J Biomed Eng Med Devic 2017, 2:2 (Suppl)
DOI: 10.4172/2475-7586-C1-003

Single chain antibodies fragment (scFv) coupled by trimerization domains, an alternative to monoclonal antibody therapy

Lionel Zapata
University of Concepcion, Chile

Many different monoclonal antibodies (mAbs) are already available for cancer, chronic infections and autoimmune diseases therapy. Over the last few years biopharmaceutical companies have experienced a huge improvement in the mAbs manufacturing process, however the antibody based therapies still remain too expensive. Because of the molecular nature of mAbs, the recombinant expression in mammalian cells cannot be replaced by less expensive alternatives such as bacteria or yeast culture. Here, we propose a molecular alternative for therapeutic mAbs, based on fragments of single chain antibodies (scFv) coupled through trimerization domain of tetranectine. Trimeric scFvs share with Mabs the domains involve in target recognition and have a similar molecular weight. These trimeric scFv can be successful expressed in cheaper platforms as yeasts or bacteria culture. To obtain scFvs coupled trough trimerization domains the yeast expression vector pPSHG20-tetra was constructed by restriction cloning. This vector codes for the expression of an anti-VEGF scFv coupled by the tetranectine trimerization domain following by a poly histidine tag was added to N-terminal, to facilitate identification and subsequent purification. MP-36 yeast strain was transformed with the plasmid-tetra pPSHG20 by electroporation and transformed cells were isolated by auxotrophy selection and the insertion was checked by Southern blotting. Trimeric scFv expression was performed with 0.7% methanol during 4 days. Then trimeric scFvs were purified using affinity purification by IMAC. The protein expression yield and the ability to trimerize were analyzed by SDS-PAGE, Western blotting and densitometry, in reducing and non-reducing conditions. Finally, the bioactivity of trimeric scFv was confirmed by *in vitro* and *in vivo* models. As a result, we demonstrated that the expression of high yields of trimeric scFv fragments in yeast is possible. The estimated molecular weight of trimeric scFv was similar to the IgG antibody isotype. Furthermore, trimers of scFv fragments were obtained with high purity (95%) and remained their trimeric structure. The trimeric anti-VEGF scFv antibody inhibits the biologic activity of human VEGF in *in vitro* and *in vivo* assay. This results suggests that functional scFv coupled trough trimerization domains production is possible. The trimeric scFv could be reliable alternative to the mAbs based therapy with similar bioactivity but with a lower production cost.

lionelzv@gmail.com

Notes: