



Research article

## In vitro and in vivo anticancer activity of hydroalcoholic extract of *Ipomoea carnea* leaf against Ehrlich Ascites Carcinoma cell lines

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### Abstract



The plant *Ipomoea carnea* Jacq (Family: convolvulaceae) have been traditionally used for many ailments including cancer, though information from organized search of published literature does not provide sufficient evidence for its antitumor activities, so we made an attempt to use Hydroalcoholic Extract of *Ipomoea carnea* (HAEIC) for studying anticancer activity by using both *in-vitro* and *in-vivo* method. The *In-vitro* anticancer activity of HAEIC was evaluated by the MTT assay method using Ehrlich Ascites Carcinoma (EAC) cell lines. Later the extract subjected to *in-vivo* anticancer therapy using EAC tumor model. The activity was assessed increase in life span, average increase in body weight, change in food intake, tumor weight, tumor volume, viable cell count, non viable cell count, packed cell volume, hematological and biochemical parameters. The potency of the extract was compared with standard 5-flurouracil (20mg/kg i.p). *In-vitro* anticancer activity of HAEIC exhibit significant cytotoxicity against EAC cell lines at different concentration. Oral administration of HAEIC at the dose of 250 and 500mg/kg, significantly ( $p<0.001$ ) increased the survival time, non viable cell count and decrease in body weight, food intake and viable cell count of the tumor bearing mice. After 14 days of inoculation, HAEIC was able to reverse the changes in the hematological parameters, protein and packed cell volume. The results indicate that HAEIC possess significant antitumor activity on dose dependent manner.

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### Key Words

*Ipomoea carnea*, Anticancer activity,  
Hydroalcoholic extract,  
Ehrlich Ascites Carcinoma.

## INTRODUCTION

Natural Products, especially plants, have been used for the treatment of various diseases for thousands of years. Terrestrial plants have been used as medicines in Egypt, China, India and Greece from ancient time and an impressive number of modern drugs have been developed from them. The first written records on the medicinal uses of plants appeared in about 2600 BC<sup>1</sup>

Cancer is a general term applied of series of malignant diseases that may affect different parts of the body. These diseases are characterized by a rapid and uncontrolled formation of abnormal cells; which may mass together to form a growth or tumor or proliferate throughout the body, initiating abnormal growth at other sites.<sup>2</sup> An extremely promising strategy for cancer Prevention today is chemoprevention, which is defined as the use of synthetic or natural agents (alone or combination) to block the development of cancer in humans. Plants, vegetables and herbs used in the folk and traditional medicine have been accepted currently as one of the main source of cancer chemoprevention drug discovery and development.

The plant *Ipomoea carnea* is a large, diffuse or struggling shrub with milky juice, leaf ovate chordate, entire, acuminate, flower large campanulate, pale rose, pink or light violet in lax, dichotomously branched axillaries and terminal, pedunculate cymes; Fruits glabrous capsule; Seed silky, belonging to family Convolvulaceae<sup>3,4</sup>. The leaves of *Ipomoea carnea* contain 1-3 flavonol glycosides and Ergine (D-Lysergic acidamide)<sup>5</sup>. Chromatographic

separation of the leaf extract resulted in the isolation of swainsonine, 2-epilentiginosine, calystegines B (1), B (2), B (3) and C (1) and N-methyl-trans-4-hydroxy-l-proline and beta sitosterol<sup>6,7</sup>. Plant derived natural products such as flavonoids, terpenoids, and steroids etc have received considerable attention in recent years due to their diverse pharmacological properties including antioxidant and antitumor activity. Antioxidants play an important role in inhibiting and scavenging radicals, thus providing protection to humans against infection and degenerative diseases. Though the plant *Ipomoea carnea jacq* have been extensively used in the folklore medicine, information from organized search of published literature does not provide sufficient evidence for its antitumor activities, so we made an attempt to use Hydroalcoholic Extract of *Ipomoea carnea* (HAEIC) for studying anticancer activity.

## MATERIALS AND METHOD

### Chemicals

5-fluorouracil(Biochem pharmaceuticals Ltd) was purchased from local medical shop; all other chemicals used for the study were of analytical grade.

### Collection of specimen

The species for the proposed study, *Ipomoea carnea jacq* leaves were collected in the month of March 2013 from the village Karatupallayalam of Erode district, TamilNadu, India.

### **Taxonomical identification**

The species was identification and authenticated as *Ipomoea carnea jacq* by Dr.P. Satyanarayana scientist & Head of the office Government of India, Botanical survey of India, Southern Circle, T.N.A.U. Campus, Lawley Road, Coimbatore-641 003.A voucher specimenNo.BSI/SRC/5/23/2011/Tech-1824.

### **Extraction of *Ipomoea carnea* leaf**

The fresh plant materials are washed and were dried in a shade drying and ground to powder. About 250gms of dried powdered *Ipomoea carnea* leaf was taken in Soxhlet apparatus and extract with measured volume of solvent (80:20 ethanol-water) 72 h. The temperature was maintained at 55°C-65°C. Extract was concentrated by distilled and solvent was recovered. The final solution was evaporated to dryness.

### **Animals**

Male Swiss albino mice weighing 25-30g were used in the present study. All rats were kept at room temperature of 22-25°C in the animal house. All the animals were followed the internationally accepted ethical guidelines for the care of laboratory animals. Prior to the experiments, rats were fed with standard food for 1 week

in order to adapt to the laboratory conditions. All animal procedures were performed after approval from the institutional ethics committee. The experimental protocol has been approved by institutional animal ethics committee, The Erode College of Pharmacy & Research Institute, Veppampalayam,Erode.PCOL/03/2013/IAEC/ECP.

### **Acute toxicity study**

Aqueous extracts of *Ipomoea carnea* leaves were studied for acute toxicity at doses of 5mg/kg, 50mg/kg, 300mg/kg, 500mg/kg and 2000mg/kg. As per OECD 420 guideline dose of 2000mg/kg showed the toxic symptoms, so according to OECD guideline 420, it is considered as a LD50 cutoff value. Doses selected for pharmacological studies by fixed dose methods are 250mg/kg and 500mg/kg<sup>8</sup>.

### **Tumor cells**

Ehrlich Ascites Carcinoma (EAC) cell were obtained from Amala cancer research center, Trissur, Kerala, India. The cells were maintained in vivo in Swiss albino mice by intraperitoneal transplantation. EAC cells aspirated from the peritoneal cavity of mice were washed with saline and given intraperitoneally to develop ascetic tumor.

## ANTICANCER ACTIVITY

### *In vitro* anticancer activity<sup>9,10</sup>

MTT assay: MTT (3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyl tetrazolium bromide) assay. Cells were grown in RPMI-1640 medium at 37°C under 5% CO<sub>2</sub> in a humidified incubator for 6-7 h. Cells were harvested, counted ( $3 \times 10^4$  cells/ml) and transferred into a 24 well plate, and incubated for 24 h prior to the addition of test compound. Serial dilutions of test samples were prepared by dissolving compounds in DMSO followed by dilution with RPMI-1640 medium to give final concentration at 100, 250, 500, 1000 µg /ml. Stock solutions of samples were prepared. Sample at 10 µl and cell lines at 90 µl were incubated for 72 h. MTT solution at 5mg/ml was dissolved in 1ml of Phosphate Buffer Solution (PBS), and 10 µl of it was added to each of the 24wells. The wells were wrapped with aluminum foil and incubated at 37°C for 4 h. The solution in each well containing media, unbend MTT and dead cells were removed by suction and 150 µl of DMSO was added to each well. Then the plants were shaken and optical density was recorded using a micro plate reader (spectrophotometer) at 595nm. DMSO as a blank. Controls and samples were assayed and replicated for each concentration and

replicated three times for each cell line. After 24 h incubation of the mononuclear cells with plant extracts, the cytotoxicity on the cancer cell lines was evaluated using MTT assay. The cytotoxicity was obtained by comparing the absorbance between the samples and control. The values were then used to calculate the concentration of plant extracts required to cause a 50% reduction (IC<sub>50</sub>) a growth (cell number) in EAC cell lines.

$$\text{Cell viability (\%)} = \frac{\text{Mean OD}}{\text{Control OD}} \times 100$$

### *In vivo* anticancer activity

Ehrlich's Ascites Carcinoma cells were cultured in the peritoneal cavity of healthy albino mice weighing between 25 to 30 g by injecting a suspension of EAC cells ( $1 \times 10^6$  cells/ml) intraperitoneally. The cells were aspirated aseptically from the peritoneal cavity of the mice on day 15 and washed with normal saline and centrifuged for 15 min at 1,500 rpm in a centrifuge. The pellet was re-suspended with normal saline and the process was repeated three times. Finally, the cells were suspended in a known quantity of normal saline and the cell count was adjusted to ( $2 \times 10^6$  cells/ml). Sample showing more than 90 % viability was used for transplantation. Each animal received 0.5 ml of tumor cell suspension

containing  $2 \times 10^6$  cells intraperitoneally<sup>11</sup>.

### **Experimental design<sup>12, 13, 14</sup>**

Male Swiss albino mice were divided in to 5 groups and 12 animals for each group. The entire groups were injected with EAC cells ( $2 \times 10^6$  cells/mouse) intraperitoneally except for the normal group.

Group I: Normal with sodium Suspension (0.1%)

Group II: Induced EAC cell ( $2 \times 10^6$ ) with sodium Suspension (0.1%)

Group III: Induced EAC cell ( $2 \times 10^6$ ) with 5fluorouracil (20mg/kg i.p)

Group IV: Induced EAC cell ( $2 \times 10^6$ ) with HAEIC (250mg/kg p.o) with sodium suspension (0.1%)

Group V: Induced EAC cell ( $2 \times 10^6$ ) with HAEIC (500mg/kg p.o) with sodium suspension (0.1%)

All groups were treated with respective drugs 24 h after inoculation, once daily for 14 days after the last dose and 24 h fasting, six mice in each group were sacrificed. The antitumor activity of the extract was measured in EAC animals with respected to the following parameters. Tumor volume, tumor weight, PCV, viable cell count,

non viable cell count, percentage of viable cell, percentage of non viable cell, increase in life span (ILS), average increase in body weight, change in food intake.

### **ANTITUMOR PARAMETERS<sup>15,16,17</sup>**

#### **Percentage increase life span**

The effect of HAEIC on tumor growth was monitored by recording the mortality daily for a period of 6 weeks and percentage increase in average life span was calculated.

$\% \text{ ILS} = \{(\text{life span of treated group} / \text{life span of controlled group}) - 1\} \times 100$

#### **Body weight analysis**

Body weights of the experimental mice were recorded both in the treated and control groups at the beginning of the experiment (day 0) and sequentially on every 5<sup>th</sup> day during the treatment period and calculated on 15<sup>th</sup> day.

#### **Changes in food intake**

Feed consumed by 6 animal/cage/week = Total quantity of feed offered during that week (gm) – Feed left over on last day of week (gm).  
Feed consumed by individual animal/week = Feed consumed by 6 animal per cage per week/6. Feed consumed by individual animal/day =

Feed consumed by individual animal per week/7.

### **Determination of tumor volume and weight**

The mice were dissected and the ascitic fluid was collected from the peritoneal cavity. The volume was measured by taking it in a graduated centrifuge tube and weight immediately.

### **Tumor packed cell volume**

The ascitic fluid was collected from the peritoneal cavity. The packed cell volume was measured by taking it in a graduated centrifuge tube and by centrifuging at 1000 rpm for 5 min.

### **Tumor viable cell count**

The ascitic fluid was taken in a WBC pipette and diluted 100 times. Then a drop of the diluted cell suspension was placed on the Neubauer counting chamber. The cells were then stained with trypan blue (0.4% in normal saline) dye. The cells that did not take up the dye were viable and those that took the stain were nonviable. These viable cells were counted in the 64 small squares.

### **Determination of Hematological parameters**

Blood was drawn from each mouse by the retro orbital plexus method and the white blood cells (WBC), red blood cells (RBC), hemoglobin and protein were determined. Serum preparation of the sample blood use to evaluated the biochemical parameters.

### **Statistical analysis**

All values were expressed as mean  $\pm$  SEM. Statistical analysis was performed with one way analysis of variance (ANOVA) followed by Tukey Kramer multiple comparison tests. *P* value  $< 0.001$  was considered as highly significant and  $< 0.05$  were considered significant when compared to control.

## **RESULTS**

### **In vitro anticancer activity**

From MTT assay, after treatment with various concentrations of HAEIC leaves parameters like percentage cell viability, percentage cytotoxicity were compared with untreated (control) cells. Decrease in cell viability and increase in cytotoxicity by HAEIC leaves was observed in EAC cell lines in dose dependent manner, but a significant decrease in cell viability ( $p < 0.001$ ) was observed for 1000 $\mu$ g/ml dose, 500  $\mu$ g/ml, 250  $\mu$ g/ml and 100  $\mu$ g/ml dose of HAEIC leaves produced significant growth inhibition. The growth inhibitory activity of *Ipomoea*

*carnea* leaves was more significant as the concentration of HAEIC leaves increases in case of EAC cell lines (Tab 1).

**In vivo anticancer activity**

Antitumor activity of HAEIC leaves against EAC tumor bearing mice was assessed by the parameters such as tumor volume, tumor weight, viable and non viable cell count, mean survival time and percentage increase in life span, average increase in body weight, change in food intake.

to  $33.83 \pm 0.31$  (% ILS = 76.41%) and  $27.83 \pm 0.30$  (% ILS = 63.57) on administration of HAEIC leaves at the dose of 500mg/kg and 250 mg/kg respectively. Finally, the change in body weight of the animal suggests the tumor growth inhibition property of *Ipomoea carnea* leaves. Food intake also reduced in the tumor control animals. All these results clearly indicate that the HAEIC leaves showed significant antitumor activity in EAC bearing mice (Tab 2).

**Tab1. In vitro cytotoxic activity of HAEIC on EAC cell lines**

Concentration of HAEIC ( $\mu\text{g/ml}$ )	% of cell viability	% of cytotoxicity
Control	100	-----
HAEIC (100)	69.751	30.249
HAEIC (250)	54.398	45.602
HAEIC (500)	30.872	69.128
HAEIC (1000)	19.985	80.015

The tumor volume, tumor weight, and viable cell count were found to be significant ( $p < 0.001$ ) increased and non-viable cell count, increase in body weight and food intake was significantly ( $p < 0.001$ ) low in EAC control animal when compared with normal control animals. Furthermore, the mean survival time was increased

**Hematological parameters**

Hematological parameters of tumor bearing mice on day 15 were found to be significantly different as compared to the extracts of treated group. In tumor bearing mice, it was found that, there was an increase in WBC count

and decrease in Hb count and RBC count. In differential count of WBC, presence of neutrophils and monocytes increased while the lymphocytes count decreased in the EAC control group. Treatment with HAEIC leaves at the dose 250mg/kg and 500mg/kg significantly increased the Hb and RBC count to normal levels. The total WBC count was found to be increased significantly in the EAC control group when compared to normal group. Administration of *Ipomoea carnea* leaves extracts (250mg/kg and 500mg/kg) in EAC bearing mice significantly ( $p < 0.05$  and  $p < 0.01$ ) reduced the WBC count as compared with EAC control (Tab 3).

### Biochemical parameters

Various biochemical parameters like Total Protein, SGPT, SGOT, LPO, ALP, GSH, SOD, CAT value of HAEIC at the dose of 500mg/kg were compared to the standard drug 5- fluorouracil (20mg/kg) (Tab 4).

### DISCUSSION

Cancer is often associated with increased risk of death and the toxic side effects caused by the modern medicine, many cancer patients seek alternative and complementary methods of treatment such as usage of phytomedicine<sup>18</sup>. The result of the present study on a hydro alcoholic

extract of *Ipomoea carnea* against EAC cells indicates the cytotoxic activity increases with concentration. MTT assay is based on the reduction of MTT (3-(4-5-dimethyl thiazolyl)-2-5-diphenyl-tetrazolium bromide) by mitochondrial dehydrogenase to purple formazan product and it showed 80.01% of cytotoxicity Thus IC<sub>50</sub> values of hydroalcoholic extract showed significant anticancer activity by MTT assay. The criteria for judging the value of any anticancer drug are the prolongation of life span, inhibition of gain in average body weight and the decrease in WBC<sup>19, 20</sup>. The result of the present study showed an anticancer effect of HAEIC against EAC in Swiss albino mice. A significant ( $p < 0.001$  and  $p < 0.05$ ) enhancement of The MST and decrease of gain in average body weight were observed.

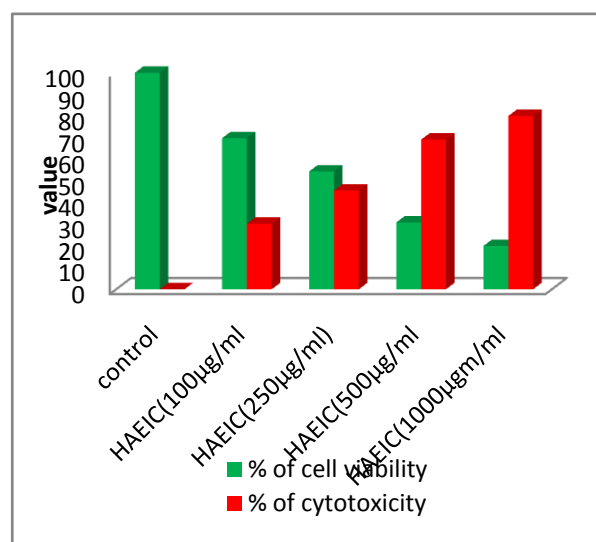


Fig 1. *In vitro* cytotoxic activity of HAEIC on EAC cell lines



**Tab 2. The Effect of the HAEIC on MST, % ILS, the average increase in body weight, change in food intake, tumor volume, tumor weight, PCV, viable and non-viable cell count in EAC bearing mice**

<b>Parameters</b>	<b>EAC</b> (Control)	<b>5-FU</b> (20mg/kg)	<b>HAEIC</b> (250mg/kg)	<b>HAEIC</b> (500mg/kg)
<b>Avg. increase in body weight (gm)</b>	14.21 ± 0.41	4.25 ± 0.54**	8.61 ± 0.65	7.13 ± 0.78*
<b>Change in food intake (gm)</b>	20.53 ± 1.85	50.36 ± 2.24***	36.36 ± 1.28***	43.52 ± 1.54***
<b>MST(Days)</b>	18.53 ± 0.21	36.16 ± 0.40***	27.83 ± 0.30***	33.83 ± 0.31***
<b>ILS(%)</b>	44.65	82.33**	63.57**	76.41**
<b>Tumor volume (mL)</b>	8.78±0.53	3.3±0.23***	6.9±0.13**	5.48±0.15***
<b>Tumor weight (gm)</b>	10.77±0.54	5.37±0.41***	9.20±0.27*	7.72±0.14***
<b>PCV</b>	6.61±0.22	1.76±0.28***	5.63±0.19***	3.9±0.15***
<b>Viable cell (×10<sup>6</sup> cells/ml)</b>	8.46±0.08	2.46±0.07***	3.62±0.05***	3.30±0.10***
<b>Non viable cell (×10<sup>6</sup> cells/ml)</b>	0.66±0.16	4.12±0.10***	2.33±0.20***	3.45±0.17***
<b>Viable (%)</b>	94.45	22.13**	42.43**	33.59**
<b>Non viable(%)</b>	11.89	92.36**	45.58**	67.44**

n=6 animals in each group, Values are represented as mean ± SEM of six animals \*  $p < 0.05$ , \*\* $p < 0.01$  and \*\*\* $p < 0.001$  between disease control and treated groups. Analyzed by ANOVA Tukey- Kramer multiple comparison tests.

**Tab 3 . The Effect of HAEIC on hematological parameters of EAC bearing mice**

Treatment	PARAMETERS					
	Hb (gms)	RBC (10 <sup>6</sup> cells/mm <sup>3</sup> )	WBC (10 <sup>3</sup> cells/mm <sup>3</sup> )	Differential Counts (%)		
				Lymphocytes	Neutrophils	Eosinophils
Normal	14.25±1.23	5.36±1.23	7.26±0.88	7.26±0.88	41.56±0.69	4.89±0.68
Tumor control	6.64±0.98	2.16±0.85	14.62±0.93	28.64±0.81	50.34±0.73	7.58±0.64
5-FU (20mg/kg)	12.38±0.74***	4.85±0.82***	09.32±0.87***	53.65±0.38***	38.63±0.82***	5.85±0.59***
HAEIC (250Mg/kg)	8.82±0.82*	3.67±0.79*	12.53±1.02	34.95±0.76	32.39±0.49*	6.24±0.35*
HAEIC (500Mg/kg)	10.05±1.02**	4.23±0.58**	11.36±0.98*	44.25±0.61***	35.64±0.51**	4.89±0.51***

N=6 animals in each group, Value are represented as mean ± SEM of six animal`s \**p*<0.05, \*\**p*<0.01 and \*\*\**p*<0.001 between disease control and treated group. (Analyzed by ANOVA Turkey-Kramer multiple comparison test)

**Tab 4. Effect of HAEIC on biochemical parameters of EAC- bearing mice**

Design of Treatment	Normal	Tumor Control	5-FU (20mg/Kg)	HAEIC (250mg/Kg)	HAEIC (500mg/Kg)
SGPT (U/I)	29.34±1.52	55.13±2.31	35.46±1.02***	43.25±1.35**	39.31±1.51***
SGOT(U/I)	38.53±1.24	70.36±1.61	42.56±1.28***	52.63±1.35*	46.38±1.64***
ALP (IU/L)	85.26±1.35	108.35±1.78	91.36±1.39***	99.86±1.51*	95.36±1.36***
Total Protein (mg/dl)	11.25±1.32	17.76±1.52	12.64±1.53***	15.56±1.64**	13.46±1.25***
LPO (Mol mda/mg protein)	1.56±0.23	4.14±0.36	2.13±0.27***	3.54±0.31*	2.86±0.16***
GSH (Mg/g wet tissue)	2.53±0.16	0.99±0.12	2.04±0.36***	1.23±0.26**	1.94±0.13***
SOD (U/mg protein)	5.26±1.81	2.35±1.47	4.43±1.58***	3.12±1.53**	4.98±0.61***
CAT (U/mg protein)	23.37±0.5	10.19±0.85	20.43±0.72***	17.52±0.6**	19.84±0.71***

N=6 animals in each group, values are represented as mean ± SEM of six animals. \* $p < 0.05$ , \*\* $p < 0.01$  and \*\*\* $p < 0.001$  between disease control and treated group (Analyzed by ANOVA Turkey-Kramer multiple comparison test)

Phytochemical studies indicate that the presence of tannins, flavonoids, phenols, alkaloids, saponins amino acids, etc. Polyphenolic compounds might inhibit cancer cells by xenobiotic metabolizing enzymes that alter metabolic activation of potential carcinogens, while some flavonoids could also alter hormone production and inhibit aromatase to prevent the development of cancer cells. The mechanism of action of anticancer activity of phenols could be by disturbing the cellular division during mitosis at the telophase stage. It was also reported that phenols reduce the amount of cellular protein and mitotic index and colony formation during cell proliferation of cancer cells.<sup>21</sup>

The Ehrlich tumor cells are one of the rapidly growing carcinoma with very aggressive behavior and are able to grow in almost all strains of mice<sup>22</sup>, Ascitic tumor implantation promotes local inflammatory reactions leading to increase in vascular permeability and results in intense edema formation, cellular migration and progressive ascitic fluid formation. Ascitic fluid is essential for tumor growth. Since it constitutes the direct nutritional source for tumor cells. Thus HAEIC decrease the tumor volume, tumor weight, packed cell volume of solid tumor. Reduction in

the viable cell count and increased non viable cell count towards normal in tumor host suggest antitumor effect against EAC cells in mice.<sup>23</sup>

The study shows that HAEIC increase the non viable cell count up to 45.58% and 67.44% at a dose of 250mg/kg and 500mg/kg respectively. This suggested that crude extract has a direct relationship with tumor cell. As these tumor cells have absorbed the anticancer drug by direct absorption and this anticancer aren't lysis the cells by direct cytotoxic mechanism.

Biological agents such as interferons and interleukins provide non-specific activity immunity (indirect cytotoxic action), whereas monoclonal antibodies provide passive immunity (direct cytotoxic action). Interferons are the small protein synthesized by the immune cells in response to various stimuli such as viral infection and cytokines inhibits viral replication and promote cellular immune response (T cell). Interferons acts via JAK-STAT pathway to stimulate the formation of specific proteins which mediate their actions. Their antitumor action is complex and includes antiproliferative effects, promotion of differentiation, immune modulation, alteration in tumor cell surface, antigen expression, inhibition of oncogene activation and angiogenesis. The

cytokines produced in the body by the lymphocytes are known as interleukins and they mediate cytotoxic action through the cell surface receptors in target cells.

Interleukins stimulates the growth and activity of immune cells, which target cancer cells. It acts as an antitumor agent by increasing the cytotoxic T lymphocytes and natural killer cells and by increasing the gene expression responsible for encoding the lytic component of cytotoxic granules.

Reduction in the viable cell count and increased non-viable cell count towards normal in tumor host suggested that HAEIC extract stimulate the growth and activity of immune cells by the production of interleukins. The reduced volume of EAC and increased survival time of the mice suggests that the extract might have exerted a delay in vascular permeability to the cells.<sup>24</sup>

While the standard drug 5-Fluorouracil as a pyrimidine analogue is transformed inside the cell into different cytotoxic metabolites which are then incorporated into DNA and RNA eventually inducing cell cycle arrest and apoptosis by inhibiting the DNA synthesis. The standard drug activity thus indicates a gene

expression mechanism is indirect cytotoxicity. The antitumor activity of the extract appears to also follow this mechanism.

Anemia and myelosuppression have been frequently observed ascites carcinoma.<sup>25</sup> Anemia encountered in ascites carcinoma due iron deficiency, either by a hemolytic or myelopathic condition which finally lead to reduce RBC count. Treatment with ethanol HAEIC brought back the hemoglobin content (Hb), RBC and WBC count more or less to normal level. Thus the study indicates that the hydroalcoholic extract of *Ipomoea carnea* shows a statistically significant increase in RBC level and a decrease in WBC level, which improves the hemopoietic system.

On the other hand the free radical scavenging system, SOD and catalase are present in all oxygen metabolizing cells. The SOD and catalase level was decreased in EAC bearing mice leads to decrease in total SOD activity in liver. The inhibition of SOD and CAT activity results in tumor growth<sup>25</sup>. The administration of HAEIC at different dose (250mg/kg and 500mg/kg) increased the SOD and CAT level, which may indicate the antioxidant and free radical scavenging property. The SGPT, SGOT and ALP level get increased in EAC bearing mice. After

the administration of HAEIC at different dose decreased the SGPT, SGOT and ALP level which improves the liver function.

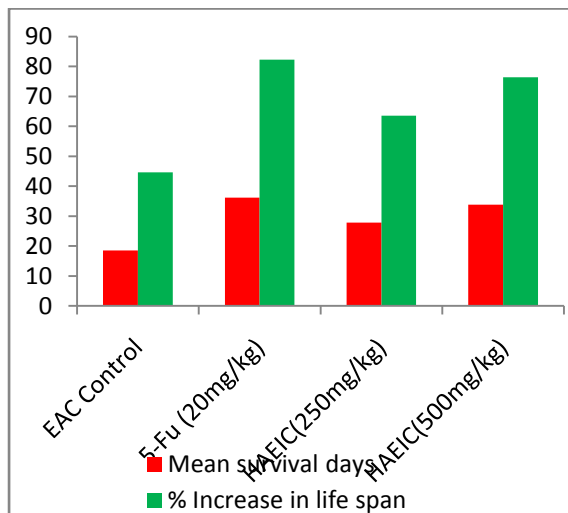


Fig 2.The effect of HAEIC on mean survival time and % increase in life span

## CONCLUSION

On the basis of the above result it was suggested that, the in-vitro and in-vivo anticancer activity of hydroalcoholic extract of *Ipomoea carnea jacq* leaves possess significant anticancer property with the dose dependent effect. This may probably due to the presence of phytochemicals such as alkaloids, phenols and flavonoids. Further isolation and purification of bioactive compound from *Ipomoea carnea* may reveal the presence of potent novel anticancer agent and also to explore the exact mechanism of action of the activity.

**Abbreviations used:** HAEIC= hydroalcoholic extract of *Ipomoea carnea*; EAC= Ehrlich Ascites Carcinoma; T.N.A.U=TamilNadu Agricultural University; OECD= Organization of Economic Co-Operation and Development; MTT=3(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyl tetrazolium bromide) assay; SGOT= Serum Glutamate Oxaloacetate Transaminase; SGPT= Serum Glutamate Pyruvate Transaminase; ALP =Alkaline Phosphatase; LPO= Lipid peroxidation levels; GSH= Glutathione Peroxidase; SOD=Superoxide dismutase; CAT= Catalase ; DMSO= Dimethyl sulfoxide; MST= mean survival time; OD=optical density; PCV=packed cell volume; ILS= increase in life span; WBC= white blood cells; RBC= red blood cells ; Hb= hemoglobin; JAK-STAT =Janus Kinase- signal Transducer and activator of transcription.

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