REPORT

Report on participation of the ICMR International Fellow (ICMR-IF) in Training/Research abroad.

1. Name and designation of ICMR- IF : **Dr.V.Balachandar**
   Assistant Professor, Bharathiar University

2. Address : **Department of Human Genetics and Molecular Biology**
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3. Frontline area of research in which training/research was carried out :
   "Human Induced Pluripotent Stem Cell Research"

4. Name & address of Professor and host institute
   Dr. Morten Meyer, Ph.D.
   Associate Professor, Department of Neurobiology Research
   Institute of Molecular Medicine, University of Southern Denmark
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   Phone +45 65503802 (office), +45 65503810 (laboratory)
   E-mail mmeyer@health.sdu.dk

5. Duration of fellowship : **6 Months (October, 2018 to April, 2019)**

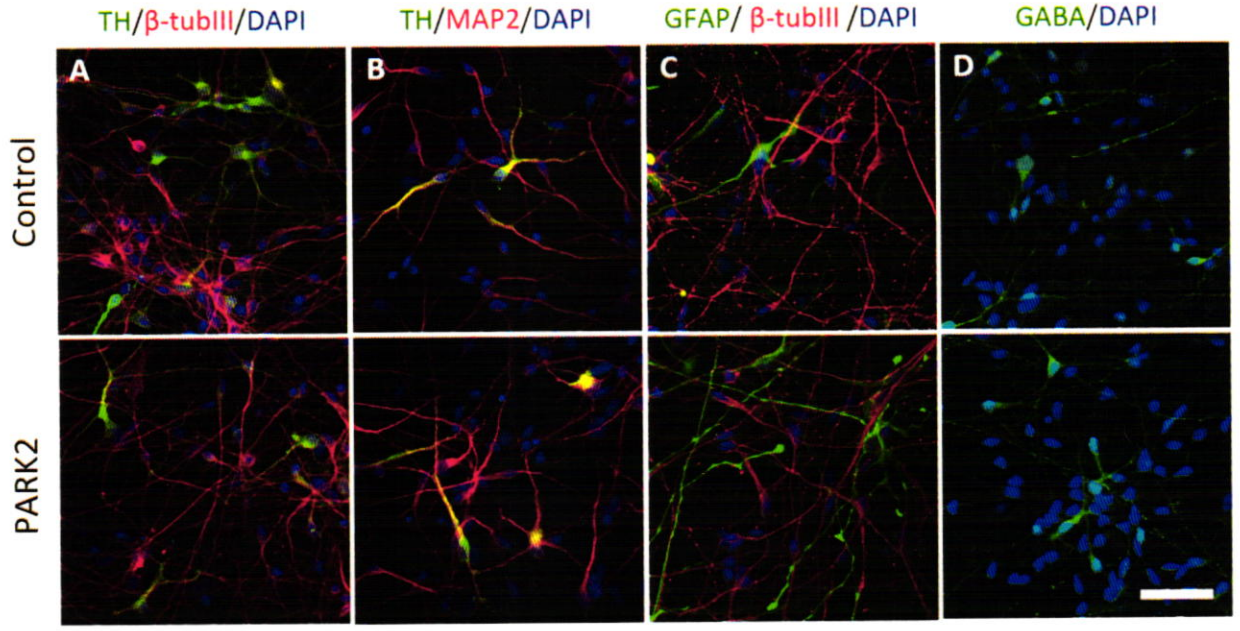
6. Highlights of work conducted

   i) Technique/expertise acquired :

- **Conducted research using human induced pluripotent stem cells (hiPSCs) with familial Parkinsons disease (PD)**
- **Validated the stem cell derived human dopaminergic neurons in hemi-parkinsonian rat model of Parkinson's Disease.**
- **Differentiation of neural stem cells and hiPSCs.**
- **Identification of molecular and biochemical signals regulating cell fate decisions, emphasizing the formation of functional dopaminergic neurons with mid brain characteristics in Parkinson's disease.**
- **Early cellular changes that underlie the onset of neurodegeneration in familial Parkinson's disease (cells with and without PARK2 mutations). This includes the use of isogenic stem cells in combination with advanced molecular assay.**
- **Characterization of stem cells derivatives by immunocytochemistry, western blotting, morphometry and quantitative analysis.**

ii) Research results, including any papers, prepared/submitted for publication :

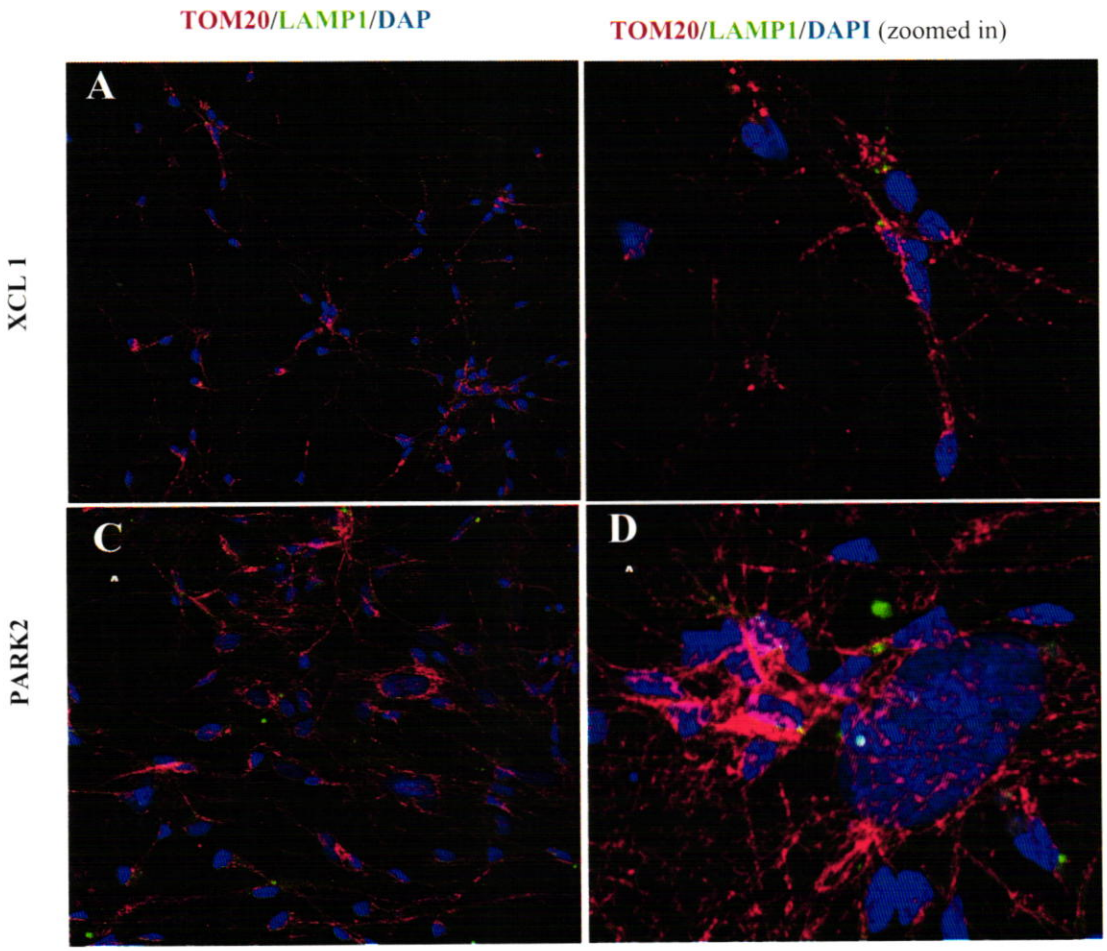
General characterization of XCL1 and PARK2 (PD) iPSC-derived neuronal cultures day 25 of differentiation



Markers used

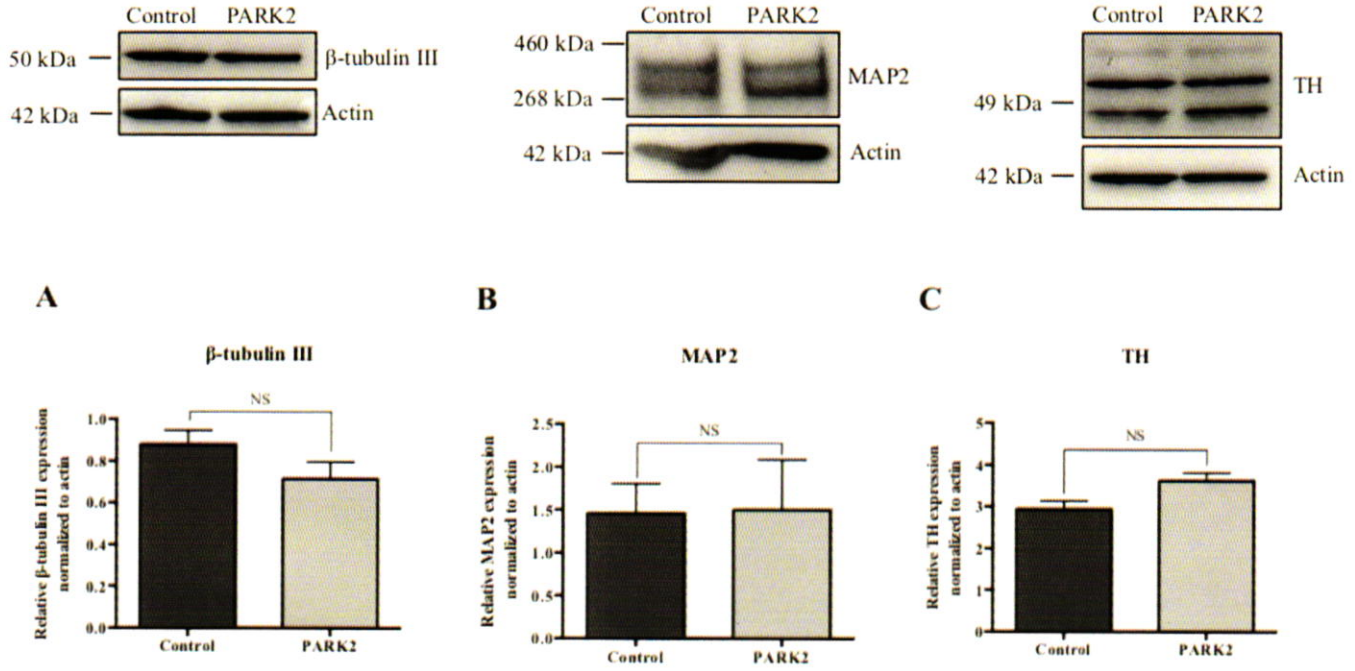
TH – marker for dopaminergic neurons;
TUJ1 – marker for newly formed neurons;
MAP2 – marker for mature neurons;
GABA – marker for GABA'ergic neurons;
GFAP – marker for astrocytes

Stainings visualising the mitochondria and lysosomes in iPSC-derived dopaminergic neurons with (C, D) and without (A,B) *PARK2* mutation. Accumulation of mitochondria and a swollen nuclei visible in *PARK2* mutated cell line (D).



Markers used:
TOM20 - Marker for the outer mitochondrial membrane;
LAMP1 - lysosomal marker.
DAPI - Marker for cell nuclei

Densitometry analysis of western blots for **ß-tubulin III, MAP2 and TH Protein expression in neuronal *PARK2* KD and isogenic control cultures.** ***PARK2*** KD and isogenic control iPSC-derived NSC cultures were differentiated for 25 days and western blotting performed to evaluate A) ß-tubulin III, B) MAP2 and C) TH Protein expression. Densitometry quantification of band intensities was performed using Image Lab software (Bio-Rad) and Protein expression levels were normalized to actin.



Prepared the review paper entitled **"Kynurenine pathway (KP)- A promising therapeutic target in Parkinson's disease** (under final revision) which highlights the

1. KP is one of the most promising metabolic pathways in PD, as its enzymes and metabolites act as modulators in metabolic levels of PD.
2. Studies on metabolites and molecular biomarker research are limited to detect PD in its early stage, thus emphasis on the development of novel KP based biomarker in PD.
3. Genetic studies related to KP would enlighten the inner mechanisms of pathogenesis in PD.
4. Targeting KP metabolites and enzymes is evolved as a best targeting approach in PD.

Figure 1: Overview of Kynurenine pathway

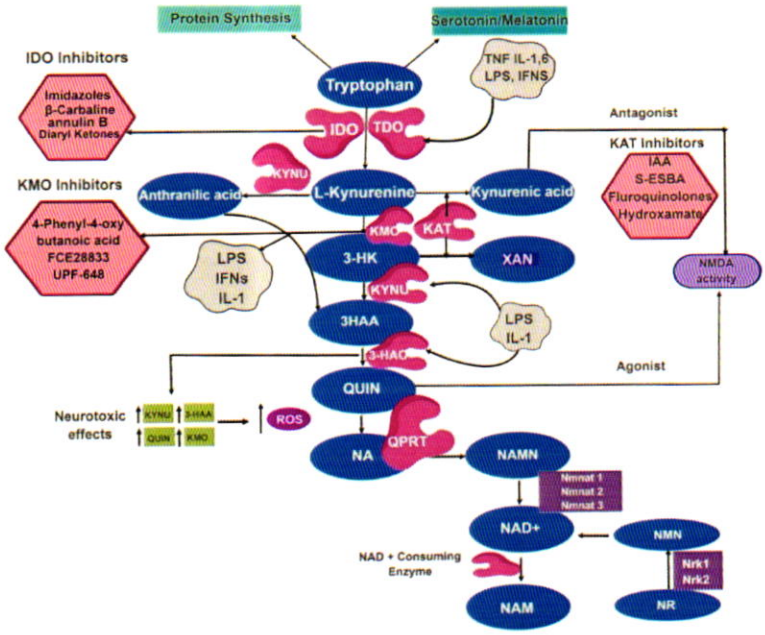
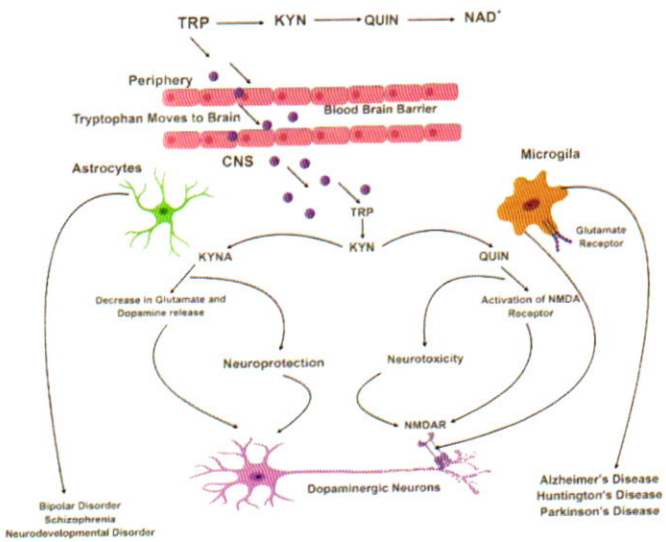


Figure 2: Effects of kynurenine pathway in periphery and central nervous system



Conference and Symposium

1. Participated the conference organized around the 2018 Brain Price Winner Prof.John Hardy **(13th March, 2019)** at University of Southern Denmark/Odense University Hospital, Odense, Denmark.
2. Participated the Symposium on Brain Inflammation (**12th April, 2019**) at the Institute of Molecular Medicine, SDU, Winsløws Vej 25, Odense, Denmark.

iii) Proposed utilization of the experience in India :

ICMR- IF is an excellent opportunity for me to become expertise in the area of stem cell research. I am very well trained in characterization and differentiation of neural stem cell and induced pluripotent stem cells (iPSCs). I am technically expertised in immunofluorescence, confocal microscopic techniques, western blotting and morphometric analysis. Through this gained experience, it is possible for me to establish the hiPSC technology in my parent institute (Bharathiar University, India). The knowledge I gained through this visit is enormous, which will bring out many innovative ideas in neuro- diseases which is applicable for current research and brings out innovative approaches in treatment options.

ICMR Sanction No. INDO/FRC/4 52N -47 1201 8-19-lHD Dated: 9th October, 2018

24/04/2019

Signature of ICMR-IF