Carotenoid Content of the Varieties *Jaranda* and *Jariza* (*Capsicum annuum* L.) and Response during the Industrial Slow Drying and Grinding Steps in Paprika Processing

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Fruits of the pepper varieties *Jaranda* and *Jariza* (*Capsicum annuum* L.) ripen as a group, enabling a single harvesting, showed a uniform carotenoid content that is high enough (7.9 g/kg) for the production of paprika. The drying system at mild temperature showed that fruits with moisture content of 85−88% generated a dry product with carotenoid content equal to or higher than the initial one. Those high moisture levels allowed the fruits to have a longer period of metabolic activity, increasing the yellow fraction, the red fraction, or both as a function of what biosynthetic process was predominant. This fact indicates under-ripeness of the fruits in the drying step. The results obtained allow us to establish that both varieties, *Jaranda* and *Jariza*, fit the dehydration process employed, yielding a dry fruit with carotenoid concentration similar to that the initial one. During the grinding step of the dry fruit, the heat generated by the hammers of the mill caused degradation of the yellow fraction, while the red fraction is maintained. The ripeness state of the harvested fruits and the appropriateness or severity of the processing steps are indicated by the ratio of red to yellow (R/Y) and/or red to total (R/T) pigments, since fluctuations in both fractions and in total pigments are reflected in and monitored by these parameters.

Keywords: *Capsicum, carotenoids, moisture, paprika processing, pigment reactions, slow drying*

INTRODUCTION

The numerous studies on carotenoid pigments highlight their attractive properties. These are a result of their molecular structure, giving them eye-catching colors, antioxidant capacity (Britton, 1995; Burton, 1989), and, in some cases, provitamin A activity (Simpson, 1983; Underwood, 1994). The pericarp of the pepper fruit (*Capsicum annuum* L.) is a plant tissue that accumulates considerable amounts of a wide variety of carotenoid pigments, giving the ripe fruit an intense color of red (Levy et al., 1995; Almela et al., 1991; Deli et al., 1996), black (Deli et al., 1992), and even yellow (Matus et al., 1991). This makes it the raw material for the production of food colorants denominated paprika and oleoresins.

Paprika is prepared in two operations: drying and grinding (which gives the product its final appearance). The first operation can be carried out in hot air-dryers or in drying chambers where the heat source is the burning of oak logs. The smoke given off by oak logs impregnates the fruits with an aroma and taste very highly prized by the consumer.

In both processes, the dehydration conditions must respect the fruit carotenoid content. The first studies on the effects of drying, with either hot air or burning logs, and subsequent grinding on the initial carotenoid concentration of the fruit were carried out on the varieties most common at that time, *Bola* and *Agridulce*, (Mínguez-Mosquera et al., 1994; Mínguez-Mosquera and Hornero Méndez, 1994b). These authors observed an effect of the drying procedure on the carotenoid content depends on the variety of pepper used. They demonstrated the metabolic complexity of carotenoid synthesis in the pepper fruit, involving two biosynthetic pathways, one or the other dominant depending on the ripeness stage. Thus, in the variety *Bola*, drying the fruit in log-burning dryers yields an increased concentration of certain pigments. The explanation is a continued biosynthesis of carotenoids in the fruits entering the dryer, caused by their incomplete ripeness, so that the metabolic pathways of pigment formation continue to exist. This leads to a total carotenoid content, with an increase in the concentration of red carotenoids at the expense of their precursors, the yellow ones. By contrast, the variety *Agridulce*, which is harvested when completely ripe, shows losses in total carotenoids after drying. Drying in industrial hot air-dryers reduces total carotenoid concentration in both varieties (Mínguez-Mosquera et al., 1993).

The varieties *Bola* and *Agridulce* present certain drawbacks as crops. The fruits of the *Agridulce* variety are used for the production of spicy, highly colored paprikas. However, they do not ripen at the same time so that more than one harvest is required. The fruits of the variety *Bola*, used for the production of sweet paprika, have two disadvantages: low productivity, caused by problems of ripening on the plant itself, and little color due to the low carotenoid content. As a result, and because of the lack of appropriate raw material for the production of nonpungent paprika, breeding trials were carried out between 1990 and 1992 to obtain varieties from *Agridulce* that were nonpungent, and had a high coloring capacity (Rodríguez et al. 1993). This resulted in the varieties *Jaranda* and *Jariza*, whose

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Table 1. Total Carotenoid Content in Fresh Fruit, Dehydrated Fruit, and Paprika, for the Varieties Jaranda and Jariza (*Capsicum annuum* L.). Effect between Phases of Processing and Overall Yield

		total carotenoids (mg/kg)				percentage of loss during			
		fresh fruit							
code[a]	moisture	W.B. [b]	D.B.[c]	dry fruit	paprika	drying	grinding	total	% seed[e]
				Variety *Jaranda*					
FD1	84.4	1055.5 ± 113.1[d]	6759.1 ± 724.4	8117.1 ± 158.6	5046.0 ± 217.4	−20.1	37.8	25.3	30
FD2	86.0	1141.2 ± 90.5	8151.7 ± 646.7	7948.0 ± 180.7	4686.0 ± 177.2	2.5	41.0	42.5	36
FD3	80.3	1611.7 ± 35.9	8177.4 ± 182.3	6579.8 ± 113.6	3986.9 ± 126.0	19.5	39.4	51.2	37
FD4	83.9	975.58 ± 36.8	6059.5 ± 228.3	6349.1 ± 220.6	3451.5 ± 119.9	−4.8	45.6	43.0	37
				Variety *Jariza*					
FZ5	86.7	974.08 ± 74.6	7318.4 ± 531.1	7452.7 ± 448.4	4270.6 ± 59.1	−1.8	42.7	41.6	36
FZ6	87.8	895.76 ± 123.9	7342.3 ± 598.7	7493.1 ± 268.9	4166.7 ± 75.5	−2.1	44.4	43.0	37
FZ7	82.6	1311.9 ± 124.8	7522.1 ± 712.5	5861.4 ± 205.1	4021.0 ± 68.0	22.1	31.4	46.5	30
FZ8	84.0	1080.8 ± 48.5	6755.2 ± 297.4	6135.0 ± 116.3	4020.8 ± 100.1	9.2	34.5	40.0	31

[a] FD, Jaranda farm. FZ, Jariza farm. [b] W.B. = carotenoid concentration (wet base). [c] W.B. = carotenoid concentration (dry base). [d] Mean ± standard deviation of six determinations. [e] Estimated % of seed added in the grinding.

fruits ripen together, allowing a single harvesting operation which might be mechanized. Furthermore, it was necessary to check whether these varieties produced an appropriate plant material (yielding a high carotenoid content) and whether the drying process affected the carotenoid content.

The present work concentrates on characterizing the carotenoid richness in the varieties *Jaranda* and *Jariza* and on determining how drying with heat generated by burning logs and subsequent grinding affect the initial pigment concentration of the fresh fruit after processing. An extensive broad field study was carried out with fruits of the same variety, but different locations in order to take into account the variability of the raw material and to compare behavior during drying and grinding. The total carotenoid contents and fluctuations in the isochromic fractions of red and yellow pigments were measured in the three processing stages: fresh fruit, dehydrated fruit, and paprika.

MATERIALS AND METHODS

Selection of Starting Material. Ripe fruits of two varieties of pepper *Capsicum annuum longum* L. were obtained from eight farms. On four of these farms the variety named *Jaranda* (FD1 to FD4) was grown, and on the other four, the variety *Jariza* (FZ5 to FZ8). The farms FD4 and FZ8 are located in Valle del Alagón. The other farms are in Valle del Tiétar.

Fruits ripen at the same time, enabling almost all to be harvested in a single operation. Morphologically the fruits are very similar: some 14 cm in length, 2 cm in maximum diameter, and 0.5 cm thick. The fruits are dried on the farm where they were harvested, in stone constructions of two stories separated by a wooden platform allowing ventilation. The fruit (up to 3000 kg) is placed in the upper story and heated by oak logs burning in the lower story. The mean temperature reaches 40−50 °C. Dehydration of the fruits is completed in 10 days, and the product of each dryer is ground in local mills. Eight batches of paprika were so obtained in this way, one from each farm and dryer.

Sampling Procedure. Three types of sample were collected. For fresh fruit, a 2 kg sample was taken from an initial amount of 10 kg at each farm. Seeds and stalk were removed, and the rest was chopped and homogenized. Six subsamples each of 10 g were used for pigment analysis. For dehydrated fruit, 1 kg samples were taken from 3 kg of each batch, seeds were removed, and the rest was chopped and homogenized. Six subsamples each of 2 g were used for the extraction and analysis of pigments. For paprika, each farm provided 1 bag of 100 g. Six samples each of 1.5 g were analyzed in each case.

Pigment Extraction. The pigments were extracted with acetone, with the sample blended in a beater (Ultraturrax, model T25) and vacuum-filtered through a Büchner funnel. The residue was re-treated with acetone, and the operation was repeated until the filtrate was colorless. Three extractions, or at most four, were enough to extract all the color. The filtrates were passed to a 500 mL decanting funnel; 150 mL of ethyl ether was added, and the funnel was shaken for 30 s. NaCl solution (10%) was added to separate the phases. The upper phase, containing the pigments, was washed several times more to remove the acetone. Any color in the washing waters was recovered with ether and added to the initial organic solution.

The filtrates, containing the pigments in ether solution, were combined, and 50 mL of KOH (10%) in methanol and an appropriate amount of β-apo-8′-carotenal (as internal standard for the quantification) were added. The mixture was left to saponify for 1 h, being shaken every 10 min. An aqueous solution of NaCl (10%) was added to separate the phases. The organic phase was washed repeatedly until neutral, filtered through a bed of solid Na_2SO_4, and dried in a rotavapor. The solid residue, containing the de-esterified pigments, was collected in acetone and kept in the freezer at −20 °C until its analysis.

Pigment Analysis and Identification by HPLC. The carotenoids were separated, identified, and quantified according to the method described by Mínguez-Mosquera and Hornero-Méndez, 1993. The analysis was performed in a high-pressure liquid chromatograph (Waters, model 600), fitted with a quaternary pump and diode array UV−vis detector (model 996). Sample injection was performed manually using a Rheodyne injector (model 7125) fitted with a 20 μL loop.

RESULTS AND DISCUSSION

Table 1 shows the results of total carotenoid concentration in the fresh fruit (as well the corresponding moisture content), dry fruit, and paprika, for each farm and variety. It also presents the response, positive or negative, in carotenoid concentration (expressed as percentage variation) after each process step in each case, together with the percentage of seeds included in the grinding steps, and the overall yield of the process.

Characteristics of Investigated Raw Material. As can be observed in Table 1, the total carotenoid concentration in the fresh fruit varies from farm to farm. The variety *Jariza* shows the best uniformity, and total concentration does not exceed 7.6 g/kg, while *Jaranda* fluctuates more widely: from 8.2 to 6.1 g/kg. However, this variability in fresh fruit is not high enough to cause significant differences (Duncan test, $p < 0.05$) based on total carotenoid comparison of each variety. The results were grouped without distinguishing the farm of origin. The total pigment concentrations of the

Table 2. Carotenoid Content by Isochromic Red and Yellow Fractions in Fresh Fruit, Dehydrated Fruit, and Paprika, for the Varieties Jaranda and Jariza (*Capsicum annuum* L.). Effect between Phases of Processing

	red fraction[b] (mg/kg)			percentage of loss		yellow fraction[c] (mg/kg)			percentage of loss	
code[a]	fresh fruit	dry fruit	paprika	drying	grinding	fresh fruit	dry fruit	paprika	drying	grinding
				Variety *Jaranda*						
FD1	3718.9 ± 406.6[d]	4154.3 ± 223.5	2956.7 ± 122.1	−11.7	28.8	3040.2 ± 326.1	3962.9 ± 129.9	2089.3 ± 95.3	−30.4	47.3
FD2	4536.3 ± 372.9	4487.7 ± 128.5	2736.6 ± 106.7	1.1	39.0	3615.4 ± 285.2	3460.2 ± 64.0	1949.5 ± 70.5	4.3	43.7
FD3	4767.4 ± 113.2	3660.5 ± 129.4	2310.2 ± 74.7	23.2	36.9	3409.8 ± 79.3	2919.3 ± 44.4	1676.7 ± 100.7	14.4	42.6
FD4	3329.2 ± 144.8	3727.3 ± 137.0	1931.7 ± 158.5	−12.0	44.2	2730.3 ± 114.5	2621.8 ± 120.5	1519.8 ± 38.6	3.9	42.0
				Variety *Jariza*						
FZ5	3964.1 ± 178.6	4007.8 ± 154.1	2404.4 ± 43.8	−1.1	40.0	3354.3 ± 383.8	3444.9 ± 297.7	1866.2 ± 15.3	−2.7	45.8
FZ6	4110.3 ± 369.0	3870.5 ± 158.3	2434.8 ± 55.4	5.8	37.1	3232.0 ± 283.8	3622.6 ± 110.6	1732.0 ± 20.1	−12.1	52.2
FZ7	4458.4 ± 436.6	3375.8 ± 113.2	2422.0 ± 10.0	24.3	28.3	3063.7 ± 277.2	2485.6 ± 92.3	1598.9 ± 67.0	18.9	35.7
FZ8	3834.2 ± 258.4	3534.3 ± 24.3	2385.3 ± 47.7	7.8	32.5	2921.1 ± 101.3	2600.7 ± 138.5	1635.5 ± 52.4	11.0	37.1

[a] FD, Jaranda farm. FZ, Jariza farm. [b] Red fraction = capsorubin + capsanthin-5,6-epoxide + capsanthin + *cis*-capsanthin. [c] Yellow fraction = violaxanthin + anteraxanthin + cucurbitaxanthin A + zeaxanthin + *cis*-zeaxanthin + β-cryptoxanthin + β-carotene + *cis*-β-carotene. [d] Mean ± standard deviation of six determinations (dry base).

varieties *Jaranda* and *Jariza* are expected to be very high, which allows an efficient production of paprika. The different moisture contents according to the source farm are also shown. A low moisture content reflects an over ripeness stage of the fruit and consequently a vital stage on the decline. A high moisture content implies a fruit not totally developed and allows biosynthetic activity during drying.

Effect of Drying on the Total Carotenoid Content. One of the most important characteristics of the type of drying process employed is that it is carried out under mild conditions of temperature, so that potentially degradative reactions minimally affect the carotenoid content. At the same time, the fruit moisture content, together with the variables temperature and drying time, determine the changes (whether positive, negative, or none) in the fruit carotenoid concentration. In the dryers FD3 and FZ7, raw material moisture levels were 80.3% and 82.6%, respectively, and total carotenoids decreased by 20%; with fruit moisture contents of 84−88%, total carotenoids in dryer FD1 increased by 20% and remained with little fluctuations in the rest except in dryer FZ8 (decrease 9.2%).

These results indicate that, in general, the process using log burning, which is slow and at mild temperature, does not affect fruit carotenoid content and that the conditions are appropriate for dehydration of fruits with high levels of moisture.

In conclusion, grouping all the dryers the response to the process is positive, since for the variety *Jaranda* there was an increase of 2% in the carotenoid concentration. For *Jariza* the process generated a loss of only 7% in the concentration. Therefore it is concluded that the drying step slightly affects the total carotenoid content of both varieties, offering a good response to the drying temperature and time employed. Comparison (Duncan test, $p < 0.05$) of the total carotenoid content of the two varieties after drying shows no significant differences, so that the final result for this step of processing was the same for the two varieties.

Effect of Grinding on the Total Carotenoid Content. The dry fruit, together with its seed, was ground to give it the characteristics required for consumption: a fine, brilliant, highly colored powder. The oil from the seeds produces this effect and also confers stability by its content of natural antioxidants. The seeds included at grinding are inert material with regard to their color contribution and act as a dilution factor for the total carotenoid content. The percentage of seed in these fruits is around 30−35%, which is the theoretical dilution factor. The real percentage in each batch of paprika analyzed can be estimated in accordance with the fatty acid content of the dry fruit, seed, and resulting paprika (Pérez-Gálvez et al., 1999).

Table 1 includes both the real dilution after grinding and the dilution calculated from percentage of seed. The loss of total carotenoids found is always higher than that due to the dilution effect of the seed included, indicating either that the grinding process causes losses of carotenoid or that the producer dilutes the dry fruit even more to standardize the final product.

The paprikas obtained from the variety *Jaranda* show significant differences in their total carotenoid content, while those from *Jariza* are more uniform. When the results by variety are combined, the Duncan test shows no significant differences, demonstrating that the process yields similar products whatever the source variety.

Biosynthetic/Degradative Balance According to Isochromic Fractions of Pigments after Drying. *Biosynthesis and Degradation*. Table 2 shows the red and yellow carotenoid concentration of the fresh fruit, dry fruit, and paprika, completed by the percentage of variation that these fractions show after drying and grinding steps.

The total pigment content is changed or constant after drying as a function of the fruit's moisture level. This fact indicates that the slow drying at mild temperature allows under-ripening that can modify, by biosynthetic processes, initial concentration of the yellow fraction, the red fraction or both. If fruits, metabolically active, maintain complete their enzymatic capacity, yellow pigments can be generated from their biological precursors until with the synthesis of red pigments. Different metabolic conditions of the fruit can promote the complete synthesis step (from precursors to yellow pigments and finally to red pigments), the first one (precursors to yellow pigments accumulation), or the second one (transformation from yellow pigments to red pigments). Degradation processes take place when the moisture levels do not allow metabolic activity. These reactions of pigment synthesis, and degradation are summed up in Figure 1. The generation of yellow pigments occurs from their biological precursors by biosynthetic processes.

Transformation. The transformation step takes place from the yellow pigments to red and degradation reactions from yellow or red pigments to uncolored products. To know which process, biosynthesis, transformation, or even degradation, has occurred primarily it is necessary to break the total carotenoids down into isochromic

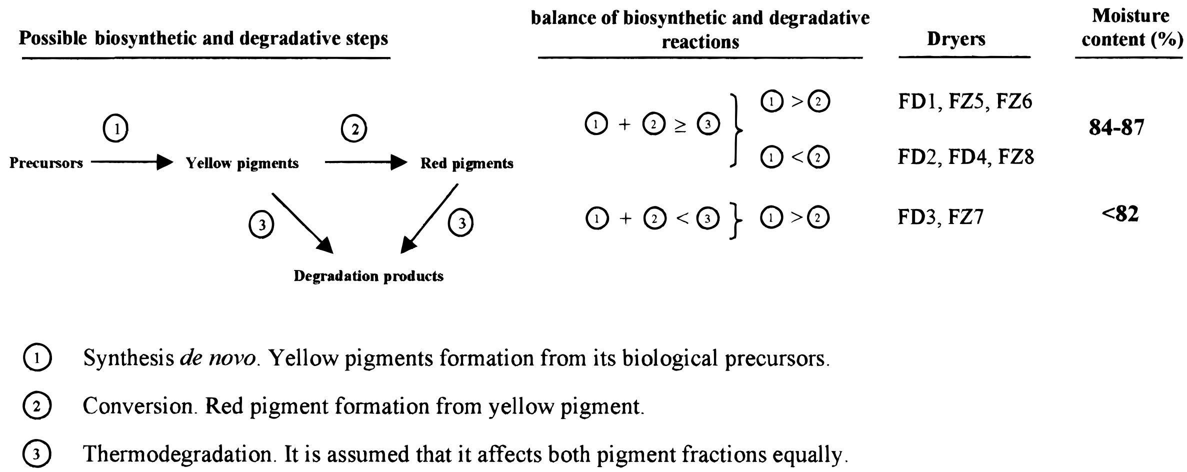

Figure 1. Total or partial biosynthesis and degradation reactions during the pepper fruits drying. Balance and yield process.

fractions. Therefore, as can be seen at Table 2, in the dryers where an increase of the yellow fraction is observable (always greater than the increases of the red fraction, that may even decrease), the first biosynthetic stage occurs primarily. This leads to an accumulation of yellow pigments and causes an increase of the total carotenoid content. Such a situation occurs easier in fruits with high moisture content (85−88%), indicating under-ripeness of the fruits (although in dryer FZ8 a slight decrease is observed). The fruits have a longer lasting period of metabolic activity related to their high level of moisture, so ripening continues during drying.

Only in one case (FD4) was there a net increase in red pigment concentration, accompanied by a slight decrease of the yellow fraction. The red pigments, final substances of the carotenogenesis enhanced their concentration while the enzymes responsible for their formation were active. In this particular case, the biosynthetic processes also lead to an increase of total carotenoid content, caused by the transformation of yellow pigments into red pigments being the most active step. In some dryers, although degradative reactions affect both pigment fractions, higher losses in red pigments concentration are reached because the final yellow pigments concentration is the result of the balance between the initial synthesis and degradative reactions.

In conclusion, the drying conditions used to fruits of high moisture content work together to induce biosynthetic processes during the step in which these could show metabolic activity, so that the end of the process shows a good balance or even gains. It is advisable to shorten the drying step of fruits with low moisture content.

The comparative study (Duncan test, $p < 0.05$) of the concentrations of red and yellow isochromic pigment fractions of the two varieties after drying indicates that there are significant differences occur only in the red fraction, of lower mean concentrations in the variety *Jariza* (3697.1 mg/kg) than in *Jaranda* (4007.5 mg/kg).

Effect of Grinding on the Isochromic Pigment Fractions. Table 2 includes the percentage of decrease for each isochromic fraction after grinding. In both fractions, this should in theory coincide with the estimated percentage of seed. In the red fraction it does and is similar to the dilution loss. In the yellow fraction, however, there is no such similarity, and the increase with respect to the theoretical dilution is attributed to loss by oxidation during grinding. The heat generated by the hammers of the mill raises the temperature, which may reach 80 °C causing a greater thermoxidative effect on the yellow pigments. Several authors attribute their lower stability to the polyunsaturated fatty acids with which they are esterified and are thus more susceptible to oxidation (Daood and Biacs, 1986; Mínguez-Mosquera and Hornero-Méndez, 1994a).

Table 3. Effect of the Steps in Paprika Processing on the Parameters R/Y and R/T

	R/Y[b]			R/T		
code[a]	fresh fruit	dry fruit	paprika	fresh fruit	dry fruit	paprika
			Variety *Jaranda*			
FD1	1.224[c]	1.094	1.415	0.550	0.522	0.586
FD2	1.225	1.345	1.404	0.556	0.577	0.584
FD3	1.398	1.263	1.382	0.583	0.558	0.580
FD4	1.220	1.423	1.273	0.549	0.587	0.559
			Variety *Jariza*			
FZ5	1.191	1.167	1.288	0.543	0.538	0.563
FZ6	1.274	1.068	1.406	0.560	0.516	0.584
FZ7	1.454	1.358	1.515	0.593	0.576	0.602
FZ8	1.313	1.359	1.459	0.567	0.576	0.593

[a] FD, Jaranda farm. FZ, Jariza farm. [b] R = red fraction. Y = yellow fraction. T = R + Y. [c] Mean values of six determinations.

Control Parameters of the Process. The variables which determine changes in the fruit carotenoid composition considered have been fruit moisture, and duration and temperature of drying. As carotenoid concentration is what determines product quality, it is of interest to have parameters which are indicators of the state of the fruits after each step of the process.

The ratio between isochromic pigment fractions (R/Y) and the ratio between red and total pigments (R/T) should remain invariable if the process does not affect the two fractions, or if it affects in the same extent. Loss of constancy in the ratio will indicate a selective effect of the process on one of them. Table 3 shows the data of the two parameters for the two varieties studied and at each step of the process. In fresh fruit of the two varieties studied, the initial ratio R/Y varies between 1.191 and 1.454. The higher values indicate totally ripe fruit in which the biosynthetic process (high content in red pigments) has finished, and lower values indicate fruits that have not yet ripened fully. In fact, the data of high R/Y correspond to low moisture values, so it seems that the level of hydration and the parameter R/Y might indicate degree of ripeness.

The drying process slightly changes the ratio R/Y, depending on the dominance of or competition between biosynthetic and degradative processes in the fruits which are still metabolic active. The effect on one or both pigment fractions depends on the state of ripeness. In the variety *Jaranda*, this ratio rises and falls indistinctly; in *Jariza* it maintains or decreases. In the final paprika, the ratio R/Y increases in all cases and in both varieties (except for a decrease of 10.5% for the farm FD4), indicating a loss of yellow pigments during grinding. Changes in these parameters during the processing of other varieties are much more clear. In *Bola*, the ratio R/Y varies from 1.28 in fresh fruit to 2.96 in dehydrated fruit, but remains constant in the variety *Agridulce* (1.76) (Mínguez-Mosquera and Hornero-Méndez, 1994b). The fact that these parameters change little while paprika is being obtained, shows the appropriateness of the process, and of the usefulness of a slow, mild drying, to conserve the carotenoid content, which is the most important quality in pepper fruits.

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