windows with venetian blinds that could be adjusted to reduce the sunlight when necessary (Fig. 1*A*). No nutrients were added to the media before or during the rooting procedure. Once a week, the pots were immersed in tap water for 10 minutes, and then drained. The cuttings were sprayed with pesticides about once a month to combat fungi and insects.

Sample cuttings were removed periodically and examined for any evidence of rooting. The first cutting to strike roots was observed 7 weeks after potting. Mortality of both diseased and healthy cuttings was high in the first 4 months. Twenty-one cuttings rooted within 8 months (Fig. 1*B*), and eight more rooted within 12 months. From the original 100 cuttings, 29 rooted, of which 14 were from diseased and 15 from healthy trees.

The rooted cuttings were planted individually in pots, 5 inches in diameter, containing a 1:1 mixture of Perlite and surface soil from a hardwood stand. The cuttings were kept moist and partially shaded in a greenhouse. In late September 1959, nine of the rooted cuttings (six from a diseased tree and three from a healthy tree) were lined out in a nursery, located approximately 3 miles from the site of the parent trees. The cuttings were planted in the same type of media in pots, were placed side by side in the nursery, and thus were growing essentially in identical sites.

In late July 1960, the diseased mother tree developed initial needle blight symptoms. Examination of the six rooted cuttings from this tree, planted 3 miles away, revealed a simultaneous development of needle blight on the new foliage, with the injury originating in tissue of the same maturation as in the mother tree. In addition, the rate of distal spread of foliar necrosis was comparable in the cuttings and in the parent tree. The three healthy cuttings were unaffected, as was the mother tree. Examination of the extent of the root systems of the diseased and healthy cuttings 1 year after they struck roots revealed no apparent differences between the two (Fig. 1, *C–D*).

From the above observations it may be concluded: (i) that the excessive mortality of main root tips which has been found to occur in needle-blighted trees (*5*) probably is a consequence of the disease and is not primarily associated with the cause of needle breakdown; and (ii) that symptom development which occurred simultaneously in the same local area on rooted cuttings and their diseased parent tree is further evidence strengthening the hypothesis that susceptibility to the unfavorable conditions which contribute to needle blight is inherent in the individual (*6*).

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Transmembrane Action Potentials from Smooth Muscle in Turtle Arteries and Veins

Abstract. Transmembrane potential and active tension were measured in isolated segments of turtle aorta and inferior vena cava. Changes in tension were associated with action potentials, but the potentials had a different pattern in the two tissues. When the frequency of the action potentials increased, the contractions summated, resulting in a steadily maintained contraction.

The activity of smooth muscle in blood vessels has usually been estimated indirectly by measuring the diameter of intact vessels, the tension in isolated strips of vessel, the compliance in the vessel wall, or the resistance to flow. The results of such measurements have often been difficult to interpret because it was impossible to determine the relative contributions of active changes in the muscle and passive changes due to the physical properties of the wall.

A more direct way of assessing muscle activity in blood vessels would have many advantages. One method would be to measure the electrical changes accompanying activity in single cells. Unfortunately, muscle cells in blood vessels are generally very small and are usually surrounded by considerable amounts of fibrous and elastic tissue, especially in larger vessels. Consequently, the impalement of these cells with microelectrodes and the measurement of their membrane potential is very difficult. Turtle arteries and veins contain relatively little fibrous and elastic tissue, which makes the impalement of their muscle cells somewhat easier. The present report describes some relationships between changes in tension and membrane potential in the smooth muscle of these vessels.

Excised segments of the abdominal aorta and the inferior vena cava were cut open lengthwise and mounted in an organ bath filled with frog Ringer's solution at 24°C. The transverse tension exerted by the opened segment was measured with an RCA mechanoelectronic transducer (No. 5734). Single muscle cells were impaled through the intimal surface with "floating" microelectrodes (*1*).

The time courses of membrane potential and contractile tension during rhythmic activity in aorta are shown in Fig. 1. In this experiment the resting potential was about 35 mv. It can be seen that the increase in tension was just preceded by a single heartlike action potential, which consisted of a

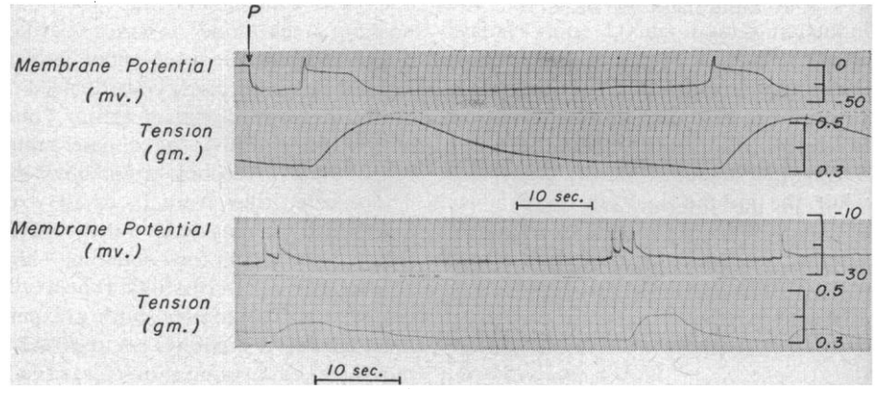

Fig. 1. Action potentials in single smooth muscle cells of turtle aorta (upper record) and turtle inferior vena cava (lower record) associated with changes in smooth muscle tension. At *P*, the arterial smooth muscle was impaled with a microelectrode.

rapid spike of depolarization followed by a long plateau. The spike, but not the plateau, overshot the zero potential level. In this example the membrane repolarized within about 12 seconds, but some action potentials lasted up to 40 seconds. Often, a small prepotential was seen.

Although the shape of the action potential resembles somewhat that of ventricular myocardium, the two types of muscle differ in other respects. The resting potential and the action potential are higher in heart than in arterial muscle. A single contraction lasts much longer in arterial (30 to 200 sec) than in ventricular muscle (1 to 3 sec). In heart muscle the cell membrane remains depolarized practically throughout the contraction, whereas in arterial muscle the membrane usually repolarizes before maximum tension is reached. The relatively shorter refractory period in the artery permitted summation of contractions when the rate of firing was increased, resulting in steadily maintained contractions. As in cardiac muscle an increase in the frequency of the action potentials decreased their duration.

A different pattern was seen in the veins. Some spontaneous contractions occurred, but they were less rhythmic and more variable in strength than the arterial contractions. Each contraction was preceded by one or more action potentials (Fig. 1). These consisted of a spike of depolarization. However, repolarization was much more rapid than in the arteries, so that the plateau following the spike was often barely perceptible. Action potentials were often followed by a period of hyperpolarization. When trains of spikes followed one another closely, the contractions summated to produce a fairly steady contraction.

The differences between the action potentials seen in the artery and the vein emphasize the danger of considering the configuration in one type of vessel as representative of that in all vascular smooth muscle. However, spikelike action potentials were recorded in the inferior vena cava of the guinea pig, and long action potentials similar to those in turtle arteries were recorded with difficulty in the aorta of the frog (*2*).

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Repression of Ornithine Transcarbamylase Protein Formation by Arginine

Abstract. Arginine-repressed cells of *Escherichia coli* W do not form a protein immunologically related to ornithine transcarbamylase. This was determined by lack of cross-reactivity of repressed cell extracts with rabbit antisera prepared against purified ornithine transcarbamylase. The results indicate that arginine acts by blocking the synthesis of the entire enzyme-protein.

Escherichia coli W cells grown in the presence of arginine contain only negligible amounts of ornithine transcarbamylase (OTC), an enzyme in the biosynthetic pathway leading to arginine formation. It has been shown (*1*) that arginine acts as a repressor of the synthesis of OTC activity in these cells, and recent evidence (*2*) suggests that perhaps ornithine competes with arginine for the site of repressor action. When arginine is removed from the growth medium, a 100-fold increase in OTC activity occurs in *E. coli* W; this increase in activity has been correlated with new protein synthesis (*3*).

In order to investigate the function of arginine in the regulation of ornithine transcarbamylase synthesis, it was first necessary to determine whether arginine blocks the synthesis of the entire OTC-protein or, alternatively, changes the configuration of the active site or prevents synthesis of a small portion of the molecule. If these cells form an enzymatically inactive OTC-like protein in the presence of arginine, this altered protein should be detectable by immunological techniques. With the use of specific antisera to the inducible enzyme, β-galactosidase, it has been shown (*4*) that uninduced cells contain no antigenically active protein related to this enzyme. Experiments described in the present report suggest that in like manner arginine represses the formation of the entire OTC-protein.

By use of a 100-fold purified OTC preparation from *E. coli* W (*5*), an antiserum was prepared from rabbits according to the method of M. Cohn (*6*). One milligram of purified OTC emulsified with Freund's adjuvant containing mycobacterium was injected into the rabbits intramuscularly under the scapula. The injection was repeated weekly for 30 days, and after two additional weeks the rabbits were bled. The serum obtained from rabbit blood was clarified by centrifugation, and the globulin fraction was collected by precipitation with ammonium sulfate at 33 percent saturation. The precipitation of a 100-fold purified OTC antigen with the prepared antisera is shown in Fig. 1. At the equivalence point about 0.24 mg or 212 units of OTC are precipitated per milligram of antisera protein in the precipitate. It was possible to demonstrate 30 percent of the original OTC activity in the washed precipitate by running it gently through a thin-nosed pipette. However, it was not possible to release the enzyme

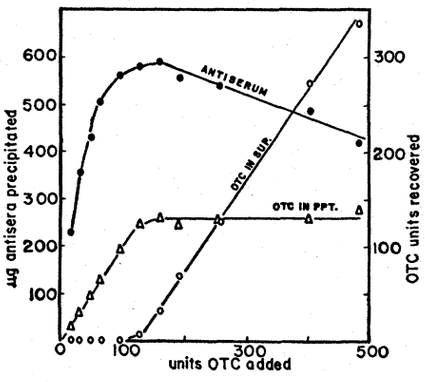

Fig. 1. Relationship between amount of rabbit antiserum precipitated and amount of purified ornithine transcarbamylase added. Precipitation reactions were carried out as follows: 0.1 ml of rabbit antiserum prepared as explained in the text; increasing amounts of purified OTC (specific activity = 873 μmole of citrulline produced per minute per milligram of protein) were added; final volume of each tube to 1.7 ml with 0.15*M* NaCl; incubated at 3°C for 3 days; centrifuged; precipitates washed three times in 0.15*M* NaCl. Curve •: micrograms of antiserum protein precipitated per tube calculated by subtracting the antigen precipitated from the total protein precipitated. At the point of maximum antiserum precipitation (590 μg), 129 units of OTC were precipitated. Curve o: units of OTC remaining in the supernatant determined as reported previously (*3*). Curve △: units of OTC in the precipitate calculated by subtraction of curve o from total OTC added. OTC activity was also demonstrated in the precipitates as discussed in the text.