cytoplasm. The granules possibly originate within the nucleus and sooner or later accumulate in the cytoplasm. At any rate, the hypertrophied nucleus eventually breaks down and disintegrates, at which time the granules may be seen distributed throughout the nucleoplasm and cytoplasm of the host cell.

Until the hypertrophied nucleus disintegrates it seems to be held intact or at least to be supported within the nuclear membrane by the fat globules pressing against all sides of the nucleus. Such a situation is not so apparent in the case of *Peridroma* granulosis. In the *Junonia* cells, however, the cytoplasm is almost entirely occupied by the globules of fat (Fig. 1, D, E) leaving very little room for the granules which, in the *Peridroma*, accumulate in large numbers in the cytoplasm of the cell.

In the later stages of the infection there seem to be fewer fat globules in the cytoplasm, which now may be fairly well filled with granules. Finally the cell membranes themselves break down, liberating the granules and other contents of the cells into the body cavity of the host insect. A few chromatin remnants may be seen scattered about in the almost completely disintegrated tissue. It is at this time that the insect dies.

Shortly after the *Junonia* granulosis was found, a similar infection was discovered in specimens of the salt-marsh caterpillar, *Estigmene acraea* (Drury) collected in Albany, California. Whether the disease is caused by the same or another virus is not yet known.

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A Filter Paper "Chromatopile"[1]

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The application of one- and two-dimensional paper chromatography to the separation of small amounts of many kinds of compounds has become a well-established and useful technique (*1, 2, 3, 5, 6*). In general these methods do not handle sufficiently large quantities of materials for isolation and chemical identification. Large columns of starch and other materials have been used satisfactorily for handling larger quantities of mixtures (*4*). Starch columns proved to be unsatisfactory, however, for isolation of certain substances under investigation in this laboratory. As a consequence, a new and simple apparatus and technique have been developed, using a pile of filter paper disks as the absorbing column.

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A diagrammatic sketch of a cross section of the column is shown in Fig. 1. It consists essentially of three parts:

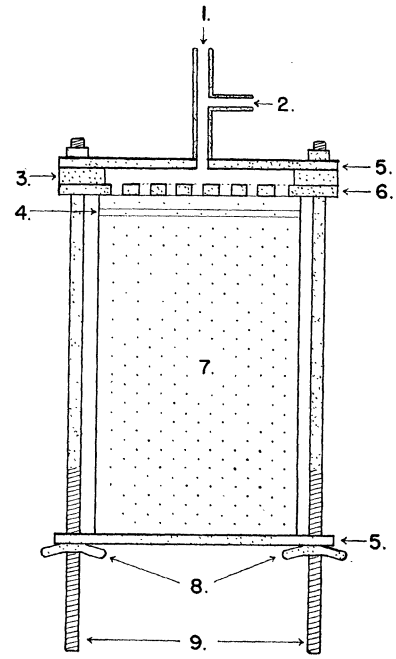

FIG. 1. Diagram of the filter paper pile column. 1. Connection for a rubber tube for filling the siphon. 2. Connection for the siphon tube. 3. Rubber gasket. 4. Filter paper disks containing the sample. 5. Stainless steel plates. 6. Perforated stainless steel plate. 7. Filter paper disk pile. 8. Wing nuts. 9. Bolts at four corners of steel plates.

the pile of filter paper itself (No. 7 in Fig. 1), a clamp for packing the paper tightly, and the solvent distributor at the top of the pile. The metal parts are constructed from stainless steel. The solvent distributor is connected by a siphon of rubber tubing to a 2-l flask containing about 1 l of solvent mixture. The flask is adjusted so that the level of the solvent is even with the top of the paper pile. In operation the column is placed upright on the ends of the clamp bolts in a 9 in. × 12 in. battery jar containing about 200 ml of solvent mixture. The jar is covered, with a hole left for the siphon tube.

The results of a trial run on known compounds will serve to illustrate the utility of the apparatus. Fifty mg each of adenine, tryptophane, phenylalanine, *p*-aminocinnamic acid, and anthranilic acid was dissolved in 20 ml of 0.1 N HCl and the solution was placed in the lid of a 100-mm Petri dish. Disks of 9-cm Whatman No. 1 filter paper were immersed in the solution, allowed to drain, and hung up to dry in air at room temperature. Twenty-five sheets were required to take up the solution and rinsings. To prepare for packing the column, the bottom plate of the clamp was removed and the clamp placed in an inverted position with the solvent distributor down. Forty filter paper disks (9 cm diam) were then placed carefully in the center of the perforated plate, followed by the 25 dried sheets containing the sample. The remainder of the column was made up of a pile of

8 packages of filter paper (9 cm, Whatman No. 1; total, 865 sheets). After careful alignment of the paper disks, the bottom plate of the clamp was placed on the pile and the wing nuts tightened as much as possible without mechanical aid. The column was then placed in the battery jar with the solvent distributor up. The distributor was then filled with the solvent mixture (3 parts *n*-butanol; 1 part *tert*-butanol and 1 part of 0.1 N HCl) with a pipette, and the siphon from the solvent container was connected and filled. After 28 hr the solvent front had descended 13.2 cm. The column was then removed from the jar and the pile taken out in sections. After the approximate locations of the five compounds had been determined by qualitative means, disks were taken 6, 10, or 20 at a time and extracted with hot 0.1 N HCl for adenine, tryptophane, and phenylalanine and hot 0.1 N NH_4OH for the remaining two compounds. Adenine, tryptophane, and *p*-aminocinnamic acid concentrations in the extracts were determined with the Beckman Spectrophotometer. Phenylalanine was determined colorimetrically with ninhydrin, and anthranilic acid fluorometrically with a Coleman Photofluorometer. The total recoveries of compounds from the column sections analyzed were: adenine, 41 mg; tryptophane, 46 mg; phenylalanine, 46 mg; *p*-aminocinnamic acid, 44 mg; and anthranilic acid, 46 mg. Thus, without considering losses in sheets removed for qualitative tests, 223 mg was recovered from the original 250 mg in the mixed sample. The distribution of the compounds on the column is shown in Fig. 2. As noted in the figure, the solvent traveled from the last sheet of the mixed sample through 730 filter paper disks or a distance of 11.8 cm. In the case of the sharpest peak (phenylalanine) more than 95% of the compound recovered was found in 36 filter paper disks representing a thickness of a little less than 6 mm. With such a degree of resolution it is clear that the solvent front movement is remarkably uniform in this type of column. Color tests made directly on sample disks showed a slightly more rapid movement of solvent at the edges, but the difference in rate is apparently negligible.

The simplicity and ease of operation of the filter paper pile column provides a practicable method for isolations without requiring complicated equipment. One feature which is most desirable is the ease with which a sample can be removed and incorporated into a new pile. Thus, a section of disks containing a desired compound can be taken out and placed in a new pile for use with a different solvent mixture.

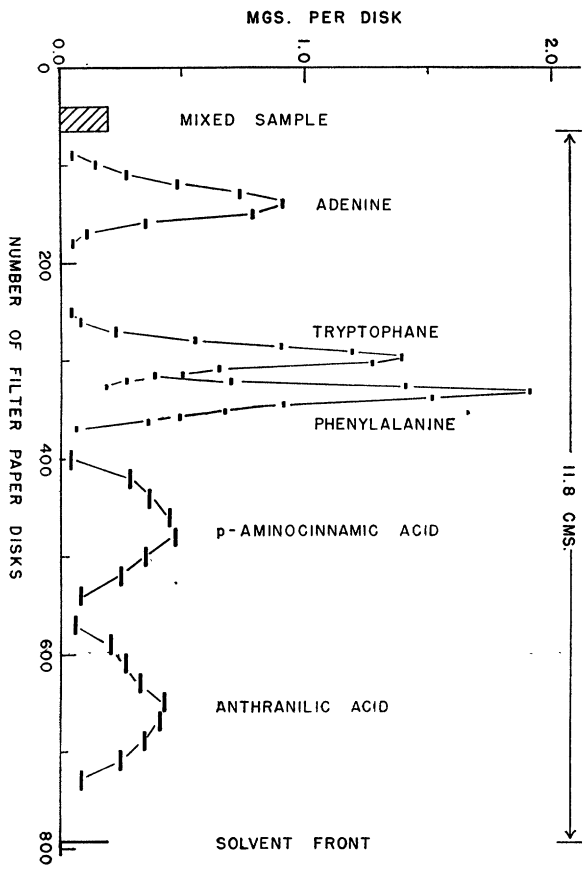

FIG. 2. Results of an experiment showing the separation of compounds of a known mixture. The length of the horizontal bars used for experimental points indicates the number of paper disks extracted for each analytical sample.

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Tryptophane as an Intermediate in the Synthesis of Nicotinic Acid by Green Plants

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The paper by Beadle, Mitchell, and Nyc (*2*) demonstrating that certain selections of *Neurospora* are able to synthesize nicotinic acid from tryptophane suggested to the writer that higher green plants might also have this ability, but only recently was it possible to set up such an experiment. That tryptophane is a precursor of nicotinic acid has now been quite conclusively demonstrated for a number of organisms. Nason (*7*) has very recently demonstrated that corn embryos are able to synthesize this vitamin when supplied with tryptophane and vitamin B_6. Several investigators have shown that animals can also use tryptophane in the synthesis of nicotinic acid (*6, 8, 9*), and there seemed to be no good reason why green plants could not do likewise.

[1] Paper No. 890 from the Department of Botany of the University of Michigan. This investigation has been aided financially by the U. S. Public Health Service, and some of the chemicals have been supplied by Merck and Co.