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Book Issue

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Stratagene has streamlined agarose gel electrophoresis and blotting. The system decreases the time required, from sample loading to prehybridization ten fold.

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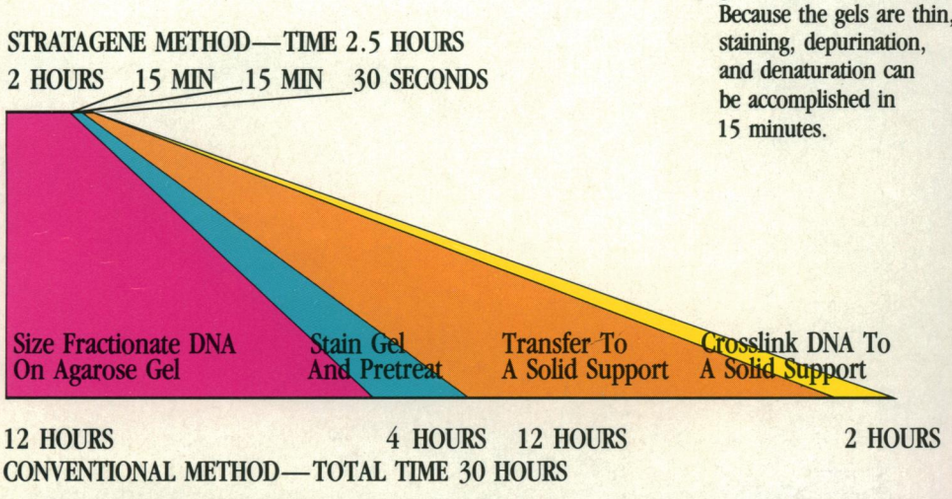


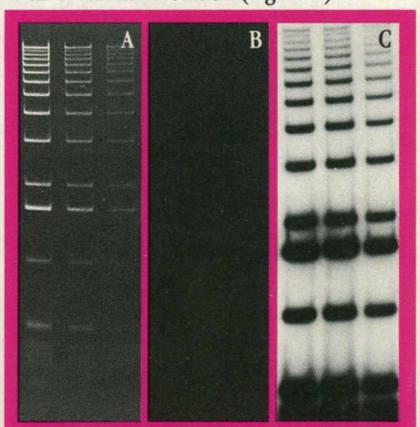
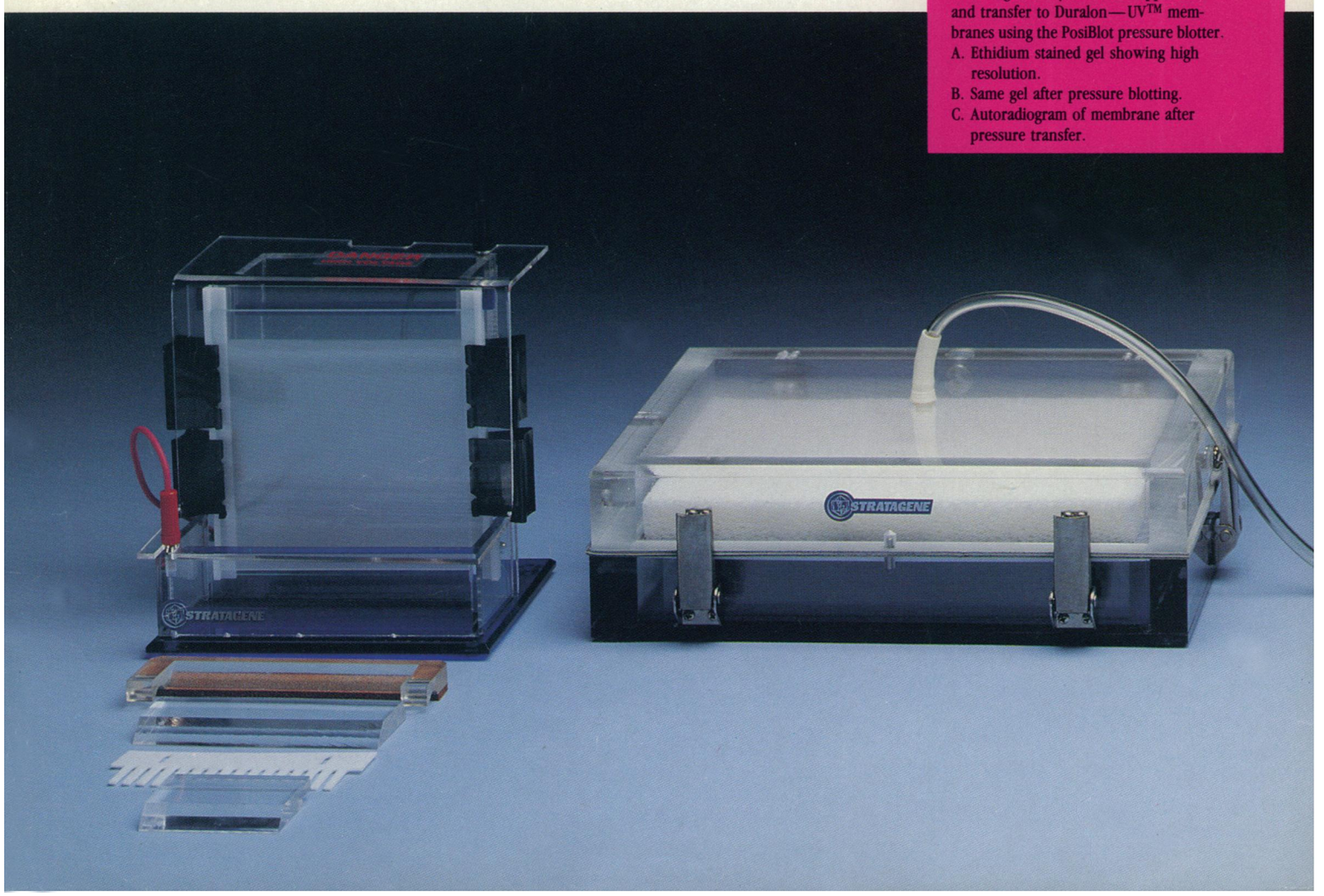


FIGURE 1:
Figure Legend: Fractionation of end labeled DNA markers on 3mm thick 0.8% agarose by the VAGE apparatus and transfer to Duralon—UV™ membranes using the PosiBlot pressure blotter.
A. Ethidium stained gel showing high resolution.
B. Same gel after pressure blotting.
C. Autoradiogram of membrane after pressure transfer.



PosiBlot™* Pressure Blotter

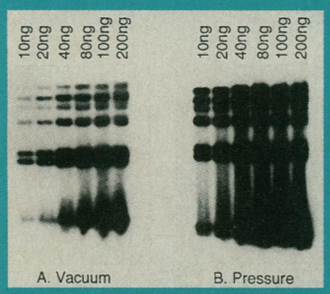


FIGURE 2:
Figure Lengend: ^{32}P end-labeled lambda Hind III markers were electrophoresed in 0.8% agarose. The DNA was then transferred to a nylon membrane with a vacuum blotter at 30mm Hg below atmospheric or with the PosiBlot pressure blotter at 100mm Hg above atmospheric. Both transfers were carried out for 15 minutes. As can be seen, pressure blotting transferred significantly more DNA in the same period of time, especially in the higher molecular weight range (largest band is 23 kilobases).

The PosiBlot™ positive pressure blotter permits the transfer of nucleic acids in 1/3 the time of vacuum blotters and 1/50 the time of capillary blotting (Figure 2). Pressure blotting does not dehydrate gels as do other methods. This allows the use of substantially higher pressure differentials, compared with vacuum blotting, without gel collapse. The PosiBlot apparatus reduces blotting time to 15 minutes.

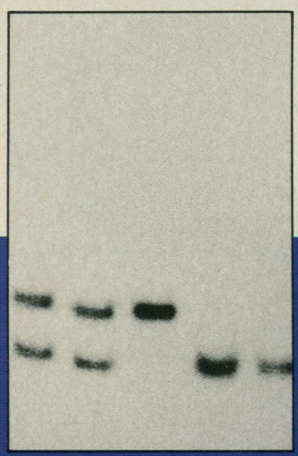


FIGURE 3:
Figure Legend: Autoradiogram showing the resolution of 2.8 and 1.3 Kb Msp I RFLP alleles revealed by a cystic fibrosis human DNA probe using the VAGE, PosiBlot and Stratalinker all in 2.5 hours.

Stratalinker™* UV Crosslinker

The Stratalinker™ UV Crosslinker fixes nucleic acids to solid supports such as nitrocellulose or nylon membranes, in less than one minute. This compares favorably to vacuum baking, which requires 2 hours. The Stratalinker actually monitors the ultra violet energy flux and deactivates the light source upon reaching the user-programmed energy level (Figure 4). Figure 3 shows an autoradiogram of a human genomic Southern blot performed using the VAGE, PosiBlot and Stratalinker all in 2.5 hours.

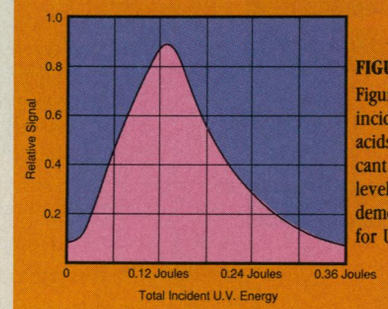


FIGURE 4:
Figure Legend: The effects of altering the incident energy for crosslinking nucleic acids to nylon membranes. The significant drop in signal intensity at energy levels below and above 0.12 Joules demonstrates the limited optimal range for UV treatment.

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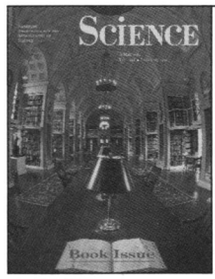

COVER Reading room, Boston Athenaeum. Housed in a building that has been described as combining "the best elements of the Bodleian, Monticello, the frigate *Constitution*, a greenhouse, and an old New England sitting room," the Athenaeum 183 after its founding "remains a retreat for those who would enjoy the humanity of books." See page 870. [Photograph by B. Metta/Uniphoto]

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Monitoring Mercury's magnetosphere

SODIUM in the atmosphere of Mercury may trace a profile of the planet's magnetosphere (page 835). Potter and Morgan have found, using two-dimensional spectral images of sodium emissions across Mercury, that the distribution of sodium varied from day to day and that frequently sodium was concentrated in two high latitude regions, one in the planet's northern hemisphere and the other in the southern hemisphere. They speculate that Mercury's magnetic substorms bring ions and electrons to the surface at high latitudes; ions caught in magnetic field lines would spiral along the field lines toward the poles until they are lost in space or come in contact with the surface. At the surface, either through sputtering or from ion neutralization, neutral sodium atoms would be generated. Although the magnetic field of the planet would determine where the ions and electrons will surface, this could vary along with variations in the solar wind.

Chelating superconductor surfaces

CREATION of new and better electronic devices and fine tuning of available ones depends on detailed knowledge of steric and electronic "landscapes" of semiconductor surfaces. One way to evaluate the structure of a surface is through "chelate effects," because the binding of a chelating agent to surface sites can perturb the surface electric field and this changes the optical properties of the interface. Lisensky *et al.* demonstrate that the photoluminescence of an electronic surface is enhanced when a chelating agent is added (page 840): semiconducting surfaces made of cadmium sulfide and cadmium selenide were exposed to amines dissolved in hydrocarbon solutions, and chelation between cadmium ions and the amines would be expected to occur at the interface of semiconductor and solution. Because the changes in photoluminescence could only be brought about if certain structural configurations pertain at the surface, the molecular form of the surface could be inferred from its chemical reactivity. This type of probe provides a nondestructive in situ technique for characterizing electro-optical devices.

Normal function for heat shock proteins

HEAT shock proteins (Hsp 70) are constituents of normal cells; they are also produced in abundance in cells exposed to various forms of metabolic stress. Thus, Beckmann *et al.* propose that, despite their name, Hsp 70 molecules actually are essential players in a routine process of normal cells, protein synthesis and assembly (page 850). Studies in nonstressed human cells show that Hsp 70 molecules bind transiently to newly synthesized portions of other proteins; as the nascent proteins mature, Hsp 70 molecules dissociate to allow protein folding, the assembly of monomeric subunits into multimers, and the movement of the protein through the proper pathways of the cell. In stressed cells, Hsp 70 molecules and nascent proteins remain bound together longer, perhaps because amino acid analogs that have been incorporated into the proteins prevent the protein from folding properly; the prolonged association may keep the complex soluble until the protein can be degraded. If free and bound forms of Hsp 70 molecules are normally in equilibrium inside cells, the overproduction of Hsp 70 molecules by stressed cells may represent a cellular response to the inaccessibility of bound Hsp 70 molecules and an attempt to reestablish the equilibrium.

Prokaryotic molecular chaperone

MANY proteins that are destined for export from bacterial cells bind to SecB, a molecular chaperone. SecB facilitates the movement of proteins through the export machinery of the cell to the cell's surface or outside. Maltose-binding protein is one of the proteins of *Escherichia coli* whose export is facilitated by SecB. It, like most exported proteins, has an attached amino-terminal "leader" segment that is essential for export but is later removed. SecB was found to bind to maltose-binding protein at a site that was separate from the leader sequence (page 860). It could bind to and form a stable complex with unfolded precursor molecules as well as with the mature unfolded protein. Randall *et al.* suggest that the leader sequence modulates protein folding such that other parts of the protein (not the leader itself) are exposed for recognition by and attachment to the molecular chaperone.

Immunosuppressing twisted amide surrogate

THE pharmaceutical FK506 is a powerful new immunosuppressive substance that may have uses in treating autoimmune diseases and in preventing graft rejection after transplantation. The drug binds to the receptor FKBP, a rotamase enzyme that can rotate the hindered bond between proline and another amino acid. Rosen *et al.* have synthesized FK506, cloned and produced the rotamase, and studied the interaction of the two spectroscopically (page 863). An understanding of how the two interact could lead to the design of even more potent immunosuppressors and should also provide insights into the molecular events that promote or block the activation of cells of the immune system. The interaction of FK506 and rotamase is completely reversible: no covalent bonds are established. Instead the α-keto amide of FK506 acts like a twisted amide bond, occupying the site for such a structure in the active site of the rotamase. It is interesting that another immunosuppressive agent, cyclosporin A, which is not structurally similar to FK506, also induces immunosuppression by binding to a rotamase.

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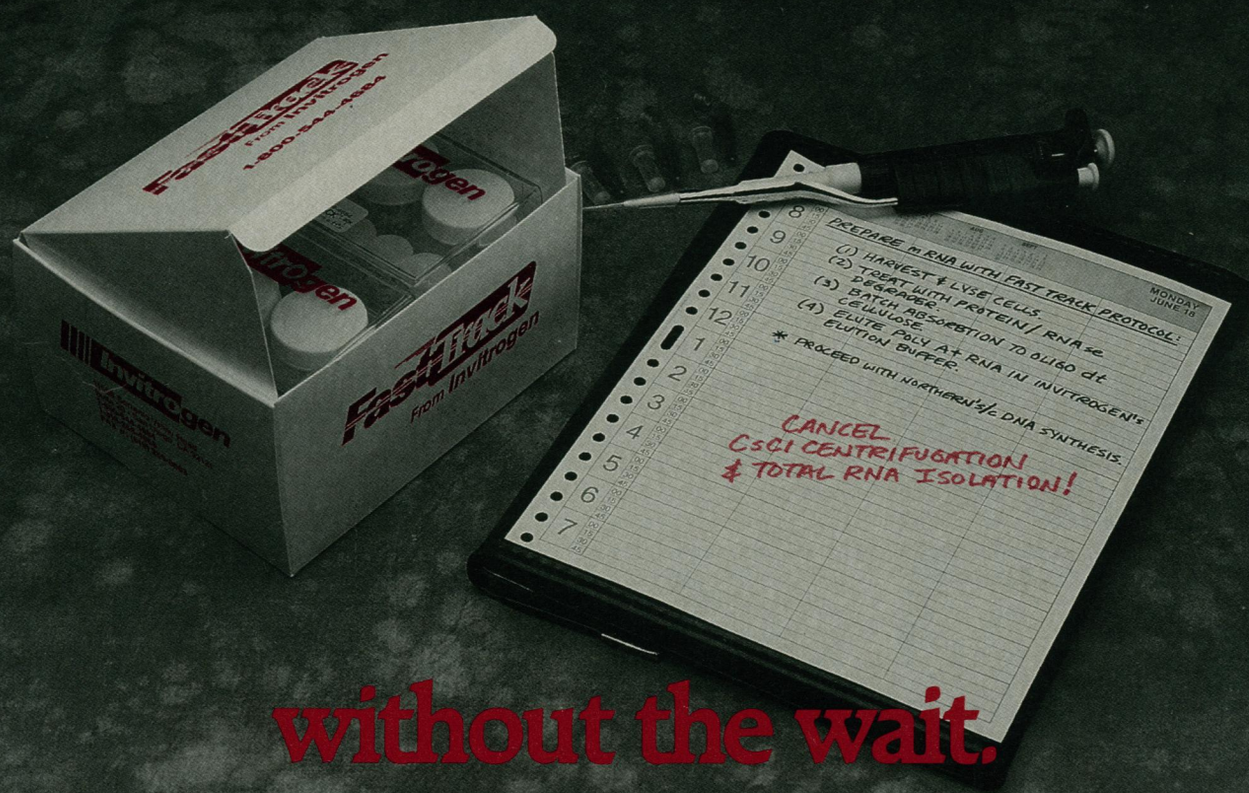

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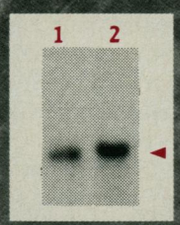

Figure 1.
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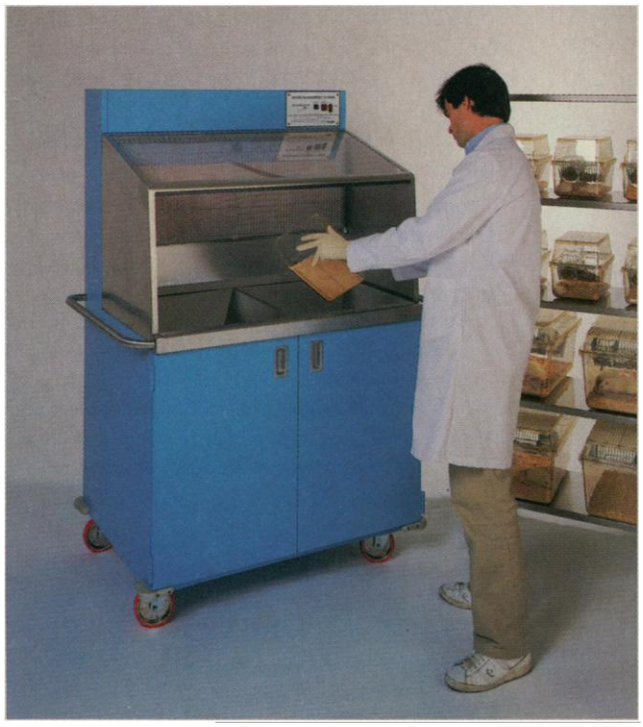

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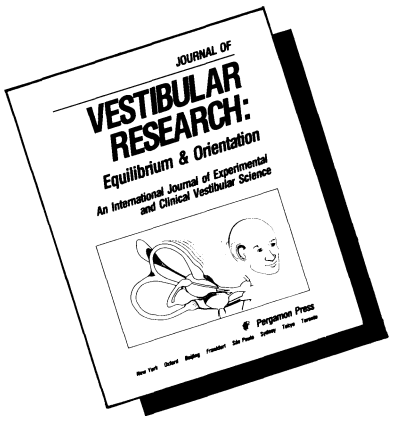

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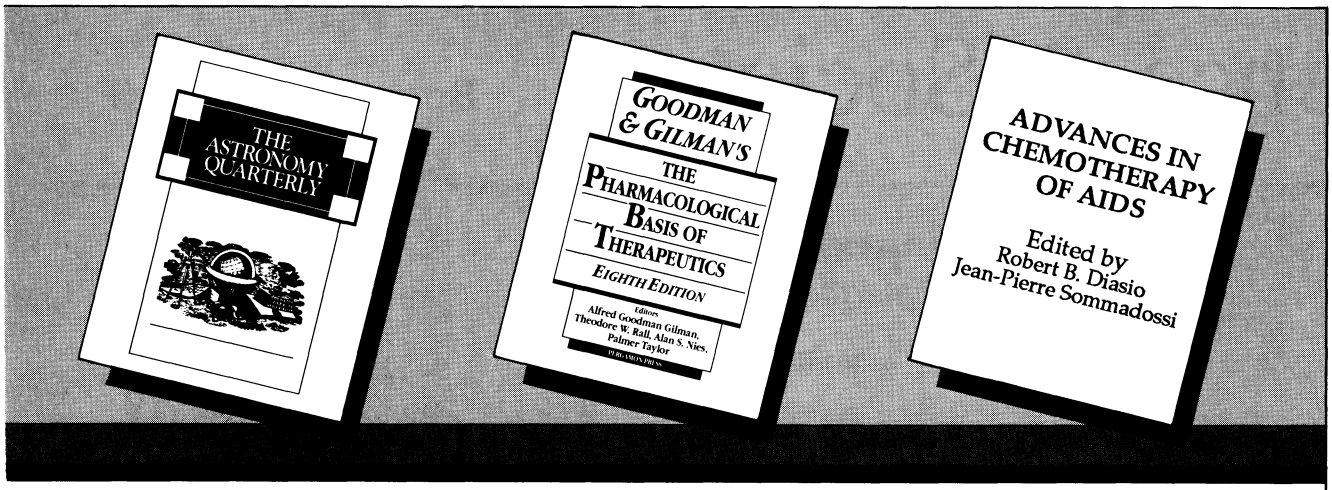

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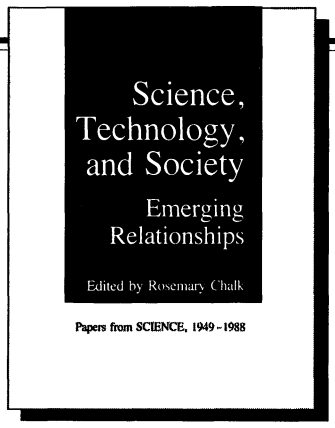

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Edited by Rosemary Chalk

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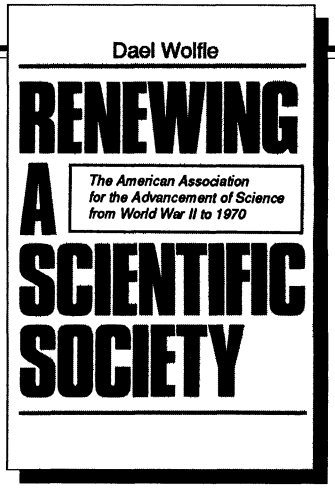

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Dael Wolfle

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