Production of fox FGF2 for fox pluripotent stem cell culture

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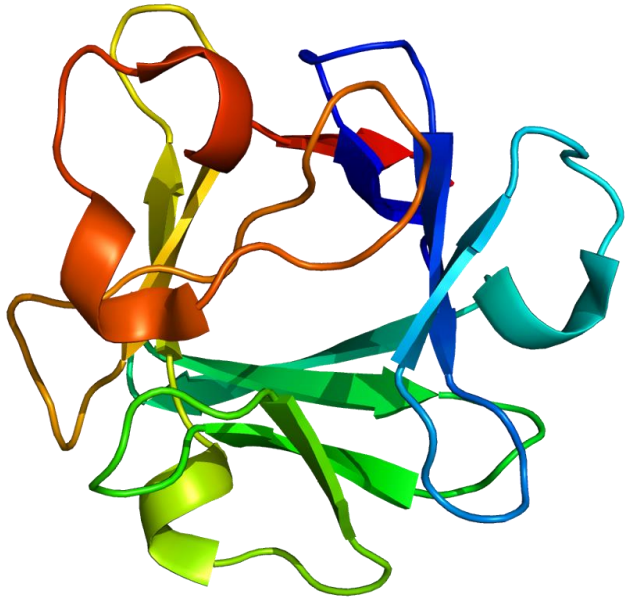
Embryonic stem cells and induced pluripotent stem cells of non-human species may require species-specific growth factors such as LIF and FGF2.

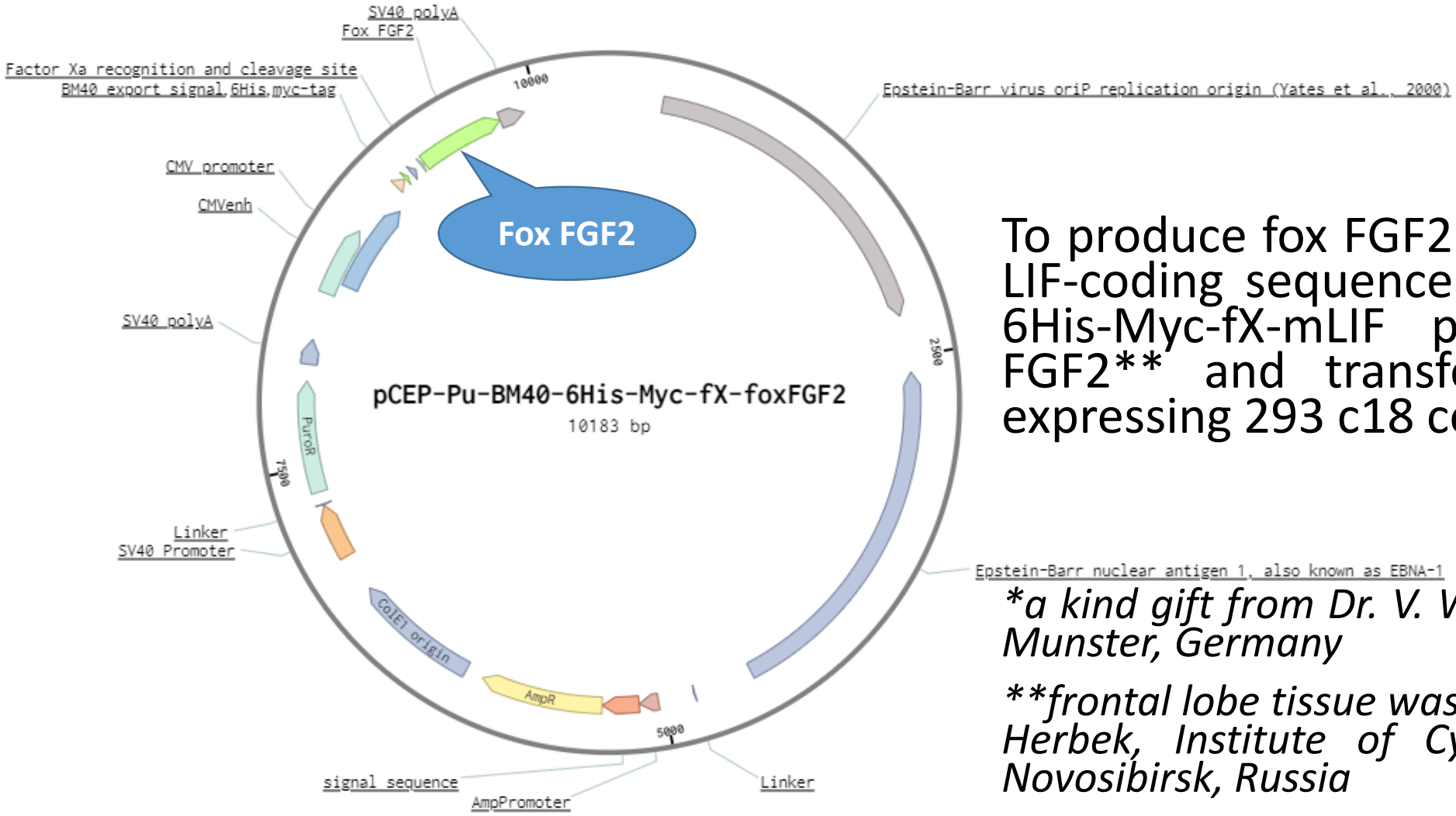
We decided to develop a system for **fox FGF2** production for fox pluripotent stem cell culture.



Human FGF2

https://commons.wikimedia.org/wiki/File:Protein_FGF2_PDB_1bas.png

HEK293-EBNA1-based system for protein production



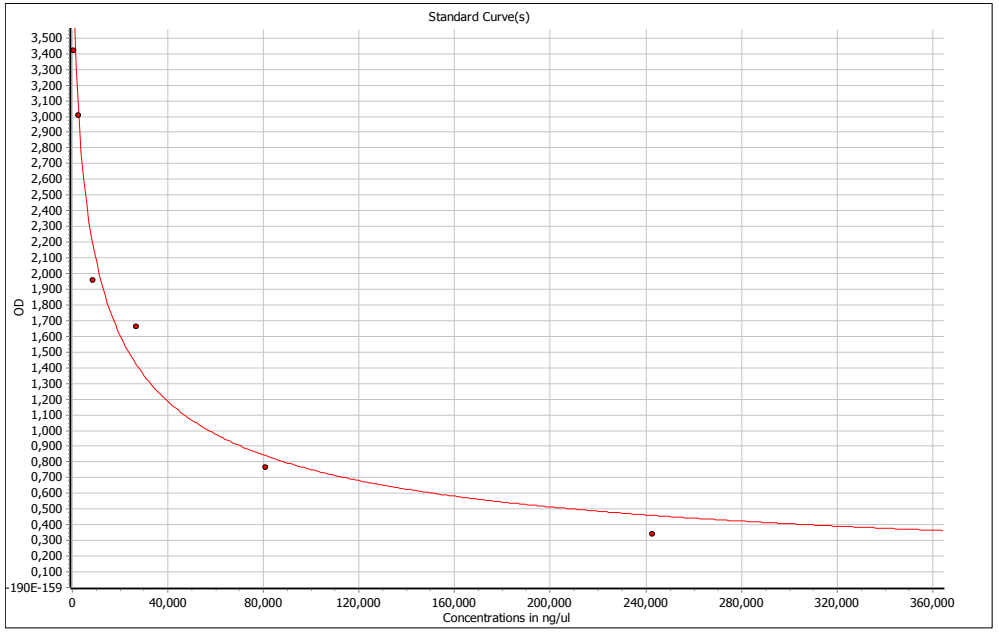
To produce fox FGF2 we substituted the LIF-coding sequence in pCEP-Pu-BM40-6His-Myc-fX-mLIF plasmid* with fox FGF2** and transfected into EBNA1 expressing 293 c18 cell line

**a kind gift from Dr. V. Wixler, the University of Munster, Germany*

***frontal lobe tissue was a kind gift from Dr. Yu. Herbek, Institute of Cytology and Genetics, Novosibirsk, Russia*

Fox FGF2 concentration in conditioned medium

His Tag ELISA Detection Kit Standard Curve



We determined the fox FGF2 protein concentration in DMEM/F12 medium conditioned by the FGF2 producing cells using His Tag ELISA Detection Kit (GenScript, USA). It was 19 ng/ml. Human pluripotent stem cells are cultured with 10 ng/ml, so 19 ng/ml may be considered low.

Overall, we have produced a cell line with fox FGF2 expression. The protein yield in the conditioned medium is low for the direct usage with fox pluripotent stem cells. Additional steps of concentration and purification are necessary.

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