

Twelve percutaneous renal biopsies were performed in patients with acute, subacute and chronic glomerulonephritis as well as lupus nephritis; biopsy specimens were processed simultaneously for light microscopy, immunohistology, electron microscopy and tissue culture.

A piece of cortex approximately 1 mm × 6 mm was chopped, washed, subjected to Trypsin and EDTA action for 20 minutes. Dissociated cells were cultured in Leighton tubes with growth media containing 30% fetal calf serum, incubated at 37° and 5% CO<sub>2</sub> in air.

One drop of cell suspension was stained with rabbit antihuman 7S gamma globulin and antihuman complement ( $\beta_{1c}-\beta_{1a}$ ) labelled with fluorescein isothiocyanate prior to culture.

Subcultures were prepared every week by 1:2 dilution and monolayers, grown on coverslips, stained with immune stains at weekly intervals.

Specific peripheral staining for 7S gamma globulin as well as for complement was observed on dispersed cells prior to culture.

Staining for antihuman 7S gamma globulin continued for several passages while staining with anti ( $\beta_{1c}-\beta_{1a}$ ) disappeared rapidly.

Six control biopsies from individuals with non-immunological renal diseases did not show any immune staining on the first day or when propagated in the tissue culture under the same condition. (SPR)

**130 The Use of Azathioprine in Nephrotic Syndrome not Amenable to Steroid Therapy.** WILLIAM T. KNIKER\*,

WILLIAM J. FLANIGAN\* and GEORGE L. ACKERMAN\*, Clin. Study Center, University of Arkansas Medical Center, Little Rock, Ark. (introduced by E.R. Hughes).

Twenty-two patients (11 children and 11 adults) with nephrotic syndrome were treated with azathioprine (Az) for periods of 2 to 31 months (average 14). All had been unresponsive to prednisone, required undue dosage, or could not tolerate the drug. Az was given daily in a dose of 3 mg/kg; steroids were used concomitantly in a third of the cases. Serial renal biopsies were obtained on each patient, the first before starting Az. Thirteen patients had proliferative glomerulonephritis, six rapidly progressive nephritis (RPN), two membranous changes, and one no histologic alterations.

Twelve cases did well, achieving sustained clinical remission and complete or nearly complete chemical remission. Two cases, both with proliferative disease, were partially improved. Eight (four proliferative; four RPN) manifested progressive disease. Six of the latter failed to respond to Az. In each of these, irreversible renal damage probably had occurred before Az therapy, as indicated by a C<sub>cr</sub> below 20 ml/min/1.73 m<sup>2</sup> and/or vascular changes in the pre-Az biopsy. In one case, Az-induced pancytopenia led to fatal sepsis. In another patient, abrupt cessation of Az led to rapid renal deterioration and death. In summary, sustained remissions followed the use of azathioprine in 12 of 15 patients treated before irreversible renal damage had occurred. (SPR)

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