In Fig. 1, the total impedance is plotted logarithmically as a function of k. The soil suction, in bars, is indicated at the top. One bar is 10^6 dyne/cm^2, or 0.987 atmosphere. The impedance was calculated from the diffusion pressure deficit (δ) and soil suction measurements made at about 9:00 A.M. The average daily transpiration rate was used as the value for q.

Two distinct regions are shown in Fig. 1. For values of conductivity above about 5×10^{-3} cm/day, the impedance is relatively independent of the conductivity. As the conductivity decreases below about 10^{-3} cm/day, the impedance tends to increase. As the lowest values for the conductivity decrease, the impedance tends to increase linearly on the log-log plot. If I_p were negligibly small, the data should lie on a straight line of the same slope as that shown.

It appears that when the soil suction is less than about 0.6 bar, virtually all of the total potential drop (initially about 10 bars) occurs at some point in the plant. We have presented evidence that the impedance in the roots is small compared with that in the soil (*7*). Therefore this impedance must occur elsewhere in the plant. As the soil suction increases, an increasingly greater proportion of the total potential difference occurs in the soil. When the suction is above a few bars, virtually all of the potential drop (about 20 bars) occurs in the soil. When the suction is low, movement of water to the roots must take place in the liquid phase since the conductivity is adequate to supply the necessary water with a very small suction gradient. When the suction is higher and the soil conductivity does become limiting, the agreement of the observed relation with the predicted relation between the impedance and the conductivity supports the conclusion that the water movement still takes place in the liquid phase. The nature of the water outflow process from the pressure membrane apparatus gives strong evidence that under isothermal conditions virtually all of the water movement takes place in the liquid phase when suction is less than 15 to 20 bars (*6*).

The suction at which the soil conductivity becomes limiting depends upon the plant and the texture of the soil. However, many soils of widely different textures exhibit about the same value of unsaturated conductivity when the suction is from 0.5 to 2 bars. Results similar to those shown in Fig. 1 are to be expected for many different soils.

An increase in I_s due to a decrease in the extent of the root system would shift the transition zone between plant limiting and soil limiting to the right, toward higher conductivity and lower suction. An increase in I_p would move the horizontal line upward and result in a shift of this point toward lower conductivity and higher suction. It is not possible to determine from Fig. 1 whether I_p is constant, for only very large changes in I_p would be apparent (*8*).

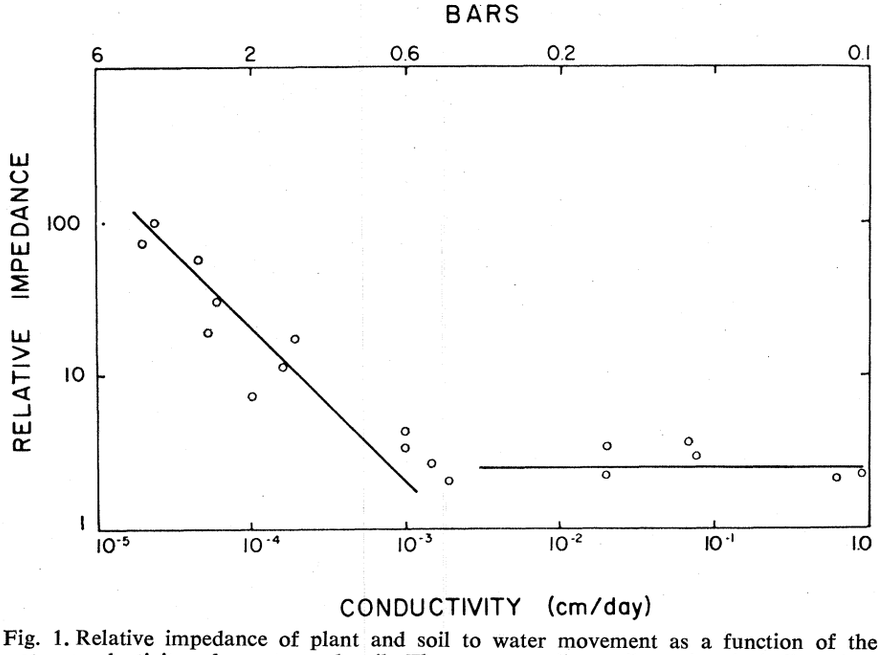

Fig. 1. Relative impedance of plant and soil to water movement as a function of the water conductivity of unsaturated soil. The corresponding soil suction is indicated at the top. Circles represent experimental data. The line on the left has a slope of 45 degrees and represents the expected relationship for negligible impedance in the plant. The horizontal line indicates constant plant and negligible soil impedance.

W. R. GARDNER
C. F. EHLIG

Salinity Laboratory,
U.S. Agricultural Research Service,
Riverside, California

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Angiotensin II: Its Metabolic Fate

Abstract. Randomly labeled tritiated angiotensin has been prepared with a specific activity of 300 μc/mg and with undiminished pressor and oxytocic activity. After infusion, angiotensin accumulated in the kidneys, adrenal glands, and uterus. Thirty minutes after infusion high levels of radioactivity appeared in brain, but the electrophoretic mobility differed from that of angiotensin II. Incubation of angiotensin with hemolyzed human red blood cells or diluted human plasma rapidly inactivated the pressor activity with production of metabolic products separable by paper chromatography. But if undiluted plasma is used with incubation up to 6 hours, no loss of activity occurs.

The biological fate of a peptide labeled with iodine-131 on the tyrosyl residues may be determined in biological systems only if the peptide remains unhydrolyzed and if the addition of iodine to the tyrosyl ring does not alter its biological activity. The I^{131}-labeling technique has been applied to angiotensin II, the octapeptide L-aspartyl-L-arginyl-L-valyl-L-tyrosyl-L-isoleucyl (or L-valyl-)-L-histidyl-L-prolyl-L-phenylalanine (*1*). The biological half-life of about 10 hours obtained when I^{131}-labeled angiotensin II is used (*2*) seems unusually long compared with that reported for oxytocin, vasopressin, and bradykinin (*3*). Further, the pressor response to angiotensin II is completed in a few minutes.

Tritiation of a peptide, however, permits the introduction of a radioactive label which would not be expected

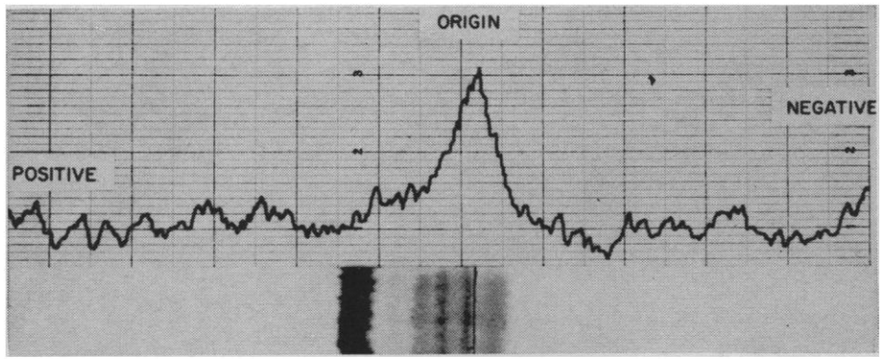

Fig. 1. Paper electrophoretic pattern of tritiated angiotensin added to heparinized human plasma developed in Spinco B 2 barbital buffer (*p*H 8.6) at 6 hours. Angiotensin shows one symmetrical peak migrating with the β-globulins.

to alter biological reactions. Also, this random label will permit study of the peptide and all its fragments after it is hydrolyzed. Radioactive oxytocin (*4*) was recovered after tritiation in 28 percent yield, but the specific radioactivity was low and the specific biological activity was less than that of the starting peptide. Vasopressin (*5*) gave a product with higher specific radio- and biological activity. However, the yield was not given. Tritiation of peptides might be expected to cause extensive fragmentation and racemization and, therefore, to give a poor yield of biologically active product. We report the successful tritiation of angiotensin II in high yield which has 300 μc of radioactivity per milligram and undiminished pressor and oxytocic activity. This labeled peptide has been used to study the tissue distribution and biologic half-life of angiotensin II. An attempt has been made to identify some of the metabolic fragments.

The starting material, asparaginyl^1-valyl^5-angiotensin II (*6*), had 11,500 pressor units per milligram of peptide (*7*). The peptide (16.4 mg) was placed in a boat at the bottom of a V-shaped cell. The cell was evacuated to less than 1 mm of mercury pressure, and 5.76 c of tritium gas was added to increase the pressure by about 1 cm. An electric arc was passed through the cell for 30 minutes (*8*). Then the peptide was dissolved in 0.1*M* acetic acid solution, and the solvent was evaporated to dryness under a stream of nitrogen. The residue was redissolved in 0.1*M* acetic acid and taken to dryness three more times to remove all exchangeable tritium. The sample was then dissolved in a minimum volume of 0.01*M* acetic acid (about 8 ml) and put onto a 24- by 750-mm column of carboxymethyl cellulose which had been equilibrated with 0.01*M* acetic acid. The column was developed with a gradient of increasing acetic acid concentration obtained by placing 500 ml of 0.01*M* acetic acid in a flask; as the solvent from this flask flowed onto the column it was replaced by an equal volume of 3.5*M* acetic acid. Fractions of 4.1 ml were collected, and the angiotensin appeared in fractions 91 to 110. This product yielded one spot which reacted with Pauly reagent on paper electrophoresis at *p*H 2.1 (*9*). However, the electrophoretograms indicated three minor radioactive substances near the spot of application which were not removed by repeating the chromatography on carboxymethyl cellulose. These substances were removed by streaking the peptide on 6-to-8-inch-wide sheets of Whatman No. 3 paper which were developed by electrophoresis at *p*H 2.1. The peptide was eluted from the paper with 0.1*M* acetic acid for 5 days and yielded 6.5 mg of tritiated angiotensin II (39.6 percent yield) which has 12,000 pressor units per milligram and a specific activity of 300 μc/mg. This product exhibited only one major spot by paper electrophoresis and paper chromatography in *n*-butanol, acetic acid, and water (4:1:5) (R_F 0.35). A very minor radioactive peptide spot was detected in both of these systems. Since this spot was absent in the sample after column chromatography it must have been formed during the period of elution from the paper after electrophoresis. Angiotensin II is known to be stable under the conditions used, so it is believed that this peptide is formed from angiotensin II as a result of the radioactivity.

Female rats (200 to 250 g) were infused with tritiated angiotensin at a rate of 4.4 μg/min for 20 minutes. This amount of angiotensin initially raised the blood pressure by 90 mm-Hg. Tachyphylaxis developed, as was shown by lack of response to a standard dose of peptide. The animals were killed by bleeding either immediately after infusion was completed or 30 minutes later, when blood pressure had usually returned to normal. Organs were removed within 3 to 5 minutes, weighed, and extracted with 0.5*N* perchloric acid (*10*). The perchlorate was removed as insoluble potassium perchlorate, and the filtrate was evaporated to dryness under reduced pressure at temperatures not over 40°C. The residue was dissolved in 0.1*M* acetic acid. Paper electrophoresis was conducted as before at *p*H 2.1 (*9*), and paper chromatography was conducted in *n*-butanol, acetic acid, and water (4:1:5). Total radioactivity was determined with a Tracerlab liquid scintillation counter; the paper strips were

Table 1. Distribution of tritiated angiotensin in a rat. Radioactivity is given in counts per minute, either per gram of tissue or per milliliter, as appropriate.

Organs	End of infusion		30 min after end of infusion	
	Radioactivity	Electrophoretic mobility (cm)	Radioactivity	Electrophoretic mobility (cm)
Heart	15,000		15,000	
Lung	20,000		18,000	9.7
Kidney	127,000	14.8	227,000	8.8 & 12.3
Adrenal	143,000	16.0	77,000	
Liver	13,000		18,000	
Spleen	16,000		15,000	
Brain	5,000		69,000	12.0
Blood	36,000	16.0	8,000	8.9 & 15.8
Urine	4,500	11.2 & 15.2	342,000	12.1
Uterus	128,000	15.5	5,600	21.0
Aorta	11,500		19,300	
Skeletal muscle	8,800	15.5	10,000	
Angiotensin		15.5		15.6

scanned in a 4π paper scanner (Fig. 1).

Immediately after infusion there was high radioactivity in kidneys, adrenal glands, and uterus; the radioactive substance had the electrophoretic mobility of angiotensin. Thirty minutes after the end of the infusion, the radioactivity of the brain had increased significantly, but high levels remained in the kidneys and adrenals. However, the mobility of these radioactive products was different from that of angiotensin (Table 1).

Because of the close physiological association of angiotensin with the kidneys and adrenal glands (*11*), it is significant that high levels of angiotensin accumulate initially in these organs. Within 30 minutes angiotensin can no longer be recovered. Hence its half-life is similar to the half-life of vasopressin, oxytocin, and bradykinin (*3*), but not to the half-life obtained when I^{131} is used as tracer (*2*).

Incubation of tritiated angiotensin with undiluted human plasma containing heparin for 6 hours showed only slight destruction of pressor activity, or none at all, and no change in electrophoretic or chromatographic mobilities. Plasma diluted 1:10 with phosphate buffer (*p*H 7.4) or saline destroyed 70 percent of the pressor activity in 10 to 20 minutes. A new peak appeared in the chromatogram with an R_F of 0.15 to 0.20. Incubating angiotensin with hemolyzed red blood cells destroyed pressor activity completely in 60 to 90 minutes; at the same time a new peak with an R_F of 0.71 to 0.76 appeared.

We assume, therefore, that two different "angiotensinases" exist, one in red blood cells and the other in plasma (*12*).

P. A. KHAIRALLAH, IRVINE H. PAGE,
F. M. BUMPUS, R. R. SMEBY
Research Division, Cleveland Clinic Foundation, Cleveland, Ohio

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24 September 1962

Relation of Infrared Spectra to Coordination in Quartz and Two High-Pressure Polymorphs of SiO_2

Abstract. Infrared spectra of the four-coordinated quartz and coesite polymorphs of SiO_2, the rutile six-coordinated (stishovite) polymorph of SiO_2, and the quartz and rutile polymorphs of GeO_2 show that a change from tetrahedral to octahedral cation coordination results in (i) a 23-percent increase in the wavelength of the main absorption band for both the SiO_2 and GeO_2 polymorphs and (ii) a significant increase in the force constant of the same magnitude for the SiO_2 and GeO_2 polymorphs. The quartz and the rutile isostructural pairs for SiO_2 and GeO_2 show that the effect of increasing mass is to increase proportionally the wavelength of the respective main absorption bands. The infrared data for the rutile form of SiO_2 fit the empirical equation of Dachille and Roy relating cation coordination, mass, atomic number, valence, and main absorption wavelength.

The infrared spectra of four polymorphs of SiO_2 and two polymorphs of GeO_2 were investigated independently by Lippincott *et al.* (*1*) and Dachille and Roy (*2*). The SiO_2 polymorphs included quartz, tridymite, and cristobalite, all of which have silicon in fourfold coordination, and coesite, a high-pressure polymorph of SiO_2 whose coordination scheme was not known at that time. The GeO_2 polymorphs were the hexagonal four-coordinated quartz form and the tetragonal six-coordinated rutile form (*3*). Both groups of investigators showed that the main Si–O stretching frequency is the same for all the SiO_2 polymorphs and concluded that coesite had fourfold coordination, a conclusion since confirmed by the single-crystal x-ray study of Zoltai and Buerger (*4*). They also found that the main Ge–O stretching frequency for the quartz form of GeO_2 is about 23 percent greater than that for the rutile form, and they ascribed this shift to the difference in primary coordination of the cation. Before the latter relationship can be used as a general indicator of coordination in simple compounds, additional pairs of polymorphs related by reconstructive transformation must be examined. With the exception of GeO_2, however, no other pairs of suit-

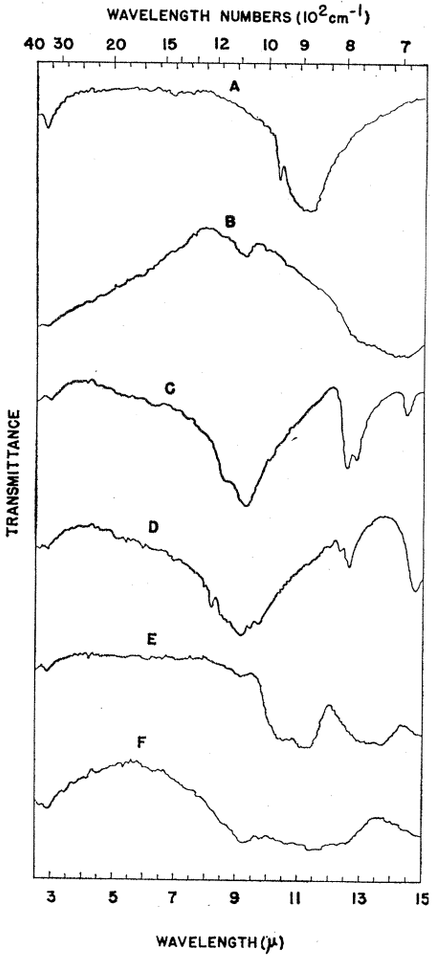

Fig. 1. Infrared absorption spectra of GeO_2 and SiO_2 polymorphs. (A) GeO_2 (quartz form), Fisher reagent; (B) GeO_2 (rutile form), prepared at 50 kb and 1000°C from the quartz form in a belt-type apparatus (*7*); (C) Quartz, high-purity single crystal from Hendersonville, North Carolina; (D) Coesite, prepared at 50 kb and 800°C from hydrated silica gel in a belt-type apparatus; (E) Stishovite from Meteor Crater, Arizona (*8*); (F) Mixture of stishovite and coesite prepared at 120 kb and >1000°C from hydrated silica gel in a girdle-type apparatus (*7*).