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Suppression of Human Papillomavirus Type 16 E7 Expression in Human Keratinocytes by Alpha Interferons. J.D. Gangemi, M.A. Khan, and L. Pirisi, Departments of Microbiology and Pathology, USC School of Medicine, Columbia, SC, 29208, USA.

The precise mechanism by which interferons can control infection by human papillomaviruses (HPV) and inhibit the proliferation of HPV-transformed cells is not known. In this study normal human keratinocytes (HKc) and HKc immortalized with human papillomavirus type 16 DNA (HKc/HPV16) were used to investigate the effects of alpha interferon (IFN-alpha) on HPV16 gene expression, and on the growth of HPV16-immortalized human epithelial cells. Normal HKc and HKc/HPV16 were treated with several recombinant human IFN-alpha subtypes (IFN-alpha B, D and αB/D hybrid). These IFN-alpha subtypes inhibited proliferation of both normal HKc and HKc/HPV16 in a dose-dependent fashion; however, while 1,000 to 10,000 units/ml of IFN-alpha were required to inhibit growth of normal HKc, HKc/HPV16 were substantially growth inhibited by only 100 units/ml. In addition, 100 units/ml of IFN-alpha B/D inhibited transformation of normal HKc by HPV16 DNA. Northern blot analysis showed no effect of IFN-alpha on the mRNA levels of the HPV16 E6 and E7 open reading frames. However, immunofluorescence studies of the HPV16 E6 and E7 proteins with anti-E6 and anti-E7 monoclonal antibodies showed significant inhibition of E7 protein expression in cells treated with IFN-alpha, while E6 protein expression was not altered. These results suggest that IFN-alpha may inhibit HPV16-mediated transformation of HKc, and proliferation of HKc/HPV16 through an inhibition of HPV16 E7 protein expression.

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Anticarbohydrate monoclonal antibodies inhibit flavi- and bunyaviruses: molecular aspects. H. A. Blough, D. Kefauver, D. Hack, T. P. Monath (National Naval Medical Center, Bethesda, MD. 20889 & U. S. Army Medical Research Institute of Infectious Diseases, Frederick, MD. 21702). H. Clausen, J-S. Hansen (Department of Infectious Diseases, Hvidovre Hospital, Denmark).

These studies were done to test the hypothesis that specific linkages and/or sequences of carbohydrates play a role in arboviral neutralization. Monoclonal Antibodies (MAbs), either dialyzed hybridoma supernates or chromatographically purified, were interacted with ca. 50 pfu of sandfly fever (SFS), yellow fever (YF 17-D), Rift valley fever (RF), and Japanese encephalitis (JEV) viruses. Seed virus was propagated in Vero or Aedes albopictus (C6-36) cells. MK-2, Vero & C6-36 cells were used for challenge. Virus was harvested at various times & titrated on Vero cells. Two Mabs directed against alpha-Tn (Tn=GalNAc-Ser/Thr) & alpha-sialosyl-Tn (NeuNAc-GalNAc-Ser/Thr) at concentrations of 0.03 ug/pfu completely neutralized SFS & YF from MK-2 cells; YF propagated in C6-36 cells was not neutralized. Pre-treatment of virus particles with neuraminidase and B-galactosaminidase as well as the hapten, GalNac-Ser or -Thr, completely abolished the activity of the Mabs; cell controls, treated in a similar fashion and challenged with SFS, were unaffacted. A comparison of uninfected & SFS-infected MK-2 cells, interacted with anti-CHO MAb and biotinylated anti-mouse MAbs, failed to reveal these carbohydrate epitopes on uninfected cells. Computer generated molecular models were used to confirm the energy constraints of these interactions. These studies confirm that carbohydrate neoantigens are incorporated into virions (possibly as *O*-linked glycans) and can be used as potential targets for broad spectrum vaccines or immunotherapy.