Plant Physiol. (1974) 53, 154–157

A Failure to Detect an Influence of Magnetic Fields on the Growth Rate and Circadian Rhythm of *Neurospora crassa*[1]

Received for publication July 27, 1973 and in revised form September 10, 1973

D. MICHAEL BITZ[2] AND MALCOLM L. SARGENT
Department of Botany, University of Illinois, Urbana, Illinois 61801

ABSTRACT

Low strength magnetic fields, 6.36 and 32.25 gauss, were found to have no effect, with one questionable exception, on the circadian rhythm and growth rate of *Neurospora crassa*. This was true whether the fields were continuous, pulsed 20 minutes daily, or on a 12 : 12, on-off cycle.

The effects of magnetic fields on animal orientation, biochemical and biophysical aspects of metabolism, and biological rhythms have been a subject of some interest for the past 30 years. The last subject has been of particular controversy because of the uncertainty as to whether rhythms are driven by endogenous mechanisms or by oscillations in subtle geophysical factors. Brown and his co-workers (3, 14) were led to suspect an exogenous influence on daily (solar and lunar) and monthly (lunar) rhythms by a correlation between circadian rhythms of oxygen production by potatoes in hermetically sealed containers and oscillations in the intensity of primary cosmic radiation. Other experiments found the same relationship between cosmic radiation and rhythms in *Fucus, Uca*, carrots, oysters, and rats (7). Later investigation showed that the rhythms could be correlated with variations in atmospheric electromagnetic fields and to the earth's field. This is reasonable as the primary cosmic radiation flux would vary inversely with the strength of the earth's field (6).

Further studies, especially with the changing of the lunar orientation rhythms of *Dugesia* and *Narsarius,* demonstrated that rhythms could be affected by changes in magnetic field strength (4, 9, 10, 12, 13, 15, 16). In addition, there are reports of positive relationships between the circadian rhythms of earthworms (2), gerbils (24), and humans (25), and changes in magnetic fields.

The experiments described in this paper were designed to test the effects of magnetic fields of varying strengths and duration on the growth rate and circadian rhythm of conidiation of the *band* strain of *Neurospora crassa*. This work represents part of a broader effort to ascertain the mechanisms responsible for circadian rhythms in *Neurospora* (21–23).

MATERIALS AND METHODS

The *band* strain (MLS 41-4) of *Neurospora crassa* was grown in straight Pyrex tubes (61 cm in length, 0.6 cm in diameter, and closed at both ends with rubber stoppers) containing 10 ml of the glucose-arginine medium (21). Inoculation took place at 1700 hr one day and the growth tubes were left in constant light until the next day when they were placed in the dark to set the phase of the rhythm in all cultures simultaneously. At the same time the cultures were put into continuous darkness, in which the rhythms would be expected to free-run, they were also placed in the magnetic fields. The experiments were run in a light-tight darkroom with the only illumination being ruby-red safety lights (GE BBX, 40W) used for marking the growth front. The tubes were aerated with 10 cc of humid air per minute to stimulate conidiation (22). The position of the growth front was recorded at 2-day intervals, and the average duration of growth was 9 days. The period of the rhythm was calculated by correlating the average distance between the conidial bands and the growth rate (21).

Two apparatuses of essentially identical construction were used to produce the magnetic fields of desired strength. They consisted of solenoids of Bakelite cylinders either 2 and one-eighth or 2 and eleven-sixteenths inches in diameter close-wound with number 20 copper wire, and powered by voltage-regulated (better than 0.5%) power supplies. The solenoids were cooled by an air flow of 45 cubic feet/min. Two solenoids having fields of the same magnitude, but with direction reversed, were placed parallel to each other.

A control cylinder was connected in-line to each of the solenoids so that they would be cooled by the airflow from the solenoid. Two growth tubes were placed in each of the solenoids and control cylinders. Measurement of the fields in the solenoids with a Bell 640 Incremental Gaussmeter determined that they were homogenous (better than 5%) lengthwise from the center of the solenoids to a distance one diameter inside of the end of the wire windings, and radially from the axis to the maximal radius. Comparison between the two solenoids used for either apparatus showed a center-axial field difference of less than 1.5%.

In all but one of the experiments the solenoids and control cylinders were oriented east-west. This was done to have the vector of the earth's field at a right angle to the artificial fields so that the field in both solenoids would have the same magnitude. At the experimenter's location the magnetic field of the earth is 0.5794 to 0.5806 gauss at an angle of 71.08° to 71.13° from horizontal and with a declination of 3° east of true north (18). As the growth fronts progressed in the same direction (east) in all eight tubes, one would expect the effect of the earth's field to be the same in all cases, so that the only variable influence would be the fields of the solenoids. This arrangement allowed a comparison between the *Neurospora* grown in the west-directed field and that in the east-directed field, and both of these to the controls.

Field strengths of 6.36 and 32.25 gauss (hereafter referred to as 6 and 32 gauss, respectively) were employed, and the du-

[1] Supported by National Institutes of Health Grant GM-16541.

[2] Present address: Washington University School of Medicine, St. Louis, Mo. 63110.

12. FRANZ, G. AND H. MEIER. 1969. Biosynthesis of cellulose in growing cotton hairs. Phytochemistry 8: 579–583.
13. HEATH, E. C. 1971. Complex polysaccharides. Annu. Rev. Biochem. 40: 29–56.
14. LAINE, R. A. AND A. D. ELBEIN. 1971. Steryl glucosides in *Phaseolus aureus*. Use of gas-liquid chromatography and mass spectrometry for structural identification. Biochemistry 10: 2547–2553.
15. LELOIR, L. F. 1971. Two decades of research on the biosynthesis of saccharides. Science 172: 1299–1303.
16. MARX-FIGINI, M. 1966. Comparison of the biosynthesis of cellulose *in vitro* and *in vivo* in cotton bolls. Nature 210: 754–755.
17. O'KELLEY, J. C. AND P. H. CARR. 1953. Elongation of the cotton fiber. *In:* W. E. Loomis, ed., Growth and Differentiation in Plants. Iowa State University Press, Ames. pp. 55–68.
18. ORDIN, L. AND M. A. HALL. 1967. Studies on cellulose synthesis by a cell-free oat coleoptile enzyme system: inactivation by airborne oxidants. Plant Physiol. 42: 205–212.
19. ORDIN, L. AND M. A. HALL. 1968. Cellulose synthesis in higher plants from UDP glucose. Plant Physiol. 43: 473–476.
20. SPENCER, F. S., B. ZIOLA, AND G. A. MACLACHLAN. 1971. Particulate glucan synthetase activity: dependence on acceptor, activator, and plant growth hormone. Can. J. Biochem. 49: 1326–1332.
21. TALMADGE, J. W., K. KEEGSTRA, W. D. BAUER, AND P. ALBERSHEIM. 1973. The structure of plant cell walls. I. The macromolecular components of the walls of suspension-cultured sycamore cells with a detailed analysis of the pectic polysaccharides. Plant Physiol. 51: 158–173.
22. TSAI, C. M. AND W. Z. HASSID. 1971. Solubilization and separation of uridine diphospho-D-glucose:β-(1→4) glucan and uridine diphospho-D-glucose:β-(1→3) glucan glucosyltransferases from coleoptiles of *Avena sativa*. Plant Physiol. 47: 740–744.
23. UPDEGRAFF, D. M. 1969. Semimicro determination of cellulose in biological materials. Anal. Biochem. 32: 420–424.
24. VILLEMEZ, C. L. AND J. S. HELLER. 1970. Is guanosine diphosphate-D-glucose a precursor of cellulose? Nature 227: 80–81.

chosen on the basis of Brown's work (5, 15) with the mud snail, *Nassarius obsoletus*, and the planarian, *Dugesia dorotocephala*. For both organisms the orientation response varied as the field strength was changed from 0.04 to 10 gauss. Six gauss was chosen for our work because at this field strength the responses of both species were large and in the same direction. We have also used a 30 gauss field to determine the effects of stronger fields (30 gauss was the largest field for which air cooling was felt to be sufficient). Palmer using *Volvox* has also shown a significant orientational response with a field strength of 5 gauss (19).

Three time durations were employed since Brown (11) has shown that the response of *Dugesia* to an imposed magnetic field is initially large and that it shows a transient accommodation to the field for about 35 min, after which the effect again increases. Possible phase-response characteristics of *Neurospora* were also tested by varying the time at which the field came on for the 12:12, 6-gauss system. This was done to determine if the organism's sensitivity to either imposition or removal of a magnetic field might vary with the time of day, as does its response to light pulses (23).

Our inability to detect an effect of magnetic fields on *Neurospora* will unfortunately do little to resolve the controversy as to the existence or nature of the effects of such fields on biological rhythms. Several workers maintain that such effects can be demonstrated. For example, Brown *et al.* (7) have shown direct or inverse correlations of the rhythms of fiddler crabs, *Fucus*, potatoes, carrots, oysters, and rats to variations in the flux of primary cosmic radiation and thus, presumably, to variations in the earth's magnetic field. More recently, Brown (4, 9, 10, 12, 13, 15, 16) has worked primarily with the orientation rhythms of the mud snail, *Nassarius obsoletus*, and the planarian, *Dugesia dorotocephala*. With these systems he has shown effects of rotation of the apparatus through 90° or 180° and/or imposed magnetic fields on solar day (24 hr), lunar day (24.8), and semimonthly and monthly rhythms. In one of his most conclusive studies Brown demonstrated that the lunar month (28-day) rhythm in *Dugesia* released in a northern direction was 180° out of phase with the rhythm of those released toward the south. A comparison of the data for the northerly directed planarians in the geomagnetic field to those northerly directed, but with a reversed field of 0.05 gauss, showed the same 180° phase shift.

Stutz (24) has also found that the maximal daily activity of male, Mongolian gerbils in a 12:12 LD cycle could be correlated to the time of day, 1500 to 1800 hr, in which the magnetic field of the earth increases about 5×10^{-4} gauss toward the mean from its maximal negative deviation. Bennet and Huguenin (2) investigated the withdrawal reflex of the earthworm, *Lumbricus terrestrius*, under the earth's magnetic field and zero field strength. The earthworms in the earth's field withdrew under the stimulus of light significantly faster at 1200 than at 2000 hr. The difference in reaction time between 1200 and 2000 hr for those in the zero strength field was not significant. In addition, Wever (25) has examined the effects of a weak electromagnetic field of 10 cps on circadian rhythms in man. A shortening of the circadian period by 1.27 hr was average. He also found that the mean circadian rhythm of his subjects in a room shielded from magnetic fields was 25.65 hr, where it had been 25 hr for nonshielded conditions. Internal synchronization of the activity rhythm to physiological functions was seen to be the lowest in the shielded conditions and a maximum under the imposed field.

In contrast to these reports of positive findings, there are several, such as our own, in which an effect could not be detected. For example, Beischer (1) exposed men for up to 2 weeks to very weak magnetic fields, less than 5×10^{-4} gauss, and tested space perception, visual-spatial memory, hand-eye coordination, equilibrium, and time estimation. A comparison of the results for men in the low field conditions and men under geomagnetic influence demonstrated no differences.

An ideal method to determine if organisms receive clues from variations in the earth's field would be to set up an experiment in which the earth's field was present but did not vary. Hamner *et al.* (17) attempted to create these conditions by mounting various experimental subjects on a rotating platform near the earth's geographical South Pole. They examined the daily activity rhythm of the Syrian golden hamster, *Mesocricetus auratus*, the zonation of *Neurospora crassa* strain 21863, the eclosion of *Drosophila pseudo-obscura* Chiricahua-8, leaf movements of the bean plant *Phaseolus vulgaris*, and the activity of the cockroach, *Periplaneta americana*. The results indicated that none of these organisms was receiving information from variations in the earth's field. Realizing that they might be criticized on the basis that the experiments had not been performed at the magnetic pole, similar experiments were conducted at Los Angeles in which a varying external field was applied by placing a magnet at the side of the turntable. Here a changing field of either 25 or 0.68 gauss did not disturb the circadian rhythms of *Drosophila*, *Neurospora*, or bean plants.

Comparison of the previous reports with respect to magnetic field strength, duration of applied field, time of field imposition, period of the rhythm, type of rhythm expression, or class of organism yields no particular pattern apparent to us to resolve the conflict between the presence and absence of magnetic field effects. Our results, however, make the hypothesis that circadian rhythms are a response to subtle changes in the earth's magnetic field seem less plausible.

Acknowledgments—The authors would like to thank M. dePeo of the Physics Department for technical assistance and E. Lyman for providing laboratory space during the summer of 1972.

LITERATURE CITED

1. Beischer, D. E. 1972. The null magnetic field as a reference for the study of geomagnetic effects in animals and man. Ann. N. Y. Acad. Sci. 188: 324-330.
2. Bennet, M. F. and J. Huguenin. 1969. Geomagnetic effects on a circadian difference in reaction times of earthworms. Z. Vergl. Physiol. 63: 440-445.
3. Brown, F. A., Jr. 1957. Responses of a living organism under 'constant conditions' including pressure, to a barometric-pressure-correlated, cyclic, external variable. Biol. Bull. 112: 288-304.
4. Brown, F. A., Jr. 1962. Responses of the planarian, *Dugesia* and the protozoan, *Paramecium* to very weak horizontal magnetic fields. Biol. Bull. 123: 264-281.
5. Brown, F. A., Jr. 1966. Effects and after-effects on planarians of reversals of the horizontal magnetic vector. Nature 209: 533-535.
6. Brown, F. A., Jr. 1968. 'Endogenous' biorhymicity reviewed with new evidence. Scientia 103: 245-260.
7. Brown, F. A. Jr. 1969. A hypothesis for extrinsic timing of circadian rhythms. Can. J. Bot. 47: 287-298.
8. Brown, F. A., Jr., F. H. Barnwell, and H. M. Webb. 1964. Adaption of the magnetoreceptive mechanism of mud-snails to geomagnetic strength. Biol. Bull. 127: 221-231.
9. Brown, F. A., Jr., M. F. Bennet, and H. M. Webb. 1960. A magnetic compass response of an organism. Biol. Bull. 119: 65-74.
10. Brown, F. A., Jr., W. J. Brett, M. F. Bennet, and F. H. Barnwell. 1960. Magnetic response of an organism and its solar relationships. Biol. Bull. 118: 367-381.
11. Brown, F. A., Jr. and Y. H. Park. 1965. Duration of an after-effect in planarians following a reversed horizontal magnetic vector. Biol. Bull. 128: 347-355.
12. Brown, F. A., Jr. and Y. H. Park. 1965. Phase-shifting of a lunar rhythm in planarians by altering the horizontal magnetic vector. Biol. Bull. 129: 79-86.
13. Brown, F. A., Jr. and Y. H. Park. 1967. Association-formation between photic and subtle geophysical stimulus patterns—a new biological concept. Biol. Bull. 132: 311-319.
14. Brown, F. A., Jr., J. Shriner, and H. M. Webb. 1957. Similarities between

ration of the magnetic field was also varied. The three test conditions used were a constant field, a 20-min pulse every 24 hr, and a system of 12 hr on, 12 hr off. Phase response was also tested by using a 6 gauss, 12:12 system to maximize the chances of detecting an effect and changing the time when the field came on. The field-on times were 0430, 1030, 1630, and 2230 hr. One experiment was performed using a 6 gauss, 12:12 on-off system with the apparatus oriented north-south instead of east-west, and the *Neurospora* growing toward the north.

RESULTS

The *Neurospora* cultures were grown in magnetic fields of various strengths and durations. The growth rate and period of each run were examined by one-way analysis of variance to determine if the results for west- and east-directed fields and the controls were statistically comparable. From inspection of the data in Tables I and II it may be seen that the 6 and 32 gauss magnetic fields employed in these experiments had no significant effect ($P < 0.05$) on the growth rate or period (conidiation rhythm) of the *band* strain of *Neurospora crassa*. In addition, no reproducible, visible alterations in morphology, conidiation, or pigmentation could be discerned. The variability, especially that of the growth rate, between runs of identical conditions, even if two tests were conducted simultaneously, was often large enough that the results could not be combined statistically. Slight differences in medium composition, aeration rate, and temperature were presumably responsible for the variation between runs.

To determine if there were a small variation throughout the entire experiment attributable to either the presence of a field or the field orientation, the averages of each orientation for every run (*e.g.*, run 1, west orientation, growth rate) were compared in a paired *t* test. Table III shows only one positive correlation ($P < 0.05$), that of the west to the control for growth rate. We question the significance of this single positive correlation. We would expect that if this effect were a result of the imposed magnetic fields, then there should be a positive correlation between the growth rates for the east and the control. A plausible, but unproven, explanation for the positive correlation is that the cultures in the west-directed field grew more rapidly because of a slower dehydration of the growth medium, due to a slower flow of air. Precise control of air flow through individual growth tubes was difficult.

Table I. *Effect of Magnetic Fields on the Growth Rate of Neurospora*

The probability of statistical comparability was arrived at by one-way analysis of variance for all eight growth tubes of a run.

Type of Field	Field Strength	Run	Growth Rate			Probability
			Field orientation			
			West	East	Control	
	gauss		*mm/day*			
Constant	30	1	35.00	33.10	33.10	$0.10 > P > 0.05$
		2	34.28	35.44	34.72	$0.25 > P > 0.10$
		3	36.13	36.32	36.01	$0.25 > P > 0.10$
	6	4	35.40	33.50	32.50	$0.50 > P > 0.25$
		5	33.95	34.24	32.24	$0.50 > P > 0.25$
		6	45.10	45.70	46.12	$0.10 > P > 0.05$
		7	44.15	45.00	43.74	$0.25 > P > 0.10$
Pulsed	30	8	34.39	32.98	31.24	$0.75 > P > 0.50$
		9	46.07	41.55	45.00	$0.50 > P > 0.25$
	6	10	46.08	47.00	44.78	$0.25 > P > 0.10$
		11	37.37	35.13	37.71	$0.50 > P > 0.25$
12:12 (on 1030 hr)	30	12	44.11	43.11	42.12	$P = 0.50$
		13	44.17	43.72	44.56	$0.25 > P > 0.10$
(on 0430 hr)	6	14	42.00	40.08	38.69	$0.10 > P > 0.05$
		15	39.13	41.32	39.35	$0.50 > P > 0.25$
(on 1030 hr)		16	38.55	46.21	36.50	$0.10 > P > 0.05$
		17	44.66	44.62	42.21	$0.25 > P > 0.10$
(on 1630 hr)		18	45.10	45.58	44.93	$0.25 > P > 0.10$
		19	43.94	42.37	44.81	$0.75 > P > 0.50$
(on 2230 hr)		20	49.05	46.50	46.86	$0.25 > P > 0.10$
		21	42.32	37.42	41.02	$0.25 > P > 0.10$
			North	South	Control	
12:12 (on 1030 hr)	6	1	33.25	34.84	33.92	$0.50 > P > 0.25$
		2	33.50	33.00	32.34	$0.25 > P > 0.10$

Table II. *Effect of Magnetic Fields on the Period of Neurospora*

The probability of statistical comparability was arrived at by one-way analysis of variance for all eight growth tubes of a run.

Type of Field	Field Strength	Run	Period			Probability
			Field orientation			
			West	East	Control	
	gauss		*hr*			
Constant	30	1	22.36	22.70	22.70	$0.75 > P > 0.50$
		2	21.98	22.05	22.35	$0.10 > P > 0.05$
		3	21.77	22.27	21.76	$0.25 > P > 0.10$
	6	4	20.54	20.62	20.33	$0.10 > P > 0.05$
		5	21.05	20.98	21.76	$0.10 > P > 0.05$
		6	21.67	21.69	22.07	$0.25 > P > 0.10$
		7	22.12	21.51	21.51	$0.50 > P > 0.52$
Pulsed	30	8	22.42	22.61	22.62	$0.50 > P > 0.25$
		9	21.28	21.25	20.84	$0.75 > P > 0.50$
	6	10	21.34	21.50	21.02	$0.50 > P > 0.25$
		11	22.14	21.22	21.63	$0.10 > P > 0.05$
12:12 (on 1030 hr)	30	12	21.48	20.71	20.86	$0.75 > P > 0.50$
		13	20.98	20.72	22.20	$0.10 > P > 0.05$
(on 0430 hr)	6	14	20.92	20.96	21.25	$0.50 > P > 0.25$
		15	21.12	20.59	21.27	$0.50 > P > 0.25$
(on 1030 hr)		16	21.42	22.08	20.84	$0.25 > P > 0.10$
		17	20.87	20.90	21.56	$0.25 > P > 0.10$
(on 1630 hr)		18	21.76	21.96	22.05	$0.50 > P > 0.25$
		19	21.59	21.57	21.92	$0.25 > P > 0.10$
(on 2230 hr)		20	20.83	20.60	21.16	$0.25 > P > 0.10$
		21	20.86	21.10	20.95	$0.25 > P > 0.10$
			North	South	Control	
12:12 (on 1030 hr)	6	1	21.78	21.71	22.02	$0.50 > P > 0.25$
		2	21.13	21.64	21.89	$0.50 > P > 0.25$

Table III. *Examination for Small Variations Attributable to the Presence of a Magnetic Field*

Paired *t* test analysis of the growth rate and period averages for each field orientation for all the runs.

Comparison Performed	Growth Rate	Period
West to control	$0.01 > P > 0.001$	$0.3 > P > 0.2$
East to control	$0.4 > P > 0.3$	$0.3 > P > 0.2$
West to east	$0.5 > P > 0.4$	$P > 0.5$

DISCUSSION

Since there are conflicting reports as to whether or not magnetic fields affect circadian rhythms, we attempted to use experimental conditions that would maximize the chances of detecting such an effect in *Neurospora*. Two field strengths were

daily fluctuations in background radiation and O_2-consumption in the living organism. Biol. Bull. 113: 103–111.

15. Brown, F. A., Jr., H. M. Webb, and F. H. Barnwell. 1964. A compass directional phenomenon in mud-snails and its relation to magnetism. Biol. Bull. 127: 206–220.
16. Brown, F. A., Jr., H. M. Webb, and W. J. Brett. 1960. Magnetic response of an organism and its lunar relationships. Biol. Bull. 118: 382–392.
17. Hamner, K. C., J. C. Finn, G. S. Sirohi, T. Hashizaki, and B. H. Carpenter. 1962. Studies of the biological clock at the South Pole. Nature 195: 476–480.
18. McGinnis, L. D. and P. C. Heigold. 1961. Regional maps of vertical magnetic intensity in Illinois. Illinois State Geological Survey Circular No. 324, pp. 1–12.
19. Palmer, J. D. 1963. Organismic spatial orientation in very weak magnetic fields. Nature 198: 1061–1062.
20. Sargent, M. L. and W. R. Briggs. 1967. The effects of light on a circadian rhythm of conidiation in *Neurospora*. Plant Physiol. 42: 1504–1510.
21. Sargent, M. L., W. R. Briggs, and D. O. Woodward. 1966. The circadian nature of a rhythm expressed by an invertaseless strain of *Neurospora crassa*. Plant Physiol. 41: 1343–1349.
22. Sargent, M. L. and S. H. Kaltenborn. 1972. Effects of medium composition and carbon dioxide on conidiation in *Neurospora*. Plant Physiol. 50: 171–175.
23. Sargent, M. L. and D. O. Woodward. 1969. Genetic determinants of circadian rhythmicity in *Neurospora*. J. Bacteriol. 97: 861–866.
24. Stutz, A. M. 1972. Effects of weak magnetic fields on gerbil spontaneous activity. Ann. N. Y. Acad. Sci. 188: 312–323.
25. Wever, R. 1967. Über die Beeinflussung der circadianen Periodik des Menschen durch schwache electromagnetische Felder. Z. Vergl. Physiol. 56: 111–128.

Plant Physiol. (1974) 53, 158–163

Accumulation and Radial Transport of Ions from Potassium Salts by Cucumber Roots[1]

Manuscript received March 12, 1973 and in revised form September 6, 1973

BRUCE J. COOIL
Department of Plant Physiology, University of Hawaii, Honolulu, Hawaii 96822

ABSTRACT

Accumulation of K^+ is insensitive to the anion supplied with it at a solution concentration below 1 mM. Rates of K^+ transport to the xylem from the same solutions are, however, dependent upon the anion present and decrease in the order $NO_3^- \gg Cl^- > SO_4^{2-}$. Parallel effects on rates of exudation and anion transport result from kind and concentration of anion supplied and time of exposure to the solution. When high K salt concentrations are used, only linear relationships are found between solution concentrations and transport rates. However, ion concentration in the exudate increases more than external solution concentration, while exudation rate is unaffected. It is suggested that some of the ions transported are from compartments within the cells. At high solution concentrations KNO_3 results in more exudation and in higher ion concentration in the exudate than is found with KCl.

The processes involved in transport of ions from the external medium into the xylem of roots continue to evoke controversy and have been discussed in several recent reviews (1, 18, 19, 27). Lüttge and Laties (22) showed that both radial transport and accumulation of K^+ and Cl^- have parabolic isotherms at solution concentrations below 0.5 mM. Above 1 mM uptake isotherms have several inflections, whereas transport isotherms appear linear, especially in intact plants. These results were interpreted as showing that transport of ions is limited by carriers only at low solution concentrations but is diffusive in nature in solutions above 1 mM (18, 22). A passive transport to the xylem in the high concentration range is supported by others, notably Hodges and Vaadia (12) with onion roots and Minchin and Baker (24, 25) with castor bean. Läuchli and Epstein (20), however, concluded that lateral transport of Cl^- across corn roots is mediated by dual mechanisms of ion absorption which they believe to reside in the plasmalemma. Läuchli, Spurr, and Epstein (21) suggested that ions are secreted directly into the conducting vessels by a carrier-mediated transport. This conclusion was based on the radial distribution of K^+ as detected with the electron probe and evidence of numerous membrane systems in the xylem parenchyma cells. Dunlop and Bowling (7) found that the electrochemical potential of K^+ is the same in the xylem exudate as in the vacuoles of corn root cells when external solution concentrations of KCl range from 0.1 to 10 mM, while the electrochemical potential for Cl^- is higher in vacuoles than in the xylem.

Uptake of K^+ into plant cells is unaffected by the anion supplied at low solution concentrations, but above 1 mM anion effects are great (10, 22). Counter-ion effects on radial transport of K^+ are less clear (22).

Cucumber roots can be induced to exude from the xylem profusely. These roots reduce little of the NO_3^- that is absorbed, and most of it is transported to the stems and leaves (3). As with tobacco (30, 31), cucumber roots depleted of NO_3^- respond to a new supply of this ion with increased rates of exudation and salt transport. These properties make the cucumber root ideal for studying ion transport to the xylem.

This report compares patterns of accumulation and radial transport in cucumber roots of K salt ions supplied at concentrations below 1 mM, and transport at concentrations of 0.5 to 10 mM.

MATERIALS AND METHODS

Plant Culture. Seeds of *Cucumis sativus* L. cv. Burpeeana Hybrid were soaked in deionized water with continuous aeration for 24 hr, then planted in moist vermiculite and allowed to germinate in the dark for about 65 hr at 23 to 25 C. The seedlings were next exposed to indirect light for about 24 hr. Single plants were then transferred to 4-liter glass jars of continuously areated nutrient solution consisting of 1 mM $Ca(NO_3)_2$, 0.25 mM $MgSO_4$, 0.25 mM KH_2PO_4, and 0.05 mM Fe supplied as chelate of diethylenetriamine pentaacetate. Micronutrients were supplied at one-half the concentrations suggested by Johnson *et al.* (16). On the 14th day after transfer of the seedlings to the solutions, the three macronutrient salts were added again in the same quantities as initially. Micronutrients were not replenished. Plants were grown in a glasshouse in which the usual temperature range was 20 to 35 C with occasional temperatures as high as 40 c. Experiments were performed 30 to 35 days after initiation of germination. At this time the plants were beginning to flower, and their low nitrogen status was evident from the yellow-green appearance of the leaves. Root systems varied in fresh weight from 8 to 20 g per plant.

Experimental Procedures. Roots used for ion accumulation experiments were prepared by removing the tops at the hypocotyls and rinsing in 0.2 mM $CaSO_4$. They were then transferred to a fresh $CaSO_4$ solution for 1 hr, with aeration provided, to permit equilibration to the temperature of the experiment. At the beginning of the experimental period, at least five root systems were placed in a desorbing solution of 0.2 to 0.4 mM $CaSO_4$ at 0 to 4 C. These served as zero time controls. The experimental root systems were placed in aerated solutions containing a K salt and 0.2 or 0.4 mM Ca salt. The volume of solution varied from 40 liters per root system for the lowest

[1] Journal Paper 1595 of the Hawaii Agricultural Experiment Station.