2016 International Chronobiology Summer School

Beijing, China *(August 1-6)*

INTRODUCTION

IDG/McGovern Institute for Brain Research at Peking University and Chinese Society for Biological Rhythms (CSBR) are hosting an international chronobiology summer school at Peking University, Beijing, China, on August 1-6.

The school will provide young investigators and graduate students the opportunities to learn the principles, the frontiers and the state of art techniques from the international leading scientists, and also provide the chances for mutual understanding among the participants, and develop future collaborations.

CONTENT

***** The registration is now closed, but the morning sessions (public lectures) are open to both registered and unregistered students. Welcome to join us!**

Morning Sessions (Open to both registered and unregistered students):

Public Lectures (L1 - L12)

Location: Dengyoucai Auditorium in Jinguang Life Sciences Building, PKU.

Afternoon Sessions (Open to registered trainees only):

Workshops (W1 - W3)

Location:
W1 - Conference Room 1710, Wangkezhen Building, PKU;
W2 - Luo lab, Room 511, Integrated Science Research Center, PKU;
W3 - Rao lab, Room 314, Wangkezhen Building, PKU.

Poster Session/Sponsors' Social hours

Location: Lobby in 1st floor, Jinguang Life Sciences Building, PKU.

Field trip

Location: PKU-Upenn Sleep Center, Peking University International Hospital

Evening Sessions

Discussions (D1 - D3)

Open to registered trainees only

Location: Conference Room 1113, Wangkezhen Building, PKU.

Meeting with the experts:
Open to Junior PIs only
Location: PKU-Upenn Sleep Center, Peking University International Hospital

Banquet
Open to registered trainees only
Location: PKU Global Village Hotel

INSTRUCTORS

- Joanna Chiu (*University of California - Davis*)
- Fang Han (*Peking University People's Hospital*)
- Samer Hattar (*Johns Hopkins University*)
- Qun He (*China Agricultural University*)
- Charlotte Helfrich-Förster (*University of Würzburg*)
- Ken-ichi Honma (*Hokkaido University*)
- Sato Honma (*Hokkaido University*)
- Carl Johnson (*Vanderbilt University*)
- Yi Rao (*Peking University*)
- Bill Schwartz (*University of Massachusetts - Worcester*)
- Xiaodong Xu (*Hebei Normal University*)
- Ying Xu (*CAM-SU Genomic Resource Center*)
- Erquan Zhang (*NIBS, Beijing*)
- Luoying Zhang (*Huazhong University of Science and Technology*)
- Yong Zhang (*University of Nevada-Reno*)

SCHEDULE

Day 0: Jul. 31, Sunday

12:00 - 17:00 **Registration**
Lobby, Building No.1, Zhongguanyuan Global Village, PKU

17:00 **Pick up by volunteer**
Lobby, Building No.1 & No. 9, Zhongguanyuan Global Village, PKU
Walk from the hotel to Nong Yuan Ding Hall

17:30 - 18:30 **Dinner**
Nong Yuan Dining Hall

18:45 **Assembly point**
In front of the north gate of Nong Yuan Ding Hall

19:00 **Campus tour**

Day 1: Aug. 1, Monday

08:20 - 08:30 **Opening remark**
Dr. Ying Xu
Professor, CAM-SU

08:30 - 10:00
L1. Introduction of Chronobiology
Dr. Bill Schwartz
Professor, University of Massachusetts - Worcester

10:30-12:00
L2. Ecology and Evolution of Clocks: Past, Present and Future
Dr. Carl Johnson
Professor, Vanderbilt University

13:30 - 16:30 **Workshops**
Group1 - W1
Topic: Demo of Chemical Oscillating; Fungal & Plants' Clocks
Dr. Carl Johnson
Professor, Vanderbilt University
Dr. Qun He
Professor, China Agriculture University
Dr. Xiaodong Xu
Professor, Hebei Normal University

Group2 - W2
Topic: Drosophila Behaviors: Locomotor & Beyond
Dr. Yong Zhang
Professor, University of Nevada - Reno
Dr. Joanna Chiu
Professor, University of California - Davis
Group 3 - W3

Topic: Lumicycle, & SCN Dissection
Dr. Sato Honma
Professor, Hokkaido University
Dr. Erquan Zhang
Professor, NIBS, Beijing

19:00 - 21:00 **D1. Discussion**
Topic: Clocks in the wild.
Dr. Charlotte Helfrich-Forster
Professor, University of Wuerzburg
Dr. Joanna Chiu
Professor, University of California - Davis

Day 2: Aug. 2, Tuesday

08:30 - 09:30
L3. Introduction of Entrainment: Pittendrigh, Daan and Aschoff
Dr. Ken-ichi Honma
Professor, Hokkaido University

09:45 - 10:45
L4A. Photoentrainment Pathways in Animals
Dr. Samer Hattar
Professor, Johns Hopkins University

11:00 - 12:00
L4B. Photoentrainment Pathways in Plants
Dr. Xiaodong Xu
Professor, Hebei Normal University

13:30 - 16:30 **Workshops**
Group 1 - W2
Group 2 - W3
Group 3 - W1

19:00 - 21:00 **D2. Discussion**
Topic: Other entrainments (temperature); Entrainment Problems
Dr. Carl Johnson
Professor, Vanderbilt University

Day 3: Aug. 3, Wednesday

08:30 - 09:30
L5A: Molecular basis of circadian rhythm generation I: TTFL in *Drosophila*
Dr. Yong Zhang
Professor, University of Nevada - Reno

09:45 - 10:45
L5B: Molecular basis of circadian rhythm generation II: TTFL in Mammals
Dr. Joanna Chiu
Professor, University of California - Davis

11:00-12:00
L6. Molecular basis of circadian rhythm generation III: New Perspectives
Dr. Carl Johnson
Professor, Vanderbilt University

13:30-16:30
Student Poster Session & Sponsors'/Social Hours
Please see *Appendix B* for the title and abstract of the posters.

17:30 - 20:30 **Banquet**
PKU Global Village Hotel

Day 4: Aug. 4, Thursday

8:30-10:00
L7. PDF and Drosophila clock circuits
Dr. Charlotte Helfrich-Forster
Professor, University of Wuerzburg

10:30-12:00
L8. The suprachiasmatic nucleus: A master circadian pacemaker
Dr. Sato Honma
Professor, Hokkaido University

13:30 - 16:30 **Workshops**
Group 1 - W3
Group 2 - W1
Group 3 - W2

19:00 - 21:00 **D3. Discussion**

Topic: Clock control of excitability

Dr. Sato Honma

Professor, Hokkaido University

Ken-ichi Honma

Professor, Hokkaido University

Day 5: Aug. 5, Friday

8:30-10:00

L9. Human circadian rhythms, Mutations, and Chronotypes

Dr. Ken-ichi Honma

Professor, Hokkaido University

10:30-12:00

L10. Circadian Mood Disorders

Dr. Luoying Zhang

Professor, Huazhong University Science & Technology

13:30 - 16:30 **Field trip**

PKU-Upenn Sleep Center, Peking University International Hospital

Dr. Fang Han

Professor, PKU People's Hospital

Dr. Ying Xu

Professor, CAM-SU

16:30-18:00

Young Chinese PIs meet with International Colleagues (CAU)

Bill Schwartz, Carl Johnson, Sato Honma, Ken-ichi Honma, Samer Hattar, Charlotte Forster, Joanna Chiu, Yong Zhang et al.

18:00-20:00

PIs meet with Chinese Sleep Society

Day 6: Aug. 6, Saturday

8:30-10:00

L11. Photoperiodism and Seasonality: Animals and Plants

Dr Bill Schwartz

Professor, University of Massachusetts - Worcester

Dr. Xiaodong Xu

Professor, Hebei Normal University

10:30-12:00

L12. A Brief History of Behavioral Neurosciences

Dr. Yi Rao

Professor, IDG/McGovern Institute for Brain Research at PKU

12:00-12:10 Conclusion Remarks

Dr. Erquan Zhang

Professor, NIBS, Beijing

CONTACT

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ORGANIZERS

IDG/McGovern Institute for Brain Research at Peking University;
Chinese Society for Biological Rhythms (CSBR);
PKU-Upenn Sleep Center, Peking University International Hospital;
Chinese Sleep Research Society;
NIBS, Beijng;
Nanjing University;
CAM-SU Genomic Resource Center.

Appendix A

Poster #1:

Kisspeptin displays sex-dependent metabolic and reproductive effects in a seasonal rodent

CÁZAREZ-MÁRQUEZ Fernando [1,2], LARAN-CHICH Marie-Pierre [1], KLOSEN Paul [1], KALSBEEK Andries [2] and SIMONNEAUX Valérie [1].

[1] *Neurobiology of Rhythms Department, Institute of Cellular and Integrative Neurosciences, Strasbourg, France*

[2] *Hypothalamic Integration Mechanisms, Netherlands Institute for Neurosciences, Amsterdam, The Netherlands*

Kisspeptin (Kp) is a hypothalamic neuropetide which increases GnRH neuron activity and peptide release. Our team reported that Kp expression displays melatonin-driven photoperiodic variation in seasonal rodents, and that chronic administration of exogenous Kp in short photoperiod (SP) adapted, sexually inactive Syrian hamster restores reproductive activity (Revel et al., 2006). In the male Siberian hamster (*Phodopus sungorus*), photoperiodic variation in Kp expression is region-specific, with higher level in the anteroventral periventricular area (AVPV) and lower level in the arcuate nucleus (ARC) in long photoperiod (LP) as compared to SP. Because Siberian hamsters not only show a reproductive inhibition but also a reduction in food intake and body weight when transferred to short day conditons, we investigated whether a kisspeptin chronic treatment (delivered by an osmotic minipump for 5 weeks) could restore reproductive and metabolic activities in SP-adapted male and female Siberian hamsters. Chronic central Kp administration reactivated both male and female reproduction as attested by a marked increase in testis and uterus mass in Kp- as compared to vehicle-treated animals. By contrast, food intake and bodyweight were significantly increased in male but not in the female hamsters, suggesting a sex-dependent effect of Kp on the central control of metabolic activity. We are currently analyzing the putative hypothalamic targets of Kp that may explain its sex-dependant differential effect on the Siberian hamster metabolism.

Poster #2:

Post-translational Modification of REV-ERB Nuclear Receptors and Transcriptional Regulation of Bmal1

KULIKAUSKAITE Justina, OHBA Yuki, TEI Hajime

Dept. Natural System, Faculty of Science and Technology, Kanazawa University, Japan

The key circadian transcription factor BMAL1 forms a heterodimer with CLOCK to drive rhythmical transcription of core clock genes in transcriptional-translational feedback loops. Transcription of *Bmal1* is activated and repressed by respective nuclear orphan receptors; RORs (RORα, β, and γ) induce the transcription of *Bmal1* and compete with the REV-ERB repressors (REV-ERBα and β) for binding to RORE within the *Bmal1* promoter. However, since both of the transcription of *Rors* (encoding transcriptional activators) and *Rev-erbs* (encoding repressors) are known to be activated by CLOCK/BMAL1 via the E boxes in their promoters and inhibited by PER/CRY, the precise dynamics of the periodic induction and repression of *Bmal1* transcription remain unclear.

In my research, I try to reveal the mechanism of the *Bmal1* regulation by examining post-translational modification of its transcriptional regulators. In this presentation, I describe the relationship between the phosphorylation of the transcriptional repressor REV-ERBs and the cellular localization, degradation, and hence *Bmal1* transcription.

Poster #3:

Crosstalk Between Circadian Clock and Brassinosteriod Signaling in *Arabidopsis*

Chenguang Zhang, Li Yuan, Xuan Cui, Liyan Shi, Hongya Xing, Min Gao, and Xiaodong Xu

Hebei Key Laboratory of Molecular and Cellular Biology; Key Laboratory of Molecular and Cellular Biology of Ministry of Education, College of Life Sciences, Hebei Normal University; Hebei Collaboration Innovation Center for Cell Signaling, Shijiazhuang, Hebei 050024, China

Endogenous circadian pacemaker synchronizes daily and seasonal behavior of organisms to the periodic changes of environmental signals, such as light, temperature, biotic/abiotic stress, soil nutrients, etc. Plant circadian oscillator is based on multiple interlocked transcription-translation feedback loops; however, the exact molecular mechanism is largely unknown. It has been reported that many hormone signaling, including auxin, ethylene, abscisic acid, cytokinin, gibberellin, jasmonic acid, salicylic acid, brassinosteroids and circadian clock are inextricably linked. Brassinosteroids (BRs) play a critical role in the plant growth and development. Exogenous application of brassinosteroid analogue could reset the clock-controlled circadian rhythm, but the mechanism is still unclear. Here we attempts to explore the crosstalk between the plant circadian clock and brassinosteroid signaling. Our data shown that the expression of both *DWF4* (*DWARF 4)* and *CPD* (*CONSTITUTIVE PHOTOMORPHOGENESIS AND DWARFISM*), core components in brassinosteroid biosynthesis, exhibits a circadian rhythm with a maximum around dusk. Light signaling and clock components, CCA1 (CIRCADIAN CLOCK ASSOCIATED 1), LHY (LATE ELONGATED HYPOCOTYL), TOC1 (TIMING OF CAB EXPRESSION 1), PRR5 (PSEUDO-RESPONSE REGULATOR 5), PRR7, PRR9 participate in the rhythmic expression of *DWF4* and *CPD*. Exogenous treatment of eBL (24- Epibrassinolide) affected the circadian rhythms, with the induced expression of *PRR9*. Taken together, clock-regulated expression of brassinosteroid biosynthesis genes *DWF4* and *CPD*, contribute jointly to the maintenance of circadian rhythms.

Key words: Circadian Clock, Brassinosteroid Signaling, Light Signaling, *Arabidopsis*

Poster #4:

Achilles is a circadian clock controlled gene that regulates innate immune function in Drosophila

Jiajia Li[1], Erin E Terry[1], Edith Fejer[2], Diana Gamba[1], Natalie Hartmann[1], Joseph Logsdon[1], Daniel Michalski[1], Lisa E Rois[1], Maria J Scuderi[2], Michael Kunst[3], and Michael E Hughes[1]

[1]Department of Biology, University of Missouri – St. Louis, St. Louis, MO 63121
[2]Department of Chemistry, University of Missouri – St. Louis, St. Louis, MO 63121
[3]Department of Genes - Circuits - Behavior, Max Planck Institute of Neurobiology, Martinsried, Germany 82152

The circadian clock is a transcriptional/translational feedback loop that drives the rhythmic expression of downstream mRNAs. Termed "clock-controlled genes," these molecular outputs of the circadian clock orchestrate cellular, metabolic, and behavior rhythms. As part of our on-going work to characterize key upstream regulators of circadian mRNA expression, we have identified a novel clock-controlled gene in *Drosophila melanogaster*, *Achilles (Achl),* which is rhythmic at the mRNA level in the brain and which represses expression of anti-microbial peptides in the innate immune system. *Achilles* knock-down in neurons dramatically elevates expression of crucial immune response genes, including *IM1* (*Immune induced molecule 1*), *Mtk* (*Metchnikowin*), and *Drs* (*Drosomysin*). As a result, flies with knocked-down *Achilles* expression are resistant to immune challenges from both gram-positive and gram-negative bacteria. Meanwhile, no significant change in core clock gene expression and locomotor activity is observed, suggesting that *Achilles* influences rhythmic mRNA outputs rather than directly regulating the core timekeeping mechanism. Notably, *Achilles* knock-down in the absence of immune challenge significantly diminishes the fly's overall lifespan, indicating a behavioral or metabolic cost of constitutively activating this pathway. Together, our data demonstrate that (1) *Achilles* is a novel clock-controlled gene, (2) *Achilles* links circadian clocks to regulation of the innate immune system, and (3) *Achilles* participates in circadian signaling from neurons to the fat body, a principal metabolic and immunological organ in flies.

Poster #5:

Circadian Oscillators are Intact in both Shoot and Root of Arabidopsis

Yue Li, Min Gao, Pengjuan Liu, Qiguang Xie and Xiaodong Xu

Hebei Key Laboratory of Molecular and Cellular Biology; Key Laboratory of Molecular and Cellular Biology of Ministry of Education, College of Life Sciences, Hebei Normal University; Hebei Collaboration Innovation Center for Cell Signaling. Shijiazhuang, Hebei, 050024, China

The transcription of circadian components shown tissue-specific in Arabidopsis. The root oscillators are relatively simple with only morning genes included. In our research, the tissue-specific expression of circadian clock is confirmed, since the period of circadian rhythm in root is longer than that in shoot as described before. Here, our data indicated that the transcriptional and translational expression of most evening genes oscillate robustly in root, such as TIMING OF CAB EXPRESSION 1 (TOC1), GIGANTEA(GI), EARLY FLOWERING 3 (ELF3) and PSEUDO-RESPONSE REGULATOR 5 (PRR5). Evening genes TOC1 and ZEITLUPE (ZTL) are essential to maintain the proper rhythms in both shoot and root. CIRCADIAN CLOCK-ASSOCIATED 1 (CCA1) could recruit to the EE region of TOC1 promoter, and evening complex components ELF3 and ELF4 show dynamic interaction in shoot and root. The entrainment by temperature, phase response curve and temperature compensation are functional in independent shoot and root. In conclusion, circadian oscillators are intact and independent in both shoot and root of Arabidopsis.

Poster #6:

The size matters: differential roles of FRQ protein isoforms in regulating the Neurospora circadian clock

Linzhang[1], Guobin Huang[1], Xianyun Chen[1], Jinhu Guo[1]*

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The FREQUENCY (FRQ) protein is a central component of the circadian clock in Neurospora crassa, which contains two isoforms arising from alternative splicing. The two FRQ isoforms, long FRQ (l-FRQ) and short FRQ (s-FRQ), l-FRQ has an additional 99 amino acids at N' terminus compared with s-FRQ. The ratio of l-FRQ and s-FRQ plays an important role in determining the periodicity and temperature compensation of the circadian clock, but the underlying mechanisms remain unclear. Here we show that splicing of frq exhibits rhythmicity. Two forms of FRQ act diversely in the positive and negative limbs of circadian clock. L-FRQ is more likely to take effect at the higher temperature, while s-FRQ at the lower temperature. In contrary to l-FRQ, s-FRQ was higher association with the white collar complex (WCC). The ability of l-FRQ to induce or promote WC-2 expression is more temperature-sensitive relative to s-FRQ. Both l-FRQ and s-FRQ support the phosphorylation of the WCC at lower temperature, while only l-FRQ at the higher temperature. Furthermore, l-FRQ proteins are phosphorylated and degraded much faster than s-FRQ. Stability analysis suggests that l-FRQ is likely to come into being a looser structure owing to the N'-terminal 99-aa region. Further analysis indicates that various regions of the N'-terminal 99-aa region of l-FRQ play differentially roles in the WCC phosphorylation and might contribute to the phosphorylation of FRQ C'-terminal distinctively. Taken together, these findings collectively suggest two FRQ isoforms play different roles in regulating the circadian clock.

Key words: Circadian clock; l-FRQ; s-FRQ; Phosphorylation

Poster #7:

Orexin signaling regulates both the hippocampal clock and the expression of Alzheimer's disease-risk genes

Zhixiong Ma[1, 2], Weiliang Jiang[3], & Eric Erquan Zhang[2]*

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Recent studies have revealed that Alzheimer's disease (AD) is a circadian clock-related human disease. However, it is not clear whether pre-symptomatic AD leads to circadian disruption, or whether reversing clock malfunction accelerates AD development. Here, with the real-time recoding the hippocampal slices ex vivo, we identified a functional oscillator that exists in the hippocampus. This oscillator receives input signals and releases output signals to maintain the hippocampal clock robustness. One of the most important inputs to the oscillator is orexin signaling, which can shorten the hippocampal clock and thereby regulate the expression of clock-controlled-genes (CCGs). A 24-h time course qPCR analysis followed by a JTK_CYCLE algorithm analysis indicated that a number of Alzheimer's disease-risk genes (AD-risk genes) are potential CCGs in hippocampus. Specifically, we found that beta-secretase 1 (BACE1) and beta-secretase 2 (BACE2), which are related to the production of the amyloid-beta peptide, are CCGs. BACE1 is inhibited by E4BP4 which is a repressor to D-box genes, while BACE2 is activated by the CLOCK: BMAL1 complex. Finally, we observed alteration of rhythmic expression patterns of the BACE2 and Apolipoprotein E (APOE) genes in the hippocampus of aged APP/PS1dE9 mice. Our results therefore indicate that the circadian oscillator in the hippocampus functions in linking orexin signaling to the development of AD.