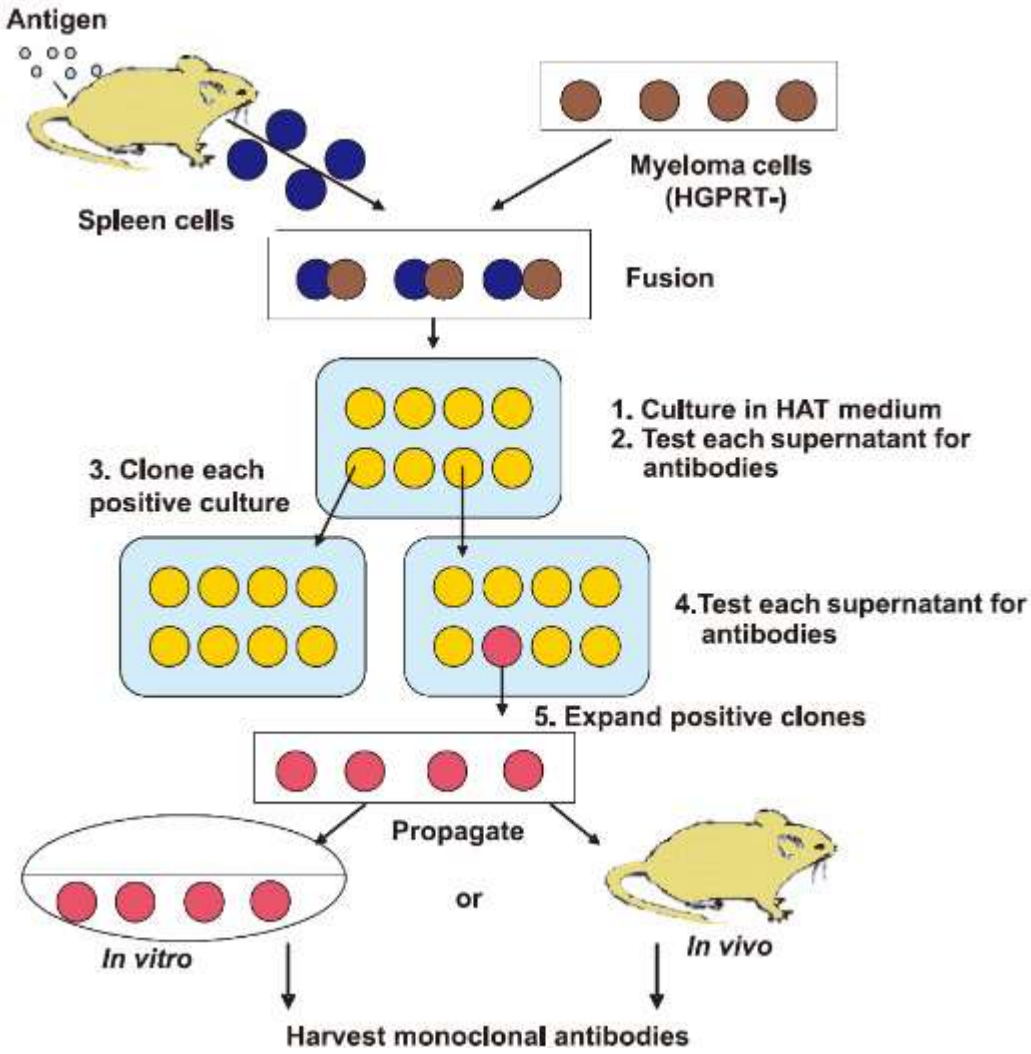
Hybridoma Technology

Hybridoma technology is a well-established method to produce monoclonal antibodies (mAbs) specific to antigens of interest. Hybridoma cell lines are formed via fusion between a short-lived antibody-producing B cell and an immortal myeloma cell. Each hybridoma constitutively expresses a large amount of one specific mAb, and favored hybridoma cell lines can be cryopreserved for long-lasting mAb production. Hybridoma technology was discovered in 1975 by two scientists, Georges Kohler and Cesar Milstein. They wanted to create immortal hybrid cells by fusing normal B cells from immunized mice with their myeloma cells.

Steps of Hybridoma:



1) Cell fusion: Adjacent myeloma and/or antibody-secreting cells are fused employing PEG (polyethylene Glycol) or by electrofusion.

2) Hybridoma screening: There are only about 1% of the starting cells are fused, and only about 1 in 105 form viable hybrids. This leaves a large number of unfused cells still in culture. Commonly, the myeloma cells have a defective HGPRT enzyme (hypoxanthine-

guanine phosphoribosyl transferase). These cells containing a non-functional HGPRT protein will die in HAT medium. Only the hybridoma cells have got the ability to divide and proliferate on the HAT medium because genome from the B-lymphocyte makes them HGPRT positive and genome from the myeloma cells they can divide indefinitely.

3) mAb production: Hybridoma antibodies can be produced in vitro and in vivo.

For *in vitro*, the hybridomas are transferred to tissue culture plates. And for *in vivo* mice are primed by intraperitoneal injection with 10^5 - 10^7 hybridoma cells.

Suggested readings: https://www.ncbi.nlm.nih.gov/pubmed/31347924