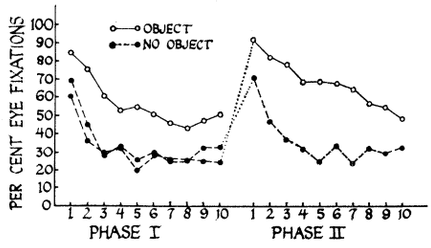

Fig. 1. Duration of eye fixations in successive 5-second intervals.

could be lowered to prevent the subject from seeing into the box. There were three experimental conditions. In the first of these conditions the door was raised and the subject's eye fixations on the empty box were recorded for 1 minute (phase 1). The door was then lowered for 2 seconds and the box remained empty. The door was again raised and the eye fixations to the stimulus were observed for another minute (phase 2). In a second condition, phase 1 was the same as in condition 1, but now when the door was lowered for 2 seconds a novel object (for example, a piece of a broken toy) was placed in the box. As in condition 1, the eye fixations to the stimulus window were observed for another minute. The third condition was restricted to phase 1 only, with an object in the stimulus box.

Three trials were run for each subject in each condition on each of 3 days. Trials were separated by 1-minute periods in which the light in the box to be presented on the next trial was turned on behind the closed guillotine door. Since light leaked around the sides of the guillotine door, the subject could see which window would be presented on the following trial. Position of the window and order of the experimental conditions were randomized.

Curves averaged over subjects are presented in Fig. 1. Analysis of variance was performed for the effects of ten 5-second intervals, the five curves, and subjects. The decrement in eye fixations over 5-second intervals was again observed ($p < .001$). Neither of the other variables interacted with intervals. There was a significant difference among the five curves ($p < .001$), with all subjects showing the highest scores in the Object conditions. The curve for the Object condition of phase 2 was significantly higher than each of the No Object curves. None of the other comparisons between curves was significant. The difference between subjects was significant ($p < .001$).

The results indicate that when a single visual stimulus is presented, the amount of eye fixation on the stimulus is maximum immediately after its presentation, and then it decreases. The orienting response of the eyes to the onset of a stimulus is thus not merely a single shift of the gaze but a series of fixations whose occurrence is graded over a short period of time. This function (*6*), together with other psychophysiological responses (for example, galvanic skin response, alpha blocking), may reflect a general phasic mobilization of the organism for the identification of a stimulus. This process is initiated by the presentation of the stimulus and probably involves arousal mechanisms in the reticular formation of the midbrain. Since the light remains on, a short-term habituation of the eye fixations to the light may also be involved. Retinal adaptation is probably not an important factor. The shape of the function is not significantly affected by individual differences or by characteristics of the stimulus, such as novelty. However, these variables appear to affect the total duration of fixation on the stimulus which is reflected in a variation of the level of the curve (*7*).

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N-Methylmetanephrine: Excretion by Juvenile Psychotics

Abstract. *Three out of 18 psychotic children excreted* N-*methylmetanephrine, a metabolite of* N-*methylepinephrine. It is not clear whether this tertiary amine plays a part in causing some forms of psychosis or is merely a secondary result of mental dysfunction. Urinary excretion of bufotenin and of 3,4-dimethoxyphenylethylamine, each of which has been reported elevated in some adult schizophrenics, was not unusual in these children.*

In recent years there has been an intensive search for biochemical causes of mental illness, stimulated in part by an awareness that an increasing number of diseases involving enzyme deficiency are associated with mental deficiency. The possibility that some forms of mental illness may be due to abnormalities in amine metabolism is an attractive one. Biogenic amines are believed to participate in synaptic functions in the central nervous system. Three tertiary amines, bufotenin, *N*,*N*-dimethyltryptamine, and psilocin, as well as mescaline, produce psychotomimetic effects in man (*1*). Recent reports state that some schizophrenic patients, in contrast to normal subjects, excrete bufotenin (*2*), or 3,4-dimethoxyphenylethylamine (*3*) in their urine. The latter amine, and other *p*-methoxylated phenylethylamines, produce striking neurological effects in animals (*4*). Mammalian tissues contain an enzyme (*5*) which converts, by *N*-methylation, serotonin and tryptamine to their psychotomimetic metabolites, bufotenin and *N*,*N*-dimethyltryptamine. Finally, a recent study by Pollin *et al.* (*6*) has shown that administration of tryptophan and methionine to schizophrenics maintained on a monoamine oxidase inhibitor in some cases provokes an exacerbation of mental symptoms. Tryptophan and methionine, since the latter might act as a methyl donor, could serve as precursors to some of the psychotomimetic amines listed above.

The present report is a preliminary survey of the amines present in the urine of 18 juvenile psychotics. The likelihood of success in detecting biochemical abnormalities causally related to mental illness seemed greater in children than in adults, both since the early onset of mental illness would favor a genetic determination and since psychotic children are less likely than psychotic adults to show the secondary

Table 1. Urinary excretion of amines before and during monoamine oxidase (MAO) inhibition, in micrograms of free base per 100 mg of creatinine. C, control; MAO, after 7 to 8 days' administration of MAO inhibitor.

Subjects	Normetanephrine		Metanephrine		*N*-methylmetanephrine		Serotonin		Tryptamine	
	C	MAO	C	MAO	C	MAO	C	MAO	C	MAO
					Patients					
1	5.0	10.8	4.2	6.3	3.4	4.3	9.1	19.4	16.2	130
2	3.3	8.3	2.7	5.0	2.6	4.7	5.8	17.4	7.1	88
3	1.9		2.6		1.9					
					*Normal children**					
Min.	0.8	6.3			0	0	1.3	4.6	3.9	50
Max.	3.3	13.8			0	0	7.7	10.6	22.0	102

* Figures given for normal children based on 20 subjects before and 6 subjects during MAO inhibition.

effects of stress and disease resulting from prolonged hospitalization. The 18 patients had been variously diagnosed by psychiatrists as having childhood schizophrenia, infantile autism, or juvenile psychosis. The group probably included several different disease entities, but each patient's behavior strongly suggested a psychotic process rather than mere mental defect. All were physically well and living at home when studied; none were receiving tranquilizers.

Urine specimens were collected over 24- to 48-hour periods, in some cases after the patient had been placed on a plant-free diet and given a monoamine oxidase inhibitor. After initial concentration and separation from the other constituents of urine, the amines were detected by two-dimensional paper chromatography (*7*).

N-Methylmetanephrine was detected in the urine of 3 of the 18 psychotic children. This amine was not found in the urines of 20 normal children and 15 mentally defective children studied in the same manner (*7*). Co-chromatography on paper of this compound with authentic *N*-methylmetanephrine resulted in a single spot in four different solvents. In addition, the urinary amine and the authentic reference compound produced identical colors when developed with diazotized *p*-nitroaniline, diazotized sulfanilic acid, and dichloroquinone chloroimide.

Further proof of identification was obtained from the behavior of the compound in ion-exchange column chromatography (*8*). Both the urinary amine and authentic *N*-methylmetanephrine emerged from a 1- by 45-cm column of Amberlite CG–50 at an effluent volume of 78 to 89 ml, when the column was eluted with 0.1*N* pyridine acetate buffer solution, *p*H 6.32, which was pumped through the resin at 10 ml per hour at a temperature of 40°C.

Two of the patients who excreted *N*-methylmetanephrine were brothers, aged 13 and 10 years, who had exhibited psychotic behavior and some degree of mental retardation since early childhood. They were hyperactive, moved about aimlessly with frequent grimacing, and their speech was usually meaningless and characterized by frequent echolalia. Electroencephalograms were diffusely abnormal in both children. The third patient who excreted *N*-methylmetanephrine was a 10-year-old boy of normal intelligence who for 2½ years had exhibited repeated episodes of hyperactivity, destructiveness, and uncontrollable rage, but whose electroencephalogram was normal. None of the three patients showed hypertension or other evidence of pheochromocytoma.

Table 1 presents amounts of *N*-methylmetanephrine and other amines found in the urine of these three psychotic children, and for the two brothers it lists amounts in urine both before and during administration of 200 mg of nialamide daily. For comparison, Table 1 includes the range of excretion of certain amines by 20 normal children, as well as the range of excretion during monoamine oxidase blockade in six of these normal subjects. Experiments in which authentic amines were added to urine showed that 25 to 30 percent of the metanephrines, 50 percent of serotonin, and 70 percent of tryptamine were recovered by the technique used. The actual urinary excretion of *N*-methylmetanephrine was therefore three or four times greater than indicated in Table 1. Monoamine oxidase blockade increased the excretion of this amine but produced no alteration in the behavior of the children. Administration of a strictly plant-free diet did not decrease the excretion of *N*-methylmetanephrine.

The pattern of excretion of identifiable amines in the remaining 15 juvenile psychotics was not remarkable. Even though marked hyperactivity characterized the psychotic behavior of five members of this group, *N*-methylmetanephrine was not detected in the urines of any of them. Their excretion of normetanephrine, metanephrine, serotonin, and tryptamine was not quantitated, but it did not appear to differ significantly from that of normal children. None of the 18 psychotic children excreted 3,4-dimethoxyphenylethylamine or *N*,*N*-dimethyltryptamine in detectable amounts. Bufotenin was found in the urine of two psychotic children during monoamine oxidase blockade, but not in greater concentration than had been found in normal children (*7*).

N-Methylepinephrine is present in the adrenal glands of several mammals, and administration of this amine to the rat results in the excretion of *N*-methylmetanephrine in the urine (*9*). *N*-Methylmetanephrine has been detected in the urine of some patients having pheochromocytomas (*10, 11*). Small amounts of this amine have appeared in the urine of normal subjects, but the excretion ratio of N-methylmetanephrine to metanephrine was approximately 1:10 to 1:20 (*11*). In the present investigation, the urines of three psychotic children contained almost as much *N*-methylmetanephrine as metanephrine, whereas in the urines of normal children the amine could not be detected at all.

Whether *N*-methylepinephrine, of which *N*-methylmetanephrine is a metabolite, may be responsible for mental symptoms in some psychotics remains unclear. Although caution must be used in interpreting the possible significance of *N*-methylepinephrine, its presence in some psychotics is of special interest, since it is an *N*,*N*-dimethylated tertiary amine, and a number of psychotomimetic amines have a similar configuration. On the other hand, the excretion of the amine might be only a secondary function of the psychosis, with anxiety causing increased catecholamine metabolism. In any case, this amine should be searched for in the urine of other psychotic patients (*12*).

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Auxin in Coleus Stems: Limitation of Transport at Higher Concentrations

Abstract. *Previous indirect evidence that the endogenous concentration of plant hormones of the auxin type is controlled by a limitation of transport over the physiological range of concentration was confirmed by measuring directly the transport of the native auxin, indoleacetic acid, through segments of* Coleus *stems.*

Indoleacetic acid (IAA) is generally considered, on the basis of its activity and occurrence, to be the major endogenous plant hormone of the auxin type. As such, its transport properties are of particular importance in understanding plant development (*1*). Concentrations of IAA in the range of 0.1 to 10 ppm have been judged to transport approximately normal physiological amounts to plant tissue, although quantitative checks of this assumption have been rare (*2*). Indirect evidence from studies of the transport of IAA in relation to growth and regeneration of xylem supported the hypothesis that there is not a steady increase in the amount of auxin transported as the amount added is increased, but rather that a plateau is reached when the concentration is in the range of 1 to 10 ppm (*1, 3*). Earlier workers, who had tested this point directly on segments from the coleoptile of *Avena,* did not find a plateau; the more recent of them concluded that "the amount transported from apex to base (normal transport) increases almost linearly with the logarithm of the applied indoleacetic acid concentration" (*4*). However, the potential importance of such a saturation effect in the normal control of plant development is sufficient (*1, 3*) to warrant making a direct test on one of the organisms from which the indirect evidence cited was obtained. The preliminary evidence already presented was not considered conclusive, since the results might have reflected an artifact in the bioassay (*1*). The present report, therefore, describes a more detailed analysis of this system.

Studies of the basipetal translocation of auxin were made from individual segments taken from the second internode of *Coleus* plants that were closely matched for developmental age, and which were of the clone used previously. Each segment, approximately 10 mm in length, was prediffused on moist filter paper for 2½ hours, a time sufficient as shown by direct bioassay for total depletion of endogenous diffusible auxin. A 4.9-mm length was then cut from the middle of each original segment and ringed with vaseline to obviate physical movement of added IAA along the outer surface of the tissue (*3, 5*). The morphological base of each segment was placed on a washed-agar block which contained 1.5 percent plain agar and whose dimensions were 11.0 by 8.0 by 1.5 mm. A similarly prepared washed-agar block of the same dimensions, but containing IAA in concentrations of 0.5, 1, 2, 5, 10, or 20 ppm, was placed on the apical end of the segment. Auxin was usually collected for a period of 2½ hours under humid conditions at room temperature in diffuse light.

The amount of auxin in the receiver blocks placed at the base of the segment was determined on the day of, or the day after, the transport. Recovery of the amount of auxin transported was measured as the full amount and also as 1 : 2 and 1 : 4 dilutions of the full amount when the IAA had been added in concentrations of 10 and 20 ppm. Dilutions were made after the receiver block had been equilibrated for 3 hours. The block was then divided into equal halves (5.5 by 8.0 by 1.5 mm).

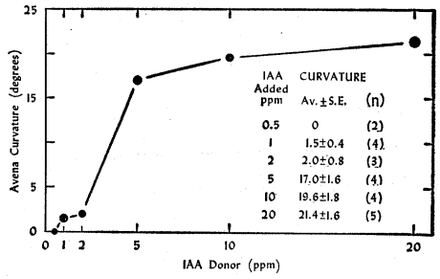

Fig. 1. Auxin recovery in undiluted, basal receiver blocks after increasing concentrations of IAA were applied to the apical ends of prediffused, 4.9-mm segments of *Coleus* stem. Transport time was 2½ hours. Values are average curvatures from bioassays (*Avena* method) of two to five separate experiments, as indicated by size of the circles and the numbers under *n*.

One of these halves was stored overnight and assayed as an index of full recovery (as compared to the assay which was made on another full-size block that had been treated similarly). The other half was stacked on a single half block of plain agar to obtain a 1:2 dilution or on a stack of three half blocks of plain agar to make a 1 : 4 dilution. The blocks were placed for 13 hours in the refrigerator in the dark in order for physical diffusion to take place. Each half block was then separated from the other and assayed independently. All blocks were assayed by the standard *Avena* curvature test (*6*). Two or more calibrations with synthetic IAA were used for each of the six tests. Results are expressed as degrees curvature.

Figure 1 shows the average curvature in the receiver blocks when the donor blocks contained various concentrations of IAA. There is a sharp increase in the amount of auxin transported as the concentration of the donor block is raised from 1 ppm to 5 ppm. An equally clear reduction in the rate of increase occurs when the concentrations of the donor block are greater than 5 ppm, although the curve does not show a flat plateau. The average curvature when the concentration of the donor block was 5 ppm was more than 1000 percent greater than when the concentration was 1 ppm, the lowest concentration resulting in transport sufficient to be detected with this method of bioassay. Yet a fourfold further increase in the concentration of the donor block (to 20 ppm) resulted in a curvature of only 26 percent more. The average curvatures from concentrations of 5, 10, and 20 ppm in the