Crystallization of Waxes in Sunflowerseed Oil: Effects of an Inhibitor

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The mechanism of action of a commercial inhibitor on the crystallization of waxes present in sunflowerseed oil was analyzed. The results showed the inhibitor favored nucleation, leading to a decrease in the amount of waxes available for the growth of the crystals already formed. The inhibitor decreased the crystal size, increased the number of crystals and possibly caused slower crystallization of waxes.

KEY WORDS: Crystallization, inhibitor, sunflower, waxes.

Sunflower seed oil obtained either by pressure or solvent extraction is refined to eliminate interferring substances (1-4). The conventional process includes elimination of gums, neutralization, bleaching, deodorization and winterization; different sequences of these processes are possible. During winterizing, the oil is cooled to eliminate substances that crystallize at low temperatures to cause turbidity (5). The precipitating fraction contains mainly waxes that were present in the husk in concentrations ranging from 1.5-3% and that remain in the oil during removal of the husk (6-8). Contents of waxes in crude oil vary between 0.02 and 0.35% (9-12). Some authors have reported values as high as 1% (3,13). Waxes and other substances producing turbidity are extracted by centrifugation or filtration of the oil. Substances that delay or prevent crystallization of waxes such as phospholipids, which are natural inhibitors of the crystallization process (14-16), can also be employed.

The aim of the present study is to analyze the mechanism of action of a commercial inhibitor on the crystallization of waxes present in sunflowerseed oil.

MATERIALS AND METHODS

Refined sunflowerseed oil, to which known amounts of wax and crystallization inhibitor were added, was used throughout this study. Waxes were obtained from the sediment found at the bottom of storage tanks and were purified by successive extractions with hexane and petroleum ether, followed by chloroform and isopropanol. Waxes were dissolved in the corresponding hot solvent and then separated by centrifuging at 3,000 × *g* for 30 min at 5°C. Purity was determined from melting points by differential scanning calorimetry (DSC). A DSC thermogram showed an endotherm at T_{max} = 74°C. This value was within the order reported by Leibovitz and Ruckenstein (17), who found a range of melting points within 76-77°C for the crystals of pure sunflowerseed waxes.

The crystallization inhibitor, which is commercially available, was prepared by esterification of fatty acids and polyglycerol. Fatty acid composition was: C12:0, 20%; C14:0, 2.8%; C14:1, 2.5%; C16:0, 26.5%; C18:0, 29.6%; and C18:1, 17.8%, as determined by gas chromatography with a Hewlett-Packard 5890A and a 10% SP 2330 column on WAW 100/120 chromosorb (Hewlett-Packard, Palo Alto, CA).

All experiments were performed with refined sunflower seed oil, containing 0.005% wax, to which known amount of waxes and inhibitor were added. Concentrations of wax and inhibitor determined by weighing were 0.15% and between 0.0125 and 0.1%, respectively. A Leitz microscope model Ortholux II (Ernest Leitz Co., Wetzlas, Germany) with a controlled temperature plate was used to determine temperature and crystal formation times, crystal size and growth (18). In the crystallization tests, the samples were melted at 100°C, placed on the slide immediately and then cooled exponentially following the equation:

$$T = T_f + (T_i - T_f)\, e^{-kt} \qquad [1]$$

where T_f is the temperature of the coolant, T_i is the initial temperature and k is the constant, which represents the heat transfer from the coolant to the sample.

Thermal histories were characterized on the basis of k, T_r (temperature of cooling chamber), T_f and T_i (18). Stage temperature was controlled by means of a Lauda TUK cryostat (Werklauda, Königshopen, Germany), which was filled with a mixture of ethyleneglycol/water (3:1), and a stage transformer (maximum load 0.7 A). A fine thermocouple of Cu-constantan of 0.05 mm diameter connected to a two-channel Gilson Potentiometer (Gilson france S.A., Villiers le Bel, France) was placed in the sample between the slides. A water-ice bath was used as reference. Times and temperatures when wax crystals appeared were recorded. Photographs of the crystals were taken with a Leitz-Vario-Orthomat during crystallization and again 1 hr later. Morphology and size distribution were also determined. Crystal size was arbitrarily considered as the longest dimension of the crystal. Observations were made under polarized light.

RESULTS AND DISCUSSION

Times and temperatures at which crystals appear, as well as their growth rate, were determined under the same cooling conditions in order to study the mechanism of action of the inhibitor on the crystallization of waxes.

Table 1 shows that the lower the final cooling temperature, the lower the times and temperatures at which the wax crystals appeared, since this corresponds to the highest supercooling attained. Moreover, the presence of the inhibitor did not significantly alter the time at which the crystals appeared.

Figure 1 shows the number of crystals per field at different inhibitor concentrations. An increase in the number of wax crystals formed was observed at higher concentrations of the inhibitor. These results suggest that the inhibitor had the ability to nucleate during cooling. Furthermore, since the relationship between the number of

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TABLE 1

Effect of Inhibitor Concentration on Time and Temperature at which Crystals Appear from Sunflowerseed Oil Containing 0.15% Wax

Inhibitor (% by wt.)	t_n[a] (sec)	T_n (°C)	T_f (°C)	$K.10^{-3}$ (sec^{-1})	T_r (°C)
0.100	178	27	21	11.7	15
0.050	174	27	20	9.1	15
0.025	167	27	20	9.3	15
0	162	28	21	12.0	15
0.100	125	23	13	12.0	5
0.050	157	24	13	10.5	5
0.025	145	25	12	10.0	5
0	128	24	12	11.9	5

[a] t_n, Time at which crystals appear; T_n, temperature at which crystals appear; T_f, temperature of coolant (cooling fluid); K, constant representing heat transfer from cooling fluid to sample; and T_r, temperature of the cooling chamber.

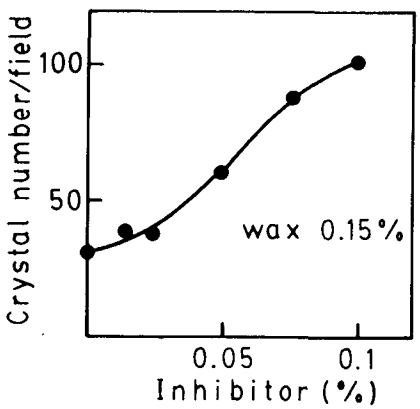

FIG. 1. Variation of number of crystals per field as a function of inhibitor concentration.

crystals formed with or without the inhibitor remained constant under different conditions of cooling, both the inhibitor and the waxes required similar supercooling conditions for nucleation to take place.

The effect of the concentration of the inhibitor on crystal size distribution, as determined 1 hr after the beginning of crystallization, can be observed in Figure 2. A shift towards smaller crystals occurred as the inhibitor content of the oil increased. These results are in agreement with those reported with phospholipids (17).

Figure 3 shows the mean size of crystals obtained under the same cooling conditions as a function of the inhibitor content of the samples. Crystal size was reduced by approximately 60% by the addition of 0.0125% inhibitor. Larger decreases (to approximately 80%) occurred as the inhibitor concentration increased.

With wax concentrations of 0.15%, two fractions of crystals can be considered—one in which the size was higher than the mode (6.4 μm), and the other with sizes lower than that value. When the inhibitor was present, the first fraction disappeared, as seen in the first part of Figure 3, and the decrease in size in the second fraction was much less marked. A constant size was not reached with the inhibitor concentrations we employed.

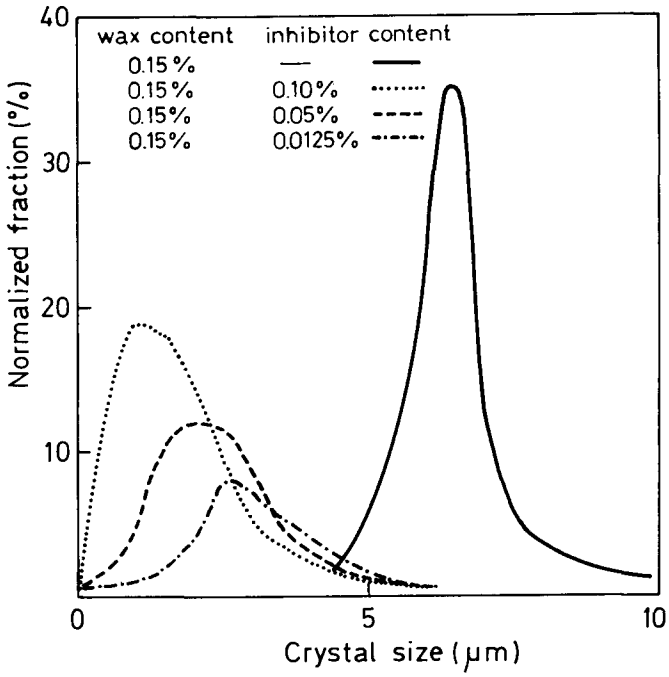

FIG. 2. Distribution of crystal size in the presence of different inhibitor contents.

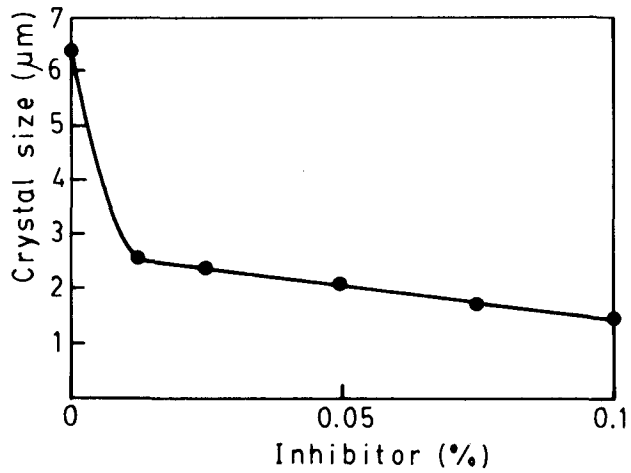

FIG. 3. Effect of inhibitor content on crystal size.

Figure 4 represents the variations in the size of wax crystals as a function of the, time of crystallization. Growth curves did not go through the origin. This can be explained by taking into account that a certain period of time is required for the formation of stable nuclei, from which growth starts. It should be pointed out that the time at which crystals appear is not equivalent to the time of nucleation, since the crystals must reach a minimum size of 2 μm to be detected. Figure 4A shows the kinetics of growth corresponding to a 0.15% wax solution for different crystal sizes, the growth rates being approximately the same in all cases. Figure 4B represents the behavior

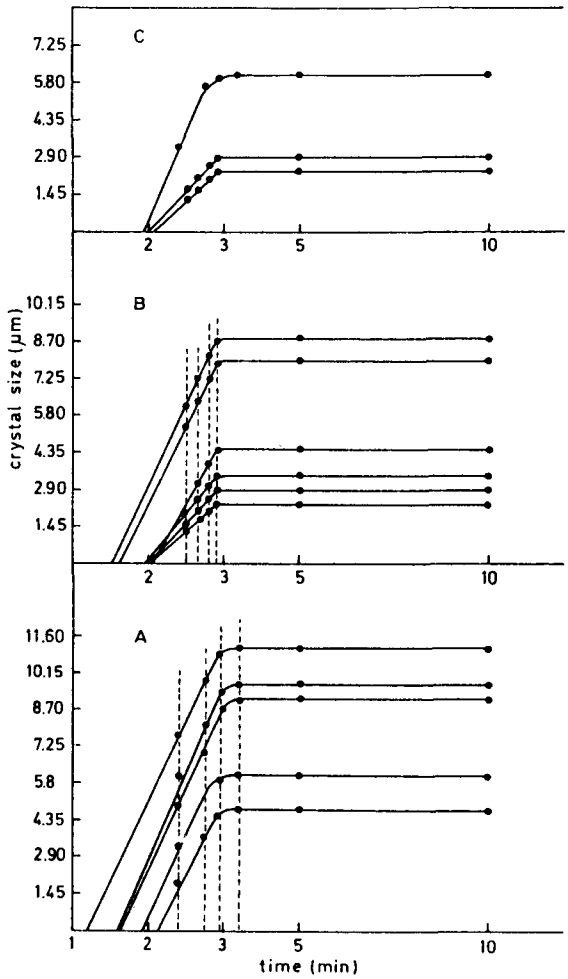

FIG. 4. Kinetics of growth of crystals of different sizes. A: Solution of 0.15% wax in sunflowerseed oil. B: Solution of 0.15% wax and 0.05% inhibitor in sunflowerseed oil. C: Solution of 0.15% wax and 0.15% wax plus 0.05% inhibitor in sunflowerseed oil.

of a solution containing 0.15% waxes and 0.05% inhibitor. Growth rate increased as crystal size increased for crystals under 4 μm in size, whereas for larger crystals growth rate was no longer a function of the crystal size. Growth rates were the same whether the inhibitor was present or not.

The growth rates of crystals of a size similar to the crystal distributions of 0.15% wax solutions were compared to those corresponding to waxes containing 0.05% inhibitor. A decreased growth rate in the presence of the inhibitor took place at these crystal sizes.

From our results it can be concluded that the inhibitor favored nucleation, thus obtaining large numbers of crystals in very short times. This leads to a decrease in the amount of wax available for the growth of the crystals already formed; therefore, these crystals were smaller than those obtained when the inhibitor was absent. The latter could favor nucleation by decreasing the energy of activation needed for the formation of crystal nuclei. This nucleating action can be interpreted in terms of the inhibitor itself providing the crystal nuclei; this would lead to heterogeneity of the resulting crystals.

Moreover, these results show that the decrease of crystal size in the presence of inhibitor was in the range of 70–80%, and that the number of crystals formed was twice as large. A lower amount of wax crystallized in the presence of the inhibitor, which implies either an interference with crystal growth or the presence of a kinetic problem. Data of growth rates suggest an interference by the inhibitor on the crystal growth, probably through a mechanism similar to that of phospholipids (17). The kinetic interference, however, cannot be ruled out, since groups of two to three crystals were formed in the presence of the inhibitor (results not shown). These groups of crystals were homogeneously distributed very close to one another, which would lead to slow diffusion of waxes. Also, a low concentration of wax existed, as a consequence of the formation of a large number of nuclei. This would lead to slow crystallization with complete crystallization occurring after longer times.

Use of this inhibitor would not only induce a reduction of wax crystal size, but would also provoke a depletion of waxes. Both effects would contribute to preventing turbidity during storing and marketing of sunflowerseed oil.

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[Received December 12, 1990; accepted June 11, 1991]