resistance ($R_I$), where $R_I^{-1} = R_s^{-1} + R_a^{-1} + R_b^{-1}$ we find that

$$R_{sb} = R_I \frac{(\alpha + 1)(\rho + 1)}{\alpha + \rho + 1}$$

Values are already available for $R_I$ and $\rho$, and the upper and lower limits for $\alpha$ may be easily calculated by assuming either an infinite resistance or a "zero" resistance, respectively, in series with the apical shaft at the MBP and then calculating the total value of $R_a$ as seen from the soma in each case (Table 1). From the previous definitions of $\alpha$, $\rho$, and $R_I$ it then follows that for $\rho = 3$, the maximum possible value for $\alpha$ is 2.0, the minimum is 0.3. For $\rho = 6.5$, the maximum value for $\alpha$ is 1.0, the minimum is 0.1.

In the least favorable case for dendritic electrotonic propagation with low $\alpha$, low $R_m$, and low $\rho$, 27.5 percent of a potential difference produced at the MBP occurred across the soma. In the most favorable case, the higher limit of 38.1 percent was calculated (Table 1).

The anatomic data showed that cells with smaller somatic surfaces gave rise to narrower major shafts. Smaller cells, in general, also gave rise to small axons. The fact that the core resistance of the major shaft relative to the input resistance ($R_{sb}$) is the principal determinant of the potential drop across $R_{sb}$ when the leakage resistances are relatively high encouraged an attempt to see whether the core resistance of the shaft was related to size of the neurons. Core resistances decreased as cell size (surfaces calculated assuming truncated cones) increased (Fig. 1D). This approximately linear relationship suggests that the apical dendritic contribution relative to soma size may be approximately the same for a wide range of pyramidal cell sizes, and that the results on large pyramidal cells may be generally applicable to smaller pyramidal cells.

Sholl (*4*) presented data on the diameter of branching dendrites as a function of distance (extending up to 240 $\mu$) from the MBP. These data, however, were not derived from large cells comparable to those whose major shafts were used in the above calculations. Nor do we have adequate data on the distant branchings of large cells, because it becomes progressively more difficult to obtain full sections of the desired material.

However, for the sake of a rough estimate, a secondary branch 250 $\mu$ long and 2.5 $\mu$ in diameter at the MBP tapering to 2.0 $\mu$ at the end would add a core resistance of 47.8 megohm. When the appropriate leakage resistances are considered, the lower and upper limits for attenuation of a potential change ($V_2$) applied to the end of the secondary branch (500 $\mu$ from the soma) would be 5.5 to 5.8 percent. A tertiary branch 250 $\mu$ long and tapering from 2.0 to 1.5 $\mu$ would add another 75 megohm of core resistance per branch, and similarly considering the range of leakage resistances, the limits for attenuation of potentials ($V_3$) (750 $\mu$ from soma) generated on this tertiary branch would range from 2 to 3 percent.

Somatic potentials of 27.5 to 38.1 percent (estimated lower and upper confidence limits) of a potential difference appearing at the major branch point of the apical dendritic shaft would occur as a result of steady-state synaptic activity in the more distant branches. If it is reasonably assumed that depolarizations of 20 mv can occur over the entire dendritic tree distal to the MBP 250 $\mu$ from the soma, then 5.5- to 7.6-mv depolarizations would occur at the soma. These values are in the same range as the peak depolarizations produced by strong stimulation of the nonspecific thalamic system, a fiber system believed to have the majority of its endings on apical dendrites in the superficial cortical layers (*6*). As depolarizations in this range would bring most neurons to or above firing level, then it can be considered that the apical dendrites, when all are excited, can play a dominant role in determining the neuronal firing pattern. Apical dendrites, 750 $\mu$ distal from the soma, if all depolarized could produce only 2 to 3 percent changes at the soma. If a 20-mv depolarization on these dendrites is assumed, then 0.4 to 0.6 mv of depolarization would occur at the soma. This is a fraction of the depolarization necessary to reach a firing level, usually 5 to 10 mv. However, even a 0.4-mv depolarization can have a marked effect on changing the frequency of firing of a neuron already depolarized close to or slightly above firing level (*7*). In this respect the distant dendrites can play an important modulating role, not so much in setting up sporadic action potentials, but in providing a refinement in the regulation of firing rates of neurons brought close to firing level by afferent systems ending close to the soma (*1*).

Finally, it should be noted that the strength of this assessment of the importance of the apical dendritic tree rests in its quantitative aspect (Table 1), wherein the values depend almost entirely upon anatomic and physiologic measurements and minimally upon factors included as assumptions.

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## Antibodies to Deoxyribonucleic Acid Irradiated with Ultraviolet Light: Detection by Precipitins and Immunofluorescence

Abstract. *Two rabbits were immunized with a complex consisting of ultraviolet-irradiated DNA and methylated bovine serum albumin. Serum antibodies reacted only with irradiated DNA, and the serological reaction was shown by immune-precipitation and immunofluorescence.*

Antibodies to DNA have been produced in rabbits immunized with a complex consisting of DNA and methylated bovine serum albumin (*1*). Levine *et al.* (*2*) described antibodies specific for photoproducts of DNA irradiated with ultraviolet light. These antibodies were demonstrated in complement-fixation reactions and were directed against thymine-associated photoproducts. We now

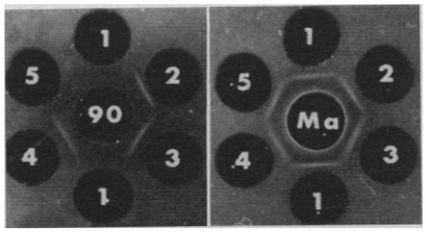


Fig. 1. Immunodiffusion in agarose demonstrating reaction of rabbit antiserum (90) only with ultraviolet-irradiated DNA while a serum of a patient with lupus erythematosus (Ma) reacts with both native and irradiated DNA. Well 1, DNA 1 mg/ml; well 2, DNA 500 $\mu$g/ml, UV, 30 minutes; well 3, DNA 500 $\mu$g/ml, UV, 60 minutes; well 4, DNA 500 $\mu$g/ml, UV, 90 minutes; well 5, DNA 500 $\mu$g/ml, UV, 120 minutes.

report that antibodies to ultraviolet-irradiated DNA can be demonstrated by immune precipitation and immunofluorescence.

Calf thymus DNA (*3*) was used at a concentration of 500 $\mu$g per milliliter in 0.15*M* NaCl, 0.01*M* phosphate, *p*H 7.0. Five milliliters of DNA in a petri dish (50 mm diameter) were irradiated for 90 minutes (*4*). The bottom of the petri dish was 5 cm from the light source, and the dish rested on a platform which was slowly rotated during irradiation. The complex of irradiated DNA and methylated bovine serum albumin was prepared by the method of Plescia *et al.* (*1*). The immunogen was homogenized in an equal volume of complete Freund's adjuvant and each of two rabbits received intramuscular doses of 500 $\mu$g of the DNA complex every week for 4 weeks.

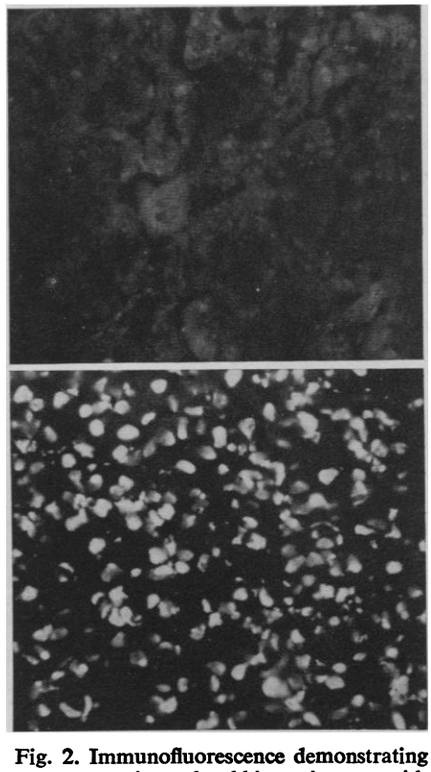

Fig. 2. Immunofluorescence demonstrating strong reaction of rabbit antiserum with nuclei of irradiated tissue section (bottom), and barely detectable reaction with nuclei of unirradiated tissue (top) (× 150).

Serologic activity in rabbit antiserums was tested by immunodiffusion in agarose (*5*) against different preparations of calf thymus DNA (Fig. 1). Rabbit antiserums did not react with unirradiated DNA but reacted with DNA irradiated for 30, 60, and 90 minutes. More extensive irradiation (120 minutes) resulted in loss of reactivity. By comparison, serum from a patient with systemic lupus erythematosus reacted with unirradiated DNA and with the preparations of irradiated DNA. The serums of three other patients with lupus have reacted similarly in this system. Other lupus patients' serums have been shown by immunodiffusion to react specifically with DNA denatured by boiling (*5*), and the antibodies are directed against purine or pyrimidine bases of single-strand DNA. Rabbit antiserums to ultraviolet DNA did not react with heat-denatured DNA by immunodiffusion.

Immunofluorescence (*6*) was also used to detect antibody activity in rabbit antiserums. The substrate, consisting of cryostat sections (4 $\mu$) of mouse kidney, was treated with antiserum, and fluorescein-conjugated sheep antiserum to rabbit gamma globulin was the indicator. Kidney sections were either untreated or irradiated for 1 minute under conditions described for DNA solutions. Antiserum reacted weakly with nuclei of untreated tissue sections, and above 1:8 dilution they were not detected (Fig. 2, top). However, they reacted very strongly with nuclei of irradiated tissues (Fig. 2, bottom), even at 1 : 512 dilution in one antiserum and 1 : 1024 dilution in the other. An important factor in the testing was duration of exposure of the tissue section to ultraviolet irradiation. Tissue sections irradiated for 2, 5, 10, and 30 minutes resulted in progressively weaker reactions with antiserum. Serums before immunization did not react with nuclei of untreated or irradiated tissue sections. In contrast to rabbit antiserums, lupus patients' serums which reacted equally with native or irradiated DNA in immune precipitation, reacted in immunofluorescence to the same serum dilution with nuclei of either untreated or irradiated tissues.

Levine *et al.* (*2*) used heat-denatured single-strand DNA irradiated with ultraviolet light as the immunogen and obtained antibodies to normal DNA bases as well as to irradiated DNA. In this study, irradiated native DNA was used for immunization, and although precipitating antibodies to irradiated DNA were detected, no precipitating antibodies were detected to unirradiated native or to heat-denatured single-strand DNA. Although immunofluorescent studies showing weak reactions with nuclei of unirradiated tissue sections suggests that there might be low titers of antibodies reacting with native DNA, the preponderance of antibodies are against DNA altered by ultraviolet light. Cyclobutane-type thymine dimers and other photoproducts are present in ultraviolet-irradiated DNA (*7*). The specific photoproduct (or photoproducts) which is the immunogenic moiety and responsible for inducing antibody production has not been characterized. However, specific antibodies to photoproducts of DNA can be produced, and the serological reaction can be studied by standard immunological techniques. Specific antibodies can thus be used as sensitive markers to detect alterations in DNA and supplement physicochemical techniques used for such studies. Immunological techniques should be useful where photoproducts of DNA are destroyed by acid or enzymatic hydrolysis.

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