THE BEHAVIOR OF A NEWLY DESCRIBED ACUTE-PHASE PROTEIN IN INFLAMMATORY JOINT DISEASE

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Abstract—Rho, a newly characterized acute-phase protein, was present in high titer in a group of 109 patients with various rheumatic diseases. Statistically significant titer elevations were demonstrated in patients with rheumatoid arthritis (RA), ankylosing spondylitis, and gout. In individual RA patients, serial titers failed to correlate with disease activity or with rheumatic seropositivity. The natural behavior of rho antigen is contrasted with that of C-reactive protein. Comments are made regarding the possible association of rubella infection with rheumatoid arthritis.

INTRODUCTION

A recently described acute-phase protein, designated rho, was initially found in the sera of patients with rubella infection, as well as in the supernatant fluids of rubella virus infected cell cultures (1,2). Although early studies seemed to indicate that rho was a structural component of the rubella virion, it later became clear that a variety of different primate species, including man, would produce rho antigen in response to a number of infectious as well as noninfectious stimuli (2,3). Its natural behavior seemed analogous to, but not identical with, that of C-reactive protein.

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The antigen migrates in the β-1 zone in immune electrophoresis and has a density of 1.305 g/ml, as measured by centrifugation in CsCl. It is present in too low a concentration to be seen by routine examination of sera. In this study and in previous studies, the antigen was measured by double diffusion, using a potent hyperimmune antiserum (2). Using this sensitive test, the antigen can be detected in most human sera.

Because of interest in the possible relationship between rubella and rheumatoid arthritis (RA), a small number of sera from patients with RA had been titrated for rho antigen soon after its discovery. Not only were elevated levels found, but it also appeared that the rho titer might be useful in distinguishing active from quiescent disease, and that the titer might also help distinguish rheumatoid arthritis from a variety of other inflammatory and noninflammatory arthritic diseases. The present study was undertaken to investigate and expand these preliminary findings. We were particularly interested in learning whether determination of the serum rho antigen titer would prove useful either diagnostically or prognostically for an individual patient with inflammatory joint disease.

**MATERIALS AND METHODS**

**Clinical Specimens.** Sera and synovial fluids were collected prospectively from patients during their routine visits to the arthritis clinics at the Veterans Administration Hospital in West Haven, Connecticut, and at the Yale-New Haven Hospital, New Haven, Connecticut. A total of 177 sera were obtained from 109 patients. A wide spectrum of rheumatic diseases was represented, and patients with acute and chronic disease as well as active and quiescent disease were available in each of the larger clinical categories. The largest single group comprised 46 patients with classical or definite RA, according to the criteria and exclusions established by the American Rheumatism Association (4). They were generally stable, with mild to moderately active disease of several months to 10 years' duration. Serial serum specimens from a number of patients permitted us to compare the rho titer with changing clinical conditions. When multiple specimens were available for a single patient, a geometric mean titer (GMT) was calculated for that patient, and this mean was used to compute the GMT for the group.

**Rho Antigen.** A sample of 130 ml human serum with a rho titer of 1:4 was centrifuged for 18 h at 22,500 rpm. The top layer containing lipid was removed. The cleared serum was dialyzed overnight against 0.0033 M sodium phosphate buffer, pH 5.2. The white precipitate that formed within the dialysis bag was separated by centrifugation, washed twice with the same buffer, and dissolved in 2 ml 0.1 M NaCl, 0.01 M Tris, pH 8.5 (Ts buffer). A 1:12 dilution of this solution was used as the rho antigen control in the standard immunodiffusion test.

**Hyperimmune Anti-Rho Rabbit Serum.** Hyperimmune serum was prepared as previously described (2).

**Titration of Rho Antigen by Immunodiffusion.** Tests were done on 75 x 25 mm glass slides covered with 2.5 ml 0.4% agarose gel in Ts buffer containing 0.1% sodium azide. Aliquots of 20 μl of sera and antisera were placed in wells 5 mm in diameter and 3 mm