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.

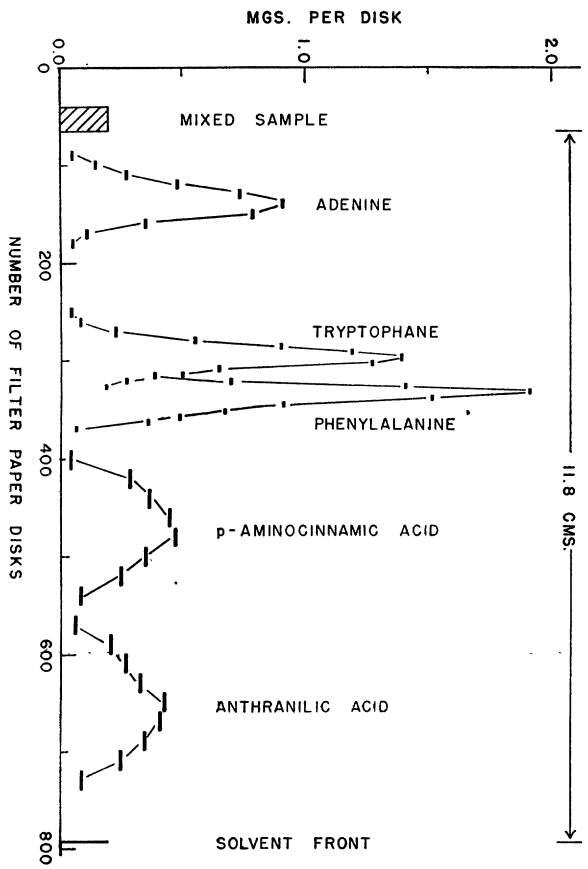

FIG. 2. Results of an experiment showing the separation of compounds of a known mixture. The length of the horizontal bars used for experimental points indicates the number of paper disks extracted for each analytical sample.

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

F. G. Gustafson[1]

Department of Botany, University of Michigan, Ann Arbor

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.

Leaves of cabbage, broccoli, and tomato plants grown in the greenhouse were used as plant material. The tryptophane was supplied to intact leaves through the petioles,

TABLE 1

SYNTHESIS OF NICOTINIC ACID BY GREEN PLANTS FROM TRYPTOPHANE*

Plant	Time in hr	Concentration of tryptophane in %			
		0	0.025	0.05	0.10
Broccoli ..	26	9.22		9.35	9.92
	47	8.18		8.53	
Cabbage ..	70	8.25	9.73	9.92	
	98	7.52	7.98		
	43	6.94		7.49	7.20
	47	6.65	8.25	7.81	7.43
Tomato ...	48	5.14	6.08		
	22	4.44	4.69		
	24	5.81	5.70	6.86	
	47	5.05	6.18	5.99	
	25	5.46	6.35	6.70	
	40	6.36	7.40	8.64	
	48	6.57	6.62	6.80	
	71	6.04			7.10
	48	5.18	6.12	7.60	7.64
	47	4.40	5.70	5.63	
	48	4.75	5.35	5.90	
	48	3.62		4.56	5.10

* The figures denote μg of vitamin/g of fresh plant material.

which were dipped into the solution. Nicotinic acid was determined by the microbiological method using *Lactobacillus arabinosus* as the test organism. The procedure as outlined in *Methods of vitamin assay* (*1*) was followed. The time allowed for synthesis to take place was usually about 48 hr, though longer and shorter periods were also used. Three days were too long, as the leaves wilted and the plants were not at their best; and usually more vitamin was obtained in two days than in one. Concentrations of DL-tryptophane ranged from 0.025 to 0.10 %

TABLE 2

SYNTHESIS OF NICOTINIC ACID FROM TRYPTOPHANE BY GREEN PLANTS IN DARK AND IN LIGHT*

Plant	Concentration of tryptophane in %					
	in light			in dark		
	0	0.05	0.10	0	0.05	0.10
Tomato ..	3.64	4.56	5.10	3.26	3.93	4.71
	4.98	5.72	6.72	4.47	4.93	6.32
	6.07	6.67	6.83	4.65	5.06	5.76
Cabbage ..	6.94	7.49	7.20	6.25	6.69	6.96

* Figures denote μg of vitamin/g of fresh material.

L-tryptophane. These concentrations may seem a little high but since the experiments cannot be run very long it has seemed best to have high concentrations and get a high rate of nicotinic acid synthesis. No attempts have been made to study the relation between concentration of tryptophane and concentration of nicotinic acid obtained. Table 1 gives the results of these experiments.

While only a few experiments were done with broccoli and cabbage and there is not a constant increase in nicotinic acid assayed with increase in concentration of tryptophane supplied to the plants, there is, nevertheless, no doubt that the three plants used have synthesized nicotinic acid from tryptophane, which is all that the writer is attempting to show. The variation in amount of vitamin in different experiments is due, as has been pointed out (*4*), to differences in age of leaves used.

Another observation, which has nothing to do directly with nicotinic acid, should be recorded. When 4–5-in. tips of tomato plants with their young leaves were put in higher concentrations of tryptophane, the young immature leaves showed unmistakable signs of response to growth hormones. They curled up much as they would if the stem had been supplied with indoleacetic or butyric acid. They had evidently synthesized a growth hormone from tryptophane. This has been shown before but only under special circumstances.

The writer has previously shown (*5*) that light influences the synthesis of thiamin and riboflavin by green plants, and unpublished data indicate this may be true also for nicotinic acid. Experiments were therefore set up in dark and in light. Table 2 presents the findings.

Evidently light is not a factor in this synthesis. Recently Bonner (*3*) has outlined a scheme for the synthesis of nicotinic acid from anthranilic acid. He does not, however, state how anthranilic acid might be formed from the products of photosynthesis.

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Relation of Sporadic E Reflection and Meteoric Ionization

V. C. Pineo

Central Radio Propagation Laboratory, National Bureau of Standards, Washington, D. C.

One of the purposes of the radio-meteor research program at the Central Radio Propagation Laboratory of the National Bureau of Standards is to determine to what extent meteoric ionization may be responsible for sporadic E reflections from the ionosphere. Various investigators (*1, 3, 4*) have proposed that sporadic E reflections are caused by ionization produced in the atmosphere by meteors. Observations made up to the present time at this laboratory fail to support this view.

Since radio-meteor observations were begun at this