Abstract
The possible correlation of Herpes Simplex virus type 1 (HSV 1), Herpes Simplex type 2 (HSV 2), and Epstein Barr virus (EBV) with skin cancer was studied by using Enzyme Linked Immunosorbent Assay (ELISA) technique based on virus specific antibodies produced by immune responses. Serums of 15 clinically diagnosed histopathologically confirmed skin cancer patients were studied for the presence of IgM antibodies against these viruses with reference to one hundred control cases. There were no correlations between the viruses HSV 1, HSV 2, and EBV with the skin cancer of the type squamous cell carcinoma (SCC), basal cell carcinoma (BCC) and melanoma most commonly found skin cancer cases in Nepal.

Key words: EBV, HSV 1, HSV 2

Introduction
Skin cancer is the most common form of cancer accounting for more than 400,000 new lesions each year in the world (Rosenbaum 1983). Skin cancer cases are very rare in tropical countries like Nepal because of the presence of higher levels of melanin in the skin. Ninety-eight percent of melanoma occurs in whites. Within the white population there is a higher incidence in light skinned individuals (Farmer 1990). The most common skin cancer reported in Nepal is squamous cell carcinoma (SCC), basal cell carcinoma (BCC), and melanoma.

The pathogenic mechanism of skin cancer has been traced to defects in single gene. Moreover, the role of environmental factors, such as sunlight, certain chemicals, food and hygienic conditions are important in the development of skin cancer (Murphy 1991). Apart from different factors viruses such as Epstein Barr virus (EBV), Human T leucocyte virus type 1 (HTLV-1), Human T leucocyte virus type 2 (HTLV-2), Varicella Zooster virus (VZV), and Human Herpes virus (HHV-6) are most often identified in relation to skin cancer (Wagner et al. 1998). Despite high prevalence of HSV 1 and HSV 2, there have been few reports of their relation to skin cancer (Wagner et al. 1998). Herpesviridae is a DNA family virus of which HSV 1, HSV 2, and EBV are important member. EBV is a common human pathogen affecting 80% of adults in the United States (Baltz 1992). HSV 1, HSV 2 and EBV are of special interest because of their multiplication character in the nucleus of the host, their high prevalence and latency in nerve cells.

Methods
Sera from one hundred fifteen patients in which seven patients having SCC, six patients having BCC, two patients having melanoma and hundred healthy patients for control were collected from different hospitals of Nepal between January 2000 and December 2000. The clinically diagnosed skin cancer case was confirmed by histology using Haematoxyline and Eosine statin.

ELISA kits (Trinity biotech, USA Jamestown, NY) were applied for the detection of IgM antibodies from the serum. In these kits viral capsid antigen of EBV, Human T leucocyte virus type 1 (HTLV-1), Human T leucocyte virus type 2 (HTLV-2), Varicella Zooster virus (VZV), and Human Herpes virus (HHV-6) were coated on polystyrene surface. Antigens bound to the solid phase were incubated with diluted serum. Excess antibody was removed by washing. This was followed by incubation with goat antihuman IgM conjugated with horseradish peroxidase. Excess conjugate was removed by washing followed by the addition of chromogenic substrate tetramethyl benzidine. The reaction was stopped by the addition of 1N H2SO4. The color produced in the microtitre plate was read in ELISA microwell plate reader and the result was processed as suggested by protocol.

Results and Discussion
The chance of getting positivity with IgG titre is high because herpesviridae family is responsible for many types of disease and IgG antibody may persist in the blood for long time. First antibody that appears in blood against any infection is IgM antibody; the levels of which rise early, peak 3-4 weeks after infection and then decline to non-detectable levels. Serum antibody
Table 1. Results of ELISA test on control and skin cancer cases

<table>
<thead>
<tr>
<th>Viruses</th>
<th>Control (%)</th>
<th>Skin cancer (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>HSV 1 +ve</td>
<td>3</td>
<td>0</td>
</tr>
<tr>
<td>HSV 1 -ve</td>
<td>97</td>
<td>15</td>
</tr>
<tr>
<td>HSV 2 +ve</td>
<td>5</td>
<td>0</td>
</tr>
<tr>
<td>HSV 2 -ve</td>
<td>95</td>
<td>15</td>
</tr>
<tr>
<td>EBV +ve</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>EBV -ve</td>
<td>100</td>
<td>15</td>
</tr>
</tbody>
</table>

against any antigen by any serological test provides no information regarding time of infection with the exception of IgM. Its presence in serum indicates recent infection. So, IgM antibodies against these viruses were subject for analysis.

Hundred control and fifteen skin cancer cases were tested for presence of IgM antibodies against HSV 1, HSV 2 and EBV. In case of HSV 1, 3% and 0% of the patients were positive in control and skin cancer cases, where as in case of HSV 2, 5% and 0% of the samples were positive for control and skin cancer cases, respectively (Table 1). This suggests that skin cancer and HSV 1 or HSV 2 is not correlated. Similarly, in case of EBV no positives samples were recorded in both the control and skin cancer cases. This again indicates that there exists no correlation of EBV and skin cancer.

Result presented in this paper indicates that HSV 1, HSV 2 and EBV do not play role in the pathogenesis of SCC, BCC, and melanoma unlike cutaneous lymphoma.

References