Effects of Ionized Silver (IM/Q610-419-01A) on 4 cancer types and PBMCs.

Starlight Laboratories

Ionized Silver

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Product

The product under testing was delivered directly to Synexa Life Sciences (Pty) Ltd as a ready-made solution and this was indicated as de-ionized water containing 18.012 ppm (18.012 mg/l) ionized silver (Ag). In parallel, a bottle containing only the de-ionized water was made available as a control for all experiments.

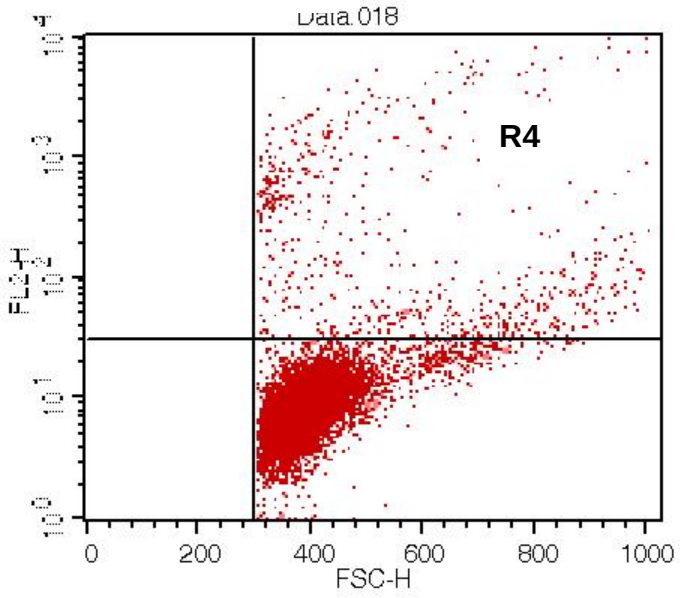
Methodology

The ionized Ag preparation was evaluated for its effect on 4 cancer cell lines at three different concentrations which included 4.503 ppm (25%), 2.252 ppm (12.5%), and 1.126 ppm (6.25%). Due to the nature of the assay and the volumes needed we were restricted to a highest final concentration of 4.503 ppm. At higher concentration interference with cell activity occurred due to the aqueous nature of the sample and the control sample.

The four cell lines used for testing the potential anti-cancer properties of ionized Ag were Jurkat (T cell leukemia), MOLT-4 (lymphoblastic leukemia), K-562 (chronic myelogenous leukemia), and HL60 (acute promyelocytic leukemia). The bio-assay involved flow cytometry based analysis of the amount of dead cells present after overnight incubation in the presence and absence of Ionized Ag at the above mentioned concentrations. The potential of each concentration of ionized Ag to induce cell death (cytotoxic) was determined by comparing the amount of dead cells to that of the corresponding water control sample. A representative plot of the results is shown in Figure 1 hereunder. If the sample is able to induce direct cell death after overnight incubation, the cells would be measured in region R4 due to the uptake of the dye used (PI).

The effect of ionized Ag on peripheral blood mononuclear cells (PBMCs) was also evaluated as per the above mentioned bio-assay to assess its *in vitro* safety profile against normal lymphoid cells. Ionized Ag was incubated overnight with both stimulated/proliferating and non-stimulated PBMCs at the above mentioned concentrations. Results were again compared to corresponding water control samples.

Figure 1



Results

The effect of the product on each cancer cell line and PBMCs are tabulated below. The amount of dead cells is expressed as a percentage of the gated cells (Fig. 1).

Jurkat		
Concentration	**Ionized Ag**	**Control H_2O**
4.503 ppm (25%)	**59.59**	**6.53**
2.252 ppm (12.5%)	**58.63**	**6.17**
1.126 ppm (6.25%)	**7.48**	**7.18**

MOLT 4		
Concentration	**Ionized Ag**	**Control H_2O**
4.503 ppm (25%)	**86.77**	**3.25**
2.252 ppm (12.5%)	**88.73**	**2.50**
1.126 ppm (6.25%)	**3.2**	**2.46**

HL60		
Concentration	**Ionized Ag**	**Control H_2O**
4.503 ppm (25%)	**40.33**	**7.56**
2.252 ppm (12.5%)	**61.85**	**10.26**
1.126 ppm (6.25%)	**8.29**	**8.31**

K-562		
Concentration	**Ionized Ag**	**Control H_2O**
4.503 ppm (25%)	**72.01**	**2.09**
2.252 ppm (12.5%)	**51.55**	**4.33**
1.126 ppm (6.25%)	**9.47**	**2.41**

Stimulated PBMCs		
Concentration	**Ionized Ag**	**Control H_2O**
4.503 ppm (25%)	**99.12**	**63.53**
2.252 ppm (12.5%)	**99.74**	**60.87**
1.126 ppm (6.25%)	**73.90**	**34.87**

Unstimulated PBMCs		
Concentration	**Ionized Ag**	**Control H_2O**
4.503 ppm (25%)	**99.89**	**6.89**
2.252 ppm (12.5%)	**99.24**	**4.98**
1.126 ppm (6.25%)	**33.19**	**8.22**

Conclusion

The results clearly show the potent cytotoxic effect of ionized Ag *in vitro* on the different cell lines. It is also evident that this activity is lost at a final concentration of 1.125 ppm for all the tested cell lines. The cytotoxic effect of ionized Ag was most pronounced on MOLT-4 and K562 cells increasing the amount of dead cells to 53% and 83% respectively at a final concentration of 4.503 ppm. Although Jurkat and HL60 cells were less sensitive to the effects of ionized Ag, its influence was still noteworthy.

The cytotoxic effect of ionized Ag was less pronounced on proliferating/stimulated PBMCs, increasing the amount of dead PBMCs in the stimulated sample to around 30% more than the control. The effect of ionized Ag seems to be much greater on non-proliferating/unstimulated PBMCs.

By comparing the effects of ionized Ag on cancer cells and normal lymphoid cells it is evident that ionized Ag has a general but varying toxic effect on different cells. Interestingly ionized Ag had potent effect on unstimulated PBMCs when one would expect the opposite, sine these cells are not proliferating. We cannot conclude whether the oral intake of the product will demonstrate the same anti-cancer activity because the *in vitro* results cannot be extrapolated to *in vivo* situations: metabolic transformation of the product, oral uptake, etc. However, it remains an interesting product to consider for further research.

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