Processing of Cottonseed.

II. Factors Determining the Distribution and Properties of Pigments in Products Prepared by Solvent Extraction

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Although direct solvent extraction has been recommended for many years (1-7) as a method for processing cottonseed for oil, it has only recently been applied on an industrial scale in the United States and does not appear to be widely practiced in other countries. The successful application of this method for processing soybean (8), as well as current trends toward the development of continuous processes requiring a minimum use of labor, have stimulated interest in the direct solvent extraction of cottonseed.

For many years meals obtained by forepressing oilseeds in hydraulic and continuous screw presses have been extracted with solvents to recover their residual oil. Cottonseed oils produced in this manner are reported (6, 7) to be less highly pigmented than the forepressed oils. Olcott (9) has recommended cooking of cottonseed prior to solvent extraction in order to obtain meals suitable for livestock feed. He reported that oil extracted with petroleum naphtha from precooked seed is comparable to expressed oils with respect to refining loss and color. However, since precooking of cottonseed has been reported (9) not to increase the yield of extracted oil, it would appear to be a superfluous operation. Cooking of cottonseed also denatures the protein, partially or completely destroys other thermally unstable, nutritive constituents of the seed, and darkens the meal so that it is unsuitable as a source of protein for industrial use. Consequently, with the development of methods for the elimination or control of color in the products, direct solvent extraction of uncooked cottonseed should offer many advantages.

Control of color is not the only problem which is encountered in the successful processing of cottonseed by direct solvent extraction, but it is an important one. The unique system of pigments in the cottonseed kernel not only differentiates it from other commercial oilseeds, but it is also responsible for many of the difficulties encountered in adapting the usual solvent extraction procedures to the processing of this seed. Recent publications (6, 7, 9, 10, 11) which have reviewed the advantages and disadvantages of solvent extraction of cottonseed have been concerned principally with the difficulties involved in avoiding or removing color in the extracted oil, and with the destruction or removal of pigments of the meal.

Pigments of Cottonseed

The pigment content of cottonseed is unusually high and variable. Gossypol, the principal pigment of cottonseed, has been found (12) to occur in concentrations as high as 2% of the weight of the kernel. Gossypurpurin (12, 13, 14) has been found to constitute as much as 0.055% of the weight of the kernel. Gossyfulvin (15, 16) has been detected in low concentrations in only a few samples of cottonseed which had been stored at high moisture content. The concentration of the yellow, oil-soluble carotenoid in cottonseed has been reported (17, 18) to vary from 0.096 to 0.219% of the weight of the kernel. Another yellow, oil-soluble pigment has recently been detected in cottonseed (12), but it has not yet been isolated in a pure state so that the constants necessary for determining its concentration in the seed have not been established. Gossycaerulin (19) has been detected only in cooked cottonseed.

The gossypol content of different samples of cottonseed may vary many-fold and has been shown (20, 21, 22, 23, 24) to be dependent upon a number of factors, including species, variety, location and year of growth, maturity, and length and conditions of storage of the seed. The concentration of cottonseed pigments other than gossypol in the seed has not yet been investigated exhaustively, but their variation seems to be of approximately the same order as that of gossypol and to be dependent upon as many factors.

Most of the complex polyphenolic pigments of cottonseed readily undergo alteration. Gossypol is so unstable after its isolation from other seed components that simple derivatives can be prepared only under mild and carefully controlled reaction conditions. Solutions of gossypol are stable for only a very short time even in relatively inert solvents. The other related pigments of cottonseed, gossypurpurin, and gossyfulvin are even more unstable than gossypol whereas the carotenoid pigment and the yellow extraglandular pigment are relatively stable.

The yellow, oil-soluble pigment has been shown (12) to occur in solution in the oil in the extraglandular tissue of the cottonseed. It also appears probable (12) that the carotenoid pigment occurs in the oil in the extraglandular tissue. On the other hand, all of the gossypol and gossypurpurin of the cottonseed kernel has been shown (12) to be segregated in the pigment glands. Since the last mentioned pigments are the most abundant, the most deeply colored, and the most unstable of the pigments of the kernel, most of the dark color observed in solvent-extracted oil and meal can be attributed to them or their decomposition products.

Behavior of the Pigment Glands

The earliest investigators (25, 26, 27, 28, 29) of the anatomy of the cottonseed noted the occurrence of the pigment glands and reported observations of their
reaction with water. Carruth (30) suggested the probable importance of these glands in the transformation which the pigments undergo during processing of cottonseed. Nevertheless, for many years the role of the pigment glands was overlooked, but the properties of the pigment gland have recently been shown (12, 14, 22, 31, 32) to determine to a considerable degree the behavior of the pigments during processing of cottonseed by either expression or solvent extraction methods.

The pigment glands, which contain all of the gossypol and gossypurpurin of the seed, possess a thick, strong, resistant wall which presumably protects the gland contents from direct contact with the components of the surrounding tissue in the intact seed. These glands possess such high mechanical strength that in seed of normal moisture content only a small fraction of them are ruptured under the pressures and shearing stresses to which they are subjected during rolling or grinding of seed preparatory to pressing or extraction. Consequently, the pigment glands containing the intraglandular pigments remain in the meal unless processing conditions are such as to rupture the gland walls.

The gland walls have been shown (12, 31, 32) to be resistant to the action of most liquids except water and a few water-miscible organic liquids of low molecular weight. Contact with water produces instantaneous rupture of the walls and the expulsion of the gland contents. The sensitivity of the glands to moisture is so great that they are affected by traces of moisture and their sensitivity increases as the temperature is increased (14).

Methanol, ethanol, isopropanol, acetone, and 1,4-dioxane have been shown (12, 31, 32) to rupture the pigment gland wall. Although the action of these solvents is fairly rapid, it is slower than that of water, and the speed with which aqueous mixtures of these solvents attack the pigment glands is proportional to the amount of water in the mixture. The pigment glands are ruptured most rapidly by mixtures containing such high proportions of water that the pigments are expelled from the glands in streams of suspended particles. Addition of more of the organic solvent accomplishes solution of the suspended pigments. Thus, rapid extraction of both the oil and the pigments is most efficiently obtained by first treating the seed with water or with a dilute aqueous solution of the organic solvent, and then adding organic solvent in sufficient quantity to dissolve the pigments and oil.

Organic liquids other than the aforementioned water-miscible solvents have been found (12, 31, 32) to fall into two categories with respect to their activity toward the glands: (a) those which completely extract the gossypol from the glands after contact for 24 hours or less, and (b) those having little or no effect on the glands even after prolonged contact. The active group includes diethyl ether, chloroform, and probably other chlorinated hydrocarbons. The slow extraction of the intraglandular pigments effected by these solvents was shown (12, 32) to be the result of their normal, slight content of moisture. Thus, the active group differs from the inert group only by the affinity of the solvents of the former group for water, and their consequent tendency to contain varying amounts of moisture. In the presence of moisture, hydrocarbons which normally do not affect the pigment glands were found (12) to extract appreciable amounts of the intraglandular pigments.

The amount of pigments extracted by organic solvents other than the water-miscible group was found (12) to be determined not only by moisture during extraction, but also by the original moisture content of the seed kernels when they were rolled or ground preparatory to extraction.

Solubility of Cottonseed Pigments in Mixtures of Oil and Solvent

The carotenoid pigment of cottonseed has been reported (17, 18) to be soluble in cottonseed oil, as well as in both polar and non-polar organic solvents such as petroleum naphtha and aqueous ethanol. The recently detected, non-acidic, yellow, extraglandular pigment of cottonseed (12) is soluble in cottonseed oil and in extracts of the oil obtained with a large number of organic solvents. Consequently, all solvent-extracted cottonseed oils will contain all of these two pigments which are present in the original seed.

Gossypol is very soluble in cottonseed oil, acetone, and 1,4-dioxane. Although it is of limited solubility in diethyl ether, benzene, chlorinated hydrocarbons, methanol, ethanol, and isopropanol, most of the gossypol from ruptured glands is dissolved in the relatively large volumes of these solvents which are normally employed for extraction of the oil.

Gossypurpurin (13) is insoluble in cottonseed oil and in most of the usual organic solvents, except acetone and 1,4-dioxane. It is slightly soluble in diethyl ether and chloroform, and its solubility appears to be enhanced by the presence of preponderant amounts of gossypol. Consequently, these solvents, as well as acetone and 1,4-dioxane, will usually extract all of the gossypurpurin from ruptured pigment glands. Because of the very limited solubility of gossypurpurin in methanol, ethanol, and isopropanol, these solvents extract all of the gossypurpurin only from seed containing very small amounts of this pigment. Since gossypurpurin is very unstable in all organic solvents except chloroform at relatively low temperatures, extracts of cottonseed usually contain gossypurpurin in the form of its yellow decomposition product.

Pigments of the Hulls

The characteristic red-brown color of cottonseed hulls has been reported (33) to be largely attributable to the presence of a stable xylan, which is extracted only by 2% alcoholic sodium hydroxide (33) and to some extent by water (34). The isolated pigment of the hull is insoluble in cold absolute ethanol, diethyl ether, and acetone (33).