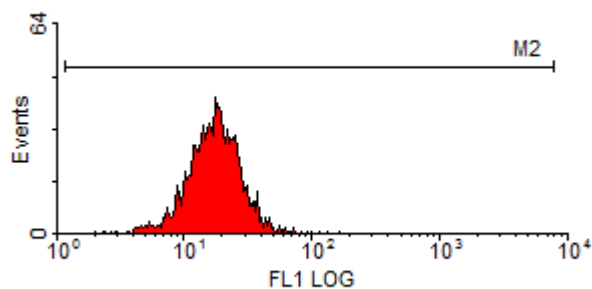


Immunomagnetic separation of hybridoma cells using target antigen-armed microspheres

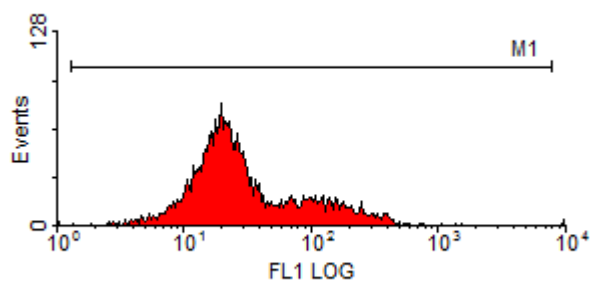
Hybridoma cell line XGY7 (Gy27), secreting anti-gliadin mAbs, was used as the model. Immunomagnetic separation of the cells was performed using biotin labeled specific antigen (gliadin) and immunomagnetic separation kit of Miltenyi Biotec.

Fractionation of hybridoma cells

Control cells GY27 – unstained, washed by PBS. Peak value = 17.26 (autofluorescence).

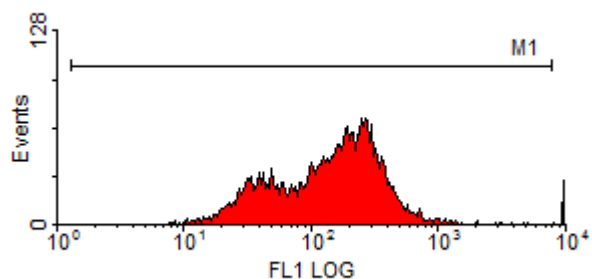


Cells GY27 were then stained by gliadin-biotin / Streptavidin-FITC. Peak average = 31.61.

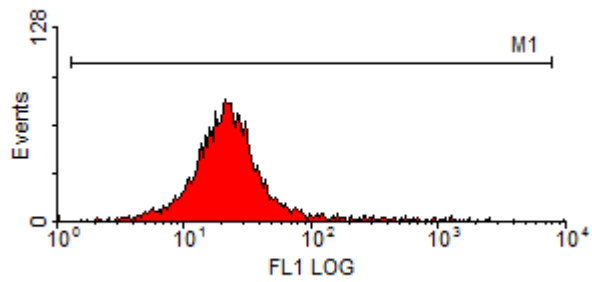


Then hybridoma cells were separated by immunomagnetic column under sterile conditions. Two fractions were obtained:

1) Antigen-binding fraction. The cells were stained by gliadin-biotin / Streptavidin-FITC. Peak average shifted to 139.39 .

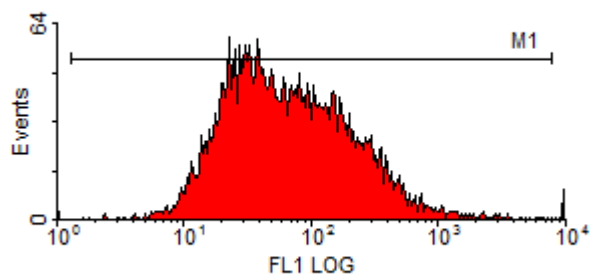


2) unbound fraction of the cells stained by the same method gave average peak value 23.95

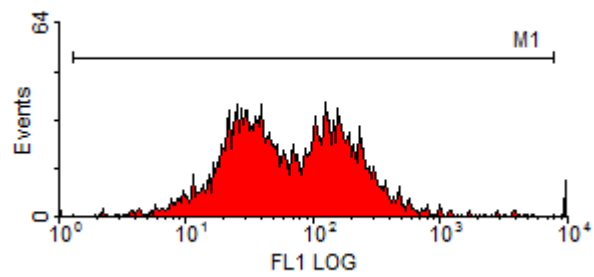


Both fractions of hybridoma cells were cultivated and re-tested after 2 weeks.

1) Antigen-binding fraction still shown increased antigen binding ability, average peak value was 66.10



2) In unbound fraction, two weeks of cultivation, caused an appearance of substantial percentage of antigen-binding cells, giving the average peak value 67.15.



We conclude that hybridoma cells can be efficiently separated by antigen binding property, however this property tends to disappear after 2 weeks of cultivation *in vitro*. We may suggest that the property to bind a target antigen is related mostly to the cell cycle than reflects the presence of different subpopulations in hybridoma cell line.