Acyclovir and Prednisolone Treatment of Acute Infectious Mononucleosis: A Multicenter, Double-Blind, Placebo-Controlled Study

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Ninety-four patients with infectious mononucleosis and symptoms \(<7\) days were randomized to treatment with oral acyclovir (800 mg 5 times/day) and prednisolone (0.7 mg/kg for the first 4 days, which was reduced by 0.1 mg/kg on consecutive days for another 6 days; \(n = 48\)), or placebo (\(n = 46\)) for 10 days. Oropharyngeal Epstein-Barr virus (EBV) shedding was significantly inhibited during the treatment period \((P = .02,\) Mann-Whitney rank test). No significant effect was observed for duration of general illness, sore throat, weight loss, or absence from school or work. The frequency of latent EBV-infected B lymphocytes in peripheral blood and the HLA-restricted EBV-specific cellular immunity, measured 6 months after onset of disease, was not affected by treatment. Thus, acyclovir combined with prednisolone inhibited oropharyngeal EBV replication without affecting duration of clinical symptoms or development of EBV-specific cellular immunity.

Primary Epstein-Barr virus (EBV) infection is usually asymptomatic. Some patients, however, have clinical disease (infectious mononucleosis [IM]), which can have mild symptoms or be fatal in persons who are immunodeficient [1]. During primary infection, EBV infects oropharyngeal or genital tract epithelium with lytic replication and subsequently spreads to B lymphocytes [2]. It has been postulated that clinical symptoms may be due to EBV-induced polyclonal humoral and cellular immunoreactivity and that only limited pathology is caused by viral replication [3].

Acyclovir is a nucleoside analogue with significant in vitro activity against EBV [4]. In several double-blind placebo-controlled studies, during treatment acyclovir suppressed EBV shedding in saliva of IM patients, although EBV replication resumed after treatment was discontinued with no significant effects on individual clinical symptoms [5-8]. However, when data were combined that included duration of fever, weight loss, tonsillar swelling, and self-assessment by the patient, a significant effect of acyclovir was evident \((P < .01)\). The effectiveness of corticosteroids was conclusively proved in placebo-controlled trials: There was significant reduction in duration of fever from a mean of 6 days to 3 days when prednisolone was given at 40–80 mg/day for 12 days [9-12], abnormal hematologic findings were reduced from 7 to 3 weeks, and there was some relief from pharyngitis symptoms. However, scattered case reports of an increased incidence of myocarditis and encephalitis have hampered general use of steroids in the treatment of IM [13, 14]. These complications were possibly due to a steroid-induced inhibition of EBV-specific cellular immune responses and increased viral replication [14, 15]. Hence, a combination of antiviral and steroid therapy in the early stages of IM might reduce clinical severity due to the immunomodulatory effect of steroid treatment without having the detrimental effects of steroid-enhanced viral replication [15].

We investigated the efficacy and safety of combined treatment with acyclovir and prednisolone on acute IM in a double-blind study. Patients with IM, who had been ill \(<7\) days, were treated with acyclovir and prednisolone or acyclovir and placebo for 10 days. Clinical parameters, oropharyngeal EBV shedding in saliva, and the development of HLA-restricted EBV-specific cellular immune reactivity were assessed.

Patients and Methods

The double-blind, placebo-controlled trial was done between 1988 and 1991 at the Department of Infectious Diseases, Danderyd and Roslagstull Hospital, Sweden, and East Birmingham Hospital (since renamed Birmingham Heartlands Hospital), United Kingdom. Patients 14-30 years old who were suspected of having IM were referred to the hospitals by primary health practitioners. Patients were eligible for the study if they had clinical features of IM (sore throat, lymphadenopathy, and general malaise), symp-
Acyclovir-Steroid Therapy of Infectious Mononucleosis

...nuclear cells in peripheral blood, and a positive heterophil antibody...tution and patients with current or past peptic ulceration, diabetes mellitus, hypertension, glaucoma, tuberculosis, or any condition in which steroid therapy is contraindicated and those who had received antiviral or steroid therapy in the preceding 2 weeks, those receiving phenytoin, phenobarbitone, ephedrine, or rifampicin, and those with impaired renal function (serum creatinine >120 μmol/L).

Two computer-generated randomization codes, one for the United Kingdom and one for the Swedish combined center, were prepared by the Department of Statistics, Wellcome Research Laboratories (Beckenham, UK). The randomization code was open to the hospital chief pharmacist but blinded to the investigators. After study entry and clinical examination, patients were allocated to oral or intravenous (iv) treatment, depending upon their ability to swallow and whether they were vomiting. Patients on oral or iv treatment were entered on opposite ends of the randomization code. About 40% of the patients were also randomly selected for the virologic assessments detailed below.

All patients were entered in sequential order according to the randomization code to receive a 10-day course of treatment with acyclovir plus prednisolone or placebo. The acyclovir treatment was 800 mg orally 5 times/day or 5 mg/kg as a 1-h iv infusion every 8 h until oral medication could be taken. The prednisolone dosage was dependent on the patient’s weight at study entry. Oral steroid treatment consisted of 0.7 mg/kg prednisolone daily (the total daily dosage was rounded to the nearest 5 mg) for the first 4 days; this was reduced by 0.1 mg/kg/day until prednisolone was withdrawn on day 10. We used an equivalent dosing schedule of continuous iv hydrocortisone, starting at 3.5 mg/kg/day, for patients who required iv therapy. Patients randomized to receive placebo were given matching placebo tablets or infusions.

Vials containing freeze-dried preparations of 250 mg of acyclovir (sodium salt) or placebo for reconstitution with 10 mL of water for injection B.P. (British Pharmacopoeia) or sodium chloride infusion B.P., tablets containing 800 mg of acyclovir or matching placebo, and 5 mg tablets of prednisolone or matching placebo were supplied, labeled, and coded by Wellcome Research Laboratories and Kabi (Stockholm) and packed in similarly labeled boxes. Hydrocortisone and matching saline for iv infusion were prepared by the hospital pharmacies.

During the trial, patients were not allowed concomitant therapy with nonsteroidal antiinflammatory drugs.

Clinical observations. Patients were examined clinically daily while hospitalized, otherwise on days 3 or 4, 7, 9, or 10 and 14 and thereafter at weekly intervals until their symptoms and signs had returned to normal. Follow-up assessments were done at the hospital at months 3 and 6. At each assessment, the following symptoms and signs were recorded and scored: general health (0 = well, 1 = slightly unwell, 2 = moderately ill, 3 = severely ill), psychologic well being (0 = normal, 1 = slightly depressed, 2 = depressed, 3 = very depressed), sore or painful throat (0 = none, 1 = mild, 2 = moderate, 3 = severe), difficulty in breathing (yes/no), difficulty in swallowing (0 = none, 1 = solids, 2 = fluids, 3 = saliva), oral temperature, body weight, tonsillar swelling (0 = none, 1 = mild, 2 = moderate, 3 = severe), tonsillar exudate (yes/no), cervical lymph nodes enlarged (yes/no) or tender (yes/no), liver enlarged (yes/no) or tender (yes/no), spleen enlarged (yes/no), and rash present (yes/no). Until symptoms and signs resolved (i.e., while weekly follow-ups continued), patients conducted daily self-assessments and recorded their symptoms on a diary card. In addition to the symptoms listed above, patients recorded tiredness (0 = none, 1 = mild, 2 = moderate, 3 = severe), difficulty in sleeping (yes/no), headache (yes/no), nocturnal sweating (yes/no), self-medications taken, and whether the day was missed from school or work (if unemployed or on holiday, whether the day would have been missed).

Serology. Blood was taken for serologic measurements on days 0 and 14 and at 3 and 6 months of follow-up. Heterophile antibodies were determined by the Davidsohn and Lee test [16]. Specific EBV serology was done for all patients and included titration of antibodies to EBV-associated nuclear antigens (EBNA) 1 and 2 and IgM and IgG antibodies to viral capsid antigen (VCA). These tests were done at the Department of Virology, Swedish Institute for Infectious Disease Control, Stockholm, and at the Regional Department of Immunology, East Birmingham Hospital. The methods, which have been described, are highly specific for diagnosing primary EBV infection [17]. The diagnosis of acute EBV infection was confirmed by demonstration of IgM antibody against VCA and titers of antibody to EBNA1 ≤1:5 on days 0 and 14.

Thirty-four patients (8, UK; 26, Sweden) were randomly selected for virologic and immunologic assessments on days 0, 7, 9 or 10, and 14 and months 3 (day 90) and 6 (day 180).

Virologic assessment. Semiquantification of oropharyngeal EBV shedding was done with throat washings. A patient was asked not to swallow for 5 min and then to rinse his or her mouth for 30 s with 10 mL of RPMI 1640 medium (GIBCO, Paisley, UK). Bacteria and detritus were removed by centrifugation at 1000 g for 10 min. Ten percent fetal calf serum (FCS) was added to the supernatants before they were frozen at −70°C. After thawing, saliva samples were diluted 10 3 , 10 2 , and 10 1 and added to microtiter wells containing Ficol-separated lymphocytes from umbilical cord blood. Aliquots of 2 × 10 5 cells in 0.2 mL of medium containing 10% FCS, 20 U of penicillin, and 20 μg of streptomycin were used, and each saliva dilution was distributed in 10 wells and cultured 4–8 weeks.

The presence of virus in oropharyngeal washings was detected by transformation of cord blood cells and by screening for the induction of EBNA in the same cord blood cells 7 days after infection with undiluted throat washing samples as described by Reedman and Klein [18]. The virus titer providing 50% transformation (TT 50 ) after 4–8 weeks was determined from the virus dilution curves. Thus, a semiquantitative measurement of the amount of oropharyngeal virus was obtained for each sample by two assays. The correlation between the TT 50 and the percentage of EBNA-positive cells was high (r = .92).

Precursor frequency of in vivo EBV–infected B lymphocytes. For a spontaneous outgrowth assay, lymphocytes were separated from patients’ peripheral blood on days 0 and 180 by centrifugation of heparinized whole blood over a Ficol-Isoaque cushion (1500 g for 30 min at 20°C) and stored in liquid nitrogen. The thawed lymphocytes were cultured in 10% FCS for 6 weeks at concentrations of 6 × 10 5–5 × 10 5 cells/well. Each dilution was distributed in 10 wells of a flat-bottomed microtiter plate. This method (de-
No. of patients measured on day 0 during treatment and at the 14-day assessment.

Evaluation of sore throat, duration of weight loss, absence from work.

The cell concentration that induced a 50% transformation rate (CT 50). The lower the minimum number of added cells required to achieve a 50% inhibition.

In vivo and could be quantified in terms of the strength of immune T cells in vivo and could be quantified with the frequency of EBV-infected B lymphocytes in vivo [15]. Cells exposed to the B95-8 laboratory strain of EBV were used as positive controls.

Immune assessment. We used an outgrowth inhibition assay to determine EBV-specific cellular immunity. Lymphocytes obtained at enrollment and on day 180 were thawed and added to microwells at four different concentrations (2 × 10^4 to 2 × 10^6 cells/well) in 10% FCS. Aliquots containing ~10^6 infectious U/mL EBV (B95-8) were added to cell suspensions that were cultured 6 weeks to allow outgrowth into lymphoblastoid cell lines. If EBV immune T cells were present in the patient's blood, they would inhibit outgrowth of EBV-infected B lymphocytes. The strength of this regression correlated with individual concentrations of EBV immune T cells in vivo and could be quantified in terms of the minimum number of added cells required to achieve a 50% inhibition of outgrowth of EBV-transformed cells (CT 50). The lower the cell concentration that inhibited outgrowth, the higher the in vivo proportion of EBV immune T cells [6].

Safety. All patients were questioned about and examined for adverse reactions. Hematologic and biochemical parameters were measured on day 0 during treatment and at the 14-day assessment.

Statistical methods. We plotted Kaplan-Meier curves for duration of sore throat, duration of weight loss, absence from work/school, and general health with respect to time, adjusted for center using survival analysis and Mantel-Cox statistics. For viral shedding, the Mann-Whitney rank test was used to compare the 2 groups. The study was designed to have sufficient power to detect a 66% reduction in median duration of viral shedding.

Results

During the 3-year study period, 94 patients (32 from Britain, 62 from Sweden) were randomized to participate in the study. Of the 94, 8 were excluded from evaluation: 6 because of wrong diagnoses and 1 each because of insufficiently elevated temperature at enrollment and symptoms present >7 days before enrollment. Of the 86 patients with the wrong diagnosis, 1 had primary herpes simplex infection, 2 had adenovirus infections, and 3 had illnesses of unknown etiology.

The diagnosis of primary EBV infection was serologically verified by the finding of IgM anti-VCA antibodies and anti-EBNA antibody titer of <1:10 on days 0 and 14 in the remaining 86 patients. Of these, 43 were randomized to acyclovir-prednisolone and 43 to placebo. Four patients later withdrew from the study (2 on day 2 and 1 each on days 7 and 10); another was given prednisolone openly by his general practitioner on day 6 because of clinical deterioration. In total, 81 patients were followed clinically until day 14. Fifty-two subjects were also seen at the 3-month follow-up; 49 were followed 6 months.

At enrollment, the 2 groups were comparable in terms of age, duration of illness, and severity of disease as assessed by clinical and laboratory parameters (tables 1, 2). The 2 groups had no differences in IgG and IgM antibody titers against VCA.

Clinical Findings

We decided not to use duration of fever as a primary end point in the study, since fever may have been directly affected by steroid treatment through an inhibiting effect on interleukin (IL)-1, tumor necrosis factor (TNF)-α, and prostaglandin E1 production. Fever was included in the general health assessment. There were no statistically significant differences between the groups in the resolution of any clinical parameter during the study period. Time to resolution of sore throat (figure

Table 1. Demographic details of clinical parameters of evaluable patients (n = 86) with infectious mononucleosis.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Acyclovir and steroids</th>
<th>Placebo</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of patients</td>
<td>44</td>
<td>42</td>
</tr>
<tr>
<td>No. of males/females</td>
<td>21/23</td>
<td>25/17</td>
</tr>
<tr>
<td>Mean age in years (range)</td>
<td>18.5 (14-29)</td>
<td>18.7 (14-25)</td>
</tr>
<tr>
<td>No. of days with symptoms before admission (range)</td>
<td>4.7 ± 1.4 (2-7)</td>
<td>4.8 ± 1.3 (2-7)</td>
</tr>
<tr>
<td>No. with severe symptoms</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>No. with mild-to-moderate symptoms</td>
<td>39</td>
<td>37</td>
</tr>
<tr>
<td>No. with sore throat score 3</td>
<td>15</td>
<td>17</td>
</tr>
<tr>
<td>No. with sore throat score 0-2</td>
<td>28</td>
<td>25</td>
</tr>
<tr>
<td>No. with palpable liver enlargement</td>
<td>14</td>
<td>20</td>
</tr>
<tr>
<td>No. with palpable spleen</td>
<td>23</td>
<td>22</td>
</tr>
<tr>
<td>No. hospitalized</td>
<td>14</td>
<td>19</td>
</tr>
</tbody>
</table>

* Evaluated by physician. For scoring system, see Patients and Methods.

† Evaluated by patient, see Patients and Methods.

Table 2. Laboratory parameters of evaluable patients with infectious mononucleosis enrollment (n = 86).

<table>
<thead>
<tr>
<th>Patients with</th>
<th>Acyclovir and steroids</th>
<th>Placebo</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heterophile antibody test positive</td>
<td>44</td>
<td>41*</td>
</tr>
<tr>
<td>Positive VCA IgM and VCA IgG, anti-EBNA &lt; 1:10</td>
<td>44</td>
<td>42</td>
</tr>
<tr>
<td>Elevated liver enzymes = ALAT &gt; 0.7 µkat/L (n = 70)</td>
<td>35</td>
<td>35</td>
</tr>
<tr>
<td>Atypical lymphocytes present (&gt;5%)</td>
<td>40</td>
<td>38</td>
</tr>
<tr>
<td>Mean (range) of white blood cell count (×10^9/L) (n = 85)</td>
<td>11.8 (4.3-32.6)</td>
<td>12.4 (3.4-21.5)</td>
</tr>
</tbody>
</table>

NOTE. VCA, viral capsid antigen; EBNA, Epstein-Barr virus (EBV) nuclear antigen; ALAT, alanine aminotransferase.

* 1 negative became positive on day 3.
Figure 1. Kaplan-Meyer life table analysis of clinical effects of combined acyclovir (ACV)-steroid treatment vs. placebo-placebo treatment. 

A, Duration of sore throat until 15 days after enrollment. B, No. of patients who had not returned to baseline weight. C, Duration of time away from school/work for 44 patients given ACV steroids and 42 who received placebo-placebo. No significant differences were noted between groups in A–C.

Sore throat. At follow-up days 2–4, 4 (9%) of 44 patients in the acyclovir-steroid group had severe pharyngeal discomfort (score = 3) compared with 10 (24%) of 41 in the placebo group. At the 14-day follow-up, 95% of each group no longer had a sore throat. The median duration of sore throat was 9 days for acyclovir-steroid patients and 7 days for placebo recipients (figure 1A).

Body weight. At day 14 of follow-up, 13 (31%) of 42 acyclovir-steroid patients had not yet regained baseline weight compared with 15 (40.5%) of 37 placebo recipients (figure 1B).

Duration of illness. In the acyclovir-steroid group, the median duration of absence from school/work was 13 days (range, 2–42; mean, 15 ± 2 days). The placebo group had a corre-
responding median time of 11 days (range, 1–98; mean, 17 ± 4 days; figure 1C).

The parameters general health, sore throat, and tender cervical lymph nodes, respectively, were improved by day 3 in 40%, 70%, and 46% of patients in the acyclovir-steroid group and in 26%, 40%, and 28% of those in the placebo group. The hazard ratio, which measured the time for improved general health in the acyclovir-steroid group relative to the placebo group, was 0.01, suggesting a trend in favor of acyclovir-steroid treatment. By day 7 and thereafter, this difference was no longer visible.

Other Clinical Complications

One patient in the acyclovir-steroid group had signs of encephalitis at enrollment. He recovered without any sequelae. One patient (also in the acyclovir-steroid group) had facial palsy at enrollment and on day 9 when the patient withdrew from the study.

Laboratory Parameters

Hemoglobin concentration, total white blood cell count, presence of atypical lymphocytes (≥5%), platelet count, prothrombin time, bilirubin, liver enzymes (alkaline phosphatase, aspartate and alanine aminotransferase), and creatinine levels were comparable in the 2 groups at baseline. Two patients had clinical jaundice at enrollment (both in the acyclovir-steroid group); 70 (83%) of 84 patients had elevated liver alanine aminotransferase levels (0.8–26.5 μkat/L; normal, <0.7 μkat/L).

The decline in the total white blood cell count was somewhat faster in the placebo group, possibly due to the well-recognized steroid effect on granulocyte chemotaxis. The blood film was followed in more detail in 54 Swedish patients (27 in each treatment arm). The number of atypical lymphocytes (mean, 20% ± 11% in both groups at enrollment) normalized more rapidly in the acyclovir-steroid group. On treatment day 7, patients who received acyclovir and steroid had 8% ± 11% atypical lymphocytes and those in the placebo arm had 12% ± 10%. Platelets also normalized faster in acyclovir-steroid recipients than in the placebo group (data not shown). Neither group had evidence of renal impairment (table 3).

Virology

Oropharyngeal EBV shedding. We found no statistically significant differences between the 2 groups in median values of oropharyngeal EBV shedding at enrollment (figure 2). Acyclovir and prednisolone treatment effectively suppressed oropharyngeal EBV replication, with a significant reduction of the EBV titer of the mouthwash samples at assessment days 7 and 10, respectively (P = .02, Mann-Whitney rank test). However, this effect was transient, and EBV shedding returned to baseline values at day 14, after acyclovir-prednisolone treatment was discontinued. There were no significant differences between the 2 groups thereafter (figure 2).
Precursor frequency of EBV-infected B lymphocytes. The frequency of EBV-infected B cells was estimated by spontaneous outgrowth test. No differences were seen in either group ($n = 18$) in the proportion of patients with spontaneous outgrowth of EBV-infected B lymphocytes on day 0 (16 [89%] of 18) or day 180 (5 [28%] in the acyclovir-prednisolone group and 4 [22%] placebo recipients). We used the spontaneous outgrowth assay to assess a limited number of patients 7, 10, and 14 days after enrollment ($n = 11$). There was spontaneous outgrowth in 70%–90% of the patients regardless of treatment. If prednisolone had induced increased viral replication or impaired the establishment of EBV-specific immunity, the incidence of EBV-infected B cells at day 180 would have been higher in the active treatment group than in the placebo group.

**Immunology**

**Humoral immune response.** The diagnosis of primary EBV infection was serologically verified in all 86 evaluable patients. All patients had IgM and IgG VCA and EBNA negativity. Of the 94 enrolled patients, 88 had a positive heterophile test on day 0. Eighty-seven cases of IM were verified with EBV-specific serology. One patient had a false-positive heterophile test and did not have a primary EBV infection. Titration of antibody levels to IgM and IgG classes against VCA was done at study entry. There were no differences between groups.

**Cellular immune response.** Previous studies have shown that almost 100% of IM patients lack HLA-specific T cell-mediated EBV immunity at onset of disease [2, 3]. This was investigated at enrollment and at 180 days. The outgrowth inhibition assay determined the incidence of HLA-restricted T cell-mediated immunity against autologous EBV-infected B cells. Before treatment only 2 of 35 patients had evidence of outgrowth inhibition. All patients investigated ($n = 31$: 16, acyclovir-steroid arm; 15, placebo arm) had evidence of T cell-mediated cellular immunity on day 180. The mean frequency of immunocompetent cells ($C_{0.5}$) was $0.1–0.8 \times 10^5$ mononuclear cells. There were no differences between the treatment groups: $C_{0.5} = 0.17–0.18 \times 10^5$ (figure 3).

![Figure 2](https://example.com/figure2.png)

Figure 2. Median virus titer providing 50% transformation ($TT_{50} \pm SD$) for Epstein-Barr virus (EBV) in mouthwash samples randomly selected from 34 IM patients: 17 treated 10 days with acyclovir-prednisolone combination and 17 with placebo-placebo. $TT_{50}$ is sample dilution providing 50% transformation of cord blood lymphocytes in wells in lymphoblastoid cell lines and verified by immunofluorescent staining for EBV nuclear antigen-positive cells. Statistically significant inhibition of viral shedding was achieved on days 7 and 10 with acyclovir-prednisolone regimen ($P = .02$, Mann-Whitney test). No differences were found on days 0, 14, 90, or 180.
Primary EBV infections with or without IM features have many complications that are primarily categorized by immunopathologic responses to the virus. Among these, autoimmune hemolysis, airway obstruction from enlarged tonsils, splenic rupture, encephalitis, and myocarditis are seen in otherwise healthy patients. Agranulocytosis, aplastic anemia, and other rare but grave features of acute progressive EBV infection occasionally arise in healthy patients and may indicate a demonstrable deficiency of EBV-specific cellular immunity. For such cases, specific antiviral treatment regimens are needed. Acyclovir has anti-EBV-specific activity [4]; however, despite that disappearance of viral shedding in saliva during treatment of acute IM, several studies have found no significant effect on individual clinical symptoms [5–8]. Therefore, it has been suggested that the clinical features of IM are caused by a characteristic T cell proliferation and activation [20, 21]. We have also found that monocytes/macrophages are constantly involved in the immunopathology of IM [22, 23]. Steroid therapy may down-regulate production of cytokines in monocytes and also inhibit activation of T cells in certain conditions. Moreover, prednisolone may inhibit EBV-induced autoantibody production, which may be responsible for some complications found in severe IM cases [24]. The current study sought to determine if combined treatment with acyclovir and steroid could down-regulate the EBV-induced polyclonal activation in acute IM and also inhibit lytic EBV replication.

Apart from lytic replication in the oropharyngeal epithelium, EBV can transform B cells to proliferating active lymphoblasts [25] that express a number of adhesion receptors, produce various intracellular cytokines, and up-regulate oncogenes, thus establishing intrinsic growth factors [25, 26]. The current concept of the pathogenesis of IM is that these cells are extremely immunogenic and trigger profound activation of the cellular immune response [26, 27]. The activation is initially polyclonal and HLA-unrestricted with limited specificity directed against EBV-derived antigens [21, 25]. We studied the local cytokine response in tonsillar tissue in patients with acute IM and found that interferon (IFN)-γ, IL-2 and -6, and TNF-α were the dominant cytokines [27, 28]. We previously found that pharmacologic steroid doses induce a dose-dependent increase in viral replication in EBV-infected cells [15]. Thus, it might be possible that addition of steroids to acyclovir for IM therapy would limit the antiviral effect of acyclovir.

Results of the present study clearly showed that this was not the case and that acyclovir combined with prednisolone efficiently inhibited EBV replication in the oropharynx. Furthermore, the number of atypical lymphocytes circulating in the blood declined more rapidly in patients treated with acyclovir and prednisolone. Similar results have been reported in studies of iv or oral acyclovir therapy alone [6, 7]. The steroid therapy also did not alter the natural cellular immunity against EBV or the precursor frequency of in vivo-infected B lymphocytes analyzed 6 months after the acute illness. Thus, in the pharmacologic doses of prednisolone used in this study, development of HLA-restricted cytotoxic T cells was not affected.

In an earlier study, the same oral dose of acyclovir was shown to significantly inhibit viral shedding in the oropharynx [7]. The major limitation of acyclovir is the low grade of absorption (10%–20%). This problem can be overcome by administration of the prodrug valacyclovir [29]. However, no augmented antiviral effect can be expected with this drug compared with results obtained with high-dose iv acyclovir [6]. Penciclovir was recently shown to effectively inhibit the productive replication cycle of EBV in vitro and may be an alternative treatment regimen [30]. However, at the acyclovir dosage used in this study, EBV replication in the oropharynx was significantly suppressed. Thus, we believe that lack of clinical efficacy in the current study was due to mechanisms other than insufficient antiviral effect on infected cells.

In the present study, we did not find any significant clinical effect on duration of overall symptoms of acute IM by treatment with a combination of acyclovir and prednisolone. This may be explained by the findings of Luedke and Cerami [31], who showed that steroid-mediated down-regulation of monokine production in macrophages could not be blocked in the presence of IFN-γ. Indeed, we recently showed that IFN-γ is elevated in serum, circulating T cells, and tonsil tissue at the onset of disease [26–28]. Thus, the induced IFN-γ expression may be an explanation for the lack of clinical effects of prednisolone in this study. However, there was a trend in favor of such treatment: Individual symptoms and signs improved during the
first few days of therapy. This could be explained by a direct steroid-mediated antiinflammatory effect in the pharynx mucosa. To see if this trend is statistically significant, further studies of severely unwell IM patients would be needed. The majority of patients in our study had moderate IM symptoms. There were no increased adverse events in patients treated with acyclovir-steroid compared with the placebo group.

We conclude that combination therapy with acyclovir and prednisolone is of limited use for patients with mild or moderate IM symptoms. In severely ill patients, however, steroid treatment may be indicated to prevent incipient airway obstruction. A study of 11 severely ill IM patients treated in intensive care units showed clinical benefits of such therapy [15]. Further studies are needed of acyclovir-steroid combination therapy in IM patients with complications requiring intensive care. Such therapy may also be of clinical value in selected cases of cytopenia caused by autoantibody production [24]. In such circumstances, the results of the present study suggest that acyclovir and prednisolone could be given without augmenting the risk of uncontrolled viral replication and without altering the delicate balance between viral latency and specific cellular EBV immunity.

Acknowledgments

We thank study nurses Berit Emilson and Pat Ogan and laboratory technicians Lena Radler and Caroline Ekberg for technical assistance, Elisabeth Berg (Karolinska Institute, Department of Medical Informatic and Medical Education, Section for Medical Statistics) for statistical evaluations, and R. Thompson (Regional Immunology Laboratory, East Birmingham Hospital).

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