Infection by *Mycoplasma pneumoniae* leads to a wide range of clinical manifestations, pneumonia being the most common (1). When pneumonia occurs it can be differentiated from most viral and rickettsial infections only by specific laboratory tests. Thus, laboratory diagnosis of *Mycoplasma pneumoniae* pneumonia is relevant for the early initiation of specific antibiotic therapy (2).

The complement fixation (CF) test has been widely used for the serological diagnosis of *Mycoplasma pneumoniae* infection. However, this test is hampered not only by its technical complexity but also by the existence of some glycolipid cross-reactions with membrane components of host tissues and other bacterial agents (3). In recent years the detection of specific IgM antibody by μ-chain capture enzyme-immunoassay (EIA) has been described and evaluated for the early diagnosis of *Mycoplasma pneumoniae* infection (2, 4, 5, 6). This type of test offers practical advantages, allowing diagnosis in the very early phase of the disease, when CF serum antibodies have not yet reached significant levels (5).

We present in this short report the evaluation of two recently developed commercial assays, representing two different approaches to the serological diagnosis of *Mycoplasma pneumoniae* infection: a microparticle agglutination (MAG) assay for titrating *Mycoplasma pneumoniae*-specific antibody and an antibody-capture, enzyme-labelled antigen immunoassay (ELA-EIA) designed to detect both specific IgG and total (IgM + IgG + IgA) antibody to *Mycoplasma pneumoniae*.

**Materials and Methods.** Two panels of serum samples were used. Panel 1 comprised 169 samples taken from 120 patients with *Mycoplasma pneumoniae* pneumonia as demonstrated by detection of seroconversion, rising titers, or single high titers (≥ 128) for specific CF antibodies. Patients ranged in age from 8 months to 76 years (103 were aged 5 to 49 years). The time elapsed from the clinical onset was known at sampling in 88 cases (128 samples). Paired serum samples were obtained from 48 patients. Panel 2 included 53 serum samples taken from 39 patients with non-*Mycoplasma pneumoniae* pneumonia (control group). The etiological diagnosis was obtained in all cases by the CF test, using a panel of CF antigens from different pathogens involved in lower respiratory tract diseases. Seventeen cases were due to influenza B virus, eight to respiratory syncytial virus, seven to *Coxiella burnetti*, three to adenoviruses (unknown serotype), two to influenza A virus and the remaining two to parainfluenza type 3 virus. Patients ranged in age from 1 month to 84
years. The *Mycoplasma pneumoniae* CF antibody titer was < 64 in all of these serum samples.

CF antibody titers to *Mycoplasma pneumoniae* and other antigens were measured according to Grist et al. (7). CF antigens were purchased from the Institute Virion, Switzerland. MAG assay antibody titers to *Mycoplasma pneumoniae* were determined by a commercial assay (Serodia-Myco II; Fujirebio, Japan). Briefly, artificial gelatin particles sensitized with cell membrane components of *Mycoplasma pneumoniae* (Mac strain) were mixed with equal volumes of twofold serial dilutions of heat-inactivated (56 °C, 30 min) test samples in standard U-shaped microtiter plates. After incubation for 3 h at room temperature, agglutination titers were recorded following the manufacturer's directions.

IgM and total antibody to *Mycoplasma pneumoniae* were measured by a commercial ELA-EIA method (MpTest; Diatech Diagnostica, Israel). Each test sample was assayed on two microtiter wells coated with anti-human IgM or Ig/GMA antibodies, respectively. After washing, specific antibodies bound to the solid phase were detected by adding an alkaline phosphatase-labelled *Mycoplasma pneumoniae* antigen. Paranitrophenil phosphate was used as chromogen. Testing of samples and interpretation of results were performed following the manufacturer's protocol. Results were expressed as the ratio of the optical density (OD) of the test sample to the cut-off OD value (EIA index). Samples giving an EIA index ≥ 1.0 were considered positive.

Selected serum samples were tested by the ELA-EIA test after fractionation by sucrose gradient ultracentrifugation according to Palmer et al. (8). The IgG and IgM content of the fractions was monitored by single radial immunodiffusion (LC Partigen; Behring Institute, FRG).

**Results and Discussion.** Figure 1 shows the results obtained by the MAG assay and the ELA-EIA on serum samples from the control group (Panel 2). On the basis of these results and the information given in the package insert of the MAG assay, we considered a MAG assay titer of ≥ 160 to be diagnostically significant. Therefore, we used this criterion to analyze the results obtained by the MAG assay on samples from Panel 1. Consequently, one sample from the control group obtained from a patient with influenza B infection gave a significant MAG assay titer (160). Positive results for specific IgM were obtained for two samples by the ELA-EIA assay (EIA index: 2.3 and 1.1 for IgM; 2.5 and 0.6 for total antibody). These samples were obtained from two patients with parainfluenza 3 and influenza B virus infections, respectively, and gave negative results by the MAG assay.

Table 1 shows the results obtained for samples from Panel 1. Among patients over 15 years, the MAG assay failed to detect three cases of infection and the ELA-EIA assay was negative for specific IgM antibody in nine cases. The results...