CALIFORNIA STATE UNIVERSITY SAN MARCOS

PROJECT SIGNATURE PAGE

PROJECT SUBMITTED IN PARTIAL FULFILLMENT
OF THE REQUIREMENTS FOR THE DEGREE

MASTER OF SCIENCE

IN

BIOTECHNOLOGY

PROJECT TITLE: Metabolic characterization of induced pluripotent stem cells from the critically endangered Northern White Rhinoceros

AUTHOR: Chenyuan Gao

DATE OF SUCCESSFUL DEFENSE: July 24th 2020

THE PROJECT HAS BEEN ACCEPTED BY THE PROJECT COMMITTEE IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTER OF SCIENCE IN BIOTECHNOLOGY.

Betsy Read	Betsy Read	8/4/2020
PROJECT COMMITTEE CHAIR	SIGNATURE	DATE
Tetsuya Kawamura		08/06/2020
PROJECT COMMITTEE MEMBER	SIGNATURE	DATE
Inigo Valiente-Alandi	DocuSigned by: Inigo Valiente Alandi 4B14F8FC72864D0...	8/6/2020
PROJECT COMMITTEE MEMBER	SIGNATURE	DATE

EXECUTIVE SUMMARY

Metabolic characterization of induced pluripotent stem cells from the critically endangered Northern White Rhinoceros

Sponsor by San Diego Zoo Institute for Conversation Research

Chenyuan Gao, Student Research Associate, M.B.

Project Defense Date: July 24th, 2020

Professional Science Master's Degree Program

California State University San Marcos

The Semester-in-Residence project was conducted in San Diego Zoo Institute for Conservation Research, conservation genetics department. The purpose of this study is to discover the metabolic properties of induced Pluripotent Stem Cell (iPSC) lines from the critically endangered Northern White Rhinoceros (NWR). We perform a series of experiments to characterize the metabolic properties of NWR iPSCs, including mitochondria detection, ATP quantification, glycolysis capacity and oxygen consumption level detection. Our results show (1) The NWR iPSCs contain high levels of viable mitochondria. (2) The intracellular ATP level in NWR iPSC is 9.55±1.53 fmol, which is less than human induced Pluripotent Stem Cells (hiPSCs). (3) NWR iPSCs and hiPSCs both primarily rely on glycolysis instead of oxidative phosphorylation for energy generation. This glycolysis and oxygen consumption data also suggest that NWR iPSCs are at primed pluripotent states. This knowledge is firstly proposed for NWR iPSCs and support a long-term international collaborative conservational project to save NWRs. There are some recommendations to enhance this study, which include: introducing more hiPSC and NWR iPSC lines; introducing positive and negative controls for the glycolysis study; including beta-oxidation study; collaborating with epigenomic study to fortify our conclusion that NWR iPSCs are at primed pluripotent states.

Metabolic characterization of iPSCs from the critically endangered Northern White Rhinoceros

Chenyuan Gao

07/30/20

SAN DIEGO ZOO
INSTITUTE FOR
CONSERVATION
RESEARCH

California State University
SAN MARCOS

The northern white rhino, population: 2

Cellular technologies apply in NWR conservation

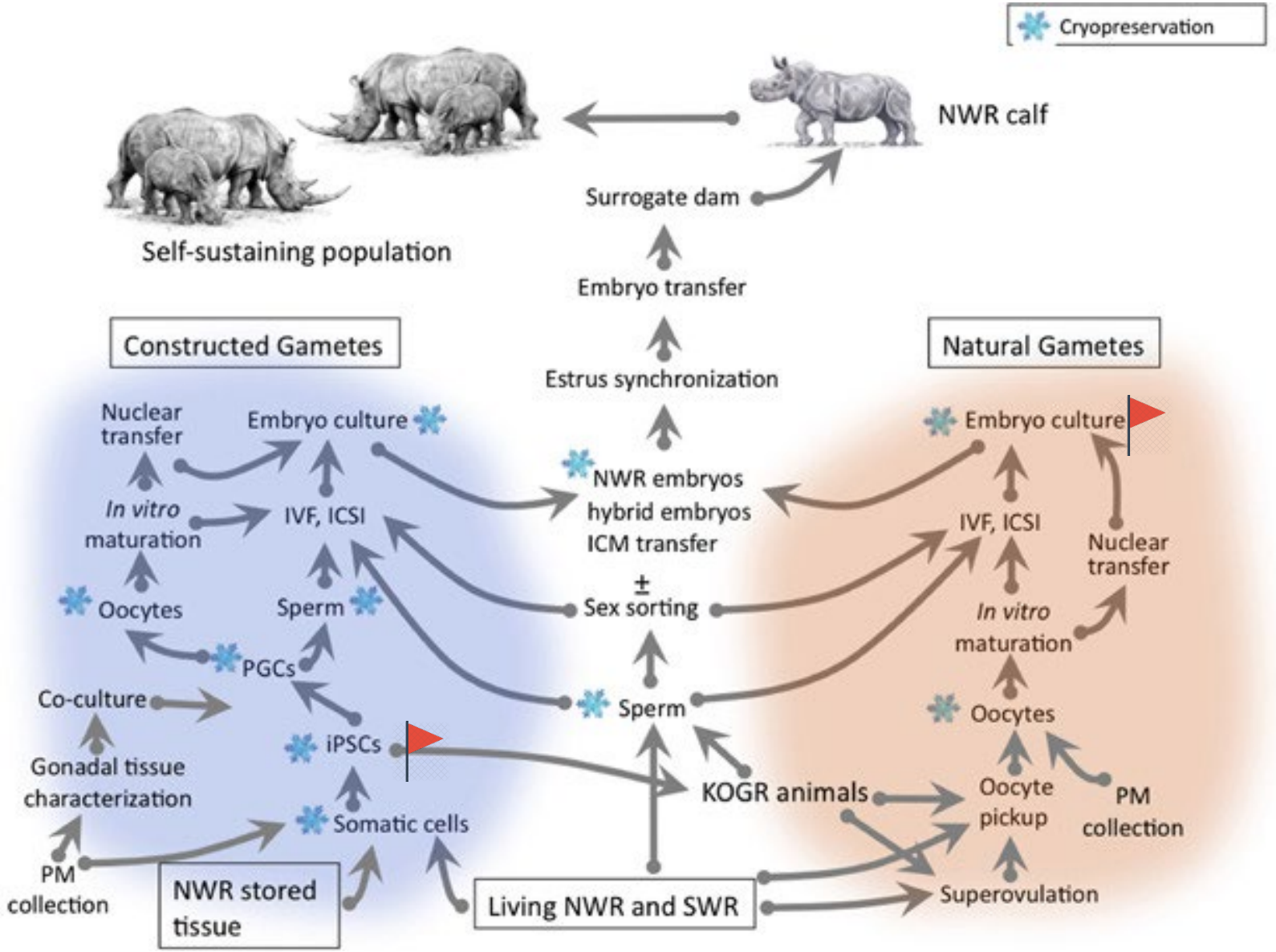
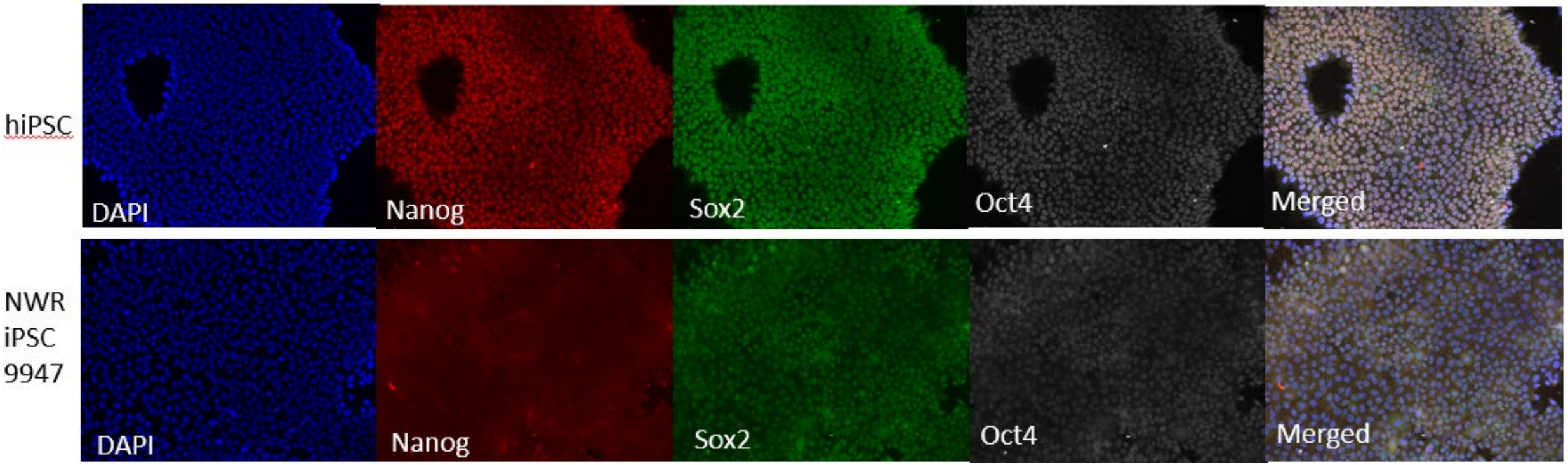
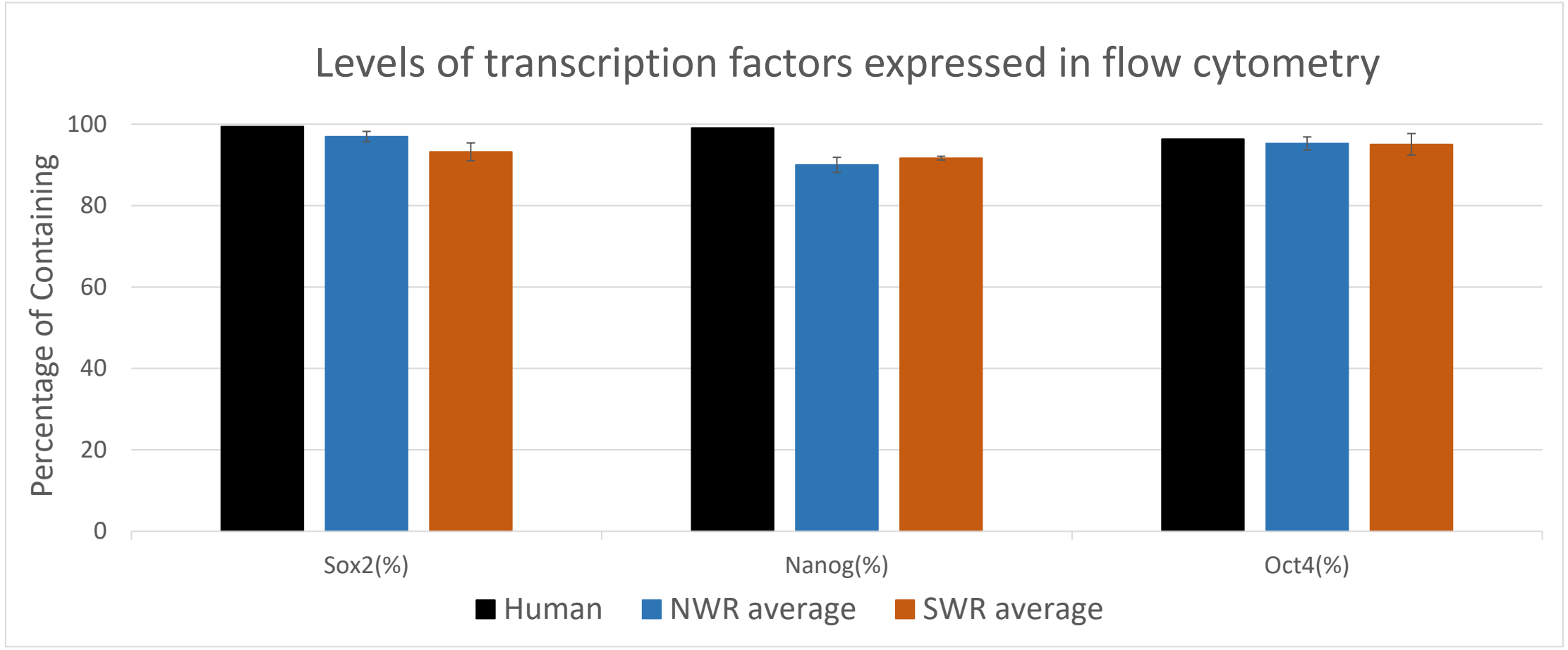


Image source: https://onlinelibrary.wiley.com/doi/full/10.1002/zoo.21284

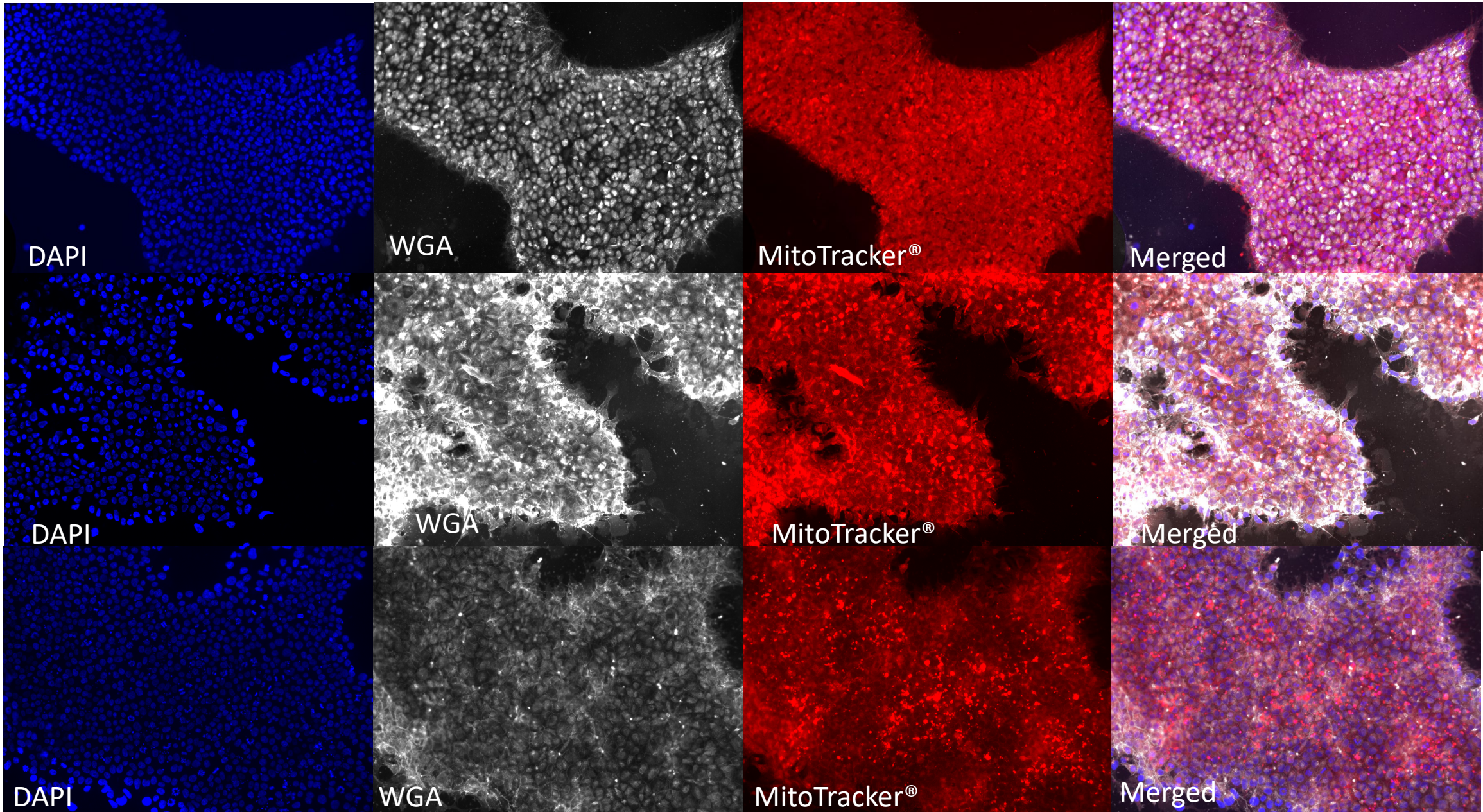
hiPSCs and NWR iPSCs expressed three pluripotency markers



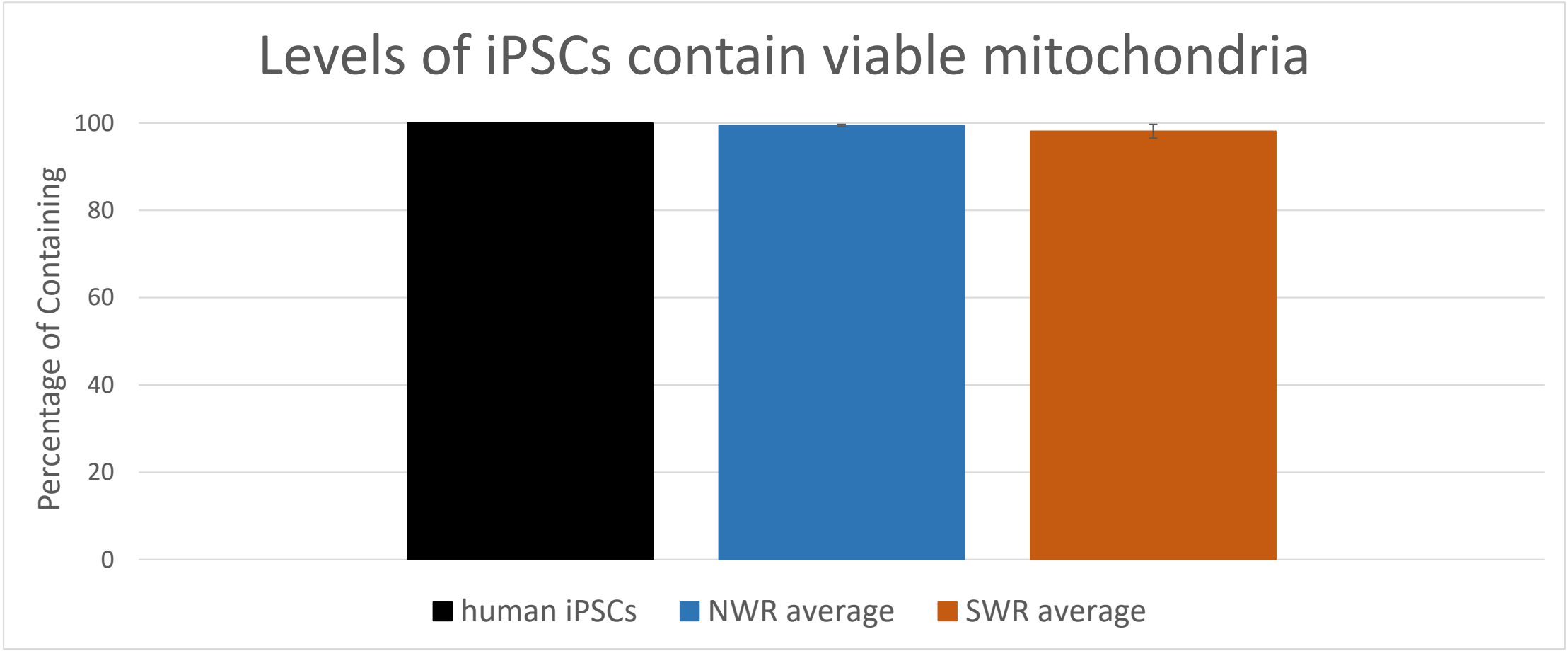
All iPSC lines expressed high level of pluripotency markers.



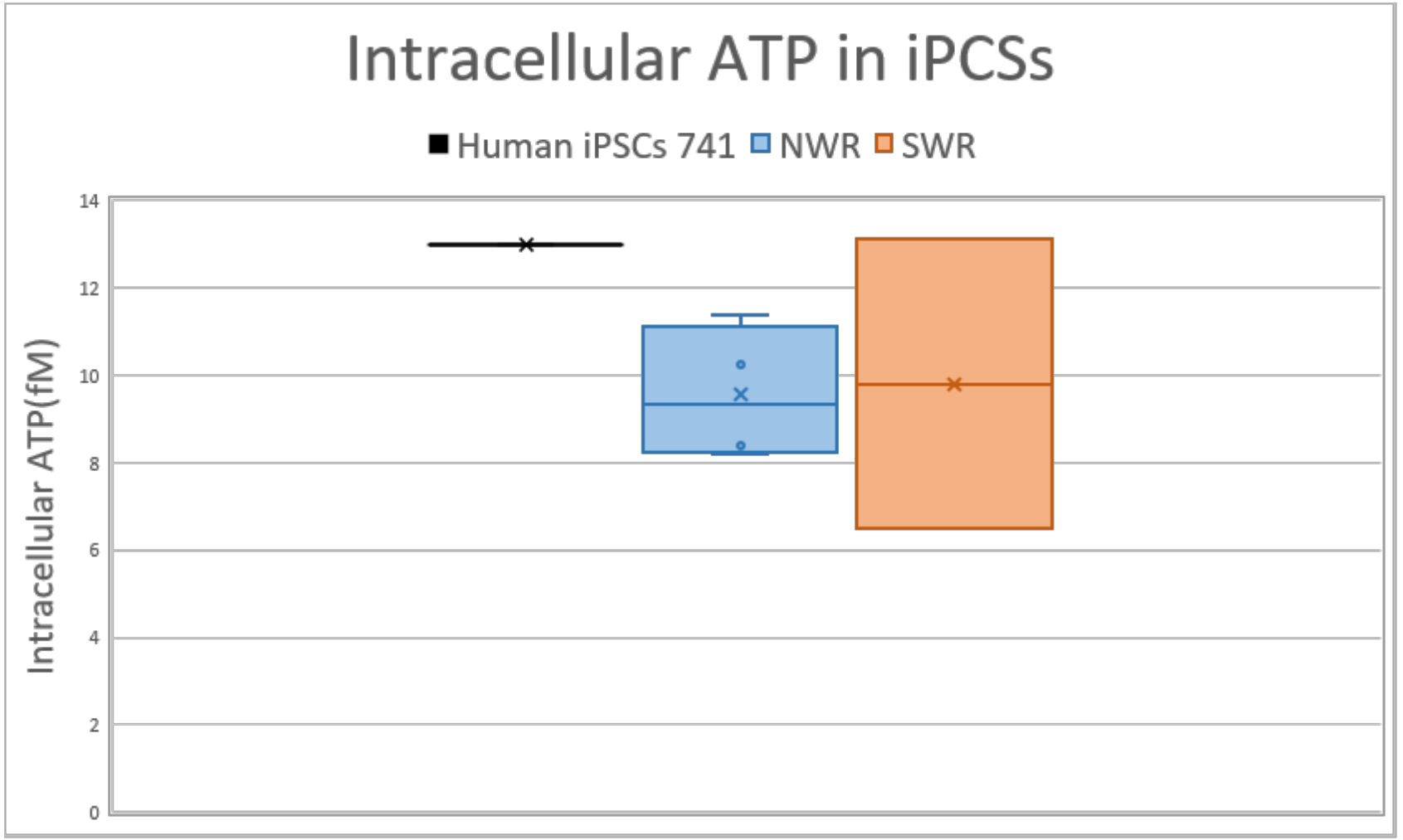
All iPSC lines contains viable mitochondria.



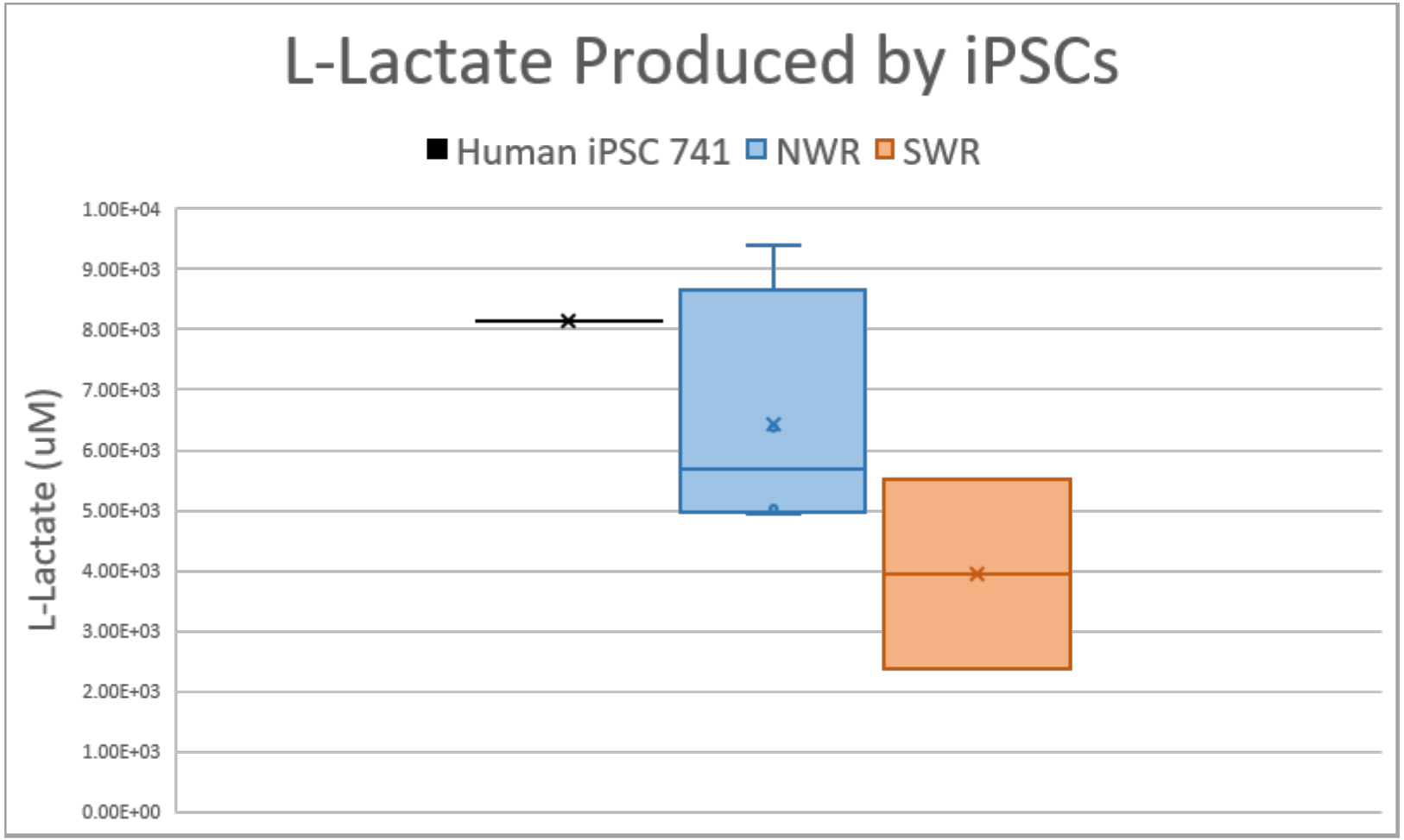
99% of the cell from most lines contained active mitochondria



Average NWR iPSC contains 9.55 fM cellular ATP.



hiPSCs produced similar level of L-lactate with NWR iPSCs



All iPSC lines shown low level of oxygen consumption tendency.