

Detection of Chromatographic Spots in Paper

Many substances which show no fluorescence or phosphorescence at room temperature do so on cooling. Accordingly, it was found that many substances which give no visible chromatographic spots do so if the paper is cooled in liquid nitrogen. The method of detection consists of simply dipping the paper into liquid N₂ and then viewing it in the dark in near-ultraviolet light. Since the paper itself becomes weakly phosphorescent under these conditions, spots can also be detected as dark areas if the substance in question quenches the phosphorescence of the paper. Many substances show an afterglow.

If no liquid N₂ is available, Dry Ice can be used in some cases. The paper is placed in a glass cylinder so that it touches the glass. The cylinder thus formed by the paper is then filled with crushed Dry Ice, and the paper is then viewed without removing it from the container.

This method of detection has the advantage that it does not entail the chemical alteration of the substance to be tested. The color and intensity of the phosphorescence may help in identification.

ALBERT SZENT-GYÖRGYI
*Institute for Muscle Research,
Marine Biological Laboratory,
Woods Hole, Massachusetts*

9 August 1957

Pink Discoloration in Eggs Caused by Sterculic Acid

Pink discoloration of eggs during cold storage is associated with the feeding of cottonseed oil or cottonseed meal to laying hens. This discoloration is a result of the combination of conalbumin of the white with ferrous ion of the yolk to form a pink complex (1). Increased permeability of the vitellin membrane allows diffusion of proteins and water into the yolk. Reverse diffusion of the complex into the white accounts for the pink color of the white. The yolk enlarges and becomes apricot-colored as a result of the blending of the pink color with the natural yellow of the yolk. The causative

agent present in cottonseed has not been positively identified.

Lorenz (2) first observed that the component causing pink discoloration of eggs and the component responsible for the Halphen reaction (3) might be identical. The Halphen test is used to identify cottonseed oil since no other common oils give a positive test. Several uncommon oils, such as Kapok seed oil and *Sterculia foetida* oil, are also known to give a positive test. Kapok seed oil also causes pink discoloration of eggs. The fatty-acid composition of *S. foetida* oil was investigated recently. In addition to minor quantities of oleic, myristic, and palmitic acids, this oil contains a large proportion (70 percent) of an unusual C₁₉ acid called sterculic acid (4). The structure of this acid (I) was first elucidated by Nunn (5). Verma *et al.* (6) disagreed with this assignment and proposed structure II. Other evidence supporting structure I has been presented by Faure (7, 8), who has shown also that pure sterculic acid gives a positive Halphen reaction (Fig. 1).

In this laboratory *S. foetida* oil (9) has been fed to laying hens and found to cause pink discoloration of eggs. When the *S. foetida* oil was hydrogenated sufficiently to eliminate double bonds but not to disrupt the cyclopropane ring structure, it did not cause pink discoloration. Six individually caged White Leghorn laying hens were divided into three groups and fed *ad libitum* for 15 days. The feed was the usual complete ration mixed with or without *S. foetida* oil in corn oil as follows: group I, 1.5 percent corn oil; group II, 1.5 percent corn oil and 0.09 percent *S. foetida* oil; and group III, 1.5 percent corn oil plus 0.09 percent hydrogenated *S. foetida* oil. At the end of 1 month of storage, the eggs were opened. The eggs from groups I and III were all normal, while seven of 11 eggs from group II showed definite pink discoloration.

Sterculic acid is the major constituent fatty acid in *S. foetida* oil. It is known that the other component fatty acids do not cause pink discoloration of eggs. Sterculic acid is, therefore, apparently responsible for the increase in permeability of the vitellin membrane which leads to pink discoloration of eggs (10).

Note added in proof: In a subsequent experiment, pure sterculic acid, $n_D^{28.3}$

1.4632 [$n_D^{24.8}$ 1.4643 (8)], prepared by the urea complex method of Nunn (5) was fed to White Leghorn laying hens. Six individually caged birds were divided into three groups and, in addition to the basal ration, were fed daily by pipette the following: group I, 1 ml of corn oil; group II, 1 ml of corn oil and 0.10 g of pure sterculic acid; group III, 1 ml of corn oil and 0.025 g of pure sterculic acid. After 1 month of cold storage, eggs from groups II and III showed definite pink discoloration, while eggs from Group I were normal, thus confirming the conclusion that sterculic acid causes pink discoloration of eggs.

J. C. MASSON, M. G. VAVICH,
B. W. HEYWANG, A. R. KEMMERER
*Department of Agricultural Biochemistry,
University of Arizona, Tucson,
and Southwest Poultry Experiment
Station, Glendale, Arizona*

References and Notes

1. P. J. Schiabe, S. L. Bandimer, J. A. Davidson, *Poultry Sci.* 25, 440 (1946).
2. F. W. Lorenz and H. J. Almqvist, *Ind. Eng. Chem.* 26, 1311 (1934).
3. *Official Methods of Analysis* (Assoc. Off. Agr. Chemists, Washington, D.C., ed. 8, 1955), p. 475.
4. T. P. Hilditch, *The Chemical Constitution of Natural Fats* (Wiley, New York, 1956), p. 244.
5. J. R. Nunn, *J. Chem. Soc.* 1952, 313 (1952).
6. J. P. Verma, B. Nath, J. S. Aggarwal, *Nature* 175, 84 (1955); *J. Chem. Soc.* 1956, 2550 (1956); J. P. Verma *et al.*, *J. Indian Chem. Soc.* 33, 111 (1956).
7. P. K. Faure, *Nature* 178, 372 (1956).
8. P. K. Faure and J. C. Smith, *J. Chem. Soc.* 1956, 1818 (1956).
9. The oil was extracted from seeds provided through the courtesy of F. R. Amos, director of forestry, Manila, Philippines.
10. This report is Arizona Agricultural Experiment Station technical paper No. 430. This work was supported in part by the National Cottonseed Products Association, Inc.

8 July 1957

Rapid Symptoms in Seedling VII Sweetpotato of a Virus Always Associated with Internal Cork

The rapid mechanical transmission of a virus (1) consistently associated with sweetpotato cork virosis to Scarlett O'Hara morning glory (2) opened up a new approach to study of this virus by reducing the incubation period from about a year on sweetpotato to a week on the morning glory. This discovery posed the possibility of finding a sweetpotato plant that would respond as promptly as the morning glory with distinctive symptoms to the same method of inoculation.

After numerous transmission experiments in 1955, there was no doubt that the same mechanical technique transmitted the virus from sweetpotato or other susceptibles to the various sweetpotato varieties and seedlings under test, but the expression of symptoms in sweetpotato was poor, and the incubation

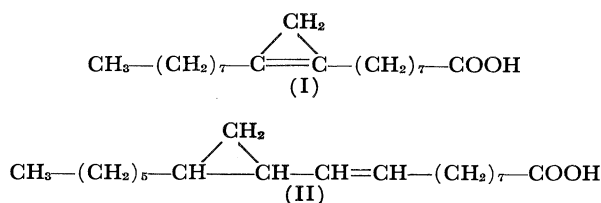


Fig. 1. Structures of sterculic acid proposed by Nunn (I) and by Verma *et al.* (II).