EFFECT OF VARIOUS FACTORS ON THE STABILITY OF THE ANTHOCYANIN PIGMENT IN PASSION FRUIT SKIN (*PASSIFLORA EDULIS*, SIMS)

by

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ABSTRACT

Since loss of anthocyanins is one of the major factors contributing to the colour deterioration in many highly coloured fruits, the effects of various factors such as temperature, oxygen, *pH*, ascorbic acid, tannic acid, thiourea and hydrogen peroxide on the stability of the anthocyanin pigment (*Pelargonidin 3-diglucoside*) naturally occurring in passion fruit skin (*Passiflora edulis*, sims). have been studied with a view to elucidate the mechanism of degradation of the pigment during refrigerated and common storage of the fresh fruit.

Storage studies demonstrated that (*i*) ascorbic acid in the presence of oxygen accelerated the deterioration of the pigment; (*ii*) thiourea decreased the rate of destruction of ascorbic acid, thus indirectly preventing the rate of anthocyanin losses; (*iii*) tannins had a stabilizing effect on the pigment; (*iv*) pH had a very significant effect on the stability of the pigment; and (*v*) high storage temperature and H_2O_2 both had destructive effect on the pigment.

Introduction

Earlier reports[1,2] indicate that purple passion fruit (*Passiflora edulis*, sims.) has a very short storage life and that the outer purple colour of the fruit deteriorates during storage. This has now recently been attributed to the degradation of the anthocyanin (Pelargonidin 3-diglucoside)[3] occurring in the skin.

The loss of anthocyanins is one of the major factors contributing to the colour deterioration in many highly coloured fruits. A multiplicity of factors individually or collectively affect the stability of the pigment during processing or storage. For instance, light, temperature, presence of oxygen, ascorbic acid, metals and change in *pH* are known to have deleterious effect on the pigment[4, 6] while thiourea[7] and tannic acid[8] exert some protective action. With a view to understand the mechanism of degradation of the pigment in passion fruit skin, it was of interest to study the effect of these factors on the pigment under the conditions of *pH*, etc., existing in passion fruit skin. This was studied as follows:

Experimental

(*i*) *Effect of Temperature*—In our present studies on the effect of temperature on the pigment, the crude passion fruit skin pigment obtained by precipitation

with ether was used. The pigment was extracted by methanolic *HCl* and the *pH* adjusted to 2·9. The extract was divided into 3 lots, packed in several sample tubes (covered with black paper), corked airtight and stored at three different temperatures; *viz.* 5-7°C; 30°C; and 55°C. The samples from each lot were removed at frequent intervals and the optical density measured at 530 mμ (absorption peak for pelargonidin). Besides, the over-all absorption spectra of the fresh extract as well as those of the extracts stored at different temperatures for ten days were obtained and are presented in fig. 1.

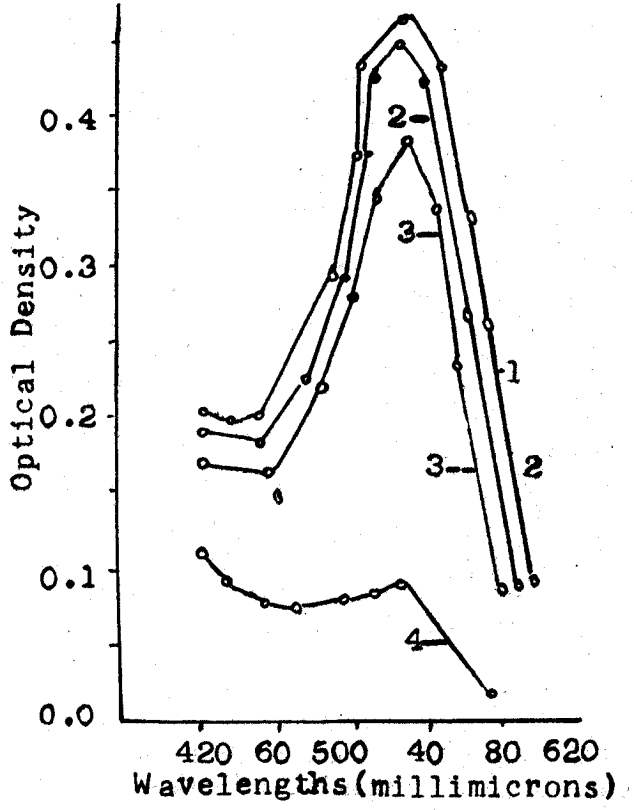

FIG. 1: Effect of Storage Temperature on the Absorption Spectra of the Anthocyanin Pigment isolated from Passion Fruit Skin.

References in the figure :

1. Initial
2. After 10 days storage at 5—7°C
3. After 10 days storage at 30°C
4. After 10 days storage at 55°C.

It was found that, while the pigment was quite stable for 3 days at 5—7°C, the degradation of the pigment had occurred at 55°C, even within 24 hours' storage. It was also noted that the room temperature (22—28°C) had no deleterious effect on the pigment for at least 24 hours. Thereafter, slight deterioration did set in.

(*ii*) *Effect of light*: Light has been reported to have little effect upon the anthocyanins in fruit packs[4,5] and if any, it is negligible in comparison with that brought about by other factors. Our studies confirmed this observation.

(*iii*) *Effect of pH*: In our present studies on the effect of *pH* on the absorption spectra of the pigment, three *pH* ranges were chosen. The pigment extract was buffered to *pH* 2·0, 3·6 and 5·5 by using Sorenson's sodium citrate buffer, and examined for the changes in the absorption spectra which are presented in figure 2. The results indicate that at low *pH*, the intensity of light absorption increased in the visible range. The colour of the pigment faded as the *pH* was raised to 3·6 and above.

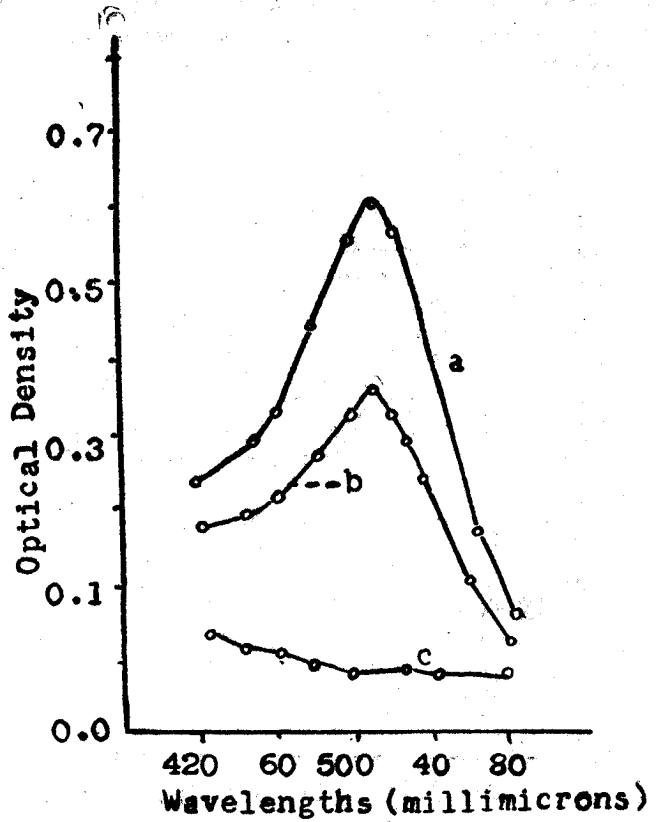

FIG 2: Effect of *pH* on the Absorption spectra of the Anthocyanin Pigment isolated from Passion fruit Skin.

References in the figure :

a = *pH* 2·0

b = *pH* 3·6

c = *pH* 5·5

(*iv*) *Effect of Ascorbic Acid and Oxygen* : The following experiment was carried out to study the effect of ascorbic acid and oxygen on the pigment :

The precipitated extract from ether was dissolved in 1% methanolic *HCl*. The ascorbic acid content of the extract was estimated. The solution was divided into 6 lots. One lot was kept as a control, while the others were treated respectively with (*a*) 50 mg. ascorbic acid/100 ml. of solution, (*b*) 50 mg. of thiourea/100 ml. of solution, (*c*) 50 mg. ascorbic acid and 50 mg. thiourea/100 ml. of solution, (*d*) 50 mg. ascorbic acid plus 0·2% tannic acid and (*e*) 0·2% tannic acid alone. Since it has been reported[6] that lack of oxygen inhibits the ascorbic acid oxidation and thus prevents marked deterioration of the pigment, no attempt was made to pack them under vaccum. The samples were packed in sample tubes corked airtight and stored at 30°C. Ascorbic acid content was estimated at frequent intervals and the optical density of the pigment recorded at 530 mμ. The changes in the pigment as affected by the above factors during 10 days' storage at 30°C are illustrated in fig. 3.

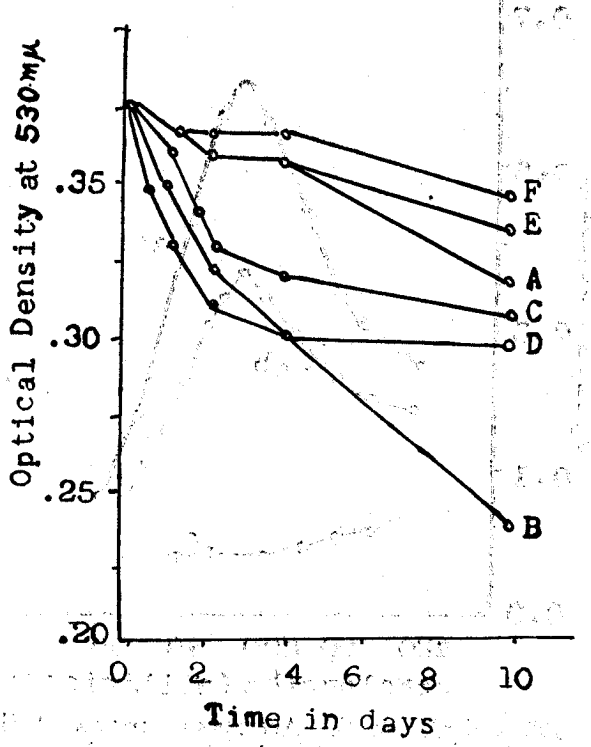

FIG 3: Effect of Ascorbic Acid Thiourea and Tannic acid on the stability of Anthocyanin Pigment isolated from Passion Fruit Skin.

References in the figure :

A = Control
B = Added Ascorbic Acid (A.A) @ 50 mg/100 ml. of the Solution.
C = Added Thiourea @ 50 mg/100 ml. of the solution.
D = Added 50 mg. A.A. and 50 mg. Thiourea/100 ml. of the solution.
E = Added 50 mg. A.A. and 200 mg. Tannic Acid/100 ml. of the solution.
F = Added 200 mg. Tannic Acid/100 ml. of the solution.

Storage studies demonstrated that greater losses in ascorbic acid and greater deterioration in pigment were encountered in samples containing added ascorbic acid, in comparison with the control samples, wherein the pigment destruction was comparatively slow. These results are in agreement with those on strawberry anthocyanins[6].

(*v*) *Effect of Thio-urea on the stability of the Pigment*: From curve C (fig. 3), it may be seen that thiourea in the absence of ascorbic acid, has no demonstrable effect on the rate of pigment destruction. These results are in agreement with those of Gockel[7]. Curve D (fig. 3), however, indicates slightly greater destruction of the pigment in the presence of added ascorbic acid and thiourea, but it is far less than that in the presence of added ascorbic acid alone (Curve B), indicating thereby that thiourea decreased the rate of pigment degradation. Besides, it was also noted to retard ascorbic acid oxidation as well.

(*vi*) *Effect of Tannins on the Stability of Anthocyanin Pigments*—Tannins are reported[8] to have a stabilizing effect on anthocyanins. In our storage studies, similar results were obtained as illustrated in Curve F (Fig. 3).

(*vii*) *Effect of Hydrogen Peroxide on the Stability of Anthocyanin Pigment*: Preliminary studies revealed that hydrogen peroxide adversely affected the stability of the pigment. It was further interesting to note that the changes in colour were almost similar to those actually observed *in situ* in passion fruit during storage at room temperature.

Discussion

The results obtained above could perhaps be utilized in partially explaining the mechanism of colour deterioration in passion fruit skin as tentatively suggested below :—

1. The colour deterioration in passion fruit skin may mostly be attributed to the degradation of the anthocyanin pigment (*Pelargonidin 3-diglucoside*) naturally occurring in passion fruit skin, which might be partly due to the storage temperature itself, since the colour deterioration in passion fruit was found to be greater at higher temperatures[1,2].

2. The degradation of the pigment might have also been accelerated by the concurrent destruction of ascorbic acid, which is usually present in high concentration[1,9] (78·3 to 166·2 mg/100g) in the passion fruit skin along with the pigment.

3. Recently, the H_2O_2 induced oxidation of ascorbic acid in passion fruit juice has been reported[10]. The transient presence of hydrogen peroxide in passion fruit skin also is a possibility and, therefore, the loss of some of the passion fruit skin pigment through reaction with this hydrogen peroxide, is a reasonable assumption. There are a number of reactions which may give rise to the production of hydrogen peroxide in passion fruit skin. It is known, for instance, that in the presence of oxygen and cupric ions, the oxidation of ascorbic acid to dehydroascorbic acid is accompanied by the formation of hydrogen peroxide[11,13].

4. Passion fruit skin does contain considerable quantity of tannic acid which should be exerting a protective action on the pigment, but probably, the destructive factors outlined above outweigh this protective factor and hence the resultant deterioration in the colour of the fruit skin.

However, in order to have a still more clear picture of the actual mechanism of colour deterioration in passion fruit skin, experiments both *in situ* and in model systems on the lines suggested above are in progress and will be reported later.

Acknowledgement

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References

1. Pruthi, J. S., *Ph. D. Thesis.* The Pb. Univ., Solan, India (1955).
2. Pruthi, J. S. & Lal, G., *Indian J. Hort.*, **12**, 4, 1955.
3. Susheela, R., *Assoc. Thesis.* C.F.T.R.I., Mysore (India) 1959.
4. Thompson, H. H., *et al.*, *Food Indust.*, **18**, 1341, 1946.
5. Tressler, D. K. & Pederson, C. S., *Food Res.*, **1**, 87, 1936.
6. Sondheimer, E. & Kertesz, Z. I., *Ibid.* **18**, 475, 1953.
7. Gockel, H., *Dent. Lebensm-Rundeschau*, 9. (C. A., 1943, 3228), 1942.
8. Sastry, L.V.L. & Tischer, R. G., *Food Tech.*, **6 7**, 264, 1952.
9. Pruthi, J. S. & Lal, G., *J. Sci. Food & Agr.* (*U. K.*), **10**, 3, 188, 1959.
10. Ross, E. & Chang, A. T., *J. Agr. Food Chem.*, **6**, 8, 610, 1958.
11. Sondheimer, E. & Kertesz, Z. I., *Food Res.*, **17**, 3, 288, 1952.
12. Dekker, A.O. & Dickinson, R. G., *J. Am. Chem. Soc.*, **62**, 2165, 1940.
13. Silverblatt, E. *et al.*, *Ibid* **65**, 137, 1943.