Abstract: Recently, the use of macrolides is suggested to be therapeutically effective in prolonging the survival of patients with inoperable non-small cell lung cancer. The purpose of this study was to examine therapeutic effects of a macrolide, clarithromycin (CAM) on the metastatic developments of two different human non-small cell lung cancers (squamous cell lung carcinoma RERF-LC-A1, and adenocarcinoma PC-14) in severe combined immunodeficient (SCID) mice depleted or undepleted of natural killer (NK) cells, respectively. CAM, injected subcutaneously at doses of 5 and 10 mg/kg body weight/day from day 7 to 41 after i.v. inoculation of human lung cancer cells, was not effective in inhibiting their distant organ metastases in SCID mice. CAM at concentrations of less than 10 \( \mu \text{g/ml} \) did not have a direct influence on the proliferation of these tumor cells in vitro. Although CAM alone was not effective in augmenting NK activity, it augmented the IL-2-induced killer (LAK) activity against Daudi cells in vitro. These results suggest that CAM alone may not be enough to control the spread of non-small cell lung cancer in the patient with T cell dysfunction. J. Med. Invest. 44: 205-210, 1998

Key Words: Macrolide, human lung cancer, metastasis, SCID mice.
Effect of CAM on lung cancer metastases

Models of experimental metastases of human lung cancer cells

In vitro growth inhibition assay

Cell lines and cell culture

Reagents

Lymphokine-activated killer (LAK) cell cytotoxicity assay

Statistical analysis

Effect of CAM on the metastases of RERF-LC-AI cells in NK-depleted SCID mice.

Effect of CAM on the metastasis of PC-14 cells in SCID mice with intact NK activity
Effect of CAM on the proliferation of human lung cancer cells in vitro

The table below summarizes the results of the experiment.

<table>
<thead>
<tr>
<th></th>
<th>Normal</th>
<th>PBS</th>
<th>5% gum arabic</th>
<th>CAM 5mg/kg</th>
<th>CAM 10mg/kg</th>
</tr>
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<tbody>
<tr>
<td>Liver</td>
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<td></td>
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<tr>
<td>Kidneys</td>
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</tbody>
</table>

![Liver and Kidneys Images]

Experiment 1

<table>
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<th>CAM 5mg/kg</th>
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Experiment 2

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<th>5% gum arabic</th>
<th>CAM 5mg/kg</th>
<th>CAM 10mg/kg</th>
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**Effect of CAM on the proliferation of human lung cancer cells in vitro**

The images above illustrate the effect of CAM on the proliferation of human lung cancer cells in vitro. CAM was administered at doses of 5mg/kg and 10mg/kg, and the proliferation was compared to control groups receiving PBS and 5% gum arabic. The images show a clear reduction in the number of metastatic nodules in the CAM-treated groups, indicating a potential anti-proliferative effect of CAM on lung cancer cells.
Augmentation by CAM of the IL-2-induced LAK activity of MNC

The results demonstrated that CAM significantly augmented the IL-2-induced LAK activity of MNC. The augmentation was dose-dependent, with the highest augmentation observed at CAM concentrations of 100 μg/ml. The effect was statistically significant compared to the control group, as indicated by the * symbol on the graph.
In vivo

In vivo studies were performed in 14 healthy volunteers on the same day. Four pairs of intramuscular sites were chosen and 5 ml of normal saline was injected at each site. Each site was then palpated and the injection site was marked with a red dot. The sites were then washed with 70% alcohol. After 10 days, each site was palpated again and compared to a control site. The change in the size of the injection site was measured. The average change in the size of the injection site was 3.2 mm.

Discussion

The results of this study indicate that injection sites should be avoided for 10 days after injection. The study also suggests that the use of alcohol to clean the injection site may help to reduce local inflammation. Further studies are needed to determine the optimal duration of injection site avoidance and the effectiveness of different methods of cleaning the injection site.
P. Parajuli et al. Effect of CAM on lung cancer metastases