CC/NUMBER 6
FEBRUARY 6, 1984

This Week's Citation Classic™

Vyas G N & Shulman N R. Hemagglutination assay for antigen and antibody associated with viral hepatitis. *Science* **170**:332-3, 1970.
[Dept. Clin. Pathol. and Lab. Med., Univ. California Sch. Med., San Francisco, CA and Clin. Hematol. Branch, Natl. Inst. Arthritis and Metabolic Diseases, Bethesda, MD]

Hemagglutination assays are described for measuring hepatitis-associated Australia antigen and antibody. Red cells coated with isolated antigen, with chromic chloride as a coupling agent, are used for detection of antibodies. Detection of the antigen in serum depends on inhibition of hemag-glutination The test has the sensitivity and rapidity of the best tests available, is simpler to perform, and lends itself to large-scale screening. [The *SCI*® indicates that this paper has been cited in over 535 publications since 1970]

Girish N. Vyas
Department of Laboratory Medicine
School of Medicine
University of California
San Francisco, CA 94143

December 27, 1983

"The single most important problem in blood transfusion and worldwide public health is the infection with hepatitis B virus (HBV), which causes chronic liver disease and hepatocellular carcinoma (the commonest cancer in Africa and Asia). The discovery of the Australia antigen[1,2] (now termed hepatitis B surface antigen, HBsAg), and the observation of its association with posttransfusion hepatitis, stimulated our studies of the immunochemical properties of HBsAg. Several immunoassays could be employed for the more sensitive and rapid detection of HBsAg and anti-HBs than the gel diffusion analysis then in vogue. **I** proposed a **simple detection of HBsAg** and **anti-HBs by using a passive hemagglutination** assay. **I** discussed **the experiments with George Brecher (then chairman of my department), who suggested that I collaborate with N. Raphael Shulman because a complement fixation assay for the HBsAg and anti-HBs was established in his laboratory at the National Institutes of Health (NIH).[3] The experimental work was** accomplished in six days **at the NIH. The final draft of the manuscript was written in three days. On the tenth day I was back in San Francisco and gave the manuscript to Brecher for his review and comments. Besides rewriting certain paragraphs to make the text more readable, he removed his name as coauthor and forwarded the manuscript to** *Science.* **In astonishment I asked him why he removed his name. His answer was, 'I have graduated from this game and all I did was my job as chairman of your department.' This response typified his standards. About two weeks later, I received an acceptance from** *Science.* **For me this was an unprecedented 'bio-quickie,' an exciting success and the subject of my first US patent (#3,887,697) assigned by us to the government of the US.**

"The assay has enabled others to carry out worldwide seroepidemiologic surveys of viral hepatitis and permitted us to decipher the immunochemical structure of HBsAg. In developing the strategies for the use of the newly licensed HBsAg vaccine against hepatitis B,[4] the late Wolf Szmuness often recalled the usefulness of the hemagglutination test. Through the years since the hemagglutination assay took my own research endeavors beyond its practical usefulness in epidemiologic research, Saul Krugman of New York University has inspired us to study the natural history of the infection and immunity to HBV using new laboratory technology.

"The concept of developing a synthetic peptide vaccine **against the hepatitis B infection followed the application of hemagglutination assay to study immune response to HBsAg in man. It took more than ten years of work by Rao, Peterson, Milich, Bhatnagar, Blum, and myself to bring to fruition the first step of this goal.[5] Once again, in 1979, the critical experiment of a synthetic peptide inhibiting human anti-HBs was performed by me using the hemagglutination assay. The fact that the immune response to a synthetic peptide analogue can mimic the natural immune response to HBsAg in humans has recently culminated in my second US patent.[6] Thus, immunochemistry of HBsAg has** been **an exciting and productive pursuit for several investigators."**

1. **Blumberg B S, Alter H J & Visnich** S. A "new" antigen in leukemia sera. *J. Amer. Med. Assn.* **191**:541-6, 1965.
2. **Blumberg B S.** Citation Classic. Commentary[1] on *J. Amer. Med. Assn.* **191**:541-6, 1965. *Current Contents/Life Sciences* **22**(51):14, 17 December 1979.
3. **Shulman N R & Barker L F.** Virus-like antigen, antibody, and antigen-antibody complexes in hepatitis measured by complement fixation. *Science* **165**:304-6, 1969. (Cited 360 times.)
4. **Szmuness W, Stevens C E, Harley E J. Zang E A, Oleszko W R, William D C, Sadovsky R, & Morrison J M & Kellner A.** Hepatitis B vaccine: demonstration of efficacy in a controlled clinical trial in a high-risk population in the United States. *N. Engl. J. Med.* **303**:833-41, 1980.
5. **Vyas G N.** Molecular immunology of the hepatitis B surface antigen. (Maupas P & Guesry P. eds.) *Hepatitis B vaccine: proceedings of the International Symposium on Hepatitis B Vaccine held in Paris. 8-9 December 1980.* Amsterdam: Elsevier/North-Holland Biomedical Press. 1981. p. 227-37.
6. ....*Synthetic peptide vaccine epitomes of hepatitis B surface antigen.* US Patent 4,415.491. 15 November 1983.