

YING YANG Effect of IL-10 in the incidence of Cervical Cancer

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Abstract

The cytokine family of protein has been shown to have a vital role in the control of normal cellular differentiation, mitosis and motility. These are important immunological mediators of cell mediated immunity against tumor. The family of cytokines includes variety of Interleukins (IL-1, IL-12, IL-10, IL-3 etc.), TNFs, IFNs, M-CSF, GM-CSF etc., each one having their specific source and target. The role of cytokine on the tumor cell is necessary to have a differentiation therapy which favor a unique and potentially effective, less toxic treatment paradigm for cancer. The most important of all cytokines is Interleukine-10 which has a dual biological role anti-inflammatory and anti-angiogenic effect. These IL 10 molecules are produced by T cells, Monocytes, Macrophages, B cells, Natural killer cells, Eosinophils, Mast cells and Keratinocytes. TH1 cells are their main target. In cervical cancer patients the incidence of IL-10 is found to increase with the severity of the disease.

Key words: IL-10, IFN- , TNF-

1999). High-grade SIL are associated with less concentration of IL-12. Thus, an immuno regulating cytokine network is developed. Immuno stimulating or Th1 type cytokine level decreases in cervical cancer patients with the substantial increase in the immuno suppressive or Th2 type cytokine level (Aagje *et al.*, 2007). A shift from Th1 type to a Th2 type cytokine response was observed when healthy control or LG-SIL was compared to HG-SIL or cervical cancer. IL-2 decreases with increasing grades of CIN. IFN- decreases from CIN III to cancer (Aagje *et al.*, 2007). IL-12 reaches maximum in CIN II and decreases as the diseases progresses from CIN III to carcinoma (Aagje *et al.*, 2007). IL-10

Introduction

Globally cervical cancer is the second most common cause of cancer death in women (Tindle and Frazer, 1995). The chronic infection of keratinocytes of the uterine cervix by the Human Papilloma Virus (HPV) is associated with the development of cervical cancer (Giannini *et al.*, 1998). HPV codes for six early proteins (E1, E2, E3, E4, E5, E6, E7) and two late proteins (L1 and L2). E6 and E7 are oncoproteins (Maria *et al.*, 2002). HPV infection alone is not sufficient for cancer development because majority of women with HPV infection do not develop cervical cancer (Tindle and Frazer, 1995 & Ho *et al.*, 1998). The role of immune system in viral clearance is not clear, but there is clear evidence that cellular mediated immune response is important in the control of HPV infection (Stephen, 1988). The occurrence and progression of tumor is due to the loss of tumor antigenicity and suppression of immune system. Cytokines contribute to this phenomenon. Tumor associated suppression of immune system may be mediated by IL-10 (Germain *et al.*, 1998). The role of IL-10 in

cervical cancer is not clear. Thus, it is necessary to analyze the biological role of IL-10.

Cytokine Profile In Cervical Cancer Patients

Progression of cervical cancer is associated with preferential constraint on the development of type 1 cellular mediated response. The initiation and maintenance of CMI is associated with CD4+ T helper cells producing either Th1 Cytokine (IL-12, IFN , TNF) or Th2 Cytokine (IL-4, IL-6, IL-10). Peripheral blood mononuclear cells (PBMC) from patients with both Squamous Intraepithelial Lesions (SIL) and cervical cancer produced decreased amount of IL-12 and IFN- and increased level of IL-4 and IL-10 (Giannini *et al.*, 1998 & Mota *et al.*, 1999). Basal level of IL-10 is augmented in PBMC of patients with SIL (Mota *et al.*,

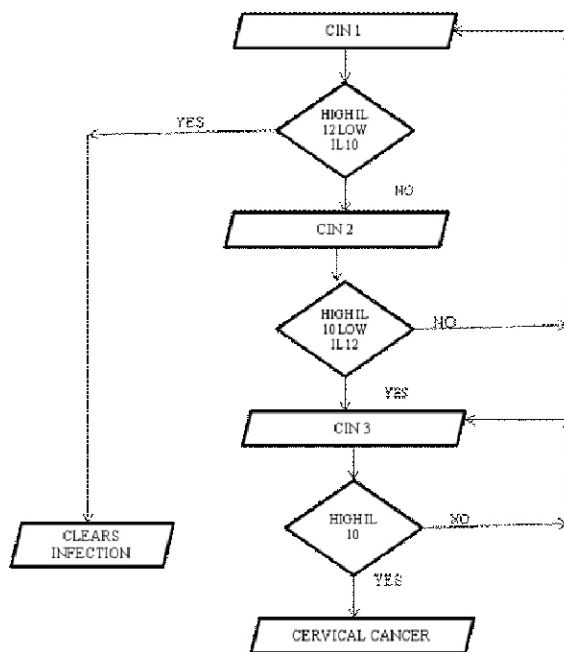


Fig. 1 Cytokine profile in Cervical Cancer development : High incidence of IL- 12 helps clear the infection with HPV. IL- 10 incidence was found to be high as the cancer progresses.

was maximum in CIN III and invasive carcinoma. This suggests a viral activation of the systemic cytokine network in HPV positive women. Development of SIL or cancer is preferentially associated with type II or immuno suppressive cytokine in particular IL-10 (Giannini *et al.*, 1998).

IL-10 and its Receptor

IL-10 is described by Moosaman in 1989. The gene, which codes for IL-10 is located on chromosome 1. IL-10 is made up of 160 amino acid residues. It is a homodimer with non-covalent inter wining of two identical oppositely oriented polypeptides (Syto *et al.*, 1998 & Donnelly *et al.*, 1999). IL-10 is produced by T cells, Monocytes, Macrophages, B cells, Natural killer cells, Eosinophils, Mast cells and Keratinocytes (Germain *et al.*, 1998). IL-10 binds to a functional tetrameric complex of two receptors consisting of two α or R1 chain which binds IL-10 (Liu *et al.*, 1994 & Konten *et al.*, 1997) and of two CRF 2-4 chains (β or R2) which initiate the IL-10 induced signal transduction events (Syto *et al.*, 1998 & Donnelly *et al.*, 1999).

Functions of IL-10

IL-10 is an immuno modulatory cytokine with both suppressing and enhancing properties on different types of the immune cells (Moore *et al.*, 1993). IL-10 functionally inhibits Antigen Presenting Cells. IL-10 blocks the cytokine synthesis of Th1 type T cells, activated monocytes and Natural killer cells. IL-10 stimulates and/or enhances the proliferation of B cells, thymocytes and mast cells and decreases the cytotoxic T lymphocytes generations (Moore *et al.*, 1993 & Yang *et al.*, 1995). It has no effect on the expression of CD80. It down regulates the expression of CD86 on monocytes and dendritic cells (Buelens *et al.*, 1995 & Creery *et al.*, 1996).

IL-10 down regulates tumor specific immune response by one of the following mechanisms:

- i. Directly suppressing IFN- and IL-12 production thereby preventing the activation of CTLs and Natural Killer Cells (Mosmann and Coffman, 1987).

- ii. Reducing MHC expression on the surface of tumor cells, thus preventing the optimal expression of binary complexes formed by tumor antigens in association with MHC molecules on the surface of such cells (Moore *et al.*, 1993).
- iii. Inhibiting tumor antigen presentation by professional APCs (Beissert *et al.*, 1995).

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IL-10 has multiple biological roles based on the fact that their receptors are expressed by diverse cell population (Vicki, 2005). Langsenlehner's study suggests that decreased IL-10 levels are associated with the risk of cervical cancer (Vicki, 2005). But in all cases of advanced cancer IL-10 level was found to be apparently high (Mota *et al.*, 1999). The dual biological function of IL-10 as anti-inflammatory (potentially cancer promoting) and antiangiogenic (cancer inhibiting agent) reflects the apparently conflicting data. HPV stimulates a chronic cytokine inflammation which sets up a

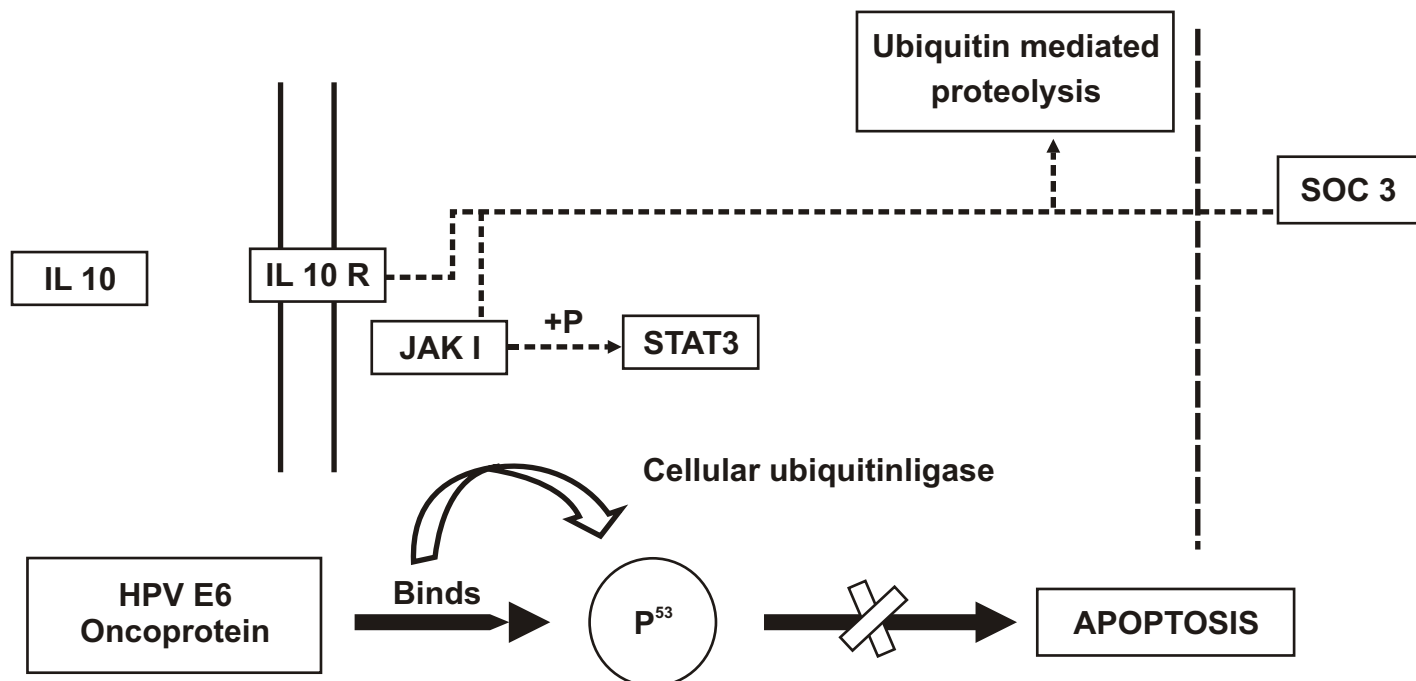


Fig II. Anti Inflammatory effect of IL-10: IL-10 competes with IFN- and activates Ubiquitin mediated proteolytic pathway. E6 protein of HPV binds with p53 and is cleaved through Ubiquitin mediated proteolytic pathway.

cascade of cytokine that pushes the chronic infection towards cancer. Individual cytokine profile would be the missing link between infection and cancer. Cervical cancer patients carry a polymorphism for enhanced IL-10 production (Vicki, 2005). It depends on the biology of each cancer and its micro environment if IL-10's anti-inflammatory or anti angiogenic effect is stronger. IL-10 generates its signal through the JAK 1 STAT 3 pathway and activates the Soc-3 (Suppressor of Cytokine signaling 3) gene of which the expression results in the inhibition of JAK/STAT signaling of Th1 cytokines (Donnelly *et al.*, 1999 & Riley *et al.*, 1999). This suppression of Th1 response is utilized by HPV for its progression.

Anti-Inflammatory effect of IL-10

Poor immunogenicity of tumor cells may be the possible reason for the failure of immune system to control tumor growth. Augmented IL-10 production is responsible for the poor immunogenicity of tumor cells (Mario *et al.* 1997). IL-10 down modulates the presentation of tumor antigens to CD8+ CTL's (Matsula *et al.*, 1994 & Beissert *et al.*, 1995). In cervical cancer patients E7 protein is considered to be tumor specific antigen. E7 specific T cell help in CD8+ CTL-mediated tumor regression. IL-10 down regulates E7-

antigen presentation to CD8+ T cells thus favoring the progression of the disease.

Another possible role of IL-10 in promoting cancer is in the inhibition of Th1 cytokine TNF. IL-10 inhibits TNF- production in macrophages in the presence of STAT 3 JAK1 and one IL-10 receptor (Joan *et al.*, 1999). TNF- is necessary for the activation of Langerhans cells and is capable of inducing apoptosis *in virus* infected cells (Wong *et al.*, 1992 & Crish *et al.*, 2000). However, it does not induce apoptosis in all cells (Mota *et al.*, 1999). Most cells are protected from apoptosis caused by TNF- by a separate proliferative pathway involving the activation of NF- B. HPV infected cells contain E6 protein which also activates the NF- κ B production thus, preventing the cells from the apoptosis caused by TNF- (Carter *et al.*, 1999, Marconi *et al.*, 1999, Carter and Hunninghake, 2000, Gentry *et al.*, 2000 & Manna *et al.*, 2000). Thus, it is hypothesized that IL-10 in the NF- B activated cells reduces the TNF- production. However the exact link between E6 protein and IL-10 is not known.

In a similar fashion IL-10 inhibits IFN- , another tumor suppressing cytokine. Oncoprotein E6 has a possible inhibiting role

in IFN- production. Inhibition of SOC-3 signal by IL-10 results in the decrease is IFN- . Efficient production of IFNs in a virally infected cell is an essential aspect of host defense. E6 mediated degradation of p53 also contributes to decreased IFN production. E7 inhibits the anti proliferative and anti inhibiting function of IFN- . Thus, IL-10 was found to function as anti inflammatory agent in the presence of HPV oncoprotein (Woodsworth *et al.*, 1992). IL-10 and HPV functions parallelly or serially in suppressing the immune system.

Anti-Angiogenic Effect Of IL-10

IL-10 in combination with IL-2, a Th1 cytokine was able to consistently increase the cytotoxicity. Administration of IL-10 in combination with IL-2 after antigen stimulation consistently increases the intracellular expression of Th-1 cytokine (Alessandro *et al.*, 2000). In an investigation of optimal culture conditions for potentiation of the cytotoxic activity of HPV-E7 specific CTL for adoptive transfusions to cervical cancer patients, it is observed that IL-10 in combination with IL-2 consistently increases the cytotoxic potential of the CTL population. The ability of IL-10 to enhance proliferation, expression of immunologically important surface molecules and Th1 cytokine production by CD8+ CTL suggests that its use in combination with IL-2 may be a valuable adjunct in the cancer therapy. But unfortunately there is no *in vivo* proof for this as there is always a decrease in Th1 cytokine level when IL-10 level is high (Alessandro *et al.*, 2000).

Conclusion

IL-10, E6 and E7 oncoproteins have the same effect on the TNF- and IFN- and CD8+ T cells. But the possible link between these three proteins is not known. If the link between these proteins is established then the exact role of IL-10 can be formulated.

However, the role of IL-10 in promoting cancer of the uterine cervix may depend upon the microenvironment around the cervix. When E6 oncoprotein activity is high IL-10 is found to promote the tumor growth. On the other hand when IL-2 level is high it suppresses tumor. It is necessary to have a

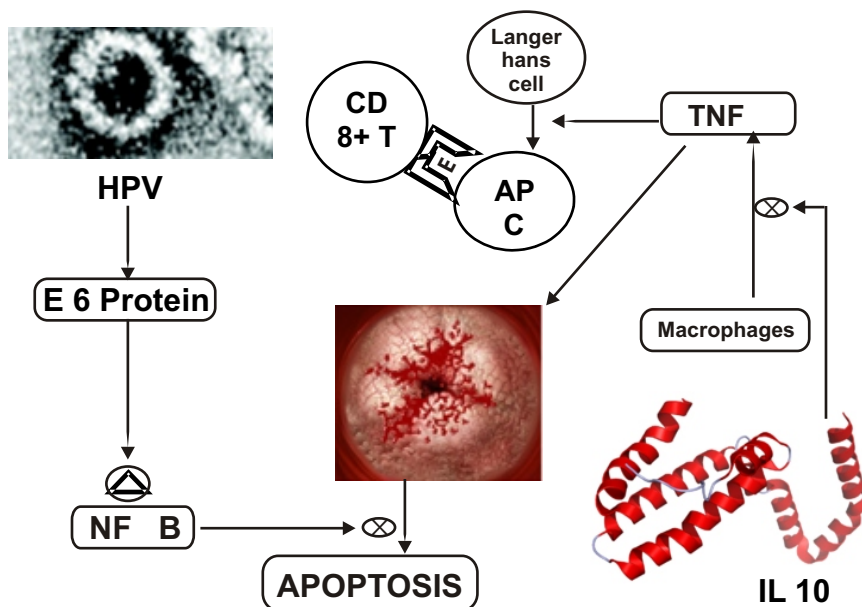


Fig III. Individual effects of IL- 10 and E6 gene in the decreased production of TNF : TNF is necessary for the activation of Langerhans cells to present antigen to the T cells. IL- 10 inhibits TNF production directly and hence inhibits antigen presentation indirectly. E6 on the other hand activates NF B pathway, which inhibits apoptosis of the cancer cells induced by TNF .

clear knowledge about the multiple roles of each cytokine so that they can be used in the treatment of cancer. When applied therapeutically cytokines reduce all adverse reactions produced by radiation therapy and chemotherapy.

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