

This Week's Citation Classic™

Evans A S, Niederman J C & McCollum R W. Seroepidemiologic studies of infectious mononucleosis with EB virus. *N. Engl. J. Med.* 279:1121-7, 1968.
[Dept. Epidemiology and Public Health, Yale Univ. Sch. Med., New Haven, CT]

This seroepidemiological study of infectious mononucleosis (IM) demonstrated antibody to Epstein-Barr virus (EBV) to be absent prior to development of IM, to appear regularly during illness, and to persist for years. This relationship was not found in other infectious diseases. EBV is the probable cause of IM [The SC® indicates that this paper has been cited in over 265 publications since 1968]

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"This study examined the possible relationship between antibody to EBV and IM in three serum collections. In one, sera collected by Jim Niederman from 362 Yale University freshmen who had been bled on entry into college from 1958 to 1963 and again at the time when 40 developed IM over the next four years were taken from our freezers and tested for EBV antibody. Of 268 students lacking antibody on entry, 19.4 percent developed IM over the next four years. In contrast, of 94 students entering with antibody, none subsequently developed IM. The second collection obtained and frozen by me was from patients with IM who had been admitted to the University of Wisconsin student infirmary, and the third collection was sera sent to the Wisconsin State Laboratory of Hygiene for heterophile antibody determinations. Of 135 sera from heterophile antibody cases of IM in all three collections, all were found to have EBV virus capsid antigen IgG antibody. Pre-illness sera available from 56 of these patients were uniformly negative for EBV antibody. Of 16 patients who were persistently negative for heterophile antibody but who fulfilled clinical and hematologic criteria for IM, six were EBV antibody positive, indicating that EBV might cause heterophile-negative cases, and nine were EBV antibody negative, suggesting that agents in addition to EBV could produce a mononucleosis like syndrome.

"This study confirmed and extended the preliminary report by Niederman, Bob McCollum, and the Henles¹ that EBV antibody was absent from the pre-illness sera and developed early in the course of IM in 29 patients, some showing a rising titer of antibody. Our study clearly indicated that EBV was implicated in the causation of IM and not

simply a passenger virus. The evidence was based on: 1) EBV antibody was consistently present in 135 cases of IM and invariably present in other infections and healthy controls, 2) the antibody was absent in preillness-sera from 58 persons and appeared during illness, 3) a rising titer of antibody was observed in some patients, 4) the absence of EBV antibody indicated susceptibility to the sub-sequent development of IM, whereas its presence indicated immunity, 5) a past history of IM was found only in students whose serum had EBV antibody, 6) EBV antibody persisted for years as expected of a true antibody and in contrast to the transient heterophile antibody, 7) the association of EBV antibody with IM was consistent with available epidemiological data on IM, and 8) the study also implicated EBV as one of the causes of heterophile-negative IM.

"My own interest in IM had begun years before this 1968 study. It started in 1946 when I had returned from service in the Army and was offered a fellowship at Yale by John R. Paul, who had discovered the heterophile antibody of IM in 193-2 with W.W. Bunnell² (the Paul-Bunnell test) and had maintained an active interest in the disease. We believed that IM was probably due to a filterable agent, but many attempts made by me^{3,4} and later by Niederman and Scotts⁵ to transmit the infection to volunteers were unsuccessful or at best equivocal. Some of the sera from volunteers were frozen and stored and when EBV antibody was discovered, we retested them and learned that almost all possessed antibody at the time we had inoculated them. Thus, working with immune subjects is not the best way to carry out such experiments. In 1950-1952, I re-entered the Army and carried out unsuccessful experiments in the Army Hepatitis Research Center in Munich, to transmit both EBV and viral hepatitis to monkeys.⁶ On leaving the service, I went to the University of Wisconsin, where I continued my interest in IM, making vigorous but unsuccessful attempts to isolate the elusive causative agent from throat washings, sera, and buffy coats from cases of IM.⁷ I did confirm Hoagland's concept⁸ that IM might be transmitted by intimate oral contact and proposed a concept of pathogenesis of the viral etiology of IM that has turned out to be substantially correct.⁷ Thus, after over 20 years of working with IM, I was well prepared, as were my associates, to evaluate the observation of Henle *et al.* that EBV might be the cause of IM.⁹ For a recent review, see reference 10."

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5. Niederman J C & Scott R B. Studies on infectious mononucleosis. Attempts to transmit the disease to human volunteers. *Yale J. Biol. Med.* 38:1-10, 1965.
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7. Evans A S. Infectious mononucleosis in University of Wisconsin students: report of a five year investigation. *Amer. J. Hyg.* 71:342-62, 1960. (Cited 95 times since 1960.)
8. Hoagland R J. The transmission of infectious mononucleosis. *Amer. J. Med. Sci.* 229:262-72, 1955. (Cited 80 times since 1955.)
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10. Evans A S & Niederman J C. The Epstein-Barr virus. (Evans A S. ed.) *Viral infections of humans: epidemiology and control.* New York: Plenum Press, 1982. p. 253-81.