April 12, 2022

BIOGRAPHICAL SKETCH

NAME: CHEN, Jun-An

POSITION TITLE: Associate Research Fellow

EDUCATION/TRAINING

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	MM/YY	FIELD OF STUDY
National Chung Cheng University, Taiwan	B.Sc.	1993-97	Chemistry
National Taiwan University, Taiwan	M.Sc.	1997-99	Biochemistry
University of Cambridge, UK	Ph.D.	2002-06	Developmental biology
Columbia University, USA	Postdoctoral	2007-12	Stem cell biology

A. Personal Statement

The focus of research in my laboratory is to elucidate how neurons establish individual identity in the developing nervous system and why only specific neuron subtypes are vulnerable in neurodegenerative diseases. We tackle these questions by studying **non-coding RNAs** and their roles during motor neuron (MN) generation and degeneration. My laboratory employs MNs generated from mouse and human embryonic stem cells (ESCs) and induced pluripotent stem cells (iPSCs), as well as mouse/chicken animal models to investigate MN development and disease. We have developed a series of stem cell lines and animal models to study the functions of microRNAs and lncRNAs by "gain-of-function" and "loss-of-function" approaches. Apart from elucidating the basic molecular mechanisms underlying neuronal diversity specification during central nervous system development, we use stem cells to study MN diseases. In particular, we are engaged in establishing patient-specific iPSC-based models of Spinal Muscular Atrophy (SMA) and Amyotrophic Lateral Sclerosis (ALS). We perform single cell multiomics on healthy and ALS iPSC-derived MNs to functionally characterize non-coding RNA pathologies in MNs. I am also a core member of the Neuroscience Program and RNA Program in Academia Sinica, which provide strong and stable consortia for inter-institutional collaborations. In summary, I employ multidisciplinary approaches, from *in vitro* stem cells to *in vivo* mouse models, to study MN development and degeneration.

1. Liau ES, Jin SQ, Chen YC, Liu WS, Calon M, Nedelec S, Nie Q*, **Chen JA*** (2022) Single-cell transcriptomic analysis unveils the diversity within mammalian spinal motor neurons. ***Nature Communications*** **14**, Article number: 46.
2. Li CJ, Liau ES, Lee YH, Huang YZ, Liu ZY, Willems A, Garside V, McGlinn E, **Chen JA***, Tian H* (2021) MicroRNA Governs Bistable Cell Differentiation and Lineage Segregation via a Noncanonical Feedback. ***Mol Syst Biol*** *(2021)17:e9945* **(Cover featured article)**.
3. Tung YT*, Peng KC, Chen, YC, Yen YP, Chang M, Thams S, **Chen JA***. (2019) Mir-17~92 Confers Motor Neuron Subtype Differential Resistance to ALS-Associated Degeneration. ***Cell Stem Cell*** Aug 1;25(2):193-209 **(Cover featured article)**. *This article has been recommended by **F1000** by Andrew Yoo: 2019. This article is highlighted by Academi a Sinica (English) (Chinese). the Academia Sinica Facebook. It is also featured in a series of newspapers, inc LibertyTimes, UDN, ChinaTimes, etc. Reported by international media: BioArt, Taipei Times, BioCentury, Asia Pacific Biotech News.*
4. Yen YP, Hsieh WF, Tsia YY, Lu YL, Liau ES ,Hsu HC, Chen YC, Liu TC, Chang M, Li J, Lin SP*, Hung JH*, **Chen JA***. (2018) Dlk1-Dio3 Locus-Derived LncRNAs Perpetuate Postmitotic Motor Neuron Cell Fate and Subtype Identity. ***eLife*** (DOI: 10.7554/eLife.38080).*This article is selected as a showcase for featured eLife Science Digests., and the "Biomedical Picture of the Day" by MRC UK.*
5. Li CJ, Hong T, Tung YT, Yen YP, Hsu HC, Lu YL, Chang M, Nie Q*, **Chen JA***. (2017) MicroRNA filters Hox temporal transcription noise to confer boundary formation in the spinal cord. ***Nature Communications*** *8. Article number: 14685 (2017). This article is highlighted by Academia Sinica (English)(Chinese), and on the Academia Sinica Facebook. It has also been featured in Asia Pacific Biotech News.*

B. Positions and Honors

Positions and Professional Experience

2019~present	**Associate Research Fellow:** Principal Investigator, Institute of Molecular Biology, Academia Sinica, Taiwan.
2012~2019	**Assistant Research Fellow:** Principal Investigator, Institute of Molecular Biology, Academia Sinica, Taiwan.
2007~2012	**Postdoctoral Fellow**: Supervised by Dr. Hynek Wichterle, Columbia University, USA.
2002~2006	**PhD student**: Supervised by Dr. Enrique Amaya, The Wellcome Trust/CR UK Gurdon Institute, University of Cambridge, UK.
2001~2002	**Teaching Instructor**: Institute of Biochemical Sciences, National Taiwan University, Taiwan.

Honors and Professional Activities

2021~2023	Academia Sinica Presidential Scholars Program (中研院特優學術研究獎)
2020~2025	MOST Frontier Science Research Program (科技部尖端研究計畫)
2020	MOST Outstanding Research Award (科技部傑出研究獎)
2019	Outstanding Alumni Award of National Chung Cheng University (中正大學傑出校友)
2019	The 15th TienTe Lee Award-Young Scientists, TienTe Lee Biomedical Foundation (李天德青年醫藥獎)
2018	Academia Sinica **Junior Research Investigators Award** (年輕學者著作獎)
2018~2022	Academia Sinica Career Development Award (前瞻計畫)
2012~2015	Taiwan National Science Council Talented Investigator Fellowship
2012~2014	Academia Sinica Investigator Fellowship (新聘學術獎)
2008~2009	Taiwan National Science Council Postdoctoral Fellowship
2002~2006	The Wellcome Trust Scholarship
2002~2005	Taiwan International PhD Scholarship (公費留考第一名)

C. Contribution to Science

1. **MicroRNAs choregraph patterning and circuit formation in the spinal cord**: The initial rostro-caudal (RC) patterning of the neural tube leads to differential expression of *Hox* genes that contribute to the specification of MN subtype identity. Using *in silico* simulation (in collaboration with Prof. Qing Nie's laboratory at the University of California, Irvine), we uncovered two feed-forward Hox-miRNA loops accounting for precocious and noisy Hoxa5 expression, as well as the ill-defined boundary phenotype of *Dicer* mutants. Moreover, we identified *mir-27* as a major regulator coordinating the temporal delay and spatial boundary of Hox protein expression. This mechanism offers a powerful strategy for achieving the precision and robustness of morphogen-mediated pattern formation. As a continuation of this study (in collaboration with Prof. Tian Hong's laboratory at the University of Tennessee, Knoxville), we applied several complementary approaches, including **1)** mathematical modeling of gene regulatory networks, **2)** single-cell RNA sequencing, **3)** embryonic stem cell differentiation simulation and **4)** microRNA (miRNA) genetic knockout mouse models to uncover the miRNA-mediated feedback mechanism underlying lineage choice of Hoxa5-Hoxc8 MNs at a tissue boundary in the mouse spinal cord. The corresponding new paradigm of miRNA-mRNA circuits is an emerging biological theme that can be applied to a wide range of systems. Our study elicited a fundamental change in how biological feedback loops are viewed as a paradigm shift in all fields of biology relating to miRNAs. In additional early neural patterning events, we also uncovered that mir-34 family ensure optimal sensory-motor circuit outputs.

    a. Li CJ, Hong T, Tung YT, Yen YP, Hsu HC, Lu YL, Chang M, Nie Q*, **Chen JA***. (2017) MicroRNA filters Hox temporal transcription noise to confer boundary formation in the spinal cord. ***Nature Communications*** *8. Article number: 14685 (2017).*

b. Li CJ, Liau ES, Lee YH, Huang YZ, Liu ZY, Willems A, Garside V, McGlinn E, **Chen JA***, Tian H* (2021) MicroRNA Governs Bistable Cell Differentiation and Lineage Segregation via a Noncanonical Feedback. ***Mol Syst Biol*** *(2021)17:e9945* **(Cover featured article)**.
c. Chang SH, Su YC, Chang M, **Chen JA***. (2021) MicroRNAs Mediate Precise Control of Spinal Interneuron Populations to Exert Delicate Sensory-to-motor Outputs. ***eLife*** (DOI: 10.7554/eLife.63768) **(Featured article)**.

2. **MicroRNA specifies motor neuron cell fate and maintains neuronal survival**: In addition to deciphering the role of miRNA during neural progenitor patterning, I also investigate the potential functions of miRNAs in postmitotic MNs. There are more than 60 subtypes of MNs positioned in different columns along the RC axis of the spinal cord, which are critical for the maintenance of body posture and coordination of complex movements. While transcription factors have been recognized as principal regulators of subtype specification, the role of posttranscriptional regulation has not been systematically established. To investigate whether miRNAs are globally involved in the regulation of postmitotic MNs, we performed a detailed analysis of MN subtype phenotype in mice in which the Dicer enzyme has been conditionally deleted in all postmitotic MNs. We found that despite an initial increase in the number of MN progenitors, disruption of Dicer function results in a loss of many limb- and sympathetic ganglia-innervating spinal MNs. Several miRNAs were uncovered to be responsible for these phenotypes, including mir-17~92 cluster. Our results provide evidence that miRNAs, together with transcription factors, are critical for the maintenance of MNs and for motor pool specification in the developing spinal cord.

a. **Chen J-A**, Huang Y-P, Mazzoni EO**,** Zavadil J**,** Tan GC**,** and Wichterle H. (2011) Mir-17-3p Controls Spinal Neural Progenitor Patterning by Regulating Olig2/Irx3 Cross-repressive Loop*,* ***Neuron****, 69(4). 721-35* **(Featured article)**.
b. **Chen J-A*** and Wichterle H*. (2012). Apoptosis of Limb Innervating Motor Neurons and Erosion of Motor Pool Identity upon Lineage Specific Dicer Inactivation, ***Frontiers in Neuroscience*** 6:69. doi: 10.3389/fnins.2012.00069. **(Featured article)**.
c. Tung YT, Lu YL, Peng KC, Yen YP, Chang M, Li J, Jung H, Thams S, Huang YP, Hung JH, **Chen JA***. (2015) Mir-17~92 Governs Motor Neuron Subtype Survival by Mediating Nuclear PTEN. ***Cell Reports***. 2015 May 20. 10.1016/j.celrep.2015.04.050. **(Cover featured article)**.

3. **Long non-coding RNA during neural development:** Although an extensive list of lncRNAs have been identified from several model organisms, it is still controversial if lncRNAs have critical functions in cell fate determination during neural development or degeneration. The major reason for this controversy remains unresolved is that most lncRNA studies are performed using *in vitro* cell lines. We thus performed a pioneering study to systematically identify lncRNAs (including poly A+ and poly A- lncRNAs, circRNAs, eRNAs, and antisense RNAs) from each stage of ESC-derived MN differentiation by strand-specific RNA-seq. We focused on detailed functional characterization of lncRNAs from the imprinted *Dlk1-Dio3* locus, given the high conservation of lncRNAs in this locus between mice and human. Our discovery of how lncRNAs in the *Dlk1~Dio3* locus regulate Hox expression and the robustness of Hox-mediated MN subtype diversification could shed light on analogous mechanisms of Hox-mediated regulation in other tissue types. Overall, our work represents one of the most comprehensive studies of the involvement of lncRNAs in motor neuron development via Hox regulation, with apparent broader impacts for other systems.

a. **Chen JA** & Conn S. (2017). Canonical mRNA is the Exception rather than the Rule. ***Genome Biology*** 18:133.
b. Yen YP, Hsieh WF, Tsia YY, Lu YL, Liau ES ,Hsu HC, Chen YC, Liu TC, Chang M, Li J, Lin SP*, Hung JH*, **Chen JA***. (2018) Dlk1-Dio3 Locus-Derived LncRNAs Perpetuate Postmitotic Motor Neuron Cell Fate and Subtype Identity. ***eLife*** (DOI: 10.7554/eLife.38080). **(Featured article)**.
c. Chen KW* & **Chen JA***. (2020). Functional Roles of Long Non-coding RNAs in Motor Neuron Development and Disease. ***Journal of Biomedical Science*** 27; Article number: 38.

4. **Develop new treatment avenues for motor neuron diseases**: The hallmark of neurodegenerative disease is that only selective cell types are susceptible to dying. Despite the tremendous efforts that have been made

in tackling this issue, the mechanism underlying this selective vulnerability remains puzzling. We adopted mouse/human embryonic stem cell differentiation approaches, together with an ALS-SOD1^{G93A} mouse model, to address this issue. We found that intrinsic MN-miRNAs can account for selective MN subtype degeneration. It was the first study to show that an MN-miRNA gene therapy is effective against neurodegenerative diseases, representing a superior approach to most studies that only test human ALS iPSCs.

a. Tung YT*, Peng KC, Chen, YC, Yen YP, Chang M, Thams S, **Chen JA***. (2019) Mir-17~92 Confers Motor Neuron Subtype Differential Resistance to ALS-Associated Degeneration. ***Cell Stem Cell*** Aug 1;25(2):193-209 **(Cover featured article)**.
b. Chen TH* & **Chen JA***. (2019). Multifaceted Roles of MicroRNAs: From Motor Neuron Generation in Embryos to Degeneration in Spinal Muscular Atrophy. ***eLife*** 2019;8:e50848.
c. Yen YP* & **Chen JA*** (2021). The m^{6}A epitranscriptome on Neural Development and Degeneration. ***Journal of Biomedical Science*** 28; Article number: 40.

Complete List of Published Work:

ORCID /PUBLON

D. Research Support

Ongoing Research Support

8/1/2020- 7/31/2025	Agency: MoST I.D.# 109-2326-B-001 -017 - Title: “Parsing the underlying molecular mechanism of long noncoding RNA mediated epigenetic regulation during neural development.” P.I.: Jun-An Chen
8/1/2020- 7/31/2023	Agency: MoST I.D.# 109-2314-B-001 -010 -MY3 Title: “Establishment of authentic microRNA biomarker for SMA disease progression and the potential gene therapy application.” P.I.: Jun-An Chen
1/1/2018- 12/31/2022	Agency: Academia Sinica I.D.# CDA-107-L05 Title: “Decipher the role of long non-coding RNA *Meg3* during motor neuron generation and degeneration.” P.I.: Jun-An Chen

Pending Research Support

1/1/2023- 12/31/2027	Agency: Academia Sinica Title: “Exploiting the mechanisms of RNA modification and developing new therapeutic avenues in ALS.” P.I.: Jun-An Chen

Completed Research Support in the last three years:

1/1/2020- 12/31/2021	Agency: Academia Sinica I.D.# AS-GC-109-03 Title: “Exploring and testing the application of microRNAs to counteract the aging process in vivo.” P.I.: Jun-An Chen
1/1/2019- 12/31/2021	Agency: NHRI I.D.# NHRI-EX108-10831NI Title: “Utilization of MicroRNA for Gene Therapy for Age Onset ALS”

P.I.: Jun-An Chen

8/1/2019-7/31/2020
Agency: MoST
I.D.# 108-2311-B-001-011 -
Title: “Using single-cell technique to decode the underlying mechanism for motor neuron development.”
P.I.: Jun-An Chen

8/1/2018-7/31/2019
Agency: MoST
I.D.# 107-2311-B-001-043-
Title: “Investigate the role of mir-34/449 during motor neuron development”
P.I.: Jun-An Chen