Hacking our body clocks to optimize health

Katja A. Lamia, PhD
Associate Professor of Molecular Medicine
<table>
<thead>
<tr>
<th>Who is Katja Lamia?</th>
<th>I grew up in:</th>
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<tbody>
<tr>
<td><img src="image1.jpg" alt="Image of children" /></td>
<td>• Los Angeles, CA</td>
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<td><img src="image2.jpg" alt="Image of Washington, DC" /></td>
<td>• Washington, DC</td>
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<tr>
<td><img src="image3.jpg" alt="Image of South Bristol, ME" /></td>
<td>• South Bristol, ME</td>
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</tbody>
</table>
Who is Katja Lamia?

I was educated at:

- UC Berkeley
- BA, Physics
- Harvard
- PhD, Biophysics
- Salk Institute
- Postdoctoral with Ron Evans
Take Home Messages

Circadian clocks promote health

Clock disruption elevates risks

We are investigating why,
...to reduce risks for shift workers
Outline

History of circadian clocks research

My early work in circadian biology

Recent work in my lab at Scripps

Q & A with YOU!
We live in a rhythmic environment

light
warmth
dryness
dark
cold
moisture
A (Very) Short History of Circadian Clocks
Plant leaves exhibit daily movements

1880:
“The Movements of Plants” by Charles Darwin

Plant leaves move to maximize sun exposure
…and they *anticipate* sunrise
Algae emit bioluminescence at night.
California grunion anticipate high tide to lay eggs above the water line.
Human physiology is rhythmic

Greatest cardiovascular efficiency and muscle strength (17:00 a.k.a. 5:00 PM)

Lowest body temperature (04:30 a.k.a. 4:30 AM)

Highest body temperature (19:00 a.k.a. 7:00 PM)
Rhythmic cycles are internally driven.

Seminal experiments led by Colin Pittendrigh at Princeton in the 1960s described behavioral rhythms in rodents and provided strong evidence for internal clocks.

“Circadian” (circa = about, -dian = daily) is used to describe biological phenomena that exhibit ~24 hour oscillations independent of external timing cues.
Rhythmic cycles are genetically determined

1971: Ron Konopka & Seymour Benzer identify flies with altered circadian rhythms due to genetic changes

Seymour Benzer

This was the first demonstration that behavior can be controlled by a single gene!
Rhythmic cycles are genetically determined

The Nobel Prize in Physiology or Medicine 2017

Jeffrey Hall
Michael Rosbash
Michael Young

Benzer’s arrhythmic mutant “per⁰”
Behavior restored by fixing the “period” gene
Rhythms are driven by “feedback loops”

Electrical Circuit

Feedback Network (Frequency control) e.g. x 1/30

Positive Feedback with Closed Loop Gain = 1

Amplifier e.g. x 30

180° Phase shift

(http://www.learnabout-electronics.org/Oscillators/osc11.php)

Genetic Circuit

“central dogma” of biology:

Transcription

Factors

DNA "transcription"

RNA "translation"

Protein

Transcriptional repressors
Rhythms are driven by “feedback loops”

Electrical Circuit

- Feedback Network (Frequency control) e.g. x 1/30
- Positive Feedback with Closed Loop Gain = 1
- Amplifier e.g. x 30
- 180° Phase shift


Genetic Circuit

- Transcriptional repression
- PER
- CRY
- Per1, 2, 3, Cry1, 2
- Repression
- Activation

Clock Bmal1

Clock Bmal1
Light-emitting proteins let us watch biology

Luciferase (~100,000X)

https://pinoytransplant.com/2011/07/07/catching-fireflies/
Light-emitting proteins let us watch biology

Green Fluorescent Protein, “GFP”
Molecular clocks are everywhere.

Per-GFP

Plautz et al., Science 1997

Yoo, Yamazaki, et al., PNAS 2004
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Q & A with YOU!
Molecular clocks are everywhere

Proteins in mouse liver nuclei:

Time of Day
(hours after lights on)

noon midnight

0 0 4 4 8 8 12 12 16 16

AMPKα1
AMPKα2
CRY1
PER2
REVERBα
Lamin A

“A rose is not necessarily and unqualifiedly a rose; that is to say, it is a very different biochemical system at noon and at midnight.”

~ Colin S. Pittendrigh

Data from Lamia et al., Science 2009
Clocks drive rhythmic gene expression

Data from Casanova et al., Molecular Metabolism 2022

Data from Zhang et al., PNAS 2014

>50% of all genes are rhythmically expressed somewhere in the body
"La fixité du milieu intérieur est la condition d'une vie libre et indépendante"

("The constancy of the internal environment is the condition for a free and independent life")

~Claude Bernard (1813-1878)
Molecular clocks modulate physiology

Bmal1

PER
CRY

Clock
Bmal1

Repression

per1,2,3, cry1,2

Activation

Transcriptional repression

Lamia et. al., PNAS 2008

Bmal1\textsuperscript{lox/+} ; Cre\textsuperscript{+/-} (Liver KO)

Bmal1\textsuperscript{lox/+}

Liver

Bmal1\textsuperscript{+/-} ; Albumin-Cre\textsuperscript{+/-} (“Control”)

Bmal1\textsuperscript{lox/lox} ; Albumin-Cre\textsuperscript{+/-} (Liver KO)
Molecular clocks modulate physiology

Blood glucose

Zeitgeber time (hr)

** Relative expression

G6pt1

0 8 16 24

Relative expression

Liver

Lamia et. al., PNAS 2008
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Q & A with YOU!
Mismatch with natural cycles is detrimental

Internal (genetic) period

20 28

External (light-dark) period

20 28

Plant Circadian Clocks Increase Photosynthesis, Growth, Survival, and Competitive Advantage

Antony N. Dodd, Neeraj Salathia, Anthony Hall, Eva Kével, Réka Tóth, Ferenc Nagy, Julian M. Hibberd, Andrew J. Millar, Alex A. R. Webb
Disruption of natural cycles elevates risk
Disruption of natural cycles elevates risk

Time Zones:
Divergence from “solar time” is highly variable
Disruption of natural cycles elevates risk

**Time Zones:**

Westward divergence from “solar time” is associated with increased cancer risk

<table>
<thead>
<tr>
<th>Cancer/subgroup</th>
<th>Rate ratio (95% CI)</th>
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<tr>
<td>All cancers</td>
<td></td>
</tr>
<tr>
<td>Oral cavity and pharynx</td>
<td></td>
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<tr>
<td>Esophagus</td>
<td></td>
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<tr>
<td>Adenocarcinoma</td>
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<td>Squamous cell</td>
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<td>Stomach</td>
<td></td>
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<tr>
<td>Colon and rectum</td>
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<tr>
<td>Liver and intrahepatic bile duct</td>
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Fangyi Gu et. al., *Cancer Epidemiol Biomarkers Prev*; 26(8) August 2017
Disruption of natural cycles elevates risk

Shift Work
Shift work increases risk of some cancers

RE: Night Shift Work and Breast Cancer Incidence: Three Prospective Studies and Meta-analysis of Published Studies

Eva S. Schernhammer

A meta-analysis including dose-response relationship between night shift work and the risk of colorectal cancer

Xiao Wang¹, Alin Ji¹, Yi Zhu¹, Zhen Liang¹, Jian Wu¹, Shiqi Li¹, Shuai Meng¹, Xiangyi Zheng¹, Liping Xie¹

Does night-shift work increase the risk of prostate cancer? a systematic review and meta-analysis

Dapang Rao, Haifeng Yu, Yu Bai, Xiangyi Zheng, Liping Xie
We model shift work using altered lighting

Control (12:12LD)  Chronic Jetlag (CJL)
Chronic jet lag (CJL) enhances tumors in mice

Papagiannakopoulos et al., 2016

KRasG12D; p53−/−

Tumor Burden

Investigating Proposed Mechanisms of Shift

To gain more insight into these increased health risks resulting from chronically alternating light cycles, we focused on the proposed mechanisms linking shift work to cancer [3]. We analyzed clock (Per1, Per2, Bmal1) and clock-controlled (Dbp, c-Myc) gene expression in liver and corticosterone serum concentrations (Figure 3) to identify alterations and desynchronization among organ-specific clocks and/or between central and peripheral clocks. In line with behavior and CBT, Per1, Per2, and Dbp hepatic gene expression re-entrained within 7 days in

Figure 1. Peak Temperature Phases under Normal and CRD Conditions

(A–C) Peak temperature phases of p53R270H/+ WAPCre animals maintained under stable LD 12:12 conditions (closed circles) or weekly alternating light cycles (open circles) (n = 5 animals per group) before start of the light inversions (A), at the first LD inversion (B), and after 18 LD inversions (C). Per graph, subsequent days are plotted from top to bottom. Time of day on the x axis is expressed as external time (ExT), with ExT 0 corresponding with mid-dark. The upper axis indicates the ExT before the LD inversion. Values represent the mean ± SEM. Diamonds indicate the average temperature peak times of animals maintained under normal LD conditions. Data are presented as double plots to help with visualizing phase shifts (day 0 + 1, day 1 + 2, day 2 + 3, etc., on consecutive lines). Gray areas indicate darkness.

Figure 2. Long-Term Health Effects Resulting from CRD Exposure

(A) Relative body weight gain of p53R270H/+ WAPCre animals exposed to a regular LD cycle (closed circles, n = 20) or weekly alternating LD cycles (open circles, n = 21) in the longitudinal study. Note the significantly stronger weight gain of animals exposed to chronically alternating light cycles compared with animals maintained under a regular LD cycle in the longitudinal study (RM-ANOVA, group: F(1, 39) = 4.950, p = 0.0319; time: F(27, 1053) = 42.48, p < 0.0001; interaction: F(27, 1053) = 3.738, p < 0.0001). Values represent the mean ± SEM.

(B) Percentage of mice with palpable tumor in normal LD cycles (n = 20; closed circles) or chronic CRD conditions (n = 21; open circles). Black color indicates mammary gland tumor, whereas red color indicates other tumor types. See Table S1 for pathology data.

Van Dycke et al., 2011

Kettner et al., 2016

Papagiannakopoulos et al., 2016
We chose to study lung adenocarcinoma.

Marie Pariollaud

Papagiannakopoulos et al., 2016
We measure lung tumors after CJL

20 weeks exposure to lighting conditions

**LSL-KrasG12D** (mild lung tumor model)

**Control group**  **CJL group**

Count Lung Tumors
Circadian disruption increased lung tumors by 68%
We used RNA sequencing to ask WHY.
We used RNA sequencing to ask WHY

DESeq2 CJL vs 12:12LD (ZT9/ZT21 confound)

**Hsp = “Heat shock protein”**

CJL activates genes that are turned on by heat

These genes are controlled by a transcription factor called “Heat Shock Factor 1” (HSF1)
HSF1 daily drop is lost in CJL

**Graph:**
- X-axis: Time of day (ZT = hours after lights on)
- Y-axis: Relative intensity HSF1/LAMIN-A (Arbitraty unit)
- Two lines:
  - Black line: 12:12LD
  - Red line: CJL

**Images:**
- HSF1 and LAMIN A Western blots for 12:12LD and CJL conditions at different time points (ZT0, ZT4, ZT8, ZT12, ZT16, ZT20).
- The HSF1 daily drop is lost in CJL conditions compared to 12:12LD.
Does body temperature influence cancer risk?

Misaligned core body temperature rhythms impact cognitive performance of hospital shift work nurses

Hylton E. Molzof\textsuperscript{c}, Aoyjai Prapanjaroen\textsuperscript{a}, Vivek H. Patel\textsuperscript{b}, Mugdha V. Mokashi\textsuperscript{b}, Karen L. Gamble\textsuperscript{b,d}, Patricia A. Patrician\textsuperscript{a,e}

Characterizing the modern light environment and its influence on circadian rhythms

Dennis Khodasevich\textsuperscript{1}, Susan Tsui\textsuperscript{2}, Darwin Keung\textsuperscript{1}, Debra J. Skene\textsuperscript{3}, Victoria Revell\textsuperscript{4} and Micaela E. Martinez\textsuperscript{5}
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Pal Westermark
Christian Metallo
Kirk Lohmueller

Many Thanks!!!
THANK YOU TO OUR GENEROUS SUPPORTERS:

Kinship Foundation
Sidney Kimmel Cancer Research Foundation
Brown Foundation
Life Sciences Research Foundation
National Institutes of Health:
National Institute For Diabetes, Digestive & Kidney Diseases
National Cancer Institute
National Institute For Environmental Health & Safety

Many Thanks!!!
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