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FINAL REPORT

1. Effects on viability of cancer cells

Several different human cancer cell lines were used: MCF-7 (breast), HL-60 (leukemia), NCI-H23 (lung) and PC-3 (prostate). In each case, cells were cultivated at $1x10^6$/ml concentration at RPMI 1640 medium with and without 10% FCS. Four different doses of tested substances were added to the culture wells and the viability of cells was tested 48 hours later by trypan blue exclusion.

2. Effects on phagocytosis

Natural modulators usually act via phagocytosis, therefore we also added evaluation of effects on phagocytosis. We used the synthetic polymeric microspheres, HEMA, since their use, dose and timing are already well established in glucan studies. We used single injection of tested material and evaluated the phagocytosis of mouse peripheral blood neutrophils 24 hours after i.p. injection. We used 50 □g and 100 □g/mouse.

3. Effect on IL-2 production

Evaluation of the effects of glucan on IL-2 production by splenocytes using a commercial IL-2 ELISA kit.

RESULTS

Proliferation

Experiments evaluated doses from 0.01 to 10 □l/well in an experimental setup allowing us to observe possible stimulation of cell growth (in samples without FCS) or inhibition of cancer growth (in samples with 10% FCS). In none of the samples we found any significant effects on cell growth.

	% positive neutrophils	pg of IL-2
PBS	31.1	1.1
50 □g	40.4	112.7
100 □g	47.2	262.4

CONCLUSION

From these experiments it is clear that the sample designed MG-LZ8 is a biologically active sample with significant effects on immune reactions. In addition, the sample showed no toxic effects towards cells *in vitro.*

Sincerely yours

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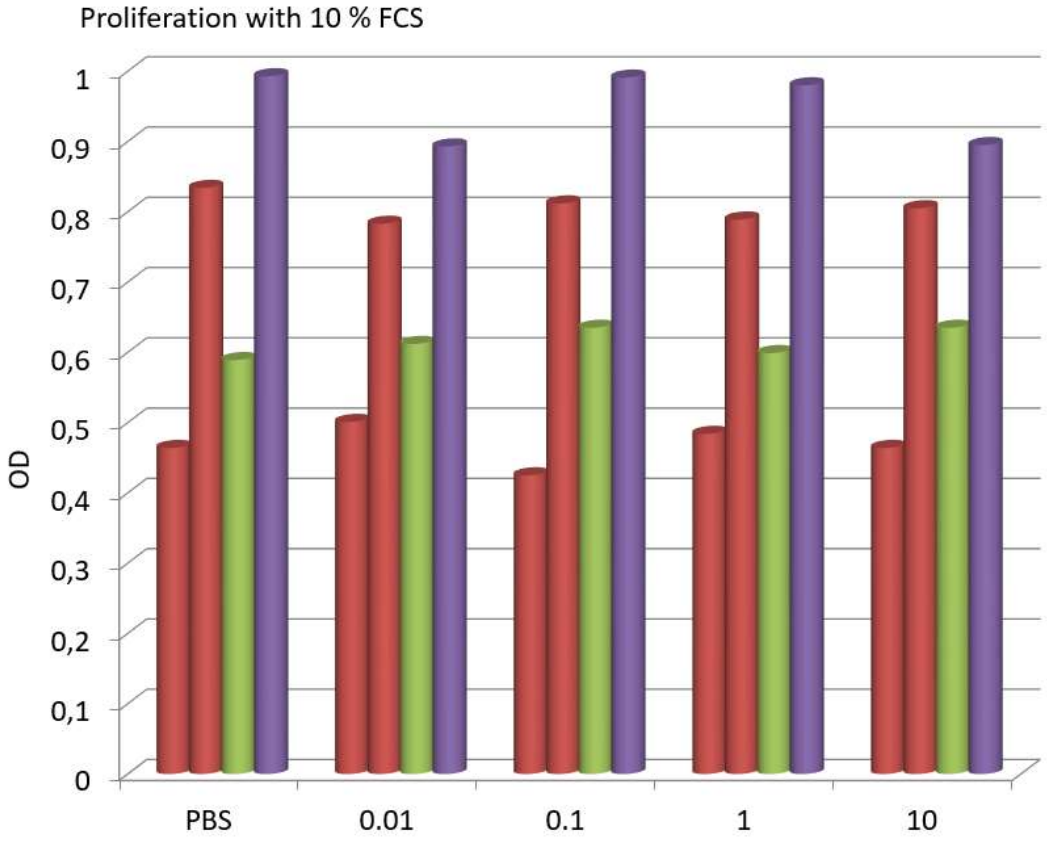
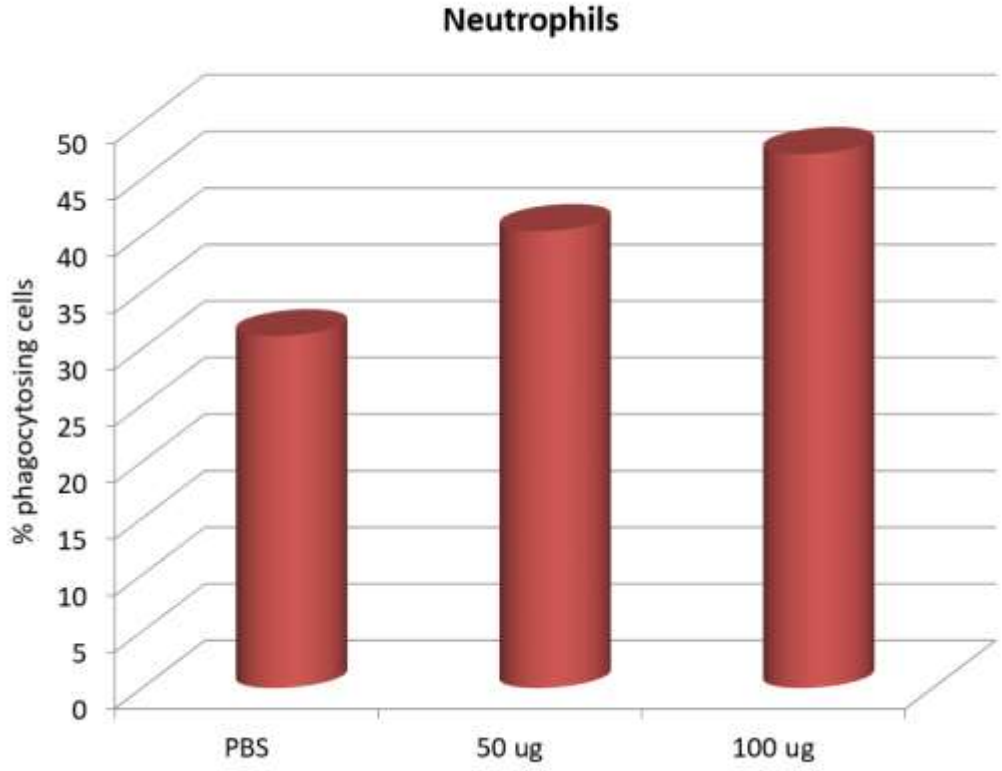
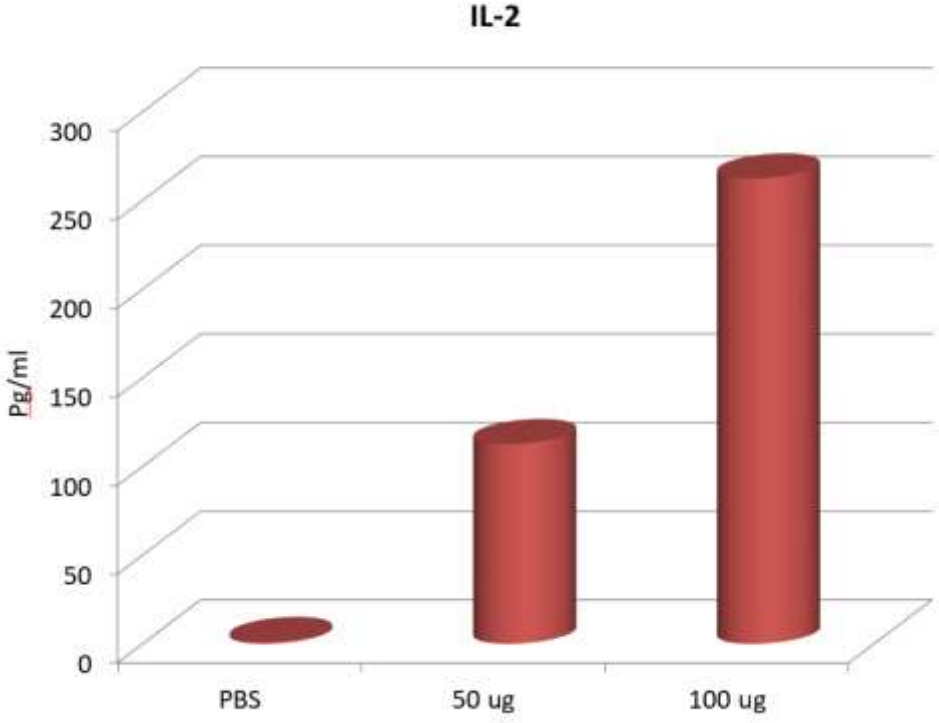
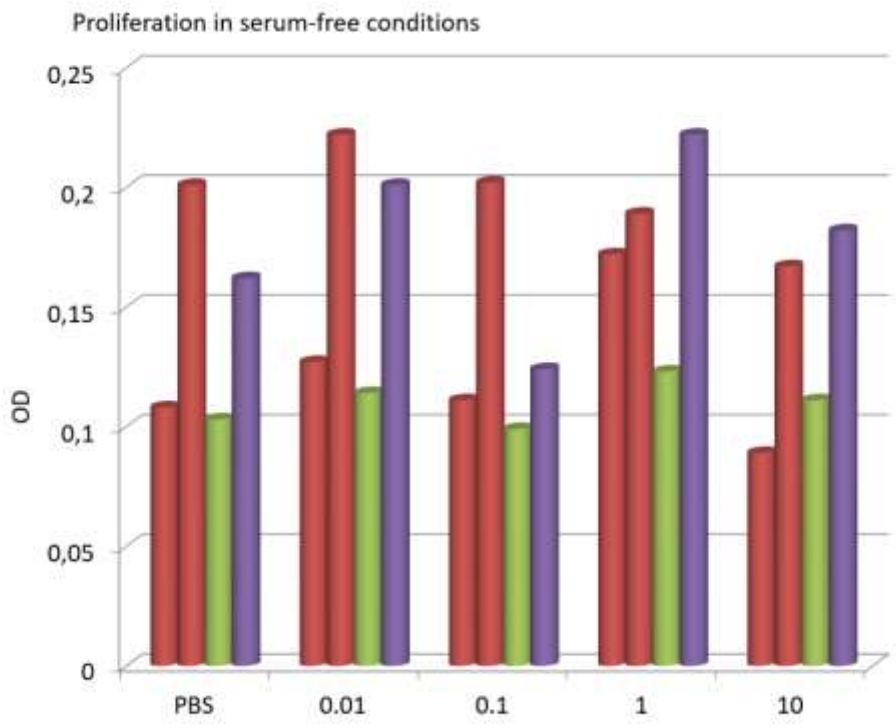
GRAPHS




Neutrophils
% phagocytosing cells
50
45
40
35
30
25
20
15
10
5
0
PBS
50 ug
100 ug


IL-2
Pg/ml
300
250
200
150
100
50
0
PBS
50 ug
100 ug


Proliferation in serum-free conditions
0,25
0,2
0,15
0,1
0,05
0
OD
PBS
0.01
0.1
1
10