

Objective 3: To determine the preventive efficacy of cannabis extract on EAC cells induced cancer.

The primary objective is to demonstrate the cancer prevention efficacy of cannabis extract. For this task, mice were first treated with 3 different concentrations of cannabis extract. Mice were pre-treated with doses of cannabis at 4 different times (3rd day, 1st week, 2nd week, and 3rd week). Cancer cells are then inoculated into mice. After 3rd days, 1st week, 2nd week, and 3rd week of inoculation, the mice were sacrificed and observed by pathological tests.

Group 1: Effectiveness of three doses of cannabis extract in preventing cancer

4.3.1.1 Body weight measurement of albino mice

The preventive effects of cannabis extract on body weight (EAC induced after 3 doses of cannabis extract) are given in table no. 71. There were no significant variations ($p < 0.05$) observed among the treatment groups of mice.

Table 71: Preventive efficacy of cannabis extract on body weight (EAC induced after 3 doses of cannabis extract)

Time interval	Treatment groups				P-value
	T0 (g.)	T1 (g.)	T2 (g.)	T3 (g.)	
3 rd day	23.93±1.01	23.60±1.30	24.70±0.89	24.08±1.57	0.61
1 st week	28.13±1.41	26.80±1.15	31.57±2.05	28.83±1.99	0.11
2 nd week	34.03±1.58	33.73±1.57	37.80±2.02	35.19±1.09	0.34
3 rd week	40.87±2.65	41.10±1.64	44.47±2.55	42.14±1.30	0.36

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.1.2 Biochemical profile

4.3.1.2.1 RBS

The preventive effects of cannabis extract on RBS (EAC induced after 3 doses of cannabis extract) are given in table no. 72. No significantly varied ($p < 0.05$) among the treatment groups on the 3rd day, 1st, 2nd, and 3rd week. The highest result was T2 ($9.46 \pm 0.84 \text{ mmolL}^{-1}$) in the 3rd week was not significantly varied ($p < 0.05$) to the 3rd days, 1st, 2nd, and 3rd week of observation respectively.

Table 72: Preventive efficacy of cannabis extract on RBS (EAC induced after 3 doses of cannabis extract)

Time interval	Treatment groups				P-value
	T0 (mmolL ⁻¹)	T1 (mmolL ⁻¹)	T2 (mmolL ⁻¹)	T3 (mmolL ⁻¹)	
3 rd day	5.11±0.20	4.22±0.54	6.22±0.74	4.48±0.56	0.45
1 st week	4.32±0.92	7.76±1.12	7.47±1.08	6.73±1.01	0.31
2 nd week	5.10±0.60	7.46±1.37	7.40±1.24	7.96±1.78	0.11
3 rd week	5.13±0.47	7.73±1.63	9.46±1.87	7.67±1.27	0.26

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.1.2.2 Serum creatinine

The preventive effects of cannabis extract on serum creatinine (EAC induced after 3 doses of cannabis extract) are given in table no. 73. No significantly varied ($p < 0.05$) among the treatment groups on the 3rd day, 1st, 2nd, and 3rd week of observation respectively. The highest result was T1 ($0.34 \pm 0.03 \text{ mg/dl}$) in the 3rd week was not significantly varied ($p < 0.05$) to the 3rd days, 1st, 2nd, and 3rd week of observation of albino mice.

Table 73: Preventive efficacy of cannabis extract on serum creatinine (EAC induced after 3doses of cannabis extract)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	0.21±0.02	0.33±0.02	0.26±0.03	0.32±0.03	0.30
1 st week	0.27±0.03	0.21±0.02	0.20±0.03	0.23±0.02	0.54
2 nd week	0.26±0.03	0.33±0.05	0.32±0.03	0.23±0.03	0.24
3 rd week	0.27±0.07	0.34±0.03	0.23±0.02	0.27±0.05	0.12

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.1.2.3 SGPT

The preventive effects of cannabis extract on SGPT (EAC induced after 3 doses of cannabis extract) are given in table no. 74. SGPT results differed significantly ($P<0.05$) among the different treatment groups of mice in 1st week and 3rd week compared to the other groups of mice. The highest result of SGPT (324.67 ± 111.36 U/L) on the T3 treatment group in 3rd week was significantly varied ($p<0.05$) to the lowest result (27.00 ± 3.61 U/L) on the T0 treatment group in 1st week. In the 2nd week of observation, SGPT value shows significant variation ($p=0.05$) compare to the other treatment groups of mice. The highest value at T2 (214.67 ± 107.53 U/L) was significantly ($p=0.05$) to the T0, T1 and T3 respectively.

Table 74: Preventive efficacy of cannabis extract on SGPT (EAC induced after 3doses of cannabis extract)

Time interval	Treatment groups				P-value
	T0 (U/L)	T1 (U/L)	T2 (U/L)	T3 (U/L)	
3 rd day	29.33±3.71	39.67±8.16	33.33±6.94	29.67±3.38	0.87
1 st week	27.00±3.61 ^a	185.00±129.13 ^b	284.00±140.22 ^c	347.00±184.72 ^d	0.04
2 nd week	27.00±5.00 ^a	222.67±118.02 ^c	214.67±107.53 ^c	195.00±104.03 ^b	0.05
3 rd week	32.00±5.29 ^a	251.67±135.31 ^{bc}	193.00±64.61 ^b	324.67±151.36 ^c	0.04

The values are Mean±SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.1.2.4 SGOT

The preventive effects of cannabis extract on SGOT (EAC induced after 3 doses of cannabis extract) are given in table no. 75. SGOT results differed significantly ($P<0.001$) among the different treatment groups of mice on 1st week of observation. The highest result of SGOT (284.33 ± 48.02 U/L) in the T3 treatment group was significantly varied ($p<0.001$) to the lowest result (32.00 ± 1.00 U/L) in the T0 treatment group in 1st week, but there was no significant difference found in T0, T1 and T2, T3 groups of mice. In the 2nd and 3rd week of SGOT value shows ($p<0.001$) significant variation among the treatment groups of mice.

Table 75: Preventive efficacy of cannabis extract on SGOT (EAC induced after 3 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (U/L)	T1 (U/L)	T2 (U/L)	T3 (U/L)	
3 rd day	44.67 $\pm$ 2.73	46.00 $\pm$ 3.05	32.67 $\pm$ 6.36	39.33 $\pm$ 7.33	0.33
1 st week	32.00 $\pm$ 1.00 ^a	84.33 $\pm$ 3.18 ^a	281.00 $\pm$ 42.50 ^b	284.33 $\pm$ 48.02 ^b	0.003
2 nd week	24.67 $\pm$ 1.20 ^a	91.00 $\pm$ 2.65 ^b	168.00 $\pm$ 17.57 ^c	157.33 $\pm$ 11.89 ^c	<.001
3 rd week	45.67 $\pm$ 4.25 ^a	133.33 $\pm$ 0.88 ^b	174.00 $\pm$ 15.37 ^c	146.33 $\pm$ 9.02 ^{bc}	<.001

The values are Mean $\pm$ SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.1.2.5 Alkaline phosphatase

The preventive effects of cannabis extract on alkaline phosphatase (EAC induced after 3 doses of cannabis extract) are given in table no. 76. Alkaline phosphatase results differed significantly ($P<0.05$) among the different treatment groups of mice on 3rd day, 1st week, 2nd week and 3rd week of observation. The highest result of alkaline phosphatase (209.67 ± 70.67 U/L) in the T2 treatment group was significantly varied ($p<0.05$) to the lowest result (53.67 ± 12.33 U/L) in the T0 treatment group in 3rd day, but there was a significant difference found in T0, T1 and T2, T3 groups of mice. The highest result of alkaline phosphatase (250.00 ± 92.59 U/L) in the T3 group was significantly varied ($p<0.05$)

to the lowest result ($69.33 \pm 13.13 \text{ U/L}$) in the T0 treatment group in 1st week. The highest result of alkaline phosphatase ($247.00 \pm 37.50 \text{ U/L}$) in the T1 group was significantly varied ($p < 0.05$) to the lowest result ($86.33 \pm 4.63 \text{ U/L}$) in the T0 treatment group in 1st week. The highest result of alkaline phosphatase ($292.00 \pm 34.83 \text{ U/L}$) in the T1 group was significantly varied ($p < 0.05$) to the lowest result ($120.67 \pm 0.67 \text{ U/L}$) in the T0 treatment group in 1st week.

Table 76: Preventive efficacy of cannabis extract on alkaline phosphatase (EAC induced after 3 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (U/L)	T1 (U/L)	T2 (U/L)	T3 (U/L)	
3 rd day	53.67 ± 12.33^a	166.67 ± 16.70^b	209.67 ± 70.67^c	169.67 ± 24.92^b	0.02
1 st week	69.33 ± 13.13^a	194.33 ± 24.39^b	229.67 ± 90.43^{bc}	250.00 ± 92.59^c	0.05
2 nd week	86.33 ± 4.63^a	247.00 ± 37.50^b	207.00 ± 60.71^b	245.00 ± 42.79^c	0.01
3 rd week	120.67 ± 0.67^a	292.00 ± 34.83^c	230.00 ± 46.06^b	243.33 ± 48.48^b	0.04

The values are Mean $\pm$ SE and ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.1.2.6 Serum Cholesterol

The preventive effects of cannabis extract on serum cholesterol (EAC induced after 3 doses of cannabis extract) are given in table no. 77. No significant value varied ($p < 0.05$) among the treatment groups on the 3rd day, 1st, 2nd, and 3rd week respectively. The highest result of serum cholesterol was T3 ($184.02 \pm 4.51 \text{ mg/dl}$) on 3rd day and the lowest result was T0 ($112.67 \pm 1.58 \text{ mg/dl}$) in 2nd week of observation.

Table 77: Preventive efficacy of cannabis extract on serum cholesterol (EAC induced after 3 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	151.67±6.47	151.67±6.99	131.33±2.23	184.02±8.51	0.42
1 st week	160.33±6.32	150.33±5.50	136.67±3.89	169.12±6.10	0.69
2 nd week	112.67±1.58	148.67±4.95	138.33±3.07	123.00±2.52	0.88
3 rd week	152.00±5.50	156.67±5.89	137.00±3.64	176.00±7.02	0.54

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.1.2.7 Serum Triglyceride

The preventive effects of cannabis extract on serum triglyceride (EAC induced after 3 doses of cannabis extract) are given in table no. 78. Significantly varied ($p<0.05$) among the observation groups at 3rd week compare to the other treatment groups of mice. The highest result of serum triglyceride was significantly ($p<0.05$) different at T3 (173.33±9.90) at 3rd week to T2 and T3 respectively.

Table 78: Preventive efficacy of cannabis extract on serum triglyceride (EAC induced after 3 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	122.00±1.53	105.33±1.44	103.33±1.89	106.33±1.84	0.28
1 st week	133.33±2.32	130.33±1.50	136.67±2.89	129.09±2.10	0.36
2 nd week	123.33±2.47	121.67±2.98	111.00±1.10	132.33±3.83	0.28
3 rd week	173.33±6.90 ^c	165.33±5.17 ^{bc}	143.00±4.00 ^a	157.83±5.04 ^b	0.02

The values are Mean±SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.1.2.8 Serum uric acid

The preventive effects of cannabis extract on serum uric acid (EAC induced after 3 doses of cannabis extract) are given in table no. 79. Significant ($p<0.05$) variation was observed at the 3rd week of observation. The highest result of uric acid was T3 ($13.67\pm4.80\text{mg/dl}$) in 3rd week of treatment was significantly varied ($p<0.05$) to the lowest result was T0 ($5.30\pm0.55\text{mg/dl}$) in the 3rd day of observation.

Table 79: Preventive efficacy of cannabis extract on uric acid (EAC induced after 3 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	5.30 ± 0.55	6.2 ± 0.72	5.33 ± 0.33	6.70 ± 0.30	0.33
1 st week	6.30 ± 1.01	6.70 ± 1.70	7.13 ± 1.61	6.87 ± 1.00	0.45
2 nd week	5.80 ± 0.76	6.10 ± 0.58	9.57 ± 0.97	6.63 ± 1.88	0.35
3 rd week	7.17 ± 1.04^a	6.00 ± 1.15^a	8.67 ± 2.03^a	13.67 ± 4.80^b	0.04

The values are Mean $\pm$ SE. ^{ab} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.1.3 Hematological profile

4.3.1.3.1 Hb

The preventive effects of cannabis extract on Hb (EAC induced after 3 doses of cannabis extract) are given in table no. 80. No significant ($p<0.05$) variation was observed among the treatment groups on the 3rd day, 1st week, 2nd week, and 3rd week respectively. The highest value of hemoglobin was not significantly varied ($p<0.05$) at T0 ($14.97\pm0.23\text{mg/dl}$) in the 1st-week and the lowest value was ($14.03\pm0.54\text{mg/dl}$) in the 2nd week of the T0 group of mice.

4.3.1.3.2 RBC count

The preventive effects of cannabis extract on Hb (EAC induced after 3 doses of cannabis extract) are given in table no. 80. The RBC test result was no significantly ($p<0.05$)

different among the treatment groups on the 3rd day, 1st week, 2nd week and 3rd week respectively. The highest values of RBC were not significantly varied ($p < 0.05$) at T1 ($5.00 \pm 0.01 \times 10^6/\text{cmm}$) in the 2nd week and the lowest value was ($3.60 \pm 0.30 \times 10^6/\text{cmm}$) in the 1st week of T2 group of mice.

4.3.1.3.3 WBC count

The preventive effects of cannabis extract on WBC count (EAC induced after 3 doses of cannabis extract) are given in table no. 80. Significant differences ($p < 0.01$) were observed among the treatment group in 1st week of observation. The WBC count was significantly ($p < 0.01$) highest in T1 ($41.37 \pm 7.21 \times 10^3/\text{cmm}$) and lowest in T0 ($4.37 \pm 0.60 \times 10^3/\text{cmm}$) treatment groups. No significant variation was observed in the T1, T2, and T3 treatment groups of mice. Significant differences ($p < 0.001$) were observed among the treatment group in 2nd week of observation. WBC count was significantly ($p < 0.001$) highest in T1 ($65.51 \pm 10.13 \times 10^3/\text{cmm}$) and lowest in T0 ($4.47 \pm 0.43 \times 10^3/\text{cmm}$) treatment groups. Significant differences ($p < 0.005$) were observed among the treatment group in 3rd week of observation. WBC count was significantly ($p < 0.005$) highest in T1 ($91.21 \pm 12.81 \times 10^3/\text{cmm}$) and lowest in T0 ($4.71 \pm 0.66 \times 10^3/\text{cmm}$) treatment groups.

4.3.1.3.4 Platelet count

The preventive effects of cannabis extract on platelet count (EAC induced after 3 doses of cannabis extract) are given in table no. 80. There were no significant variation ($p < 0.05$) observed in all treatment groups of mice at the 3rd day, 1st week, 2nd week and 3rd week respectively. The highest result at 3rd-week observation T0 ($4.46 \pm 0.41 \times 10^5/\text{cmm}$) of platelet count was not significantly ($p < 0.05$) varied to the lowest result at 3rd days T0 ($2.30 \pm 0.25 \times 10^3/\text{cmm}$).

Table 80: Preventive efficacy of cannabis extract on hematological profile (EAC induced after 3 doses of cannabis extract injection)

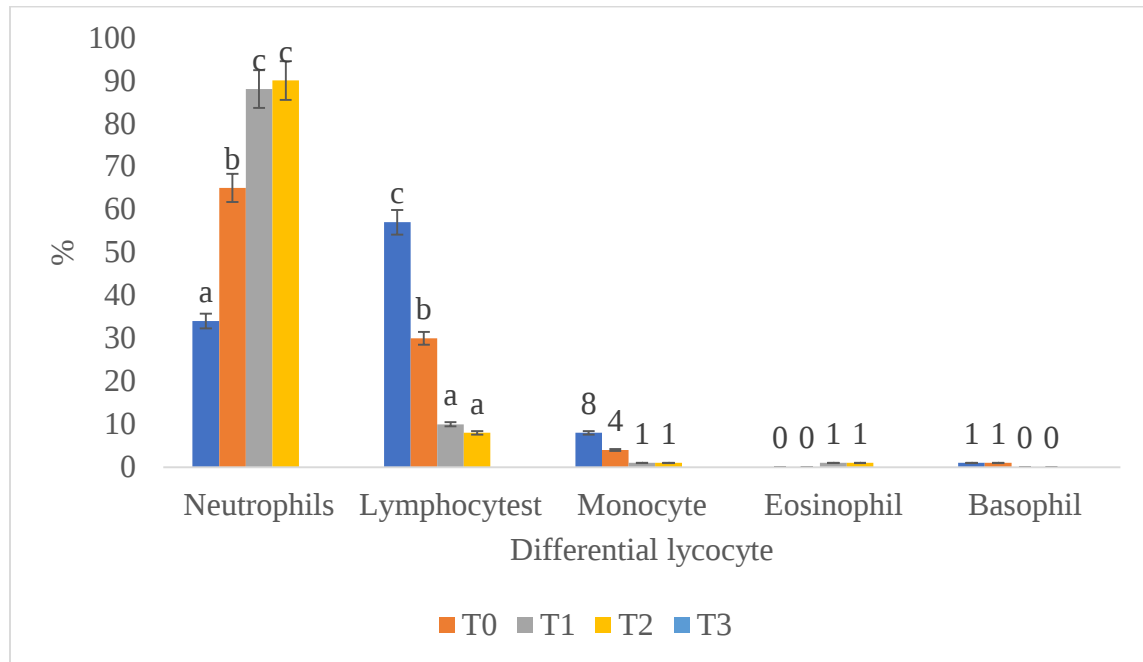
Parameters	Time interval	Treatment groups				P-value
		T0	T1	T2	T3	
Hb (mg/dl)	3 rd day	14.11±0.65	14.67±0.33	14.27±0.63	14.10±0.67	0.89
	1 st week	14.97±0.23	14.96±0.03	14.17±0.54	14.30±0.30	0.84
	2 nd week	14.03±0.54	14.76±0.39	14.70±0.30	14.60±0.31	0.24
	3 rd week	14.67±0.31	14.13±0.69	14.30±0.30	14.83±0.17	0.23
RBC (10 ⁶ /cmm)	3 rd day	4.70±0.35	4.20±0.20	4.23±0.2	4.76±0.39	0.45
	1 st week	4.67±0.33	4.93±0.07	3.60±0.30	4.63±0.32	0.14
	2 nd week	4.37±0.32	5.00±0.01	4.03±0.03	4.93±0.07	0.38
	3 rd week	4.87±0.13	5.00±0.01	4.97±0.03	4.17±0.17	0.22
WBC (10 ³ /cmm)	3 rd day	5.47±0.71	15.11±5.71	8.44±2.83	6.97±1.42	0.25
	1 st week	4.37±0.60 ^a	41.37±7.21 ^c	31.83±8.49 ^{bc}	17.82±2.76 ^b	0.009
	2 nd week	4.47±0.43 ^a	65.51±10.13 ^c	53.33±9.95 ^{bc}	40.41±4.11 ^b	0.002
	3 rd week	4.71±0.66 ^a	91.21±12.81 ^c	88.53±19.32 ^{bc}	66.96±11.02 ^b	0.005
Platelet (10 ⁵ /cmm)	3 rd day	2.30±0.25	3.57±0.36	3.27±0.04	3.23±0.54	0.14
	1 st week	3.20±0.10	3.96±1.05	2.79±0.19	2.86±0.24	0.46
	2 nd week	3.45±0.49	2.91±0.41	2.53±0.23	2.89±0.29	0.42
	3 rd week	4.46±0.41	3.70±1.04	3.27±0.53	2.83±0.27	0.37

The values are Mean±SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value

4.3.1.3.5 Differential leucocyte count

The preventive effects of cannabis extract on differential WBC count (EAC induced after 3 doses of cannabis extract) are given in figure no. 53. Significant differences ($p < 0.05$) were observed among the treatment on neutrophils count. Significantly ($p < 0.05$) highest value was found at T3 ($90.00 \pm 2.11\%$) and the lowest value was ($34.00 \pm 1.20\%$). Significant ($p < 0.05$) differences were also observed among the treatment group of lymphocyte count. The highest value at the T0 ($57.00 \pm 1.03\%$) and lowest value at T3 ($8.00 \pm 0.02\%$). No significant differences were observed for monocyte, eosinophil, and basophil count.

Figure 53: Leucocyte count in 3 days of dose application of cannabis extract for cancer prevention



^{abc} values having different superscripts differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups.

4.3.1.4 Tumor growth response

The preventive effects of cannabis extract on tumor volume, packed cell volume and tumor cell count (EAC induced after 3 doses of cannabis extract) are given in table no. 81. T1, T2 and T3 group show ($p < 0.05$) significant values. The highest value in T1 was tumor cell volume (4.13 ± 0.11 ml) was significantly varied ($p < 0.05$) to the lowest value of tumor-packed cell volume (1.43 ± 0.03 ml). The highest value in T2 was tumor cell volume (3.11 ± 0.16 ml) was significantly varied ($p < 0.05$) to the lowest value of tumor-packed cell volume (1.14 ± 0.1 ml). The highest value in T3 was tumor cell volume (2.21 ± 0.22 ml) was significantly varied ($p < 0.05$) to the lowest value of tumor-packed cell volume (0.81 ± 0.01 ml).

Table 81: The preventive effects of cannabis extract on tumor volume, packed cell volume and tumor cell count (EAC induced after 3 doses of cannabis extract)

Group	Tumor volume (ml)	Packed cell volume (ml)	Tumor cell count $\times 10^7$ cells/ml/mice		P-value
			Viable	Nonviable	
T1	4.13 ± 0.11^b	1.43 ± 0.03^a	2.15 ± 0.12^a	1.44 ± 0.2^a	0.05
T2	3.11 ± 0.16^b	1.14 ± 0.1^a	1.32 ± 0.11^a	2.14 ± 0.33^c	0.05
T3	2.21 ± 0.22^b	0.81 ± 0.01^a	1.01 ± 0.32^a	5.21 ± 0.53^c	0.05

Values= Mean $\pm$ SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, P-value= probability value.

4.3.1.5 Hematological study

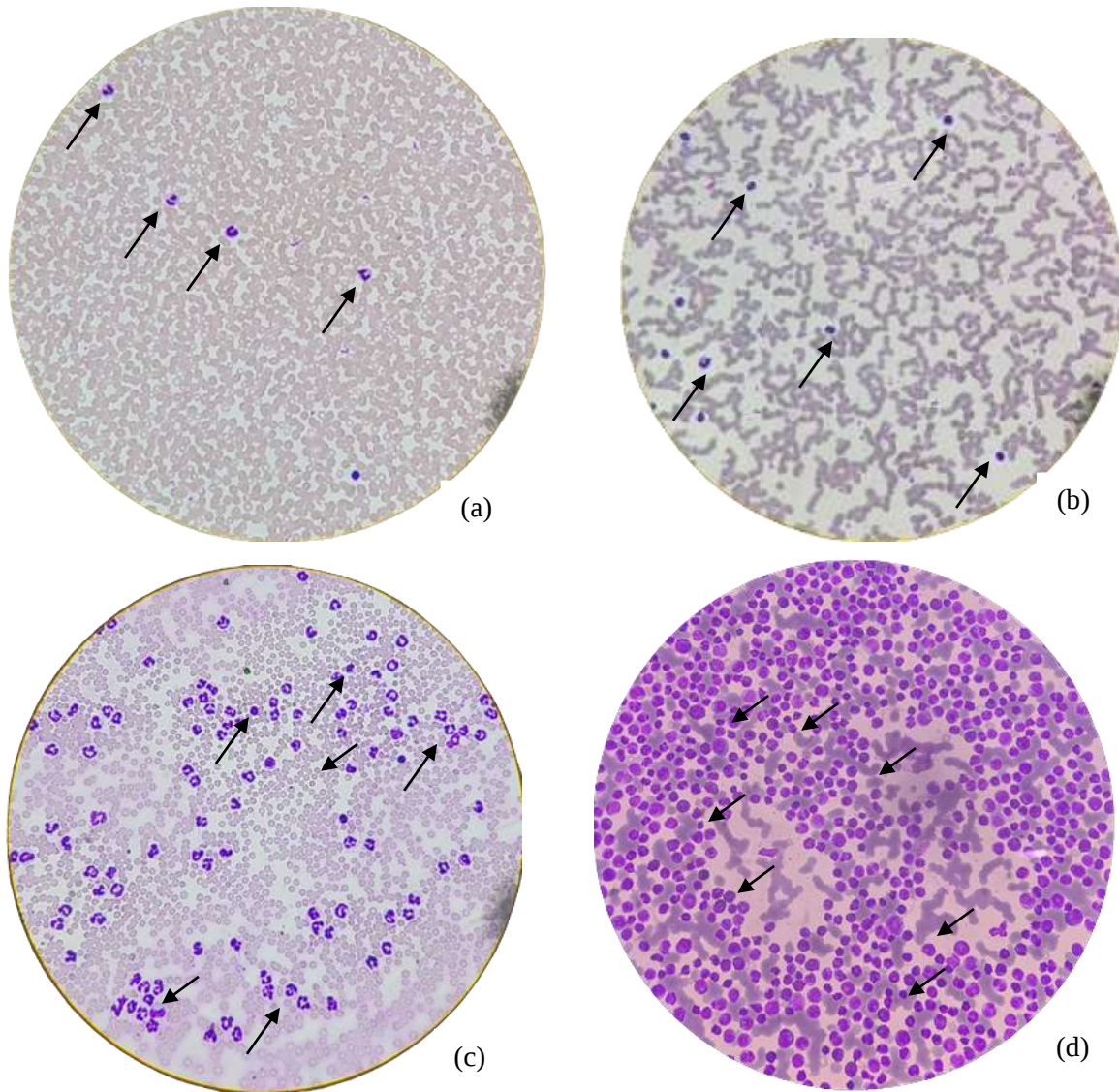


Figure 54: Preventive effects of cannabis extract on WBC count (EAC induced after 3 doses of cannabis extract injection). Hematological slide observation under light microscope 400x view (a) 3th days, (b) 1st week, (c) 2nd week, and (d) 3rd week of observation after EAC-induced cancer in albino mice. Marking area were indicated WBCs on the images. Gratuallly increased WBC counts with the increasing the observations period.

4.3.1.6 Cytopathological study of ascitic fluid

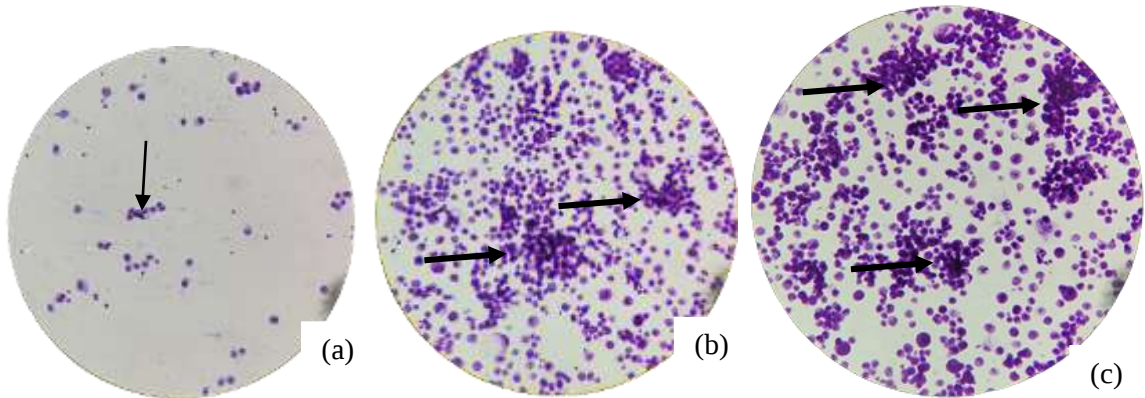
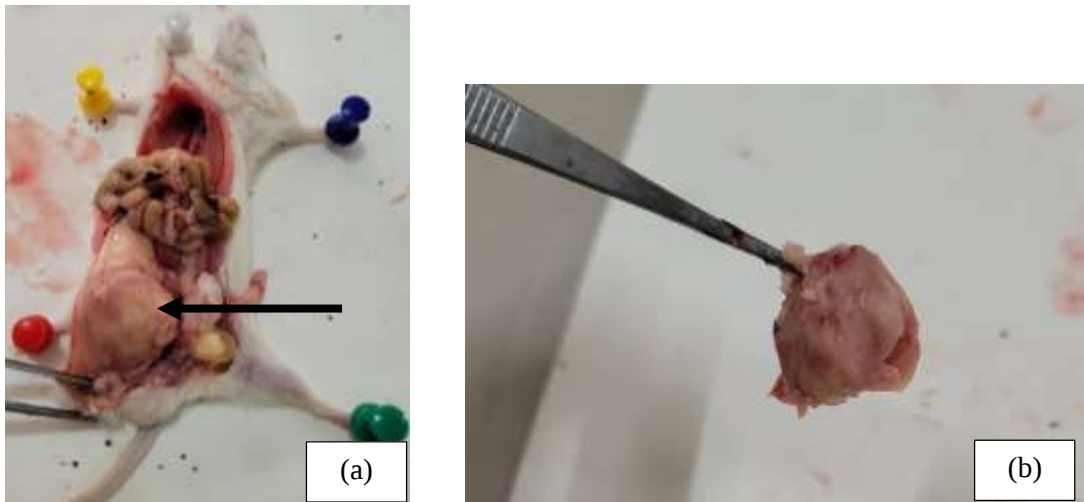


Figure 55: Preventive effects of cannabis extract on cytological changes of ascitic fluids (EAC induced after 3 doses of cannabis extract injection). Cytopathology observation using H and E dyes, and observed using a light microscope 400x view of (a) 7days after EAC inoculation, (b) 14days after EAC inoculation, and (c) 21days after EAC inoculation to the mice. Marking areas indicated the a typical cells.

4.3.1.7 Histopathological study of tumor



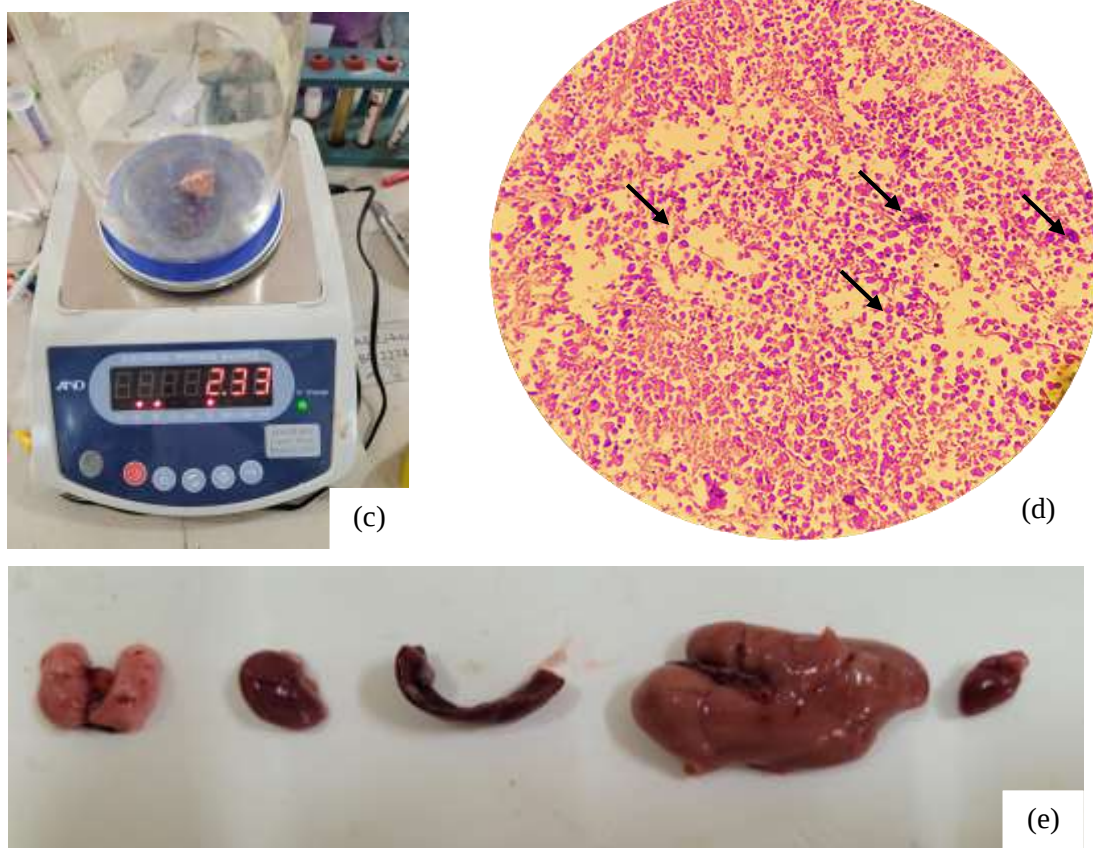


Figure 56: Preventive effects of cannabis extract on cellular changes of main organ and tumor of albino mice (EAC induced after 3 doses of cannabis extract injection). For histopathological observation (a) mice were sacrificed, marking area indicates tumor, (b) tumor was surgically removed from mice, (c) tumor was weighted using digital balance, (d) histopathological observation of tumor using H & E dyes, under light microscope 400x view, and the marking area indicates sarcoma type cancer cells (e) main organ of mouse surgically removed.

Group 2: Effectiveness of seven doses of cannabis extract in preventing cancer

4.3.2.1 Body weight measurement of albino mice

The preventive effects of cannabis extract on body weight (EAC induced after 7 doses of cannabis extract injection) is given in table no. 82. There were no significant variation ($p < 0.05$) observed among the treatment groups at the 3rd day, 1st week, 2nd week, and 3rd week respectively.

Table 82: Preventive efficacy of cannabis extract on body weight (EAC induced after 7 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (g.)	T1 (g.)	T2 (g.)	T3 (g.)	
3 rd day	23.37±0.85	23.60±1.30	24.77±0.93	23.91±0.56	0.38
1 st week	25.03±1.29	25.87±1.22	28.10±2.04	26.33±1.90	0.41
2 nd week	28.33±1.39	27.90±0.66	30.10±2.01	28.78±0.81	0.49
3 rd week	32.70±1.19	32.00±1.27	35.73±1.57	33.48±1.41	0.31

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.2.2 Biochemical profile

4.3.2.2.1 RBS

The preventive effects of cannabis extract on RBS (EAC induced after 7 doses of cannabis extract injection) is given in table no. 83. There were no significant variation ($p < 0.05$) observed among the treatment groups at the 3rd day, 1st week, 2nd week, and 3rd week respectively. The highest result was T2 ($8.46 \pm 0.47 \text{ mmol/L}^{-1}$) at 3rd week was not significantly varied ($p < 0.05$) to the lowest result was T2 ($4.40 \pm 0.91 \text{ mmol/L}^{-1}$) at 2nd week of observation.

Table 83: Preventive efficacy of cannabis extract on RBS (EAC induced after 7 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mmolL ⁻¹)	T1 (mmolL ⁻¹)	T2 (mmolL ⁻¹)	T3 (mmolL ⁻¹)	
3 rd day	6.13±0.23	5.21±0.50	6.02±0.04	5.40±0.51	0.49
1 st week	5.31±0.32	6.72±0.86	6.47±0.31	6.73±1.60	0.54
2 nd week	5.64±0.63	7.43±2.30	4.40±0.91	4.90±0.72	0.23
3 rd week	5.43±0.47	7.73±1.13	8.46±0.47	7.61±1.37	0.46

The values are Mean±SE, T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.2.2.2 Serum creatinine

The preventive effects of cannabis extract on RBS (EAC induced after 7 doses of cannabis extract injection) is given in table no. 84. There were no significant variation ($p<0.05$) observed among the treatment groups at the 3rd day, 1st week, 2nd week, and 3rd week respectively. The highest result was T3 (0.33±0.03mg/dl) at 3rd week was not significantly ($p<0.05$) varied to the lowest result T0 (0.17±0.02mg/dl) at the 1st week of observation.

Table 84: Preventive efficacy of cannabis extract on serum creatinine (EAC induced after 7 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	0.30±0.04	0.23±0.02	0.21±0.03	0.33±0.03	0.42
1 st week	0.17±0.02	0.21±0.02	0.24±0.03	0.22±0.02	0.34
2 nd week	0.23±0.03	0.31±0.03	0.30±0.03	0.27±0.02	0.24
3 rd week	0.27±0.07	0.29±0.03	0.20±0.02	0.24±0.03	0.12

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.2.2.3 SGPT

The preventive effects of cannabis extract on SGPT (EAC induced after 7 doses of cannabis extract injection) is given in table no. 85. SGPT result was differed significantly ($P<0.005$) among the different treatment groups of mice in 3rd days of observation. Lowest result of SGPT in T0 (28.33 ± 6.94 U/L) at 3rd days was significantly varied ($p<0.05$) to the highest result in T2 (77.67 ± 10.35 U/L). In the 1st week of observation SGPT test result was differed significantly ($P<0.05$) among the different treatment groups of mice. Lowest result of SGPT in T0 (25.67 ± 2.33 U/L) was significantly varied ($p<0.05$) to the highest result in T1 (221 ± 64.26 U/L). In the 2nd week of observation SGPT test result was differed significantly ($P<0.001$) among the different treatment groups of mice. Lowest result of SGPT in T0 (27.33 ± 3.71 U/L) was significantly varied ($p<0.05$) to the highest result in T1 (525.0 ± 97.94 U/L). In the 3rd week of observation SGPT test result was differed significantly ($P<0.001$) among the different treatment groups of mice. Lowest result of SGPT in T0 (37.33 ± 3.71 U/L) was significantly varied ($p<0.05$) to the highest result in T1 (932.3 ± 72.34 U/L).

Table 85: Preventive efficacy of cannabis extract on SGPT (EAC induced after 7 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (U/L)	T1 (U/L)	T2 (U/L)	T3 (U/L)	
3 rd day	28.33 ± 6.94^a	77.67 ± 10.35^c	48.00 ± 3.00^b	31.67 ± 6.36^a	0.005
1 st week	21.33 ± 5.36^a	221 ± 64.26^b	95.00 ± 22.52^{ab}	107.00 ± 31.99^c	0.05
2 nd week	25.67 ± 2.33^a	525.0 ± 97.94^c	406.33 ± 81.76^{bc}	188.0 ± 42.21^{ab}	0.01
3 rd week	37.33 ± 3.71^a	932.3 ± 72.34^c	505.67 ± 68.88^b	185.00 ± 43.79^a	<0.001

The values are Mean±SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.2.2.4 SGOT

The preventive effects of cannabis extract on SGOT (EAC induced after 7 doses of cannabis extract injection) is given in table no. 86. SGOT result was differed significantly ($P<0.05$) among the different treatment groups of mice in 3rd days of observation. The lowest result of SGOT in T0 (31.6 ± 5.81 U/L) at 3rd days was significantly varied ($p<0.05$) to the highest result in T1 (73.00 ± 4.04 U/L). In the 1st week of observation SGOT test result was differed significantly ($P<0.05$) among the different treatment groups of mice. Lowest result of SGOT in T0 (30.6 ± 8.09 U/L) was significantly varied ($p<0.05$) to the highest result in T1 (152.67 ± 17.14 U/L). In the 2nd week of observation SGOT test result was differed significantly ($P<0.001$) among the different treatment groups of mice. Lowest result of SGOT in T0 (26.6 ± 3.18 U/L) was significantly varied ($p<0.001$) to the highest result in T1 (396.67 ± 38.44 U/L). In the 3rd week of observation SGOT test result was differed significantly ($P<0.001$) among the different treatment groups of mice. Lowest result of SGOT in T0 (27.33 ± 3.71 U/L) was significantly varied ($p<0.001$) to the highest result in T1 (235.33 ± 37.33 U/L).

Table 86: Preventive effocacta of cannabis extract on SGOT (EAC induced after 7 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (U/L)	T1 (U/L)	T2 (U/L)	T3 (U/L)	
3 rd day	31.6 ± 5.81^a	73.00 ± 4.04^b	29.00 ± 9.07^a	26.67 ± 7.42^a	0.004
1 st week	30.6 ± 8.09^a	152.67 ± 17.14^b	51.00 ± 19.09^a	63.33 ± 13.22^a	0.002
2 nd week	26.6 ± 3.18^a	396.67 ± 38.44^d	205.67 ± 12.76^b	283.6 ± 15.36^c	<0.001
3 rd week	38.6 ± 2.96^a	711.00 ± 98.33^c	317.00 ± 12.50^b	599.3 ± 31.83^c	<.0001

The values are Mean $\pm$ SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, P-value= probability value.

4.3.2.2.5 Alkaline phosphatase

The preventive effects of cannabis extract on alkaline phosphatase (EAC induced after 7 doses of cannabis extract injection) is given in table no. 87. Alkaline phosphatase test result was differed significantly ($P=0.001$) among the different treatment groups of mice in 1st week of observation. The lowest result of alkaline phosphatase in T0 (52.00 ± 19.09 U/L) at 1st week was significantly varied ($p=0.001$) to the highest result in T3 (435.00 ± 45.24 U/L). There no significant variation observed among the treatment groups of T1, T2, and T3. In the 2nd week of observation alkaline phosphate test result was differed significantly ($P<0.001$) among the different treatment groups of mice. The lowest result of alkaline phosphatase in T0 (79.33 ± 11.79 U/L) was significantly varied ($p<0.001$) to the highest result in T3 (685.33 ± 63.82 U/L). In the 3rd week of observation alkaline phosphatase test result was differed significantly ($P<0.001$) among the different treatment groups of mice. The lowest result of alkaline phosphatase in T0 (115.00 ± 6.56 U/L) was significantly varied ($p<0.001$) to the highest result in T2 (919.33 ± 36.03 U/L).

Table 87: Preventive efficacy of cannabis extract on alkaline phosphatase (EAC induced after 7 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (U/L)	T1 (U/L)	T2 (U/L)	T3 (U/L)	
3 rd day	65.67 $\pm$ 10.87	191.33 $\pm$ 43.73	80.33 $\pm$ 29.36	158.33 $\pm$ 92.67	0.35
1 st week	52.00 $\pm$ 19.09 ^a	329.67 $\pm$ 8.69 ^b	329.67 $\pm$ 65.73 ^b	435.00 $\pm$ 45.24 ^b	0.001
2 nd week	79.33 $\pm$ 11.79 ^a	407.00 $\pm$ 6.56 ^b	528.33 $\pm$ 26.74 ^c	685.33 $\pm$ 63.82 ^d	<0.001
3 rd week	115.00 $\pm$ 6.56 ^a	715.33 $\pm$ 31.70 ^b	919.33 $\pm$ 36.03 ^c	882.00 $\pm$ 62.43 ^c	<0.001

The values are Mean $\pm$ SE, ^{abcd} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.2.2.6 Serum Cholesterol

The preventive effects of cannabis extract on serum cholesterol (EAC induced after 7 doses of cannabis extract injection) is given in table no. 88. No significant variations ($p < 0.05$) were observed among the treatment groups at the 3rd day, 1st, 2nd, and 3rd week respectively. The highest result of serum cholesterol was T3 (152.67 ± 15.18 mg/dl) at 1st week of treatment was not significantly varied ($p < 0.05$) to the lowest result was T0 (99.33 ± 5.75 mg/dl).

Table 88: Preventive efficacy of cannabis extract on serum cholesterol (EAC induced after 7 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	99.33 $\pm$ 5.75	132.00 $\pm$ 8.35	112.60 $\pm$ 6.08	141.00 $\pm$ 7.06	0.15
1 st week	127.67 $\pm$ 13.21	152.03 $\pm$ 19.64	143.33 $\pm$ 12.18	152.67 $\pm$ 15.18	0.54
2 nd week	120.33 $\pm$ 10.17	111.00 $\pm$ 11.84	137.33 $\pm$ 19.33	127.00 $\pm$ 13.00	0.31
3 rd week	116.00 $\pm$ 12.50	120.00 $\pm$ 11.92	139.67 $\pm$ 11.49	133.00 $\pm$ 15.84	0.14

The values are Mean $\pm$ SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.2.2.7 Serum Triglyceride

The preventive effects of cannabis extract on serum triglyceride (EAC induced after 7 doses of cannabis extract injection) is given in table no. 89. No significant variations ($p < 0.05$) were observed among the treatment groups at the 3rd day, 1st, 2nd, and 3rd week respectively. The highest result of serum triglyceride was T2 (136.01 ± 19.51) at 3rd week of treatment was not significantly varied ($p < 0.05$) to the lowest result was T0 (106.60 ± 11.17 mg/dl) at 2nd week of observation.

Table 89: Preventive efficacy of cannabis extract on serum triglyceride (EAC induced after 7 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	113.00±2.59	101.20±2.92	117.67±2.51	113.00±3.05	0.11
1 st week	112.61±6.20	111.30±8.60	123.00±9.54	123.00±12.0	0.37
2 nd week	106.60±11.17	101.33±8.18	123.33±17.17	127.67±22.3	0.12
3 rd week	126.60±12.80	127.10±13.50	136.01±19.51	133.33±14.7	0.22

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.2.2.8 Serum uric acid

The preventive effects of cannabis extract on serum uric acid (EAC induced after 7 doses of cannabis extract injection) is given in table no. 90. No significant variations ($p < 0.05$) were observed among the treatment groups at the 3rd day, 1st, 2nd, and 3rd week respectively. The highest result of serum uric acid was T0 (8.11±2.11mg/dl) at 3rd days of treatment was not significantly varied ($p < 0.05$) to the lowest result was T3 (3.36±1.35mg/dl) at 3rd week of observation.

Table 90: Preventive efficacy of cannabis extract on uric acid (EAC induced after 7 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	8.11±2.11	6.96± 0.73	7.70±1.19	5.60 ± 0.32	0.70
1 st week	7.84±0.67	7.36± 0.87	6.76±3.68	8.90 ± 1.53	0.98
2 nd week	4.13±0.86	5.33± 0.28	7.33±1.46	6.10 ± 1.01	0.48
3 rd week	3.36±1.35	6.33± 0.70	7.20±1.41	6.70 ± 1.17	0.82

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.2.3 Hematological profile

4.3.2.3.1 Hb

The preventive effects of cannabis extract on Hb (EAC induced after 7 doses of cannabis extract injection) is given in table no. 91. No significantly ($p < 0.05$) varied observed among the treatment groups of T0, T1, T2, and T3. The highest values of hemoglobin was not significantly varied ($p < 0.05$) at T1 ($15.10 \pm 0.10 \text{ mg/dl}$) in the 2nd-week and the lowest value was ($13.67 \pm 0.33 \text{ mg/dl}$) at 3rd days of T1 group of observation.

4.3.2.3.2 RBC count

The preventive effects of cannabis extract on RBC count (EAC induced after 7 doses of cannabis extract injection) is given in table no. 91. The RBC count test result was no significantly ($p < 0.05$) different among the treatment groups at the 3rd day, 1st week, and 2nd week and 3rd week respectively. The highest values of RBC was not significantly varied ($p < 0.05$) at T1 ($4.93 \pm 0.52 \times 10^6/\text{cmm}$) in the 3rd-week and the lowest value was ($4.03 \pm 0.32 \times 10^6/\text{cmm}$) in the 3rd week of T3 group of observation.

4.3.2.3.3 WBC count

The preventive effects of cannabis extract on WBC count (EAC induced after 7 doses of cannabis extract injection) is given in table no. 91. Significant differences ($p < 0.01$) observed in all the treatment groups (T0, T1, T2, and T3) of 3rd day, 1st week, 2nd week, and 3rd week respectively. On 1st week of observation, the WBC count was significantly ($p < 0.05$) highest in T2 (19.25 ± 2.18) and lowest in T0 (4.37 ± 0.60) treatment groups. On the 2nd week of observation, the WBC count was significantly ($p < 0.01$) highest in T2 (30.00 ± 3.86) and lowest in T0 (4.47 ± 0.43) treatment groups. After 3rd week of observation, the WBC count was significantly ($p < 0.001$) highest in T1 (55.00 ± 6.42) and lowest in T0 (4.70 ± 0.65) treatment groups.

4.3.2.3.4 Platelet count

The preventive effects of cannabis extract on platelet count (EAC induced after 7 doses of cannabis extract injection) is given in table no. 91. The platelet count test result was no significantly ($p < 0.05$) different among the treatment groups at the 3rd day, 1st week, and 2nd week and 3rd week respectively. The highest values of RBC was not significantly varied ($p < 0.05$) at T3 ($5.32 \pm 1.31 \times 10^5/\text{cmm}$) in the 1st week and the lowest value was ($4.10 \pm 0.10 \times 10^5/\text{cmm}$) in the 3rd day of T0 group of observation.

Table 91: Preventive efficacy of cannabis extract on hematological profile (EAC induced after 7 doses of cannabis extract injection)

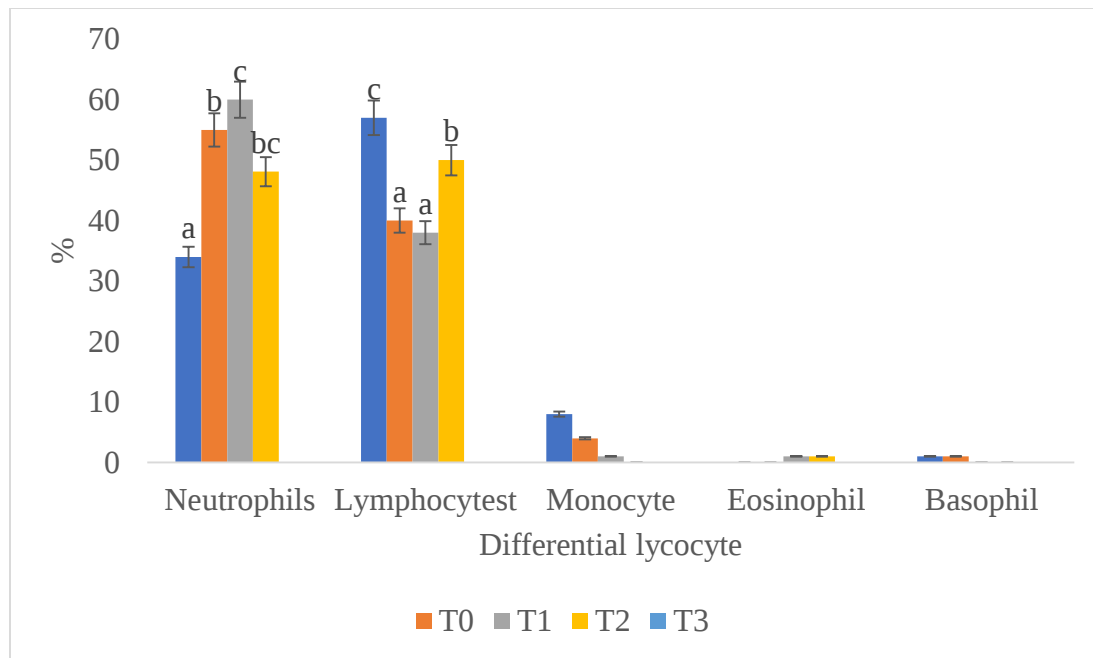
Parameters	Time interval	Treatment groups				P-value
		T0	T1	T2	T3	
Hb (mg/dl)	3 rd day	14.17 $\pm$ 0.31	13.67 $\pm$ 0.33	14.93 $\pm$ 0.51	14.70 $\pm$ 0.77	1.98
	1 st week	14.13 $\pm$ 0.62	14.07 $\pm$ 0.54	14.16 $\pm$ 0.32	14.32 $\pm$ 0.32	0.98
	2 nd week	14.00 $\pm$ 0.57	15.10 $\pm$ 0.10	14.67 $\pm$ 0.33	14.27 $\pm$ 0.63	0.41
	3 rd week	14.32 $\pm$ 0.33	14.22 $\pm$ 0.80	14.20 $\pm$ 0.41	14.50 $\pm$ 0.29	0.58
RBC ($10^6/\text{cmm}$)	3 rd day	4.11 $\pm$ 0.91	4.32 $\pm$ 0.31	4.23 $\pm$ 0.39	4.03 $\pm$ 0.32	0.35
	1 st week	4.33 $\pm$ 0.33	4.43 $\pm$ 0.35	4.20 $\pm$ 0.21	4.34 $\pm$ 0.33	0.18
	2 nd week	4.27 $\pm$ 0.33	4.68 $\pm$ 0.33	4.91 $\pm$ 0.07	4.70 $\pm$ 0.91	0.39
	3 rd week	4.23 $\pm$ 0.33	4.93 $\pm$ 0.52	4.13 $\pm$ 0.13	4.33 $\pm$ 0.33	0.20
WBC ($10^3/\text{cmm}$)	3 rd day	5.44 $\pm$ 0.71	5.14 $\pm$ 0.58	6.23 $\pm$ 1.55	6.69 $\pm$ 1.42	0.68
	1 st week	4.37 $\pm$ 0.60 ^a	14.84 $\pm$ 4.80 ^b	19.25 $\pm$ 2.18 ^c	16.82 $\pm$ 1.77 ^b	0.02
	2 nd week	4.47 $\pm$ 0.43 ^a	29.37 $\pm$ 8.09 ^b	30.00 $\pm$ 3.86 ^b	28.17 $\pm$ 3.01 ^b	0.01
	3 rd week	4.70 $\pm$ 0.65 ^a	55.00 $\pm$ 6.42 ^c	53.96 $\pm$ 4.35 ^c	41.03 $\pm$ 7.71 ^b	<0.001
Platelets ($10^5/\text{cmm}$)	3 rd day	4.10 $\pm$ 0.10	4.21 $\pm$ 0.23	4.32 $\pm$ 0.23	4.39 $\pm$ 0.19	0.40
	1 st week	4.22 $\pm$ 0.30	4.32 $\pm$ 0.22	5.10 $\pm$ 1.29	5.32 $\pm$ 1.31	0.29
	2 nd week	4.51 $\pm$ 0.31	4.45 $\pm$ 0.83	4.80 $\pm$ 1.05	4.67 $\pm$ 0.91	0.12
	3 rd week	5.27 $\pm$ 1.64	5.06 $\pm$ 1.60	4.94 $\pm$ 1.83	4.12 $\pm$ 0.87	0.21

The values are Mean $\pm$ SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.2.4 Differential Leucocyte Count

The preventive effects of cannabis extract on differential WBC count (EAC induced after 7 doses of cannabis extract) are given in figure no. 57. Significant differences ($p < 0.05$) were observed among the treatment on neutrophils count. Significantly ($p < 0.05$) highest value was found at T1 ($60.00 \pm 2.77\%$) and the lowest value was ($34.00 \pm 1.20\%$). Significant ($p < 0.05$) differences were also observed among the treatment group of lymphocyte count. The highest value at the T0 ($57.00 \pm 1.03\%$) and lowest value at T3 ($38.00 \pm 1.03\%$). No significant differences were observed for monocyte, eosinophil, and basophil count.

Figure 57: Leucocyte count in 14 days of dose applications for cancer prevention.



^{abc} values having different superscripts differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups.

4.3.2.5 Tumor growth Response

The preventive effects of cannabis extract on tumor volume, packed cell volume and tumor cell count (EAC induced after 7 doses of cannabis extract) are given in table no . T1, T2 and T3 group show ($p < 0.05$) significant values. The highest value in T1 was tumor cell volume ($4.60 \pm 0.32 \text{ ml}$) was significantly varied ($p < 0.05$) to the lowest value of tumor-packed cell volume ($1.50 \pm 0.13 \text{ ml}$). The highest value in T2 was tumor cell volume ($4.01 \pm 0.25 \text{ ml}$) was significantly varied ($p < 0.05$) to the lowest value of tumor-packed cell volume ($1.30 \pm 0.11 \text{ ml}$). The highest value in T3 was tumor cell volume ($3.20 \pm 0.16 \text{ ml}$) was significantly varied ($p < 0.05$) to the lowest value of tumor-packed cell volume ($1.01 \pm 0.08 \text{ ml}$).

Table 92: The preventive efficacy of cannabis extract on tumor volume, packed cell volume and tumor cell count (EAC induced after 7 doses of cannabis extract)

Group	Tumor volume (ml)	Packed cell volume (ml)	Tumor cell count $\times 10^7$ cells/ml/mice		P-value
			Viable	Nonviable	
T1	4.60 ± 0.32^b	1.50 ± 0.13^a	2.20 ± 0.14^a	2.11 ± 0.31^a	0.05
T2	4.01 ± 0.25^b	1.30 ± 0.11^a	2.09 ± 0.10^a	2.16 ± 0.32^c	0.05
T3	3.20 ± 0.16^b	1.01 ± 0.08^a	1.61 ± 0.05^a	4.20 ± 0.51^c	0.05

Values= Mean $\pm$ SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.2.6 Hematological study

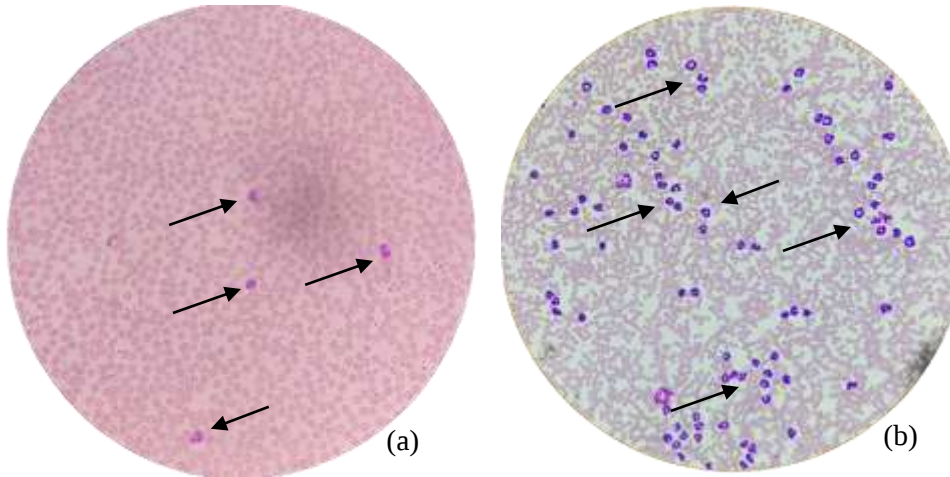


Figure 58: Preventive effects of cannabis extract on WBC count (EAC induced after 7 doses of cannabis extract injection) Hematological slide observation under light microscope 400x view (a) shows the normal WBC counting, (b) shows the increased the number of WBC counting. WBC count increased compare to the normal slide.

4.3.2.7 Cytopathological study of ascitic fluids

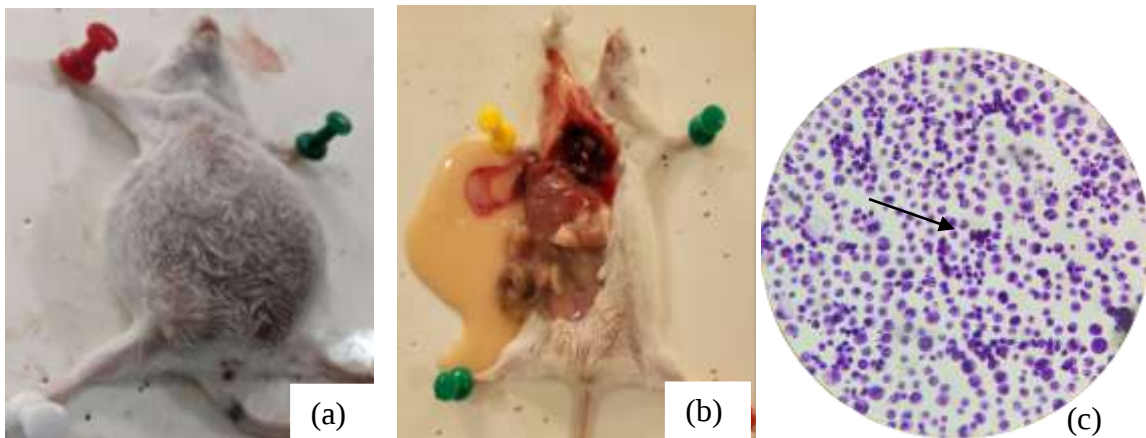


Figure 59: Preventive effects of cannabis extract on cytology of ascitic fluid (EAC induced after 7 doses of cannabis extract injection). For cytological observation (a) EAC inoculated mouse edema formation on the abdominal cavity, (b) mouse sacrificed for fluid collection, (c) cytopathological observation under microscope 400x view. Marking area was indicated the atypical carcinoma type cells.

4.3.2.8 Histopathological study of main organs and tumor

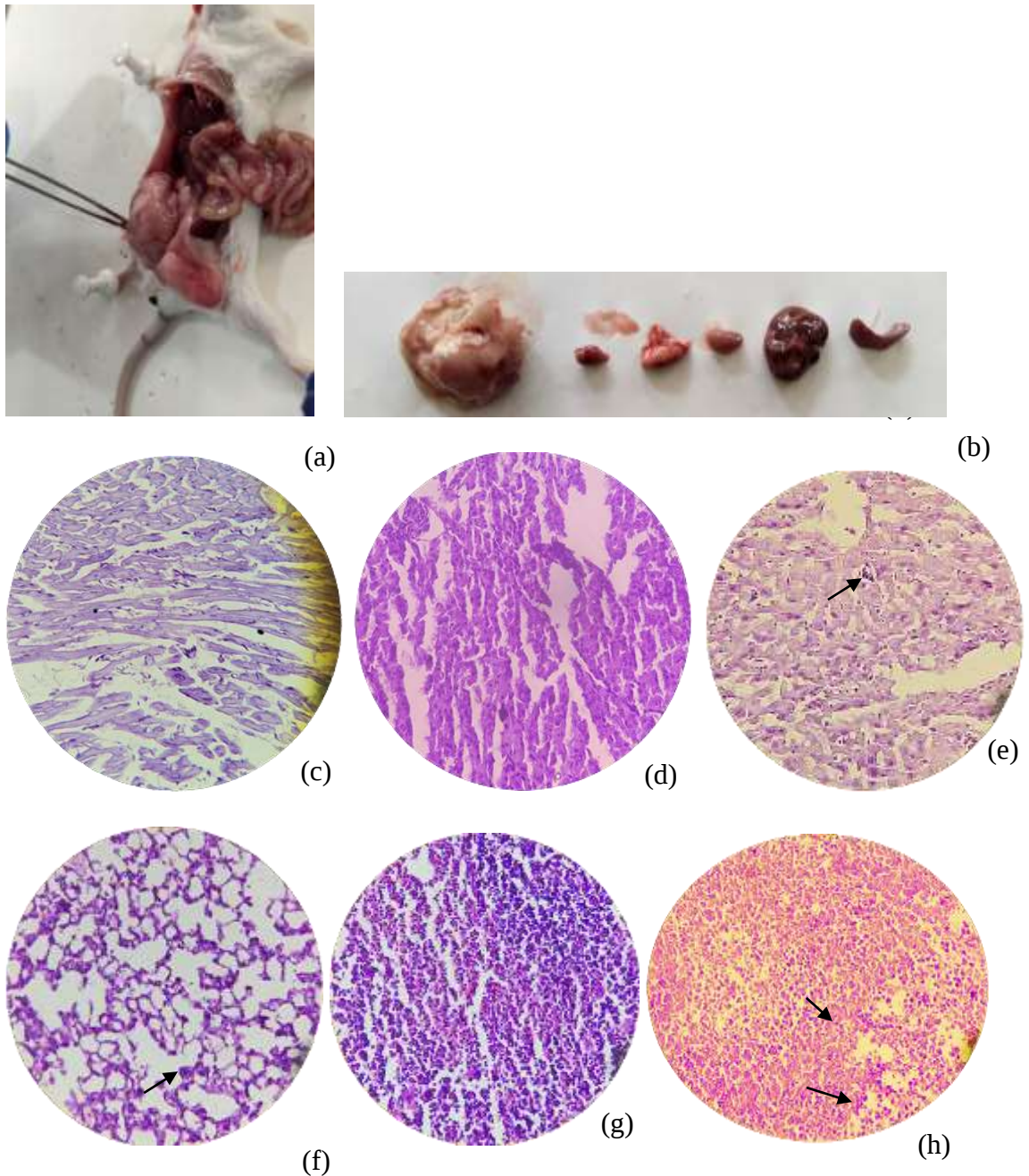


Figure 60: Preventive effects of cannabis extract on cellular changes of main organs and tumor (EAC induced after 7 doses of cannabis extract injection). For histopathological observation (a) Mouse sacrificed for organ separation, tumor indicated, (b) main organ of mice separated, (c) heart, (d) kidney, (e) liver, (f) lung, (g) pancreas, and (h) tumor microscopic observation using H and E dye, 400x view under light microscope. In the image (e) and (h) inflammatory cells.

Group 3: Effectiveness of fourteen doses of cannabis extract in preventing cancer

Three different concentrations of cannabis extract dose were used in this experiment. The doses were 5mg/kg (T1), 10mg/kg (T2), and 15mg/kg (T3) according to the body weight of mice. One dose per day was injected into intraperitoneal space into mice. After fourteen days of treatment, mice were inoculated with cancer cells (EAC). Normal food and water were provided after the inoculation of cancer cells. Mice were not given any other treatment after EAC inoculation. Only normal food and adequate water were provided. After that, the mice were killed after 3 days, 7 days, 14 days, and 21 days respectively for pathological observation in 3 replicative groups.

4.3.3.1 Body weight measurement of albino mice

The preventive effects of cannabis extract on body weight (EAC induced after 14 doses of cannabis extract injection) are given in table no. 93. No significant variations ($p < 0.05$) were observed among the treatment groups on the 3rd day, 1st week, 2nd week, and 3rd week of mice respectively.

Table 93: Preventive efficacy of cannabis extract on body weight (EAC induced after 14 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (g.)	T1 (g.)	T2 (g.)	T3 (g.)	
3 rd day	22.17±0.55	21.62±0.55	24.17±1.03	21.01±0.29	0.12
1 st week	26.13±1.09	24.80±1.02	28.10±2.04	26.34±1.90	0.41
2 nd week	29.39±1.19	27.90±1.06	30.10±2.01	27.99±1.01	0.49
3 rd week	33.70±1.09	32.03±1.57	36.71±1.87	36.08±1.34	0.31

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.3.2 Biochemical profile

4.3.3.2.1 RBS

The preventive effects of cannabis extract on RBS (EAC induced after 14 doses of cannabis extract injection) are given in table no. 94. No significant variations ($p < 0.05$) were observed among the treatment groups on the 3rd day, 1st week, 2nd week, and 3rd week of mice respectively. The highest result was T3 ($6.70 \pm 0.82 \text{ mmolL}^{-1}$) on the 3rd day and the lowest result was T0 ($4.21 \pm 0.20 \text{ mmolL}^{-1}$) on the 3rd day of observation.

Table 94: Preventive efficacy of cannabis extract on RBS (EAC induced after 14 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mmolL^{-1})	T1 (mmolL^{-1})	T2 (mmolL^{-1})	T3 (mmolL^{-1})	
3 rd day	4.21 ± 0.20	5.27 ± 0.14	5.84 ± 0.50	6.70 ± 0.82	0.11
1 st week	5.73 ± 0.24	6.25 ± 0.15	5.20 ± 0.58	5.23 ± 0.55	0.23
2 nd week	5.50 ± 0.72	6.04 ± 0.87	5.65 ± 0.31	6.03 ± 0.37	0.88
3 rd week	5.46 ± 0.48	6.33 ± 0.87	5.28 ± 0.66	5.36 ± 0.35	0.67

The values are Mean $\pm$ SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.3.2.2 Serum creatinine

The preventive effects of cannabis extract on serum creatinine (EAC induced after 14 doses of cannabis extract injection) are given in table no. 95. No significant variations ($p < 0.05$) were observed among the treatment groups on the 3rd day, 1st week, 2nd week, and 3rd week of mice respectively. The highest result was T1 ($0.49 \pm 0.38 \text{ mg/dl}$) on the 1st week and the lowest result was T3 ($0.11 \pm 0.33 \text{ mg/dl}$) on the 3rd day of observation.

Table 95: Preventive efficacy of cannabis extract on serum creatinine (EAC induced after 14 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	0.33±0.16	0.16±0.02	0.25±0.02	0.11±0.33	0.45
1 st week	0.15±0.01	0.49±0.38	0.21±0.02	0.27±0.05	0.06
2 nd week	0.22±0.04	0.31±0.03	0.28±0.03	0.25±0.13	0.64
3 rd week	0.30±0.03	0.13±0.04	0.23±0.02	0.35±0.14	0.18

The values are Mean±SE, T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.3.2.3 SGPT

The preventive effects of cannabis extract on SGPT (EAC induced after 14 doses of cannabis extract injection) are given in table no. 96. In the 3rd week of observation SGPT test result was differed significantly ($P<0.05$) among the different treatment groups of mice. Lowest result of SGPT in T0 (27.33 ± 3.71 U/L) was significantly varied ($p<0.05$) to the highest result in T1 (235.33 ± 27.33 U/L).

Table 96: Preventive efficacy of cannabis extract on SGPT (EAC induced after 14 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (U/L)	T1 (U/L)	T2 (U/L)	T3 (U/L)	
3 rd day	24.33±2.84	43.33±3.33	51.00±7.02	26.33±5.17	0.22
1 st week	39.67±1.33 ^a	52.33±11.05 ^b	80.67±13.22 ^b	146.67±16.04 ^c	0.02
2 nd week	35.00±3.51	81.33±4.21	98.00±8.51	103.67±11.83	0.10
3 rd week	27.33±3.71 ^a	235.33±27.33 ^c	189.33±16.12 ^b	279.33±20.81 ^b	0.05

The values are Mean±SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.3.2.4 SGOT

The preventive effects of cannabis extract on SGPT (EAC induced after 14 doses of cannabis extract injection) are given in table no. 97. In the 1st week of observation SGOT test result was differed significantly ($P=0.001$) among the different treatment groups of mice. Lowest result of SGOT in T0 (32.00 ± 7.00 U/L) was significantly varied ($p<0.05$) to the highest result in T1 (437.00 ± 60.65 U/L). In the 2nd week of observation SGOT test result was differed significantly ($P=0.004$) among the different treatment groups of mice. Lowest result of SGOT in T0 (30.67 ± 1.86 U/L) was significantly varied ($p=0.004$) to the highest result in T1 (770.00 ± 108.00 U/L). In the 3rd week of observation SGOT test result was differed significantly ($P<0.001$) among the different treatment groups of mice. Lowest result of SGOT in T0 (34.00 ± 3.06 U/L) was significantly varied ($p<0.001$) to the highest result in T1 (2428.00 ± 415.86 U/L).

Table 97: Preventive efficacy of cannabis extract on SGOT (EAC induced after 14 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0	T1	T2	T3	
3 rd day	24.33 ± 8.82^a	156.00 ± 71.24^b	54.33 ± 21.84^a	30.00 ± 3.61^a	0.25
1 st week	32.00 ± 7.00^a	437.00 ± 122.65^b	410.67 ± 92.48^b	166.33 ± 12.99^a	0.001
2 nd week	30.67 ± 6.86^a	770.00 ± 108.00^b	547.33 ± 77.49^b	561.00 ± 146.8^b	0.004
3 rd week	34.00 ± 3.06^a	2428.00 ± 415.86^c	777.33 ± 60.07^b	698.00 ± 39.07^{ab}	<0.001

The values are Mean $\pm$ SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.3.2.5 Alkaline phosphatase

The preventive effects of cannabis extract on alkaline phosphatase (EAC induced after 14 doses of cannabis extract injection) are given in table no. 98. Alkaline phosphatase test result was differed significantly ($P<0.05$) among the different treatment groups of mice in 1st week of observation. Lowest result of alkaline phosphate in T0 (52.00 ± 19.09 U/L) at 1st week was significantly varied ($p=0.001$) to the highest result in T2 (545.67 ± 110.18 U/L).

There no significant variation observed among the treatment groups of T1, T2, and T3. In the 2nd week of observation alkaline phosphate test result was differed significantly (P=0.007) among the different treatment groups of mice. Lowest result of alkaline phosphatase in T0 (67.67± 14.50U/L) was significantly varied (p=0.007) to the highest result in T2 (772.00± 115.69U/L). In the 3rd week of observation alkaline phosphatase test result was differed significantly (P<0.05) among the different treatment groups of mice. Lowest result of alkaline phosphate in T0 (73.33± 25.31U/L) was significantly varied (p<0.05) to the highest result in T2 (1473.33± 525.18U/L).

Table 98: Preventive efficacy of cannabis extract on alkaline phosphatase (EAC induced after 14 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (U/L)	T1 (U/L)	T2 (U/L)	T3 (U/L)	
3 rd day	73.33±28.39	170.00± 34.27	66.33±16.42	238.33±160.95	0.46
1 st week	52.00±19.09 ^a	402.67± 91.90 ^b	545.67± 110.18 ^b	465.67±102.64 ^b	0.03
2 nd week	67.67±14.50 ^a	736.33± 103.55 ^b	772.00± 115.69 ^b	409.67±64.63 ^{ab}	0.007
3 rd week	73.33±25.31 ^a	1473.33±525.18 ^b	965.67±188.57 ^{ab}	705.00±142.72 ^{ab}	0.03

The values are Mean±SE, ^{ab} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.3.2.6 Serum Cholesterol

The preventive effects of cannabis extract on serum cholesterol (EAC induced after 14 doses of cannabis extract injection) are given in table no. 99. No significant variations (p<0.05) were observed among the treatment groups on the 3rd day, 1st week, 2nd week, and 3rd week of mice respectively. The highest result of serum cholesterol was T1 (160.33± 8.50mg/dl) at 1st week of treatment and the lowest result was T3 (104.90± 8.51mg/dl) at 3rd day of observation.

Table 99: Preventive efficacy of cannabis extract on serum cholesterol (EAC induced after 14 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	137.07± 11.47	126.67± 10.99	119.33± 10.23	104.90± 8.51	0.12
1 st week	123.33± 13.32	160.33± 18.50	136.60± 12.89	119.22±11.10	0.09
2 nd week	132.21± 12.50	122.63± 9.95	131.33± 11.07	123.20±10.52	0.22
3 rd week	142.00± 12.50	146.67± 12.19	137.00± 11.64	106.00± 8.02	0.14

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, NS= non-significant, p-value= probability value.

4.3.3.2.7 Serum Triglyceride

The preventive effects of cannabis extract on serum triglyceride (EAC induced after 14 doses of cannabis extract injection) are given in table no.100. No significant variations ($p<0.05$) were observed among the treatment groups on the 3rd day, 1st week, 2nd week, and 3rd week of mice respectively. The highest result of serum triglyceride was T3 (169.03± 20.1mg/dl) at 1st week of treatment and the lowest result was T0 (103.33± 9.92mg/dl) at 3rd week of observation.

Table 100: Preventive efficacy of cannabis extract on serum triglyceride (EAC induced after 14 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	142.00±1.51	158.31± 12.41	123.30±11.80	138.33±11.81	0.65
1 st week	183.03±15.3	165.33±21.50	143.61±15.89	169.03± 20.1	0.16
2 nd week	154.30± 8.47	172.61±12.98	156.32±15.10	159.03±18.03	0.87
3 rd week	103.33± 9.92	135.33±12.12	138.00±11.10	148.32±14.13	0.12

The values are Mean±SE, T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.3.2.8 Serum uric acid

The preventive effects of cannabis extract on serum uric acid (EAC induced after 14 doses of cannabis extract injection) are given in table no. 101. In the 3rd day of observation uric acid test result was differed significantly ($P<0.05$) among the different treatment groups of mice. Lowest result of uric acid in T0 (4.70 ± 0.34 mg/dl) was significantly varied ($p<0.05$) to the highest result in T1 (8.29 ± 0.82 mg/dl). In the 2nd week of observation uric acid test result was differed significantly ($P<0.05$) among the different treatment groups of mice. Lowest result of uric acid in T2 (6.57 ± 0.07 mg/dl) was significantly varied ($p<0.05$) to the highest result in T1 (16.03 ± 4.18 mg/dl).

Table 101: Preventive efficacy of cannabis extract on serum uric acid (EAC induced after 14 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	6.10 ± 0.52^a	8.29 ± 0.82^b	9.33 ± 0.30^b	4.70 ± 0.34^b	0.02
1 st week	6.31 ± 0.80	7.72 ± 0.72	9.10 ± 1.62	9.81 ± 3.10	0.75
2 nd week	6.89 ± 0.77^a	9.10 ± 0.58^a	6.57 ± 0.07^{ab}	16.03 ± 4.18^b	0.04
3 rd week	7.10 ± 1.14	6.20 ± 1.05	9.60 ± 1.83	11.60 ± 5.21	0.48

The values are Mean $\pm$ SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.3.3 Hematological test

The hematological tests were Hb, red blood cells count, white blood cells count, differential white blood cells count, and platelets count.

4.3.2.3.1 Hb

The preventive effects of cannabis extract on Hb (EAC induced after 14 doses of cannabis extract injection) is given in table no. 102. Significant variation ($p<0.05$) observed among the treatment groups of 2nd week of observation. The highest values of hemoglobin was

significantly varied ($p<0.05$) at T3 ($14.88\pm0.34\text{mg/dl}$) to the lowest value was ($13.23\pm0.54\text{mg/dl}$) at T0 group of observation.

4.3.2.3.2 RBC count

The preventive effects of cannabis extract on RBC count (EAC induced after 7 doses of cannabis extract injection) is given in table no. 102. The RBC count test result was significantly ($p<0.05$) different among the treatment groups at 1st week of observation. The highest values of RBC was significantly varied ($p<0.05$) at T1 ($5.01\pm0.01\times10^6/\text{cmm}$) to the lowest value ($4.07\pm0.17\times10^6/\text{cmm}$).

4.3.2.3.3 WBC count

The preventive effects of cannabis extract on WBC count (EAC induced after 7 doses of cannabis extract injection) is given in table no. 102. No significant difference observed ($p<0.05$) among the treatment groups at the 3rd day, 1st week, 2nd week and 3rd week respectively.

4.3.2.3.4 Platelet count

The preventive effects of cannabis extract on platelet count (EAC induced after 7 doses of cannabis extract injection) is given in table no. 102. The platelet count test result was no significantly ($p<0.05$) different among the treatment groups at the 3rd day, 1st week, and 2nd week and 3rd week respectively. The highest values of platelet was not significantly varied ($p<0.05$) at T3 ($5.32\pm1.31\times10^5/\text{cmm}$) in the 1st week and the lowest value was ($4.10\pm0.10\times10^5/\text{cmm}$) in the 3rd day of T0 group of observation.

Table 102: Preventive efficacy of cannabis extract on hematological profile (EAC induced after 14 doses of cannabis extract injection)

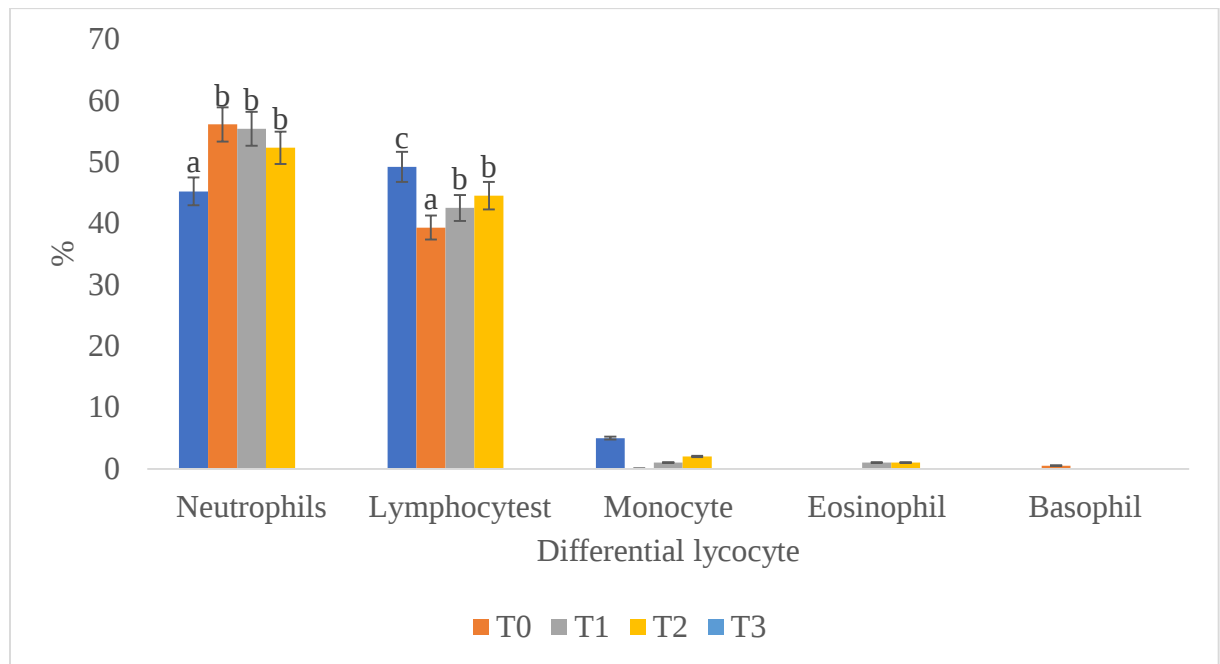
Test name	Time interval	Treatment groups				P-value
		T0	T1	T2	T3	
Hb(mg/dl)	3 days	14.13±0.62	13.88±0.54	14.07±0.33	14.13±0.87	0.11
	1 st week	14.37±1.23	13.90±0.23	13.27±0.74	14.21±0.32	0.19
	2 nd week	13.23±0.54 ^a	14.06±0.39 ^b	14.82±0.34 ^b	14.88±0.34 ^b	0.05
	3 rd week	14.67±0.31	14.13±0.60	13.87±0.31	14.81±0.08	0.24
RBC (10 ⁶ /cmm)	3 days	4.73±0.25	4.26±0.22	4.22±0.21	4.06±0.20	0.48
	1 st week	4.17±0.33 ^a	4.23±0.17 ^b	3.22±0.35 ^b	4.61±0.42 ^b	0.05
	2 nd week	4.30±0.32	5.20±0.02	4.23±0.43	5.01±0.17	0.28
	3 rd week	4.17±0.13 ^a	5.01±0.01 ^b	4.74±0.03 ^b	4.07±0.17 ^b	0.01
WBC (10 ³ /cmm)	3 days	4.80±1.04	5.27±0.38	5.77±0.50	8.17±1.92	0.44
	1 st week	5.03±0.27	9.80±0.26	9.63±0.64	6.60±0.70	0.45
	2 nd week	5.23±0.12	9.67±0.77	6.73±0.64	7.17±1.42	0.08
	3 rd week	6.17±0.16	5.03±2.09	9.73±0.90	7.80±1.10	0.48
Platelets (10 ⁵ /cmm)	3 days	5.01±0.15	4.59±0.23	4.40±0.20	4.53±0.29	0.40
	1 st week	4.21±0.32	4.05±0.51	5.10±1.29	5.02±1.03	0.54
	2 nd week	3.51±0.31	3.45±1.20	4.80±1.05	5.67±0.91	0.32
	3 rd week	5.77±1.14	5.26±1.22	5.04±1.03	4.12±0.87	0.29

The values are Mean±SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.3.4 Differential leucocyte count

The preventive effects of cannabis extract on differential WBC count (EAC induced after 14 doses of cannabis extract) are given in figure no. 61. No Significant differences ($p < 0.05$) were observed among the treatment on neutrophils count, lymphocyte, monocyte, eosinophil and basophil respectively. The highest value was found at T0 ($56.5 \pm 2.33\%$) and the lowest value was at T3 ($45.2 \pm 1.20\%$). The highest value at the T3 ($49.2 \pm 1.23\%$) and lowest value at T0 ($39.40 \pm 1.02\%$).

Figure 61: Leucocyte count in 14 days of dose applications for cancer prevention



^{abc} values having different superscripts differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups.

4.3.3.5 Tumor growth responses

The preventive effects of cannabis extract on tumor volume, packed cell volume and tumor cell count (EAC induced after 14 doses of cannabis extract) are given in table no . T1, T2 and T3 group show ($p<0.05$) significant values. The highest value in T1 was nonviable cell volume ($4.33\pm0.32\times10^7$ cells/ml) was significantly varied ($p<0.05$) to the lowest value of tumor-packed cell volume (0.77 ± 0.13 ml). The highest value in T2 was nonviable cell ($5.16\pm0.54\times10^7$ cells/ml) was significantly varied ($p<0.05$) to the lowest value of tumor-packed cell volume (0.72 ± 0.11 ml). The highest value in T3 was nonviable cell ($4.20\pm0.51\times10^7$ cells/ml) was significantly varied ($p<0.05$) to the lowest value of tumor-packed cell volume (0.41 ± 0.08 ml).

Table 103: The preventive efficacy of cannabis extract on tumor volume, packed cell volume and tumor cell count (EAC induced after 14 doses of cannabis extract)

Group	Tumor volume (ml)	Packed cell volume (ml)	Tumor cell count $\times 10^7$ cells/ml/mice		P-value
			Viable	Nonviable	
T1	1.22 ± 0.02^b	0.77 ± 0.13^a	1.11 ± 0.04^b	4.33 ± 0.32^c	0.05
T2	2.11 ± 0.25^b	0.72 ± 0.11^a	1.09 ± 0.03^a	5.16 ± 0.54^c	0.05
T3	1.20 ± 0.06^b	0.41 ± 0.08^a	0.61 ± 0.01^a	4.20 ± 0.51^c	0.05

Values= Mean $\pm$ SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, P-value= probability value.

4.3.3.6 Hematological study

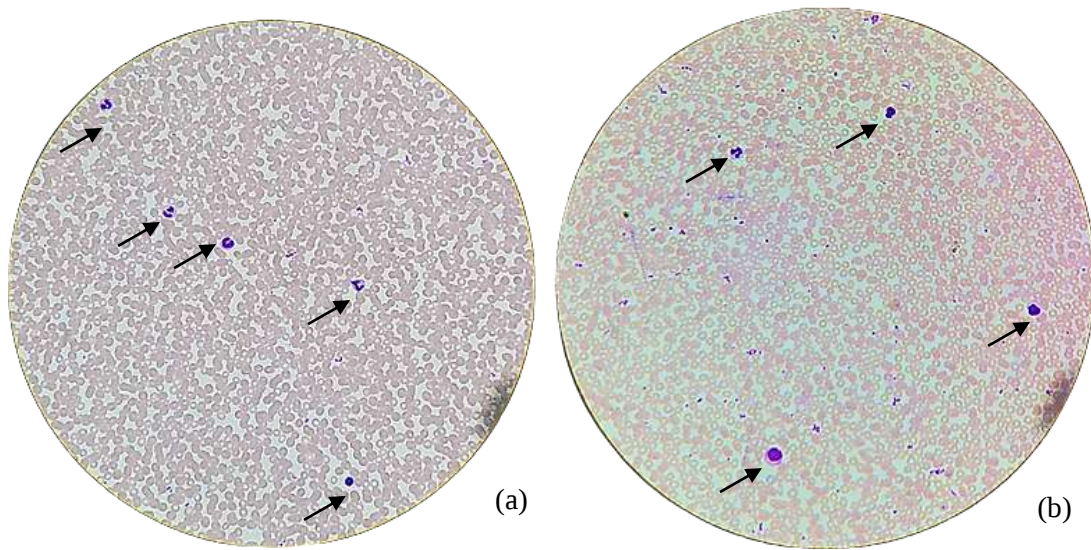


Figure 62: Preventive effects of cannabis extract on WBC count (EAC induced after 14 doses of cannabis extract injection). Image shows the slide of (a) Normal mice and (b) 14 dose's injection mice. Both slide shows similar observation of WBC count.

Group 4: Effectiveness of twenty one doses of cannabis extract in preventing cancer.

Three different concentrations of cannabis extract dose were used in this experiment. The doses were 5mg/kg (T1), 10mg/kg (T2), and 15mg/kg (T3) according to the body weight of mice. One dose per day was injected into intraperitoneal space into mice. After twenty-one days of treatment, mice were inoculated with cancer cells (EAC). Normal food and water were provided after the inoculation of cancer cells. Mice were not given any other treatment after EAC inoculation. Only normal food and adequate water were provided. After that, the mice were killed after 3 days, 7 days, 14 days, and 21 days respectively for pathological observation in 3 replicative groups.

4.3.4.1 Weight measurement of albino mice

The preventive effects of cannabis extract on body weight (EAC induced after 21 doses of cannabis extract injection) is given in table no. 104. There were no significant variation ($p < 0.05$) observed among the treatment groups at the 3rd day, 1st week, 2nd week, and 3rd week respectively.

Table 104: Preventive efficacy of cannabis extract on body weight (EAC induced after 21 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0	T1	T2	T3	
3 rd day	22.67±1.21	20.60±1.01	24.12±1.39	23.22±1.22	0.32
1 st week	25.14±1.51	24.80±1.15	29.11±1.55	25.22±1.19	0.44
2 nd week	30.54±1.58	29.43±1.57	34.77±1.72	29.09±1.48	0.20
3 rd week	36.55±1.60	35.22±1.54	40.32±2.05	36.04±1.95	0.65

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.4.2 Biochemical profile

4.3.4.2.1 RBS

The preventive effects of cannabis extract on RBS were given in table no. 105. No significant variations ($p < 0.05$) were observed among the treatment groups on the 3rd day, 1st week, 2nd week, and 3rd week of mice respectively. The highest result was T1 ($8.33 \pm 0.47 \text{ mmolL}^{-1}$) on the 3rd week and the lowest result was T0 ($4.40 \pm 0.40 \text{ mmolL}^{-1}$) on the 3rd week of observation.

Table 105: Preventive efficacy of cannabis extract on RBS (EAC induced after 21 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mmolL^{-1})	T1 (mmolL^{-1})	T2 (mmolL^{-1})	T3 (mmolL^{-1})	
3 rd day	6.21 ± 0.23	7.07 ± 0.14	5.83 ± 0.51	7.70 ± 0.82	0.26
1 st week	5.71 ± 0.24	6.05 ± 0.60	6.20 ± 0.38	6.63 ± 0.65	0.49
2 nd week	5.50 ± 0.72	5.84 ± 0.87	5.60 ± 0.30	5.33 ± 0.30	0.23
3 rd week	4.40 ± 0.40	8.33 ± 0.47	6.23 ± 0.65	8.30 ± 0.35	0.17

The values are Mean $\pm$ SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment group. SE=Standard error of the mean, p-value= probability value.

4.3.4.2.2 Serum creatinine

The preventive effects of cannabis extract on Serum creatinine were given in table no. 106. There were no significant variation ($p < 0.05$) observed among the treatment groups at the 3rd day, 1st week, 2nd week, and 3rd week respectively. The highest result was T1 ($0.39 \pm 0.08 \text{ mg/dl}$) at 1st week was not significantly ($p < 0.05$) varied to the lowest result T1 ($0.17 \pm 0.02 \text{ mg/dl}$) at the 3rd day of observation.

Table 106: Preventive efficacy of cannabis extract on serum creatinine (EAC induced after 21 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	0.23±0.06	0.17±0.03	0.21±0.03	0.17±0.03	0.75
1 st week	0.18±0.04	0.39±0.08	0.27±0.03	0.23±0.03	0.29
2 nd week	0.24±0.04	0.34±0.03	0.23±0.03	0.26±0.03	0.24
3 rd week	0.30±0.05	0.13±0.03	0.33±0.06	0.30±0.04	0.18

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.4.2.3 SGPT

The preventive effects of cannabis extract on SGPT were given in table no. 107. There were no significant variation ($p < 0.05$) observed among the treatment groups at the 3rd day, 1st week, 2nd week, and 3rd week respectively. The highest result was T3 (61.33±6.22U/L) at 3rd week the lowest result was T0 (24.00±3.73U/L) at the 1st week of observation.

Table 107: Preventive efficacy of cannabis extract on SGPT (EAC induced after 21 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (U/L)	T1 (U/L)	T2 (U/L)	T3 (U/L)	
3 rd day	31.00±5.51	33.67±5.66	41.00±6.20	43.33±6.87	0.69
1 st week	24.00±3.73	35.33±5.93	56.33±6.45	57.67±6.36	0.33
2 nd week	29.00±3.02	37.67±5.05	58.00±6.05	57.67±8.76	0.19
3 rd week	24.67±3.73	47.33±6.63	55.67±5.84	61.33±6.22	0.32

The values are Mean±SE, T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.4.2.4 SGOT

The preventive effects of cannabis extract on SGOT were given in table no. 108. There were no significant variation ($p < 0.05$) observed among the treatment groups at the 3rd day, 1st week, 2nd week, and 3rd week respectively. The highest result was T2 (105.67 ± 10.76 U/L) at 2nd week the lowest result was T0 (26.67 ± 3.18 U/L) at the 2nd week of observation.

Table 108: Preventive efficacy of cannabis extract on SGOT (EAC induced after 21 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0	T1	T2	T3	
3 rd day	31.67 ± 3.81	73.00 ± 7.04	29.00 ± 2.07	26.67 ± 1.42	0.14
1 st week	30.67 ± 3.09	52.67 ± 5.14	51.00 ± 5.09	63.33 ± 6.2	0.22
2 nd week	26.67 ± 3.18	96.67 ± 9.44	105.67 ± 10.76	83.67 ± 8.36	0.25
3 rd week	38.67 ± 2.96	71.00 ± 7.33	83.00 ± 8.49	99.33 ± 9.83	0.35

The values are Mean $\pm$ SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.4.2.5 Alkaline phosphatase

The preventive effects of cannabis extract on alkaline phosphatase (EAC induced after 21 doses of cannabis extract injection) is given in table no. 109. There were no significant variation ($p < 0.05$) observed among the treatment groups at the 3rd day, 1st week, 2nd week, and 3rd week respectively. The highest result was T0 (115.00 ± 6.56 U/L) at 3rd week the lowest result was T0 (58.33 ± 4.67 U/L) at the 2nd week of observation.

Table 109: Preventive efficacy of cannabis extract on alkaline phosphate (EAC induced after 21 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (U/L)	T1 (U/L)	T2 (U/L)	T3 (U/L)	
3 rd day	105.67±10.87	111.33±10.73	80.33±9.36	58.33±4.67	0.35
1 st week	52.00±19.09	89.67±7.69	99.67±7.73	65.00±5.24	0.21
2 nd week	79.33±11.79	77.00±6.56	77.00±6.74	68.33±5.82	0.51
3 rd week	115.00±6.56	75.33±5.70	99.33±7.03	88.00±6.43	0.44

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.4.2.6 Serum cholesterol

The preventive effects of cannabis extract on serum cholesterol (EAC induced after 21 doses of cannabis extract injection) is given in table no. 110. There were no significant variation ($p < 0.05$) observed among the treatment groups at the 3rd day, 1st week, 2nd week, and 3rd week respectively. The highest result was T1 (162.33±12.50mg/dl) at 1st week the lowest result was T3 (122.20±3.12mg/dl) at the 2nd week of observation.

Table 110: Preventive efficacy of cannabis extract on serum cholesterol (EAC induced after 21 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	151.61±11.47	146.67±11.99	129.03±12.23	124.43±4.51	0.42
1 st week	143.33±10.12	162.33±12.50	132.62±12.89	139.21±12.10	0.09
2 nd week	131.17±12.51	138.67±3.95	128.33±10.07	133.02 ±1.52	0.08
3 rd week	152.02±11.50	136.67±12.19	132.01±13.64	122.20±3.12	0.54

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.4.2.7 Serum Triglyceride

The preventive effects of cannabis extract on serum triglyceride (EAC induced after 21 doses of cannabis extract injection) is given in table no. 111. There were significant variation ($p<0.05$) observed among the treatment groups at 3rd week of observation. The highest result was T1 ($175.33\pm15.17\text{mg/dl}$) at 3rd week was significantly varied ($p<0.05$) to the lowest result was T2 ($153.20\pm13.00\text{mg/dl}$).

Table 111: Preventive efficacy of cannabis extract on serum triglyceride (EAC induced after 21 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	166.01 $\pm$ 12.51	158.33 $\pm$ 12.40	143.31 $\pm$ 13.80	136.33 $\pm$ 12.84	0.34
1 st week	163.03 $\pm$ 4.32	164.33 $\pm$ 11.50	156.60 $\pm$ 15.09	149.00 $\pm$ 12.10	0.36
2 nd week	133.31 $\pm$ 6.47	177.07 $\pm$ 12.98	141.03 $\pm$ 15.10	152.33 $\pm$ 18.83	0.10
3 rd week	163.30 $\pm$ 9.90 ^{bc}	175.33 $\pm$ 15.17 ^c	153.20 $\pm$ 13.00 ^a	157.13 $\pm$ 15.04 ^b	0.02

The values are Mean $\pm$ SE, and ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice. SE=Standard error of the mean, p-value = probability value.

4.3.4.2.8 Serum uric acid

The preventive effects of cannabis extract on serum uric acid (EAC induced after 21 doses of cannabis extract injection) is given in table no. 112. There were no significant variation ($p<0.05$) observed among the treatment groups at the 3rd day, 1st week, 2nd week, and 3rd week respectively. The highest result was T1 ($9.60\pm2.13\text{ mg/dl}$) at 1st week the lowest result was T3 ($4.50\pm0.55\text{mg/dl}$) at the 2nd week of observation.

Table 112: Preventive efficacy of cannabis extract on uric acid (EAC induced after 21 doses of cannabis extract injection)

Time interval	Treatment groups				P-value
	T0 (mg/dl)	T1 (mg/dl)	T2 (mg/dl)	T3 (mg/dl)	
3 rd day	4.50±0.55	8.23±0.71	8.03±0.35	8.73±0.33	0.32
1 st week	5.30±0.81	6.73±0.73	9.10±1.63	9.07±1.30	0.54
2 nd week	6.80±0.70	5.10±1.08	9.52±1.33	8.62±1.88	0.25
3 rd week	7.17±1.04	6.06±1.05	9.60±2.13	8.07±1.82	0.27

The values are Mean±SE. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.4.3 Hematological Profile

4.3.1.3.1 Hb

The preventive effects of cannabis extract on Hb (EAC induced after 21 doses of cannabis extract) are given in table no. 113. No significant ($p<0.05$) variation was observed among the treatment groups on the 3rd day, 1st week, 2nd week, and 3rd week respectively. The highest value of hemoglobin was not significantly varied ($p<0.05$) at T0 (14.97±0.03mg/dl) in the 1st-week and the lowest value was (13.60±0.31mg/dl) in the 3rd day of the T0 group of mice.

4.3.1.3.2 RBC count

The preventive effects of cannabis extract on Hb (EAC induced after 21 doses of cannabis extract) are given in table no. 113. The RBC test result was significantly ($p<0.05$) different among the treatment groups on the 3rd week of observation. The highest value of RBC was significantly varied ($p<0.05$) at T1 ($5.12\pm0.21\times10^6/\text{cmm}$) in the 3rd week and the lowest value was at T3 ($4.17\pm0.19\times10^6/\text{cmm}$) group of mice.

4.3.1.3.3 WBC count

The preventive effects of cannabis extract on WBC count (EAC induced after 21 doses of cannabis extract) are given in table no. 113. No significant differences ($p<0.01$) were observed among the treatment groups at 3rd day, 1st week, 2nd week and 3rd week respectively. The highest in T1 ($9.07\pm4.98\times10^3/\text{cmm}$) at 2nd week and lowest in T0 ($3.70\pm0.40\times10^3/\text{cmm}$) at 3rd day.

4.3.1.3.4 Platelet count

The preventive effects of cannabis extract on platelet count (EAC induced after 21 doses of cannabis extract) are given in table no. 113. There were no significant variation ($p < 0.05$) observed in all treatment groups of mice at the 3rd day, 1st week, 2nd week and 3rd week respectively. The highest result at 3rd-week observation T1 ($4.50 \pm 0.50 \times 10^5/\text{cmm}$) of platelet count was not significantly ($p < 0.05$) varied to the lowest result at 2nd week T0 ($2.96 \pm 0.04 \times 10^3/\text{cmm}$) group.

Table 113: Preventive efficacy of cannabis extract on hematological profile (EAC induced after 21 doses of cannabis extract injection)

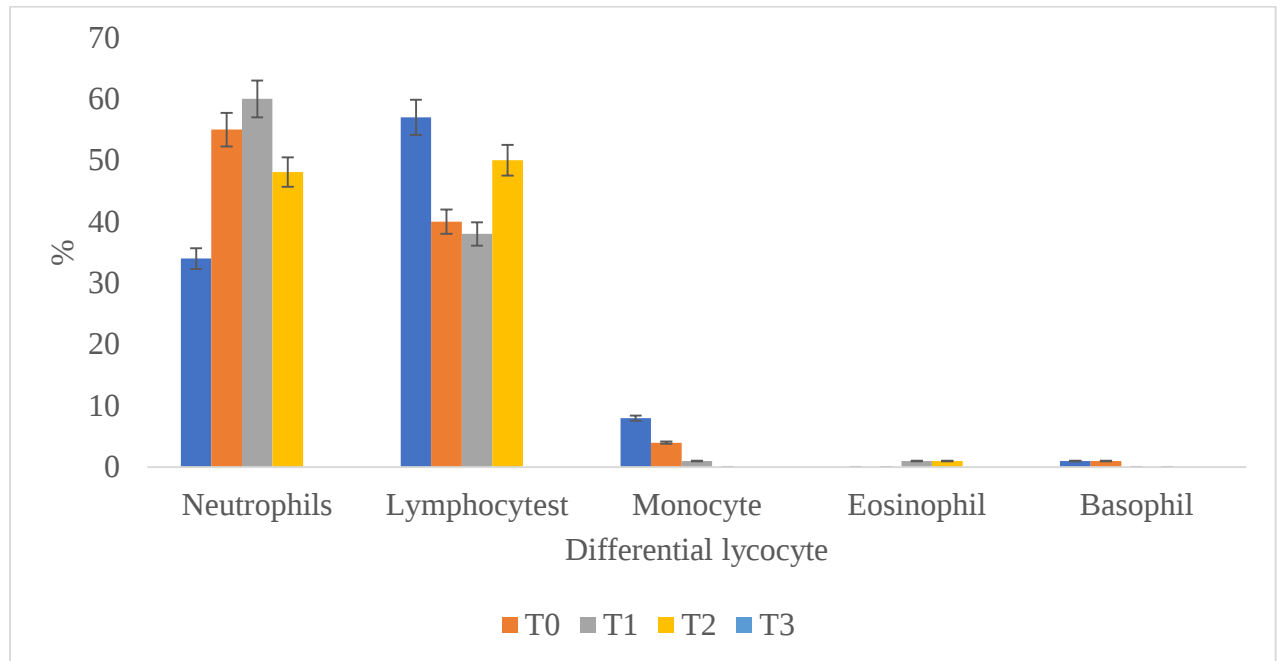
Test name	Time interval	Treatment groups				P-value
		T0	T1	T2	T3	
Hb (mg/dl)	3 rd day	14.13±0.22	13.60±0.31	14.22±0.62	14.14±0.61	0.54
	1 st week	14.97±0.03	13.96±0.13	14.97±0.14	14.31±0.44	0.10
	2 nd week	13.33±0.53	14.71 ± 0.32	14.71±0.32	14.60±0.30	0.23
	3 rd week	13.17±0.32	14.10±0.52	14.13±0.32	14.80±0.13	0.24
RBC ($10^6/\text{cmm}$)	3 rd day	4.13±0.15	4.23±0.21	4.65±0.21	4.77±0.31	0.34
	1 st week	4.61±0.30	4.23±0.21	3.62±0.32	4.60±0.33	0.14
	2 nd week	4.47±0.14	5.22±0.21	4.13±0.22	4.33±0.04	0.64
	3 rd week	4.87±0.11 ^b	5.12±0.21 ^c	4.97±0.22 ^b	4.17±0.19 ^a	0.02
WBC ($10^3/\text{cmm}$)	3 rd day	5.03±0.89	3.70±0.40	4.57±0.15	3.33±0.28	0.15
	1 st week	4.87±0.33	6.23±2.41	8.53±0.27	4.77±1.04	0.08
	2 nd week	4.47±0.29	9.07±4.98	8.13±3.59	4.50±1.20	0.09
	3 rd week	5.37±0.45	7.87±7.19	7.10±5.64	5.53±1.32	0.34
Platelet ($10^5/\text{cmm}$)	3 rd day	3.20±0.41	3.73±0.26	3.10±0.11	3.26±0.35	0.52
	1 st week	3.36±0.32	3.46±0.29	2.99±0.03	3.33±0.34	0.66
	2 nd week	3.25±0.90	3.74±0.26	3.67±0.33	2.96±0.04	0.67
	3 rd week	3.27±0.27	4.50±0.50	3.67±0.66	3.33±0.33	0.29

The values are Mean±SE, ^{abc} values having different superscripts in the same row differed significantly. T0, T1, T2, and T3 indicate the control, 5mg/kg, 10mg/kg, and 15mg/kg by body weight of mice treatment groups. SE=Standard error of the mean, p-value= probability value.

4.3.4.3.5 Differential leucocyte count

The preventive effects of cannabis extract on differential WBC count (EAC induced after 21 doses of cannabis extract) are given in figure no. 63. No Significant differences ($p < 0.05$) were observed among the treatment on neutrophils count, lymphocyte, monocyte, eosinophil and basophil respectively.

Figure 63: Leucocyte count in 21 days of dose applications for cancer prevention.



4.3.4.3.6 Hematological study

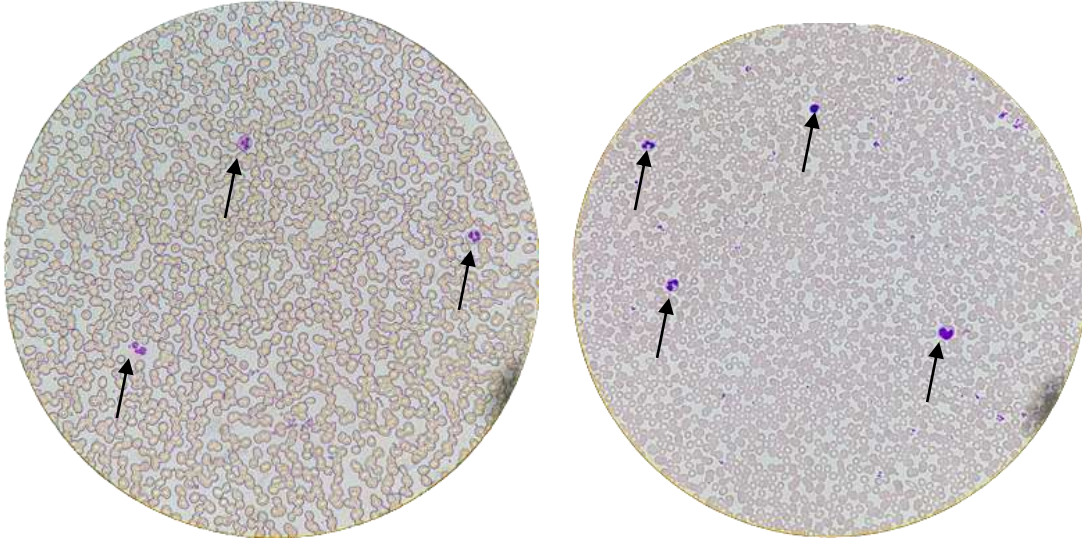


Figure 64: Preventive effects of cannabis extract on WBC count (EAC induced after 21 doses of cannabis extract injection). CBC count using light microscope (a) normal mice WBC slide, (b) 21 days dose's injection mice CBC slide. Marking area indicate WBC, both were shows the similar observation.

4.3.4.3.7 Cytopathological study

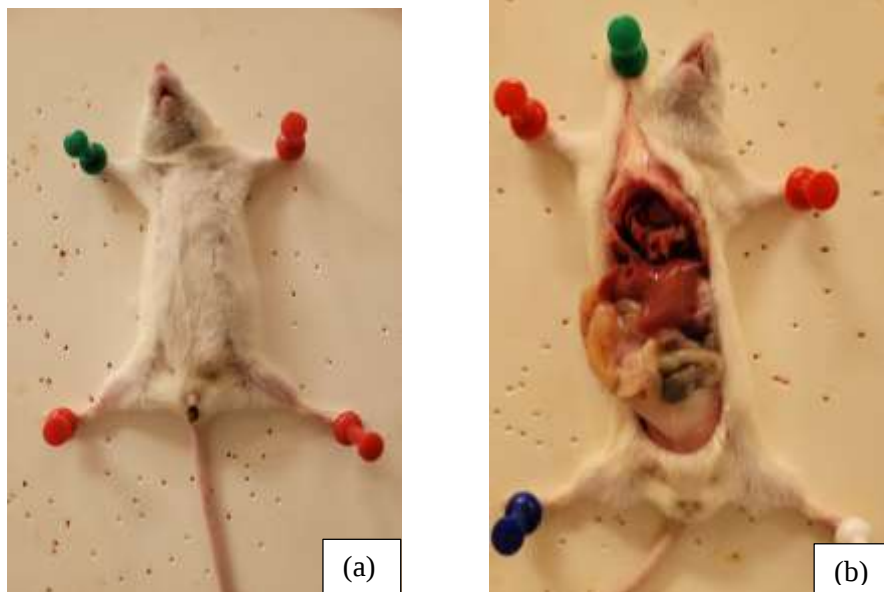


Figure 65: Preventive effects of cannabis extract on body acetic fluids (EAC induced after 14 doses of cannabis extract injection). For cytopathological observation (a) No edema form when 21 doses of treatment injection to the mice. (b) after sacrificed no abdominal fluid founded.

4.3.4.3.8 Histopathological study of main organs

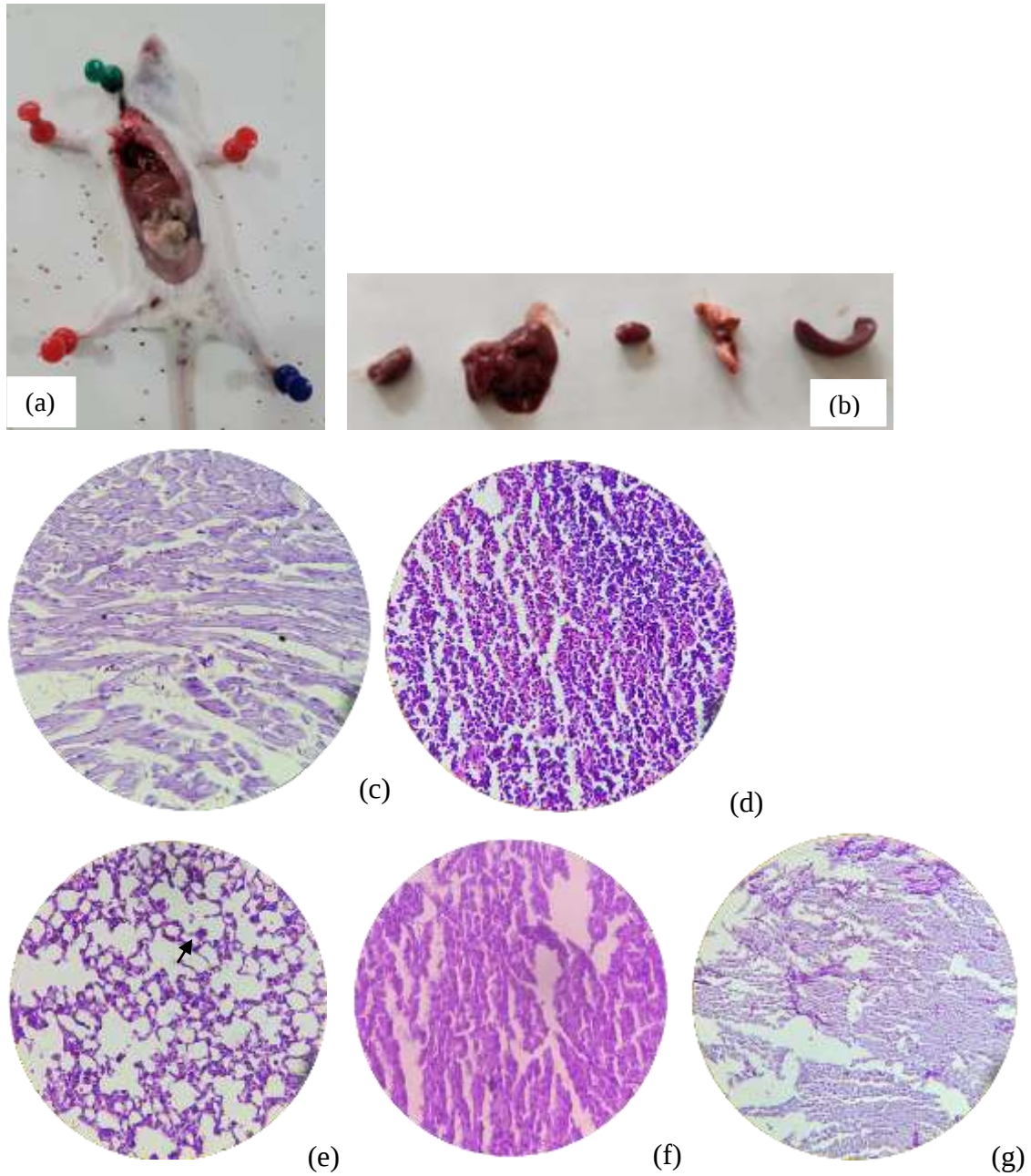
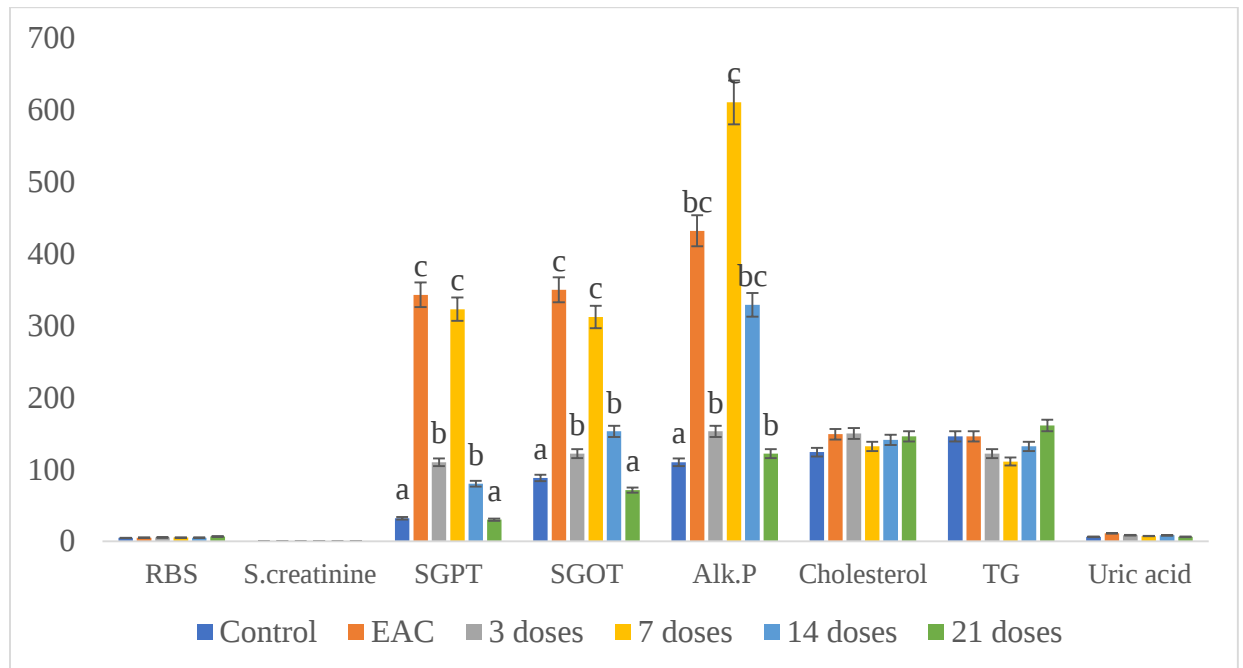


Figure 66: Preventive effects of cannabis extract on cells changes (EAC induced after 14 doses of cannabis extract injection). For histopathological observation (a) mouse sacrificed (b) main organ surgically removed, (c) heart, (d) pancreas, (e) lung, (f) kidney, and (g) liver histopathological image using H and E dyes. 400x view using a light microscope. No morphological cellular changes occur at this observation. Inflammatory cell observed at the lung.

4.3.4.4 Biochemical profile interpretations

The effects of cannabis extract as cancer prevention of biochemical profile interpretations are given the figure no. 64. In this case, RBS, serum creatinine, serum cholesterol, serum triglyceride, and serum uric acid results had not shown significant ($p < 0.05$) variations among the control, EAC, Ifodex, 3rd day, 1st, 2nd, and 3rd-week groups respectively. Significant variations ($p < 0.01$) were observed in the SGPT and SGOT values compared to the other groups. The highest value of SGPT in EAC ($343 \pm 12.4 \text{ U/L}$) was significantly different from the lowest value at 21 doses ($30 \pm 0.99 \text{ U/L}$). There were no significant ($p < 0.01$) differences observed among the group of 3 doses and 7 doses of observation. The highest value of SGOT in 3rd week ($350 \pm 31.3 \text{ U/L}$) was significantly varied from the lowest value of control ($71 \pm 2.13 \text{ U/L}$). But there were no significant differences observed in the control, 3 doses, and 7 doses of observation groups. Alkaline phosphatase value shows ($p < 0.001$) significant variation among the other groups of observation. The highest value in 14 doses ($611 \pm 32.2 \text{ U/L}$) of alkaline phosphatase was significantly ($p < 0.001$) varied from the lowest value ($110 \pm 10.2 \text{ U/L}$) in the control group.

Figure 67: Preventive effects of cannabis extract on biochemical profile interpretation.

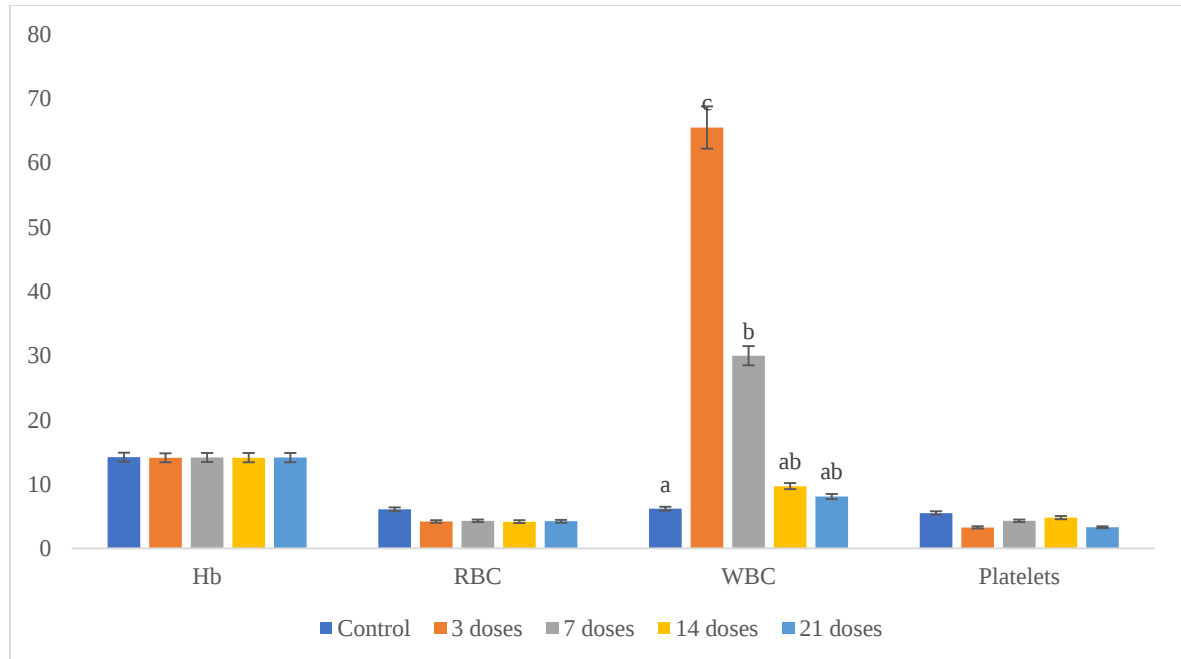


^{abc} values having different superscripts differed significantly.

4.3.4.5 Hematological profile interpretation

The preventive effects of cannabis extract of hematological profile interpretations are given the figure no. 68. There were no significant ($p < 0.05$) variations observed on Hb, RBS, and platelet count. WBC count shows ($p < 0.001$) significant variation among the groups of control, 3 doses, 7 doses, 14 doses, and 21 doses of observation. The highest value at 3 doses ($65.5 \pm 8.02 \times 10^3/\text{cmm}$) of WBC count was significantly ($p < 0.001$) varied to the lowest value at control ($6.2 \pm 0.01 \times 10^3/\text{cmm}$). There were no significant ($p < 0.001$) variations observed in the 14 doses and 21 doses of observation.

Figure 68: Preventive effects of cannabis extract on hematological profile interpretation.



^{abc} values having different superscripts in the same row differed significantly.