Competitive Adsorption of Lutein from Soy Oil onto Rice Hull Ash

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Adsorption of lutein from crude soybean oil onto acid ash prepared from rice hulls displayed Freundlich-type isotherms. Different isotherms were obtained depending on the amount of adsorbent used. Addition of isopropanol to the miscella and water deactivation of the adsorbents decreased lutein adsorption. These observations suggested competitive adsorption based on polarity. Triglycerides were adsorbed to a greater extent than lutein, probably due to their larger concentrations. These findings parallel the adsorption behavior of silicic acid.

KEY WORDS: **Adsorbents, adsorption, isotherms, lutein, soy oil.**

Removal of the carotenoid, lutein, is an important step in refining crude soybean oil, enabling the production of a light-colored oil acceptable to consumers. This is accomplished by heating the crude oil with natural or activated montmorillonite clays at high temperatures under reduced pressure. The lutein is removed by adsorption onto the clays (1). Hassler and Hagberg (2) showed that bleaching of vegetable oils follows a Freundlich isotherm, which indicates that pigments are adsorbed as a monolayer. These isotherms have been used to describe the commercial bleaching of vegetable oils (3) and more recently bleaching of rubber and melon seed oils (4).

Feuge and Janssen (5) used isotherms to explain the bleaching of cotton seed oil in hexane miscellas at room temperatures. Proctor and Snyder (6) demonstrated that lutein can be adsorbed at ambient temperature and pressure onto silicic acid from a soy oil/hexane miscella, according to a Freundlich isotherm. A different isotherm is obtained for each adsorbent dose used. Lutein binding by silicic acid is influenced by the polarity of other oil constituents. Addition of polar solvents, such as isopropanol (IPA) and water, inhibits lutein adsorption. It was suggested that competition between oil components for adsorption sites may play a role in the lutein binding process.

There is interest in examining alternatives to conventional bleaching clays. In Nigeria, the bleaching effectiveness of the local Nsu and Okija clays was investigated because montmorillonite clays were unavailable (7). Rice hulls have also been studied as a source of adsorbent because they are an inexpensive, silica-rich by-product of the cereal industry. Proctor and Palaniappan (8) found that a silicate material derived from rice hull ash was capable of binding lutein from soy oil in hexane at room temperature. Acid washing lowered the ash pH and enhanced lutein adsorption by the ash. In contrast, free fatty acids were more effectively bound by the unwashed alkaline ash, and IPA enhanced this adsorption (9).

The purpose of this investigation was to use isotherm studies to examine the mode of soy oil lutein adsorption by rice hull ash, in hexane, at ambient temperature and pressure. The adsorption pattern of soy oil triglyceride was also observed and compared with that of lutein.

MATERIALS AND METHODS

Oil and solvents. Commercially extracted crude soy oil, obtained from Capital City Products (Columbus, OH), was stored at 4°C and used throughout this investigation. Soy oil miscellas were prepared by diluting the oil with hexane. In some cases IPA was added to the miscella to increase its polarity.

Adsorbents. Rice hull ash was prepared according to the method of Proctor and Palaniappan (8). Partially combusted rice hull ash, supplied by Riviana Foods (Houston, TX), was heated at 500°C for 10 hr in a muffle furnace. Forty grams of the alkaline ash was incubated with 1 L of 20% (v/v) sulfuric acid for 5 hr at 20°C, before being washed with an excess of deionized water. This material was designated acid ash.

Lutein isotherms. Lutein was measured by optical absorbance of the miscellas at 445 nm, according to the method of Proctor and Snyder (6). Lutein isotherms were determined by preparing 100 mL of 10, 20, 30, and 40% (v/v) concentrations of soy oil in hexane. The lutein content of the miscella was determined before a known weight of acid or alkaline ash was added. The miscellas were agitated with a magnetic stirrer for 15 min in a closed vessel with 0.5, 1.0, or 2.0 g of adsorbent. The residual lutein concentration was measured, and the amount of lutein adsorbed was found by difference. Triplicate determinations were made. Isotherms were plotted as μmoles of lutein adsorbed per gram of adsorbent vs residual μmolar concentration of lutein. This process was an adaptation of the methods of Proctor and Snyder (6).

Miscella polarity. The isotherm studies were repeated with 1% (v/v) IPA in the miscella, and the results were compared with those obtained without IPA.

Water deactivation. Water deactivation was performed by adding a known volume of water to 100-g quantities of alkaline and acid ash. The flask containing the ash was rotated several times to distribute the moisture, stoppered, and left to equilibrate overnight. Deactivated adsorbents with 5%, 10% and 15% water were studied. Isotherms were prepared with 1-g quantities of deactivated ash and compared with data obtained from the active adsorbent.

Triglyceride isotherms. Triglyceride isotherms were determined by stirring 0.5, 1.0 and 2.0 g of ash with 100-mL volumes of refined soy oil miscellas for 15 min. Triglyceride concentrations were found by measurement of absorbance at 210 nm and, with the aid of a standard curve, calculated as triolein (6).

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RESULTS AND DISCUSSION

Lutein adsorption by alkaline and acid ash was very similar. In order to avoid duplication of figures, only data of acid ash adsorption will be presented. However, differences between the two adsorbents will be mentioned as appropriate.

Figure 1 shows lutein adsorption isotherms obtained with acid ash using various concentrations of soy oil/hexane miscellas. The graph shows pigment binding to display a Freundlich isotherm. A differential dose response was observed. Smaller amounts of adsorbent bound more pigment, on a per gram basis. Multiple isotherms following this trend were also obtained with silicic acid (6).

Isotherms obtained with silicic acid showed a plateau, above which there was no further adsorption. In contrast, no marked plateau was seen with ash isotherms. Increase in adsorption continued with increasing residual lutein concentrations. This may suggest an inequality of adsorption sites. Readily accessible sites may be occupied first, as shown by the dramatic increase in the pigment adsorbed at residual concentration of less than 5 μM. At larger residual concentrations there is an increase in binding, but the rate of increase is very much reduced. Possibly, a number of less accessible sites may also be available. These are only occupied as residual lutein increases and new equilibria are achieved. The variation in accessibility of adsorption sites may reflect the heterogeneity of this material's biological origins. Since silica was incorporated into a cellular structure, pores and crevices in the material may exist. Electron microscopy will be used in the future to investigate the particle structure of the adsorbent.

The effect of 1% IPA on the isotherm behavior of acid ash is in Figure 2. Isopropanol considerably reduced lutein binding. All the isotherms were essentially linear. This may suggest that the IPA occupied the most readily available sites and the remaining sites were equally accessible to lutein. Although the same trend in inhibition was observed in the alkaline ash data, inhibition was not as great. Maximum adsorbed lutein was approximately 0.3 μmoles, 0.4 μmoles and 0.5 μmoles, respectively for 0.5 g, 1.0 g and 2.0 g of alkaline ash. Isopropanol has been observed to compete with soy oil lutein for adsorption sites on silicic acid, thus inhibiting lutein sorption (2). The data suggests competitive adsorption based on polarity, as was found with silicic acid (2).

The effect of water deactivation on the adsorption behavior of acid ash is presented in Figure 3. Water depresses the isotherm in all cases. Acid ash appears to have less ability to bind pigment at a high level of moisture than alkaline ash. This is probably because acid ash adsorbs moisture more efficiently, and subsequently, on incubation with oil, lutein cannot compete as readily for sites. The inhibitory effect of moisture on adsorption is very much more marked than with silicic acid (2). Approximately 90% of the lutein adsorption capacity of the ash is lost when exposed to 15% moisture, as compared to 40% for silicic acid. It would seem that humidity is important when considering storage conditions, and may be as important as

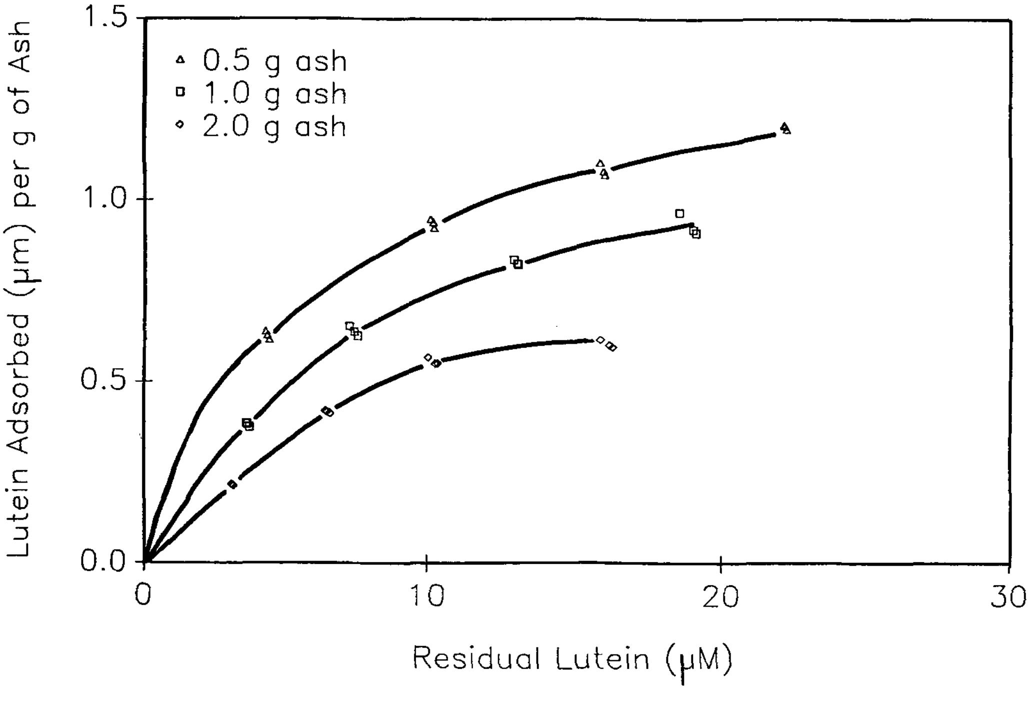

FIG. 1. Lutein isotherms were determined with variable amounts of acid ash by incubation with 100 mL of 10%, 20%, 30% and 40% (v/v) crude soy oil/hexane miscella for 15 min at 22°C. Lutein concentration was obtained by measuring absorbance at 445 nm. Triplicate determinations were made.

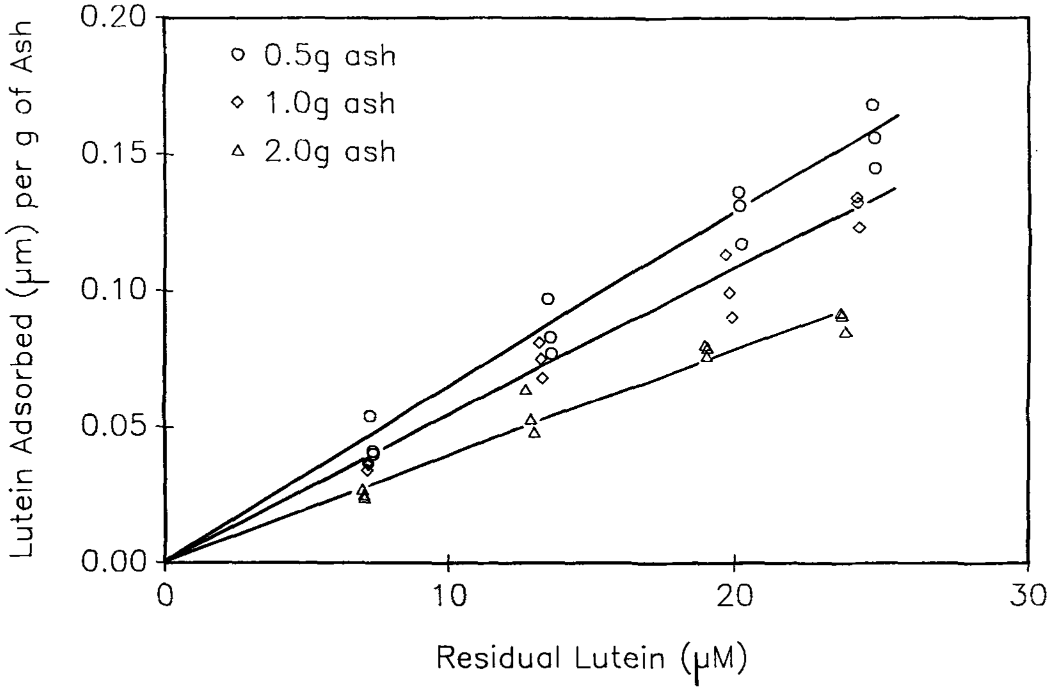

FIG. 2. Lutein isotherms were determined with variable amounts of acid ash and 1% IPA by incubation with 100 mL of 10%, 20%, 30% and 40% (v/v) crude soy oil/hexane miscella for 15 min at 22°C with variable amounts of alkaline ash. Lutein concentration was obtained by measuring absorbance at 445 nm. Triplicate determinations were made.

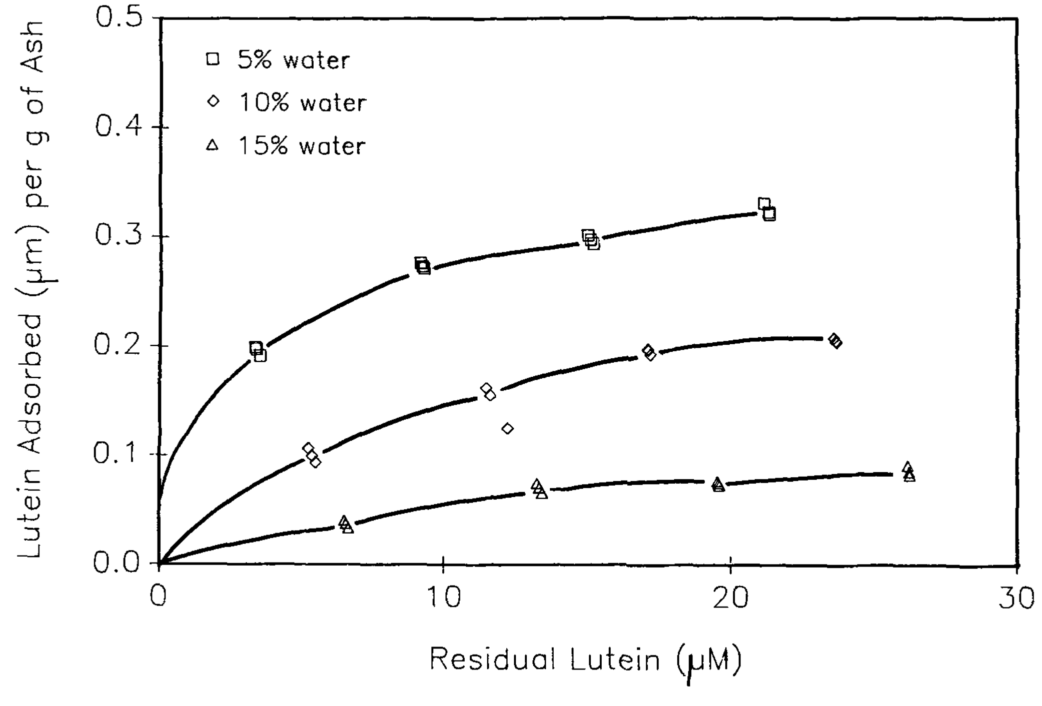

FIG. 3. The effect of water-deactivated acid ash on lutein isotherms was determined by incubation of 1 g of ash with 100 mL of 10%, 20%, 30% and 40% (v/v) crude soy oil/hexane miscella for 15 min at 22°C. Lutein concentration was obtained by measuring absorbance at 445 nm. Triplicate determinations were made.

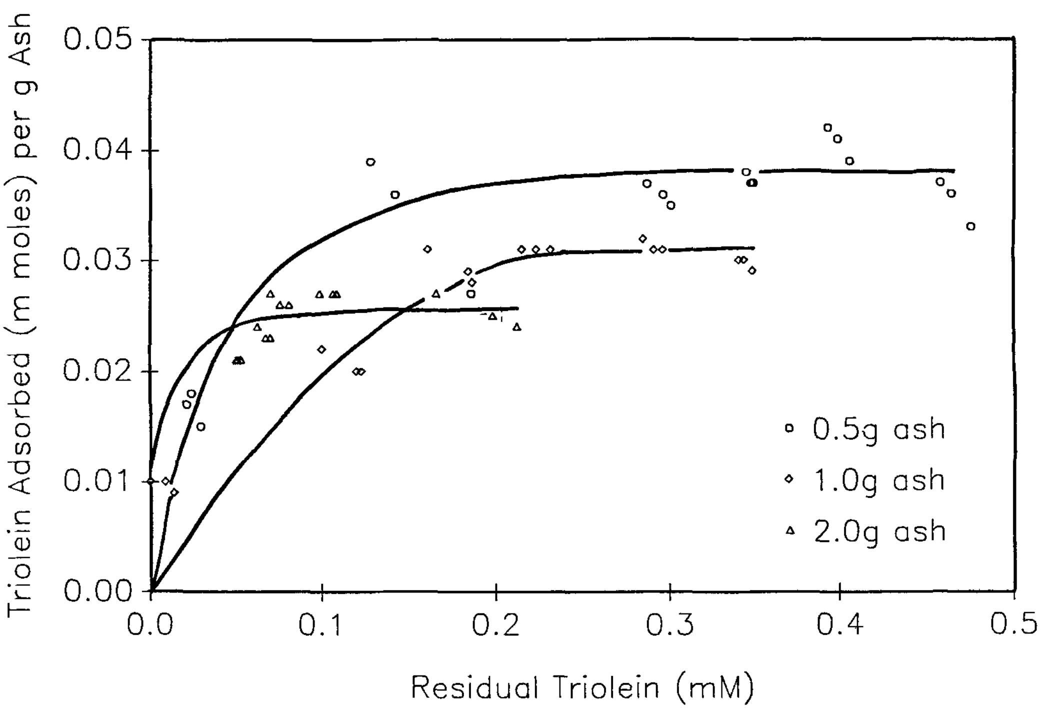

FIG. 4. Triglyceride isotherms were determined with different amounts of acid ash by incubation with 100 mL of refined oil/hexane miscella of different triglyceride concentrations for 15 min at 22°C. Triglyceride concentration was obtained by measuring absorbance at 210 nm, and was calculated as triolein. Triplicate determinations were made.

pH in controlling adsorption by rice hull ash. The studies with IPA and water-deactivated ash both suggest that polarity and possibly hydrogen bonding, rather than chemisorption, could be important in adsorption on rice hull ash. The lack of catalyzed lutein changes on the ash surface (8) supports this view.

Triglyceride adsorption was investigated because of the modification of lutein adsorption by added polar species. Obviously, triglyceride is the predominant competitor in crude oil systems for adsorption sites, and triglyceride adsorption would be an important indicator of refining loss. Figure 4 shows the triglyceride isotherms of acid ash. Smaller doses of adsorbent bound more triglyceride, on a per gram basis, than larger doses. However, triglyceride adsorption was very much greater than that of lutein, possibly due to a much higher concentration of triglyceride. Similar findings were reported for silicic acid (2).

Alkaline ash adsorbed slightly more triglyceride than acid ash for all the dose levels. More triglyceride is adsorbed at relatively low residual concentrations ($<$ 0.05mM) by alkaline ash than acid ash. A 0.5-g dose of alkaline ash adsorbs 42 μmoles of triglyceride with 1 μmole of lutein, per gram. In comparison, a 0.5-g dose of acid ash adsorbs about 38 μmoles of triglyceride with 1.2 μmoles of lutein on a per gram basis. This is considerably less triglyceride bound relative to lutein using silicic acid (2), suggesting that triglyceride molecules have more difficulty in finding accessible adsorption sites on ash than on silicic acid.

The lutein adsorption isotherm behavior of rice hull ash is very similar in principle to silicic acid. However, water deactivation reduces lutein adsorption more in the case of ash than silicic acid. In addition less triglyceride is bound by ash than silicic acid.

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