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STUDIES ON ETHYLENE AND CARBON DIOXIDE PRODUCTION BY PEARS DURING STORAGE IN AIR AT LOW TEMPERATURE AND ON SUBSEQUENT TRANSFER TO + 20° C.

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ABSTRACT

Changes in ethylene and CO_2 production were followed during storage and 'after ripening' of a variety of pears, 'Rocha', widely grown in Portugal. The ethylene production reached a maximum after five months storage at + 1° but on transfer to + 20° C. after an initial increase, the rate of production always decreased. Experiments were done with 'Rocha' and 'Conference' pears in order to follow changes in ethylene and CO_2 production during the first few hours following transfer to + 20°. The increase in respiration paralleled the increase in temperature very closely but ethylene production rose to a maximum after 6-7 hours at + 20° and subsequently fell. An atmosphere deficient in O_2, depressed the ethylene production during the warming period but respiration was stimulated.

INTRODUCTION

It is now widely accepted that the olefinic gas, ethylene, play an important rôle in the post-harvest physiology of fruits and it may be considered to act as the natural ripening hormone (Burg & Burg, 1965b). The increase in ethylene biosynthesis is an early event in ripening of the climacteric class of fruits of which the pear is a member. The increase in ethylene biosynthesis to a stimulatory concentration is the event which will initiate the «transition from growth to senescence and involving an increase in respiration and leading to the ripening of the fruit» (Rhodes, 1970). Ethylene will trigger «the many biochemical reactions in the fruit which are not necessarily causally related to each other but occur more or less in parallel and in time relation to the climacteric» (Pratt & Goeschl, 1969).

It has been shown that in fruits stored in air at low temperature the endogenous concentration of ethylene increases with time (Knee, 1971; Torres, 1971). However, it is not clearly established to what extent these changes affect the behaviour of the fruit at storage temperatures (Fidler, 1960). We know that fruits undergo a climacteric rise during low temperature storage (Kidd & West, 1937; Knee, 1971; Leonard *& al.,* 1954; Torres, 1971) and that after prolonged exposure to low temperature they often fail to ripen when transferred to room temperature (Fidler & Coursey, 1970; Wilkinson, 1970).

The production of CO_2 and ethylene by pears of the variety 'Rocha' grow in Portugal was followed during storage in air at + 1° C. and throughout a subsequent period of 7 days after transfer to + 20° C. (the 'after ripening' period). Throughout the storage period studied, the pears after the subsequent warming treatment ripened normally to produce high quality ripe fruit. Detailed studies were also made of the transitory changes in ethylene and CO_2 production which occurred during the first few hours of transfer of fruit from + 1° C. to + 20° C. The implications of these results on our understanding of the physiological state of the fruit and its metabolism during storage are discussed.

MATERIAL

'Rocha' pears picked in Portugal at the end of August of 1970 were transported to England by refrigerated van and stored for several months at + 1° C. In addition, 'Conference' pears from a commercial store at — 1° C. (Norfolk Fruit Growers Ltd., Wroxham, England) were used for some of the experiments.

METHODS

At intervals, samples of 10 'Rocha' pears stored at + 1° C. were transferred to 5 l containers and kept at + 1° C. A constant flow (of between 2-8 l/h) of air free of CO_2 (removed by passing the air through a soda lime tower) and of ethylene (removed by passing the air through a solution of mercuric perchlorate — 0.25 M mercuric oxide in 2 N perchloric acid) was passed over the pears in the container. The humidity of the air stream was maintained at 95 %. After 48 hours at + 1° C. the whole system was removed to a constant temperature room at 20° C. for an «after ripening» period of 7 days. During both phases of the experiment, samples of the gas passing over the fruit were analysed for CO_2 and C_2H_4 content using «Pye 104» chromatograms with either a 2 ft silica gel column and a katharometer detector for CO_2, or a 5 ft Porapak S column and flame ionisation detector for ethylene. From this data rates of CO_2 and ethylene production were calculated.

When the internal atmosphere of the pear was to be sampled during a temperature transition, a fine plastic tube was sealed into the core of the pear so that internal atmospheric samples could be withdrawn at intervals with a syringe (Reid, Rhodes & Hulme, 1972). The changes in the fruit temperature during such a transition was folowed by introducing thermocouples into the fruit at various positions: near to the core, under the skin or in the pulp.

RESULTS

1. Changes in 'Rocha' pears during storage

Fig. 1 shows the changes in respiration and ethylene production of pears stored at + 1° C. and subsequently transferred to + 20° C. The respiration of the pears at 1° C. stayed approximately constant at about 30 ml CO_2. 10 kg^{-1} . hr^{-1} throughout the storage period while the ethylene production rose to a maximum by the fifth month in store. Fig. 1 shows that in the «after ripening» period at 20° C. the initial rise in ethylene production increase as the pears matured in store.

Fig. 2 shows that pears kept for 5 months at + 1° C. had already reached the respiration climacteric peak since at this stage treatment of the fruit at 20° C. with 100 ppm ethylene in air had no effect on the respiration.

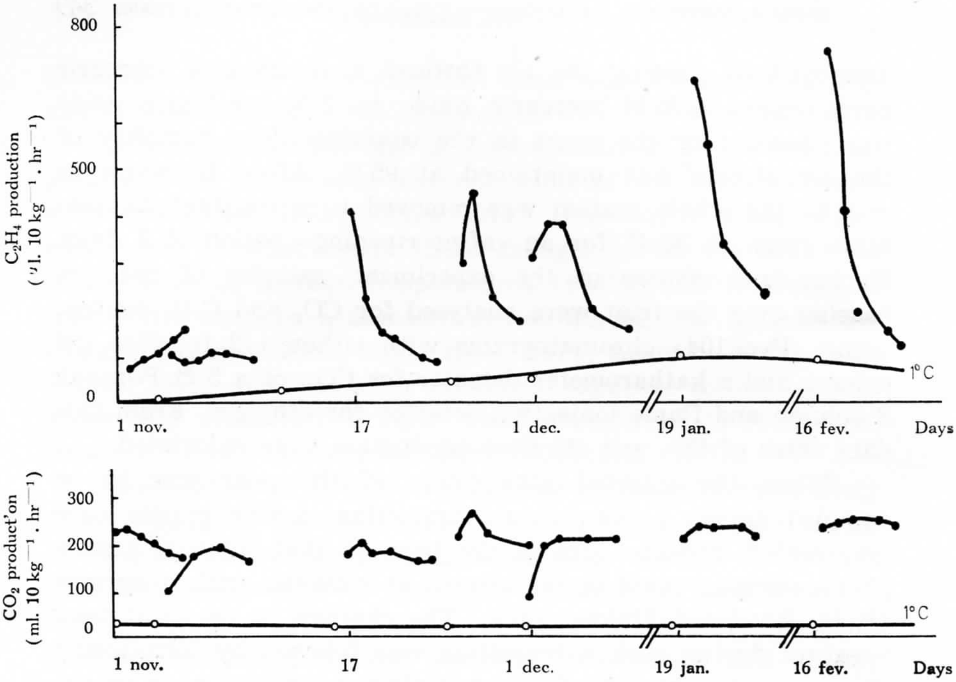

Fig. 1 — Respiration and ethylene production of 'Rocha' pears stored at + 1° C. and later transferred to + 20° C.
Respiração e produção de etileno de peras 'Rocha' armazenadas a + 1° C. e depois transferidas para + 20° C.

○ Samples at + 1° C.
Amostras a + 1° C.

● After ripening period at + 20° C.
Período de pós-maturação a + 20° C.

2. Changes in CO_2 and ethylene production during the transition from + 1° to + 20° C.

When individual pears were warmed from the storage temperature to 20° C, a slow and regular increase in CO_2 production was observed which closely followed the rise in temperature (Figs. 3 and 4). The ethylene production did not follow the rise in temperature but in 'Rocha' pears reached a maximum after **5-6** hours and started to fall before temperature equilibration was achieved (after 9 hours, see Figs. 3, 4 and 5). It is interesting that the height of the peak of C_2H_4 production over

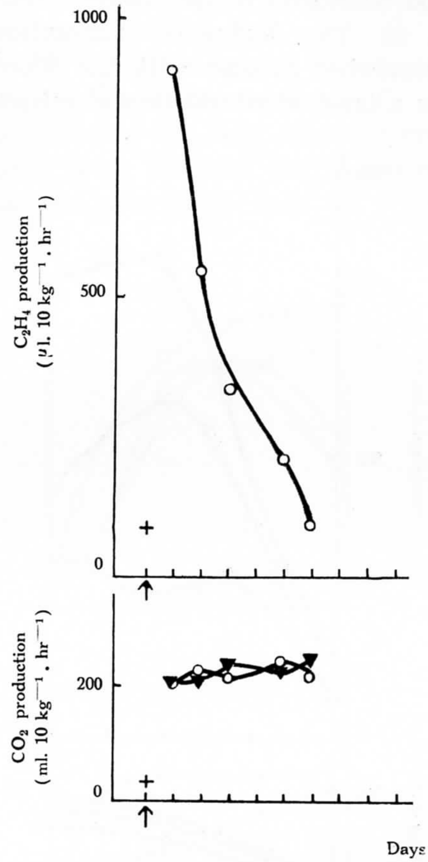

Fig. 2 — The effect of exogenous C_2H_4 on the ripening of 'Rocha' pears stored at + 1° C. for 5 months (26 Jan.).
Efeito de C_2H_4 exógeno na maturação de pêras 'Rocha' armazenadas a + 1° C. durante 5 meses (26 Jan.).

\+ In air at + 1° C.
Em ar a + 1° C.
Control in air at + 20° C.
Controle em ar a + 20° C.
▼ In air plus 100 ppm C_2H_4 at + 20° C.
Em ar mais 100 ppm C_2H_4 a + 20° C.
↑ Removed to + 20° C.
Transferidas para + 20° C.

the six-hour period decreased in the longer stored post-climacteric pears (Fig. 3). The degree of stimulation of ethylene production was somewhat greater with the 'Conference' pears (see Fig. 5) where a ten-fold stimulation of ethylene production

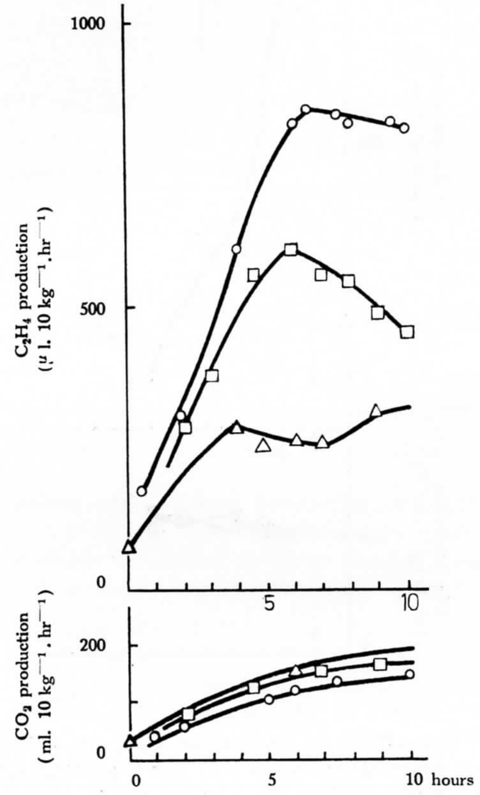

Fig. 3 — CO_2 and C_2H_4 production of individual samples of 'Rocha' pears during the temperature equilibration period.

Produção de CO_2 e C_2H_4 de amostras individuais de pêras 'Rocha' durante o período de equilíbrio de temperatura.

○ 'Rocha' pears on 2.3.71 (6 months).
Pêras 'Rocha' em 2.3.71 (6 meses).

□ 'Rocha' pears on 10.3.71 (6 ½ months).
Pêras 'Rocha' em 10.3.71 (6 ½ meses).

△ 'Rocha' pears on 24.4.71 (7 ½ months).
Pêras 'Rocha' em 24.4.71 (7 ½ meses).

occurred over a five-hour period, and suggests a Q_{10} value for ethylene production in excess of 5.

Fig. 4 shows a representative experiment with a single 'Conference' pear in which the internal CO_2 and ethylene concentrations, the rates of CO_2 and ethylene production and the flesh temperature were followed for the first 10 hours after the transfer of the pear from + 1° to + 20° C. The rate of

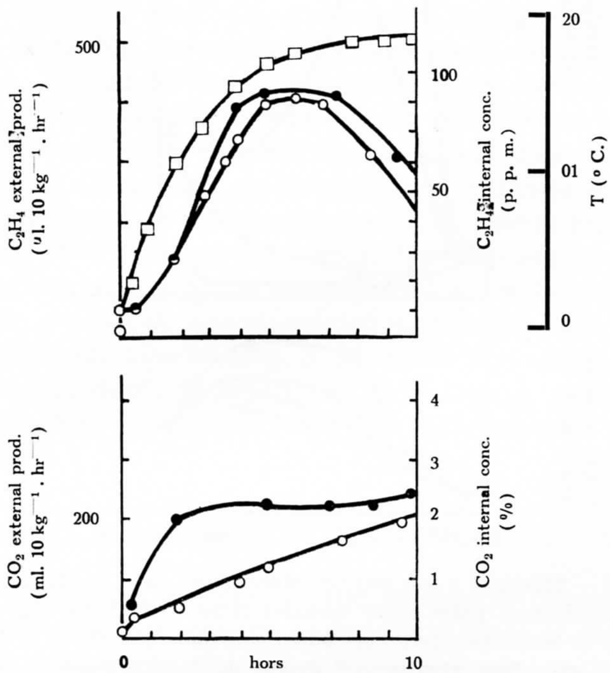

Fig. 4 — Internal atmosphere concentrations and external productions of a 'Conference' pear during the temperature equilibration period.
Concentrações da atmosfera interna e produções externas de uma pêra 'Conference' durante o período de equilíbrio de temperatura.

□ Temperature deep inside the fruit.
Temperatura do interior do fruto.

● Internal atmosphere concentrations.
Concentrações da atmosfera interna.

○ External productions.
Produções externas.

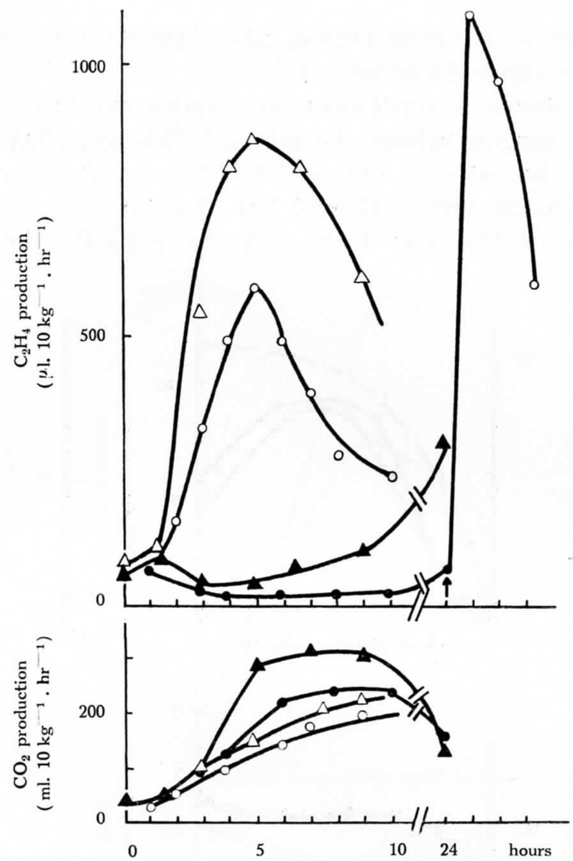

Fig. 5 — Effect of a low oxygen atmosphere (3 % O_2, 97 % N_2) on 'Conference' pears after transfer from — 1° C. to + 20° C.
Efeito de uma atmosfera com baixo oxigénio (3 % O_2, 97 % N_2) em pêras 'Conference' depois da transferência de — 1° C. para + 20° C.

● Pear in 3 % O_2 on 11.3.71.
Pêra em 3 % O_2 em 11.3.71.

○ Pear in air on 11.3.71.
Pêra em ar em 11.3.71.

↑ Removed from low O_2 atmosphere to air.
Passagem de atmosfera com 3 % O_2 para ar.

▲ Pear in 3 % O_2 on 26.4.71.
Pêra em 3 % O_2 em 26.4.71.

△ Pear in air on 2.5.71.
Pêra em ar em 2.5.71.

ethylene production and internal ethylene concentration follow a very similar pattern of change reaching a peak after 6 hours. The increase in CO_2 production follows the rise in temperature but the internal CO_2 concentration rises rapidly after the first 2 hours and then reaches a plateau. The changes in solubility of CO_2 with temperature may be important in relation to the lack of correspondence between the internal CO_2 concentrations and the CO_2 production of the fruit.

Fig. 5 shows that an atmosphere deficient in oxygen (3 % O_2, 97 % N_2) suppressed the increased ethylene production during the temperature transition but the increase in CO_2 production was stimulated. When fruit, retarded in a low O_2 atmosphere during the temperature transition, was returned to air there was a large and rapid increase in ethylene production — a ten-fold increase in 30 minutes. It is interesting that in pears which had been stored longer, the increase in ethylene production was only partially retarded in the low O_2 atmosphere but the CO_2 production was stimulated in the usual way (Fig. 5). In the second experiment (Fig. 5, 26 April) the initial increase in ethylene production during the warming up period after 5 hours at 20° was higher than in the first one (Fig. 5, 11th March).

DISCUSSION AND CONCLUSIONS

The object of the work on the storage of 'Rocha' pears is to describe optimum conditions for the storage of this newly introduced variety which is being increasingly cultivated in Portugal. The present paper describes the initial study in this project and in it we describe changes in the CO_2 and ethylene production of the fruit during storage and «after ripening». We have found that during storage the rate of ethylene production rose to a maximum after 5 months storage at + 1° C. (Fig. 1) by which time the presence of exogenously supplied ethylene had no effect on the respiration of the fruit at 20° C. (Fig. 2). Thus by the middle of January, the fruit had reached the post-climacteric stage at + 1° C.

During the temperature transition from + 1° C. to + 20° C. we have shown that the increase in CO_2 production closely followed the increase in temperature of the flesh of the fruit while the ethylene production rose to a peak after 5-6 hours before complete temperature equilibration was achieved and then fell. In several cases the rate of ethylene production continued to fall until 24 hours after transfer from the low temperature and then rose to a second peak after 24-48 hours. The changes in ethylene production were closely parallelled by changes in the internal concentration of the gas and thus changes in diffusion rate and solubility of the gas during the temperature transfer do not account for the increased ethylene emission by the fruit. The diffusion of gases from the internal atmosphere to the atmosphere follows Fick's law, i. e. the rate of diffusion of gas (equivalent to the rate of production) is proportional to the concentration difference between the interior and exterior of the fruit (Burg & Burg, 1965a). In the case of CO_2 there is no simple relationship between the internal concentrations and the rate of production during the early stages of the temperature shift and here solubility effects may be important.

It seems likely that the observed increase in ethylene production is related to the increased activity of an ethylene producing system which has developed at low temperature. The burst of production on increase in temperature is related to the developed biogenetic capacity and the availability of substrates and co-factores. The fall in activity between 5 and 10 hours is probably related to the exhaustion of the supply of either the substrates or of co-factores. Low O_2 atmospheres, which are known to inhibit the oxidative stages involved in ethylene production (Mapson, 1970), prevent the increase in ethylene production during the temperature transition while the increase in CO_2 production is somewhat stimulated. However, when temperature equilibration has been observed and the constraint of low O_2 has been removed, there is a dramatic increase in ethylene production.

Other workers (Hansen, 1941; Fidler & North, 1969) have found large and somewhat variable increases in ethylene production when pears are transferred from the cold to room temperature. Damage to tissue often results in increased ethylene production (see McGlasson, 1969) and Reid & Pratt (1972) suggested that ethylene may act as a «wound hormone». It may be that the sudden change in the flesh temperature of the fruit creates a degree of stress in the tissue and this may itself tend to stimulate ethylene production.

We have studied these transitory changes in CO_2 and ethylene production during the temperature equilibration between + 1° and + 20° C. in the hope that these changes would reflect the metabolic condition of the fruit in the store and would perhaps provide a rapid method of assessing this metabolic state. We have indications that they may prove useful but more detailed studies on this aspect are under way and when complete the value of this method of evaluation can be assessed.

RESUMO

Estudo da produção de etileno e anidrido carbónico em pêras durante a conservação em ar a baixa temperatera e na subsequente transferência para + 20° C.

Durante os períodos de armazenagem e de pós-maturação de pêras 'Rocha', variedade portuguesa largamente cultivada em Portugal, foram determinadas as produções de etileno e de anidrido carbónico. A produção de etileno atingiu um máximo aos cinco meses de armazenamento à temperatura de 1° C., mas na transferência para 20° C., depois de uma subida inicial, a velocidade de produção foi sempre decrescente. Foram feitas experiências com pêras 'Rocha' e 'Conference' para seguir as variações de produção de etileno e CO_2 durante as primeiras horas após a transferência para 20° C. A respiração aumentou paralelamente com o aumento da temperatura do fruto, mas a produção de etileno alcançou um máximo depois de 6-7 horas a 20° C. e depois baixou. Uma atmosfera deficiente em O_2 diminuiu a produção de etileno, durante o período de aquecimento, mas a respiração foi estimulada.

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REFERENCES

Burg, S. P. & Burg, E. A.

1965a Gaz exchange in fruits. *Physiologia Pl.* **18**: 870-884.

1965b Ethylene action and ripening of fruits. *Science, N. Y.*: **148**: 1190--1196.

Fidler, J. C.

1960 Naturally occuring votatile organic compounds *In:* Ruhland, W. *& al. (edit.), Handbuch der Pflanzenphysiologie.* 12 (2): 347-359. Springer Verlag, Berlin.

——————— & Coursey, D. E.

1970 Low temperature injury in tropical fruit. *Proceedings of the Conf. on Tropical and Sub-tropical Fruits:* 103-115.

——————— & North, C. J.

1969 Production of volatile organic compounds by pears. *J. Sci. Fd Agric.* **20**: 518-520.

Hansen, E.

1941 Quantitative study of ethylene production in relation to respiration of pears. *Bot. Gaz.* **103**: 543-558.

Kidd, F. & West, C.

1937 The cold storage and gaz storage of english grown William Bon Chrétien pears. *Rep. Fd Invest.* **1936**: 113-126.

Knee, M.

1971 Ripening of apples during storage. II — Respiratory metabolism and ethylene synthesis in Golden Delicious apples during the climacteric, and under conditions simulating commercial storage practice. *J. Sci. Fd Agric.* **22**: 368-371.

Leonard, S., Luh, B. S., Hinreiner, E. & Simone, M.

1954 Maturity of Bartlett pears for canning. *Fd Technol.* **8**: 478-482.

Mapson, L. W.

1970 Biosynthesis of ethylene and its control. *Proceedings of the Conf. on Tropical and Sub-tropical Fruits:* 85-92.

McGlasson, W. B.

1969 Ethylene production by slices of green banana fruit and potato tuber tissue during the development of induced respiration. *Aust. J. biol. Sci.* **22**: 489-491.

Pratt, H. K. & Goeschl, J. D.

1969 Physiological roles of ethylene in plants. *A. Rev. Pl. Physiol.* **20**: 541-584.

Reid, M. S. & Pratt, H. K.

1972 Effects of ethylene on potato tuber respiration *Pl. Physiol.* **49**: 252-255.

———————, Rhodes, M. J. C. & Hulme, A. C.

1972 In preparation.

Rhodes, M. J. C.
1970 The climacteric and ripening of fruits. **1**: 521-533. *In: Hulme, A. C. (edit.), The Biochemistry of Fruits and their Products.* Academic Press, New York.

Torres, Maria A.
1971 *Studies on cold storage of Portuguese pears.* Report for OECD.

Wilkinson, B. G.
1970 Physiological disorders of fruit after harvesting. **1**: 537-554. *In*: Hulme, A. C. *(edit.), The Biochemistry of Fruits and their Products.* Academic Press, New York.