

quent, la L-arginine libérée par l'hydrolyse du III rendra la cellule cancéreuse encore plus susceptible à l'égard de V.

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Zusammenfassung

Es wird vorgeschlagen, N- α -di-(β -chloräthyl)-carbamyl-L-arginin (III) als neues Stickstofflost auf cytostatische Wirkung zu untersuchen; dies in der Erwartung, dass III in Krebszellen in höherer Masse zum cytostatisch wirkenden Di-(β -chloräthyl)-amin (V) hydrolysiert wird als in normalen Zellen. Das gleichzeitig freierwerdende L-Arginin sollte die Krebszelle noch empfindlicher gegen V machen.

Enzymatic Synthesis and Destruction of Carotenoids in Mango Extracts

Among the carotenogenic fruits, most of the investigations on the biosynthesis of carotenoids have been carried out on tomatoes. Mangoes, which are good carotene producers, have not been examined in such detail. The present investigation was undertaken with a view to study the general factors controlling carotenogenesis in Bombay Alfanzo mangoes. An attempt has been made to prepare cell-free extracts of the fruits which can synthesize carotenoids.

The fruits were preserved in the cold room at the temperature 0–3°C, and, in the frozen condition, the outer skin was removed; then 10 g (fresh weight) of mango pulp (after immediately neutralizing) were ground in a medium containing 0.5 M sucrose in 100 ml phosphate buffer (0.1 M, pH 7.0) at 0°C for about 10 min. The supernatant solution obtained by centrifuging the pulp at $12800 \times g$ for 20 min is referred to as mango extract. 5 ml of the mango extract (containing protein concentration of about 5 mg) were incubated at 30°C in 250 ml Erlenmeyer flasks on a rotary shaker for 18 h with the desired substrates in phosphate buffer (pH 7.0, 0.1 M). Carotenes were extracted in freshly distilled ether. The ethereal extract was freed of moisture by treating with anhydrous sodium sulphate. The carotenes were transferred in 5 ml petroleum ether (B. P. 80–100°C) and they were determined as β -carotene by measuring $E_{450 m\mu}$ ($E_{1\%}^{1\text{cm}} = 2500$) in Beckman photoelectric spectrophotometer. Pure synthetic β -carotene was dissolved in minimum amount of Tween-80 and then added in the medium to study its destruction.

For estimating total acidity and sugar content of mangoes, 5 g of the pulp were completely extracted by triturating with sand and water. After filtration, from a known volume, an aliquot portion was used for the determination of sugar as glucose by Cole's method and for total acidity by titrating against 0.01 N sodium hydroxide using phenolphthalein as an indicator.

The results recorded in Table I show that ripe mangoes are about ten times richer in carotene than partly unripe ones; and the increase in carotene content of mangoes is accompanied by decrease in acid content and increase in sugar content. It was not possible to detect carotenoids, even in traces, from unripe green mangoes.

Mangoes which had reached full ripeness gave optimum results and therefore they were used in further investigations.

Table II lists the results of an experiment showing the enzymatic formation of carotenes from glucose and acetate in cell-free extracts. From the results of control experiments, it can be seen that, as in the case of carrots¹, with mangoes also, some amount of carotene originally present (at zero time) gets destroyed during incubation.

Table III includes the results of the experiments which show that synthetic β -carotene gets destroyed in presence of enzymatic extract. The destruction of β -carotene was measured by comparing the amounts present before and after reaction. In order to separate the effect of physical conditions such as light, temperature etc. synthetic β -carotene was incubated in control experiments without enzymatic extract.

Table I. Sugar, acid, and carotene content in partly unripe and ripe mangoes.

Mango	Total sugar ^a	Total acidity ^b	Carotene content ^c
Unripe	4.942	41.0	488.00
Ripe	22.390	7.6	3520.00
^a g/100 g of pulp. ^b Equivalent to 1 N acid/100 g of pulp. ^c $\mu\text{g}/100\text{ g}$ of pulp.			

Table II. Biosynthesis of carotenoids in mango extract. The test system contained final concentration of 0.1 M phosphate buffer pH 7.0, substrates in amount listed below and 5 ml of mango extract in a total volume of 30 ml in each 250 ml Erlenmeyer flask, incubated 18 h at 30°C.

Substrate	Amount added in (mg)	Carotene amount in μM		
		Zero-time	After incubation	Net change
Glucose	625	0.225	0.328	+ 0.103
Acetate	250	0.225	0.352	+ 0.127
None (Control)	—	0.225	0.127	– 0.098

Table III. Enzymatic degradation of carotenoids in mango extract. Conditions identical as shown in Table II.

Substrate	Amount added in μM	Percentage degradation of carotenoids
Synthetic β -carotene	3.938	15.710
Synthetic β -carotene (Control enzyme omitted)	3.938	1.422

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Zusammenfassung

Eine Biosynthese von Carotin lässt sich in zellfreien karotenoidhaltigen Extrakten reifer Mangofrüchte bei Gegenwart von Glucose oder Azetat beobachten. Dabei zeigt sich gleichzeitig eine Abnahme der Karotenoide.

¹ V. V. MODI and D. K. PATWA, Nature 184, 983 (1959).