In both cases chlorophyll *a* decreased faster than chlorophyll *b*. The quotient chlorophyll *a*/chlorophyll *b* decreased very rapidly towards the end (Fig. 3) and faster in pears stored at higher average temperature (C3).

If we record the measurements of chlorophyll present on a logarithmic scale (Fig. 2), the curves obtained suggest that the decay is not exponential throughout the period. It may be that there is an approximately exponential decay during the first period in store, i. e. up to the third month in C3 and until the end of the fifth month in C4.

2. *Hardness and internal atmospheres*

By the end of the second month in store pears in C2 had hardness of 4 units and 2.5 % CO_2 in their internal atmosphere. They had ripened to some extent in store and fully ripened after a period of 7 days at 20° C. Later on the peel started showing brown patches (Plate I, 3) and most of the fruit on transfer to 20° C. had an unpleasant taste, brown core and no juice. In the third month the loss in store due either to senescence or fungi attack was 38 % and it increased to 80 % during the 4th month.

Pears stored in C3 and C4 developed signs of ripening more slowly but fruit in C3 was always in a more advanced physiological state than that in C4 (see Figs. 4 and 5 and Plates).

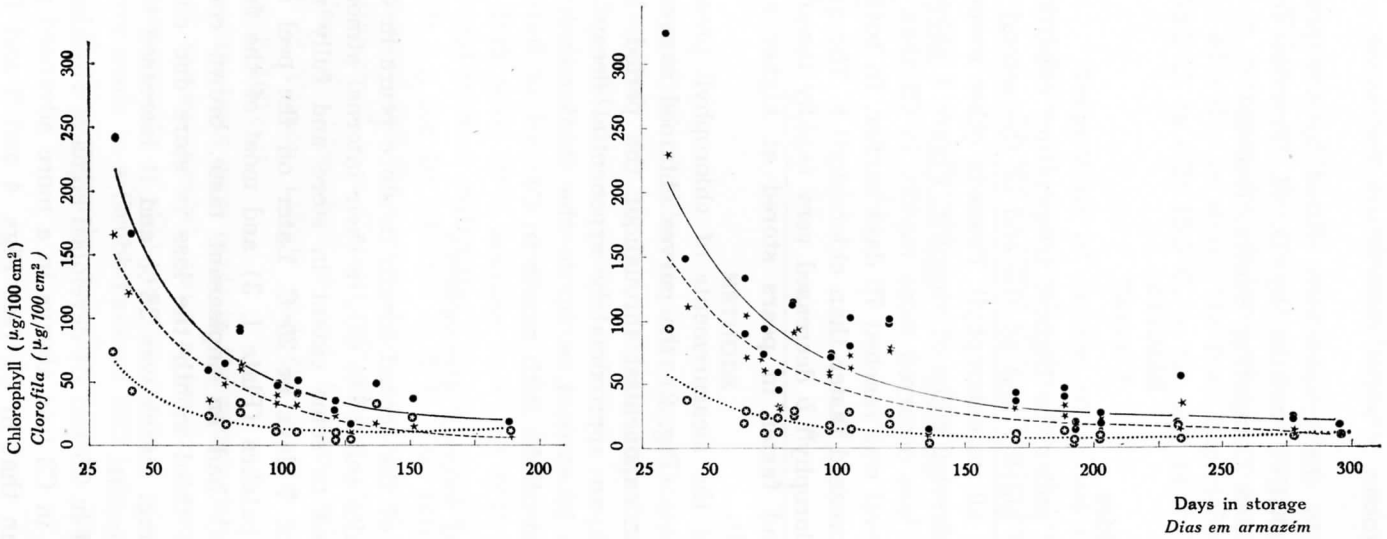

Fig. 1—Chlorophyll *a* (★---★), chlorophyll *b* (✪···✪) and total chlorophyll (●—●) concentrations in fruits in C3 (left) and in C4 (right).
Concentrações de clorofila a (★---★), *clorofila* b (✪···✪) *e clorofila total* (●—●) *em frutos conservados em C3 (à esquerda) e em C4 (à direita).*

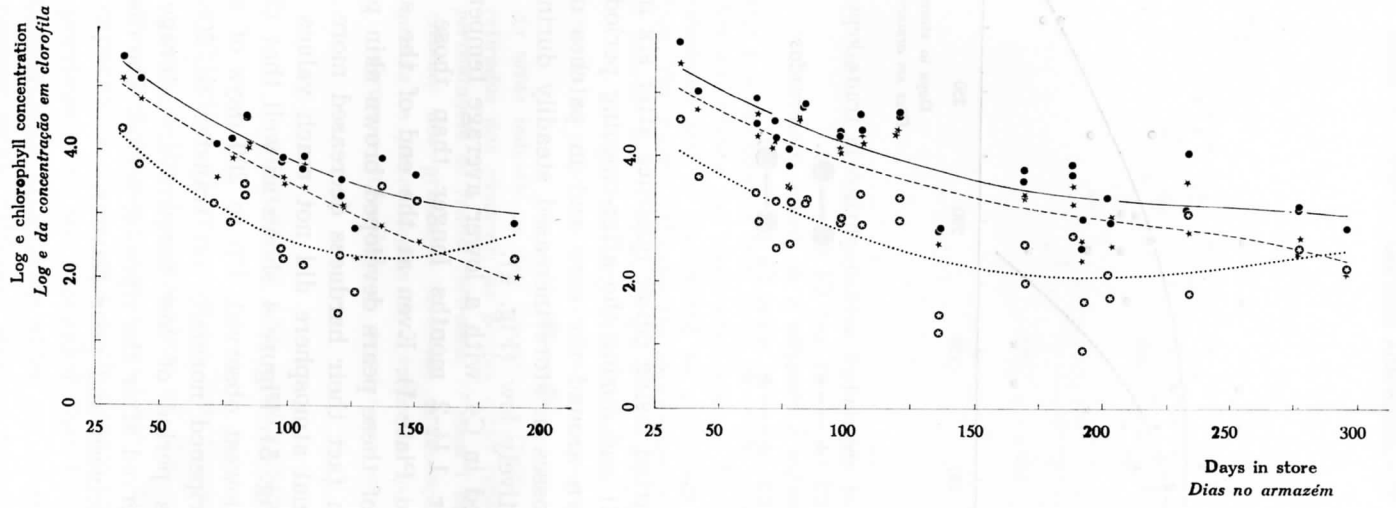

Fig. 2—Chlorophyll *a* (★---★), chlorophyll *b* (✪•••✪) and total chlorophyll (●—●) plotted in a semi-logarithmic scale. C3 on the left and C4 on the right.

Representação semi-logarítmica de clorofila a (★---★), *clorofila* b (✪•••✪) *e clorofila total* (●—●) *em frutos conservados em C3 (à esquerda) e em C4 (à direita)*

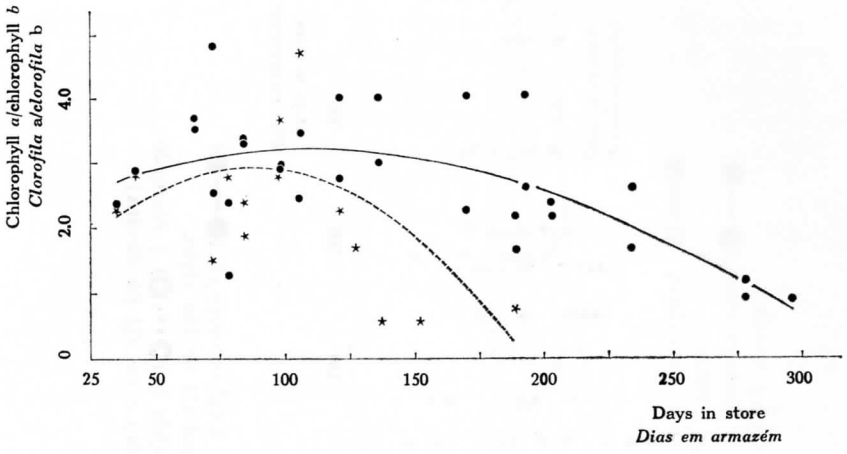

Fig. 3 — Ratio of chlorophyll *a*/chlorophyll *b* in fruits kept in C3 (★---★) and C4 (●—●).
Cociente clorofila a/*clorofila* b *de frutos conservados em C3* (★---★) *e em C4* (●—●).

Pears in C3 started losing edible qualities after six months in store (Plate I, 2) and during the after-ripening period most of them were brown around the core and in patches outside (Plate I, 1). The losses in store increased steadily during this time but were relatively low (Fig. 4).

The fruit stored in C4, with a lower average temperature, kept very well for 1 ½-2 months longer than those in C3 (Figs. 4 and 5 and Plate I). Even at the end of the storage period only a few of these pears developed brown skin patches and brown core; in fact their hardness decreased more slowly (Fig. 4) and internal atmosphere did not reach values higher than 1.3 % CO_2 (Fig. 5). Figure 4 shows as well that the loss in store were the lowest observed. Up to 200 days of storage in C4 the pears ripened normally on transfer to 20° C. for 7 days. After this period of low temperature storage some disorders were observed after the ripening period. The disorders include lack of juiciness and poor flavour.

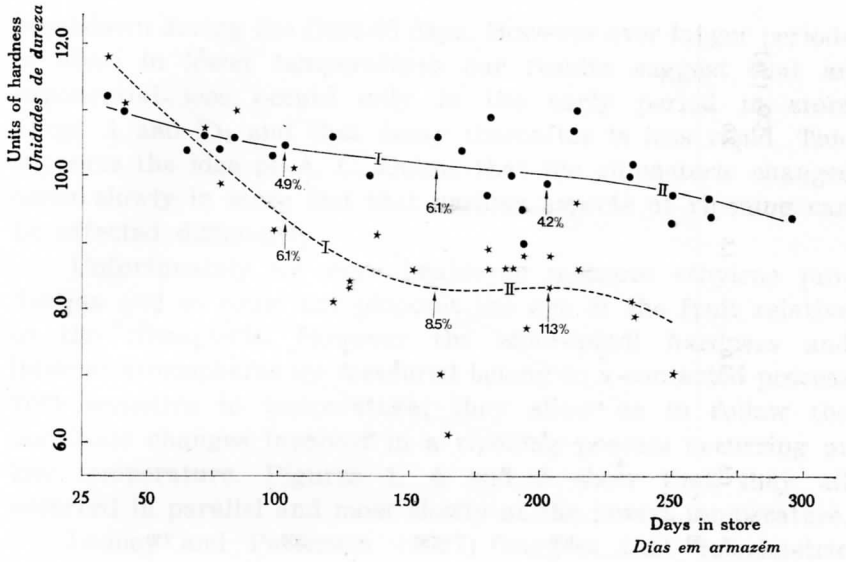

Fig. 4 — Hardness of fruit in C3 (★---★) and C4 (●—●). Arrows indicate the days when loss of fruit in store was calculated and is represented as percentage of unhealthy fruit. The mark I indicates the appearance of the first signs of disorder and II indicates when the fruit lost the capacity to ripen normally after transfer to 20° C.

Dureza da polpa dos frutos conservados em C3 (★---★) *e C4* (●—●).

As setas indicam os dias em que os frutos podres foram retirados das câmaras, as perdas estão representadas em percentagem de frutos retirados. O sinal I *indica o aparecimento dos primeiros sintomas de doença e* II *indica quando os frutos depois de transferidos para 20° C. perderam a capacidade de amadurecer normalmente.*

DISCUSSION AND CONCLUSION

The objective of research on low temperature storage of 'Rocha' pears is the determination of the conditions that will give the longest storage life compatible with the maintenance of the capacity to ripen to a high quality pear.

In previous work we concluded that fruit stored at −1° C. were commercially acceptable after up to 6 months in store. In the present work, different storage temperatures were used

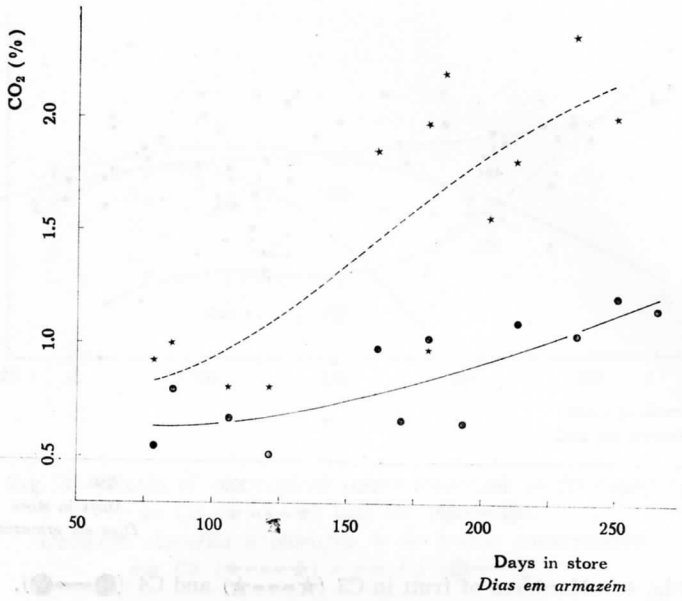

Fig. 5 — Internal atmosphere concentration of fruits in C3 (★---★) and C4 (●—●).
Concentração da atmosfera interna de frutos em C3 (★---★) *e C4* (●—●).

to re-examine that conclusion. The results were that in the highest temperature room — 4.8 ± 1.9° C. — fruits lasted only 2 ½ months, whereas at 0.6 ± 1.7° C. 6 months and at − 0.4 ± ± 2.0° C. 8 months of storage was achieved (see Table I).

Chlorophyll content decreased during storage life showing a differential response to temperature (Figs. 1 and 2). In fact chlorophyll loss during ripening has been extensively studied and Hansen (1955) found the same with 'Anjou' and 'Bosc' pears, and Laval-Martin (1969) with the 'Passe-Crassane' variety. It is known that in fruits ripening is accompanied by increased protein synthesis (Hulme, 1937), unlike the phenomena observed in abscised leaves that is accompanied by a rapid loss of proteins.

The enzyme chlorophyllase is capable of hydrolysing both chlorophylls *in vitro* and its activity is known to increase during the climacteric phase (Looney & Patterson, 1967, and Rhodes & Wooltorton, 1967). Laval-Martin (1969) found that pears in temperatures above 7° C. had an exponential chlorophyll

breakdown during the first 40 days. However over longer periods in store in lower temperatures our results suggest that an exponential loss occurs only in the early period in store (Figs. 1 and 2), and that decay thereafter is less rapid. This supports the idea of A. C. Hulme that the climacteric changes occur slowly in store and that various aspects of ripening can be affected differently.

Unfortunately we were unable to measure ethylene production and so could not pinpoint the age of the fruit relative to the climacteric. However the chlorophyll hardness and internal atmospheres we measured belong to a concerted process very sensitive to temperature; they allow us to follow the metabolic changes involved in a ripening process occurring at low temperature. Figures 1, 4 and 5 show that they all occurred in parallel and most slowly at the lowest temperature.

Looney and Patterson (1967) suggest that colorimetric scanning devices may be useful to determine fruit age; we consider that this is also appropriate with 'Rocha' pears, as the fruit is green when picked and gradually changes to yellow in any of the temperature conditions of this experiment (Plates I, 1 & 2).

We observed that when the fruit is yellow in store (lowest level of chlorophyll) the after ripening period is a much shorter and more critical period as most of the fruits will develop brown core within 7 days (Plate I, 1). The internal atmosphere concentration of CO_2, which increased to values 50 to 60 times higher than the outside atmospheric CO_2 concentration (Fig. 5) after longer periods of storage, may reflect the cellular disorganization occurring during ripening and therefore limiting diffusion. There is evidence that breakdown in pears is associated with disruption of the lamella of the chloroplasts (Bain & Mercer, 1964). Softening may be the easiest parameter to use in commercial stores to assess the age of fruit (Figure 4), together of course with the visual appearance of the fruit.

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RESUMO

Estudou-se a variação da clorofila, dureza e atmosfera interna em câmara frigorífica, de peras 'Rocha' mantidas a três temperaturas. Estes parâmetros variaram de um modo diferencial em relação à temperatura.

As peras 'Rocha' foram conservadas em boas condições durante 2 ½ meses a $4{,}4 \pm 0{,}5°$ C.; 6 meses a $0{,}4 \pm 0{,}4°$ C. e 8 meses a $-0{,}7 \pm 0{,}4°$ C.

REFERENCES

Bain, J. M. & Mercer, F. V.
1964 Organization resistance and the respiration climacteric. *Aust. J. biol. Sci.* **17**: 78-85.

Hansen, E.
1955 Factors affecting post-harvest color development in pears. *Proc. Am. Soc. hort. Sci.* **66**: 118-124.

Hulme, A. C.
1937 The metabolism of nitrogen in apple-fruits. *Rep. Fd Invest. Bd* **1936**: 126-131.

Laval-Martin, Danielle
1969 Variation de la teneur en chlorophylles au cours de la maturation et en function de la temperature chez la poire Passe-Crassane. *Physiol. vég. Paris,* **7** (3): 251-259.

Looney, N. E. & Patterson, M. E.
1967 Chlorophyllase activity in apples and bananas during the climacteric phase. *Nature, Lond.* **214**: 1245-1246.

Reid, M. S., Rhodes, M. J. C. & Hulme, A. C.
1973 Changes in C_2H_4 and CO_2 during the ripening of apples. *J. Sci Fd Agric.* (in press).

Rhodes, M. J. C. & Wooltorton, L. S. C.
1967 The respiration climacteric in apple fruits. The action of hydrolytic enzymes in peel tissue during the climacteric period in fruit detached from the tree. *Phytochemistry* **6**: 1-12.

Torres, Maria A. & Rhodes, M. J. C.
1973 Studies on C_2H_4 and CO_2 production by pears during storage in air at low temperature and on subsequent transfer to 20° C. *Agronomia lusit.* **34**: 347-359.

Vernon, L. P.
1960 Spectrophotometric determination of chlorophylls and pheophytins in plant extracts. *Analyt. Chem.* **32**: 1144-1150.

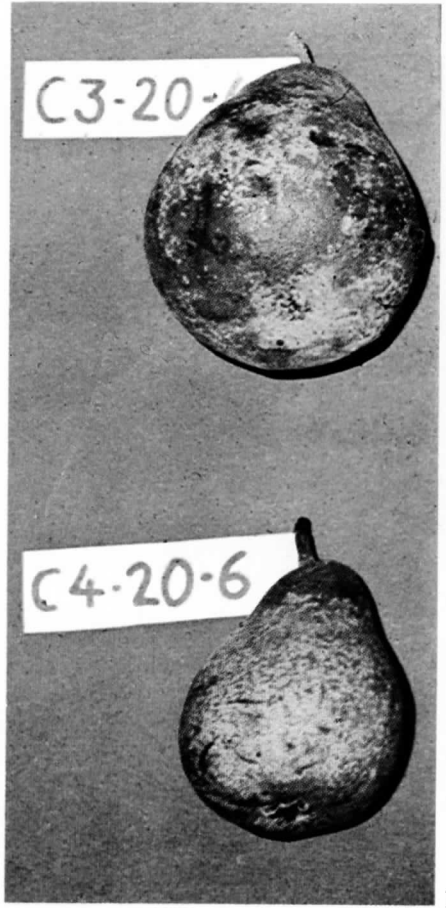
LEGEND TO PLATE I

1 — Fruits were taken from cold rooms and spent 7 days at 20° C. (1-5-1972). Note the increased incidence of brown areas on the skin in pears from C3.
Frutos que passaram 7 dias a 20° C. (1-5-1972). Notar o aumento de manchas castanhas na casca em peras provenientes de C3.

2 — Fruit during the 7th month in store (24-4-1972); whole fruit from C4 still very sound but C3 shows brown patches.
Frutos durante o 7.° mês em câmara frigorífica (24-4-1972); fruto são proveniente de C4, e fruto com manchas castanhas proveniente de C3.

3 — Pears from cold rooms during the 4th month in store (19-1-1972); notice the brown patches on the fruit from the highest temperature conditions.
Peras retiradas directamente das câmaras frigoríficas no 4.° mês (19-1-1972); note as manchas castanhas na casca do fruto proveniente da câmara a temperatura mais elevada.

PLATE I



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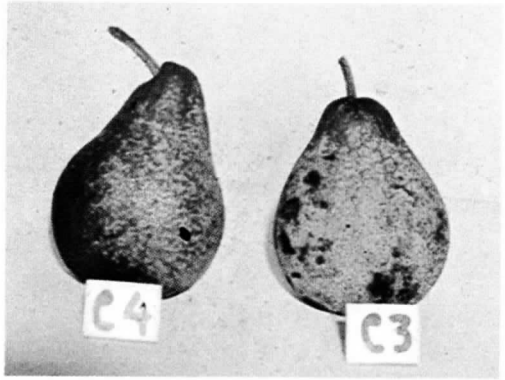

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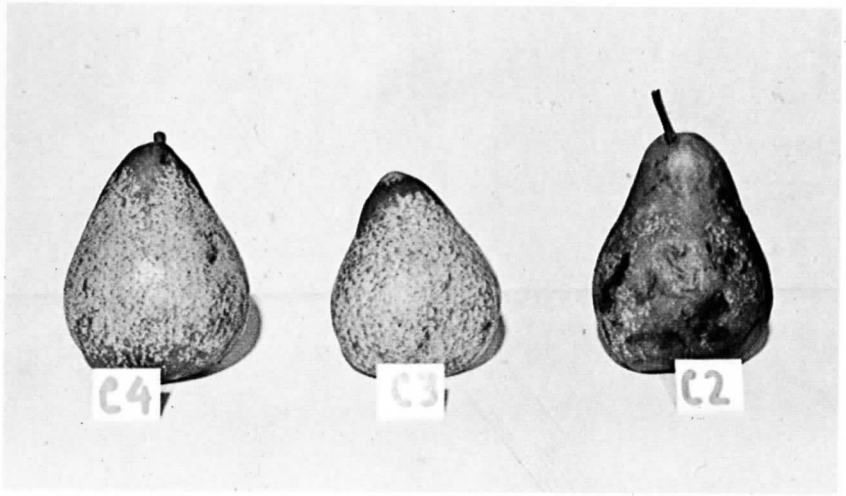

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