**Haematology and Blood Transfusion** Vol. 28
Modern Trends in Human Leukemia V
Edited by Neth, Gallo, Greaves, Moore, Winkler


# Induction of Latent Epstein-Barr Virus Information by a Serum Factor

G. Bauer and H. zur Hausen

Epstein-Barr virus (EBV) is known as the causative agent of infectious mononucleosis and is possibly associated with the development of Burkitt's lymphoma and nasopharyngeal carcinoma. Latently infected lymphoblastoid cell lines can be obtained from patients with EBV-associated diseases, seropositive donors, and by in vitro immortalization of cord blood lymphocytes with certain strains of EBV. Cell lines containing persisting EBV DNA are commonly used to study the mechanisms that regulate latency of EBV.

The expression of viral antigens in EBV genome positive lymphoblastoid cell lines is controlled by so far undefined viral or cellular regulatory mechanisms. In nonproducer lines, only Epstein-Barr virus nuclear antigen (EBNA) is expressed in virtually all cells within the population, whereas the synthesis of viral early antigens (EA) and viral capsid antigens (VCA) is blocked. The latent information may be induced to give rise to the synthesis of viral antigens by various treatments, such as culturing in arginine-deficient medium, addition of inhibitors of protein synthesis, halogenated pyrimidine analogues, antibody to human IgM, tumor-promoting phorbolesters (TPA), *n*-butyric acid, and in some lines with 5-azacytidine and intercalating agents.

Here we summarize the characterization of a humoral protein that can induce the synthesis of viral antigens and that cooperates with chemical inducers in the induction process.

## A. Inducing and Cooperative Serum Factor

### I. Effects

Serum factor induces the synthesis of viral antigens in latently infected cells. The kinetics of induction resembles those obtained by chemical inducers.

Serum factor cooperates with chemically different inducers such as TPA, *n*-butyric acid, anti-IgM, and IUdR in the induction process. Cooperation is characterized by a total induction that is much higher than the sum of inductions reached by individual inducers, and by a substantial shift of the dose-response relationship between chemical inducer and induction into a more sensitive range.

### II. Nature of the Inducing Serum Factor

Inducing serum factor has been purified from calf serum (300,000-fold enrichment). It is a relatively heat-stable protein of about 500,000 daltons. Nanograms per millimeter of the purified material are sufficient for measurable effects. The factor is composed of subunits.

### III. Activation of the Serum Factor

Ninety-five percent of serum factor is present in inactive form. Activation in vitro is carried out by treatment with alkali and acid. The activation reaction has been classified as a conformational change within the molecule. The mechanism of activation of the molecule in vivo is presently only known to exist but is not characterized.

### IV. Occurrence

Inducing serum factor has been demonstrated in the sera of all vertebrates.

### V. Specificity for Defined Cell Lines

Only several Burkitt's lymphoma cell lines and marmoset lines can be induced by the factor. Lymphoblastoid cell lines from seropositive donors or lines established by in vitro immortalization of human cord blood lymphocytes fail to show any response to the factor. The factor therefore seems to recognize cellular markers associated with the state of differentiation.

### VI. Inhibition

The action of serum factor can be inhibited by retinoic acid, in analogy to the induction by tumor promoters and by other drugs.

## B. Conclusions

The factor described here may play a central role in the regulation of EBV gene activity by virtue of both its inducing capacity and its dramatic enhancement of the action of inducing drugs. Thus it may have a central role in processes that overcome latency of EBV.

The widespread occurrence and the conserved nature of the molecule point to a more general physiological function. EBV induction may be an epiphenomenon of this hypothetical process. The existence of inactive and active forms of the factor indicates the possibility of modulation of total factor activity by an activating mechanism. The interaction of factor with chemically different inducers points to the action of the molecule at a central step in the molecular events during induction. Inhibition by retinoic acid points to the utilization of at least one common pathway in the course of induction of viral information and tumor promotion.

## References


Bauer G, Höfler P, Zur Hausen H (1982) Epstein-Barr virus induction by a serum factor. I. Induction and cooperation with additional inducers. Virology 121:184–194

Bauer G, Höfler P, Simon M (1982) Epstein-Barr virus induction by a serum factor. II. Purification of a high-molecular weight protein that is responsible for induction. J Biol Chem 257:11405–11410

Bauer G, Höfler P, Simon M (1982) Epstein-Barr virus induction by a serum factor. III. Characterization of the purified molecule and the mechanism of its activation. J Biol Chem 257:11411–11415

Wittmann P, Höfler P, Bauer G (1982) Epstein-Barr virus induction by a serum factor. IV. Ubiquitous occurrence of the factor and its specificity for defined lymphoblastoid cell lines. Int J Cancer 30:503–510