Arc1 functions in insulin producing cells to regulate sleep in *Drosophila*

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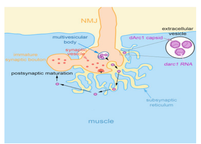
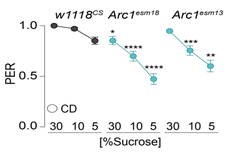
Abstract:

Neural regulation of sleep and metabolic homeostasis are critical in many aspects of human health. Despite extensive epidemiological evidence linking sleep dysregulation with obesity, diabetes, and metabolic syndrome, little is known about the neural and molecular basis for the integration of sleep and metabolic function. The gene Activity regulated cytoskeleton protein 1 (Arc1) has been linked to synaptic plasticity and metabolic function, which play crucial roles in sleep regulation and raise the possibility that it functions to control these processes. Here we characterize the effects of Arc1 on sleep duration. Flies lacking Arc1 significantly increase sleep duration by increasing the length of individual sleep episodes, raising the possibility that loss of Arc1 promotes deep sleep. The effects of Arc1 on sleep duration can be localized to neurons expressing the Drosophila insulin-like peptide (Dilp2), which has been previously implicated in the metabolic regulation of sleep depth. Silencing expression of Arc1 in these neurons significantly increases sleep duration, phenocopying Arc1 mutants, while overexpression of Arc1 significantly decreased sleep duration. We also find that Arc1 neurons are acutely required for increased sleep duration. A key hallmark of sleep depth in mammals and flies is a reduction in metabolic rate during sleep. We are currently investigating whether Arc1 functions in Dilp2-expressing neurons to regulate metabolic rate during sleep. Together, these findings will shed light on the role of Arc1 function in insulin-producing cells in sleep quality in Drosophila. Overall, this work contributes to our understanding of Arc1 function in the regulation of sleep.

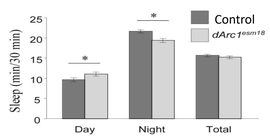
Background:



(Ashely et al 2018)



(Vaziri et al 2020)



(Mattaliano et al 2008)

Figure 1. *Activity-regulated cytoskeleton associated protein 1* (*Arc1*) is a regulator of synaptic plasticity and regulates a suite of behavioral traits.

Methodology:

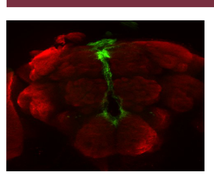


Figure 2. Dilp2 neurons were targeted using the Dilp2-GAL4 driver. The expression pattern of Dilp2-expressing neurons is visualized with GFP. Background staining is NC82 antibody (red).

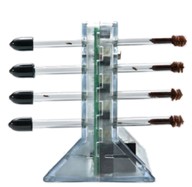


Figure 3. The Drosophila Activity Monitoring (DAM) system. Sleep was measured starting at ZT0 and averaged over 3 days (Pfeiffenberger *et al.,* 2010).

Results:

Loss of *Arc1* regulates sleep

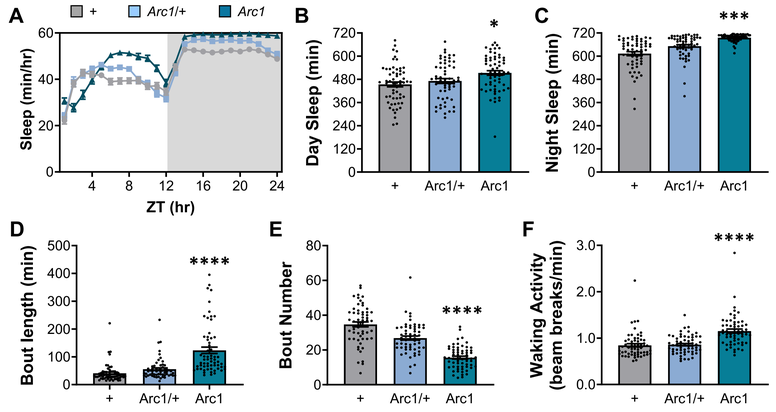


Fig. 4 Loss of *Arc1* promotes sleep. (**A-C**) Loss of *Arc1* significantly increases sleep duration during both the (**B**) day and (**C**) night. (**D,E**) Loss of *Arc1* consolidates sleep by increasing (**D**) bout length and decreasing (**E**) bout number. (**F**) Loss of Arc1 significantly increases waking activity.

Inactivation of *Arc1* neurons regulate sleep

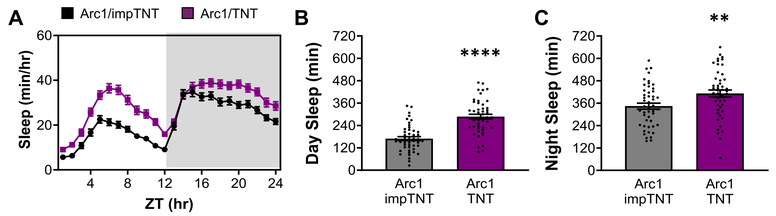


Fig. 5 Inactivation of Arc1 neurons promotes sleep. (**A-C**) *Arc1* neuron activation significantly increases sleep duration during both the (**B**) day and (**C**) night.

Arc1 functions in Dilp2 neurons to regulate sleep

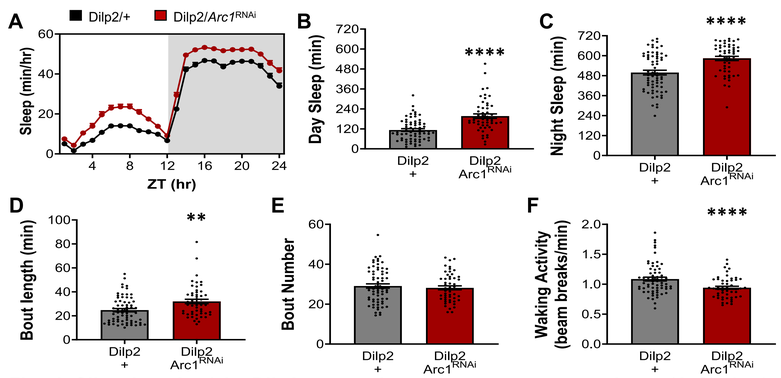


Fig. 6 Silencing *Arc1* in Dilp2-expressing neurons promotes sleep. (**A-C**) *Arc1* silencing significantly increases sleep duration during both the (**B**) day and (**C**) night. (**D,E**) *Arc1* silencing consolidates sleep by increasing (**D**) bout length, with no change in (**E**) bout number. (**F**) *Arc1* silencing significantly increases waking activity.

Results (continued):

Arc1 functions in Dilp2 neurons to regulate sleep

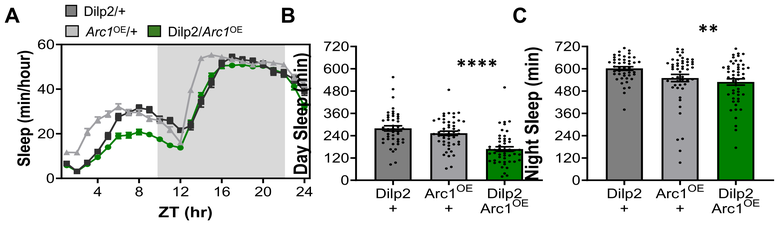


Fig. 7 Overexpression of *Arc1* in Dilp2-expressing neurons decreases sleep. (**A-C**) *Arc1* overexpression significantly decreases sleep duration during both the (**B**) day and (**C**) night.

Acute silencing of *Arc1* neurons promotes sleep

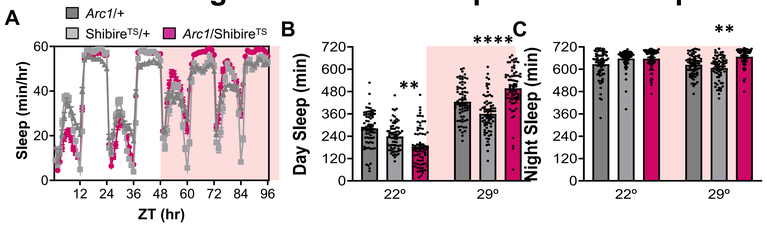


Fig. 8. Acute silencing *Arc1* promotes sleep. (**A-C**) Temporal silencing of *Arc1* neurons at the permissive temperature of 29°C increases sleep duration during the (**C**) day and (**B**) night.

Conclusion:

- Flies lacking *Arc1* have increased sleep duration and increased sleep consolidation.
- Knockdown of *Arc1* in *Dilp2*–expressing neurons increases sleep, while overexpression of *Arc1* decreases sleep, suggesting *Arc1* functions in insulin-producing neurons in a dose-dependent manner.
- Temporal silencing of *Arc1* neurons during adulthood promotes sleep, suggesting Arc1 functions acutely during sleep regulation.
- This work sets the stage for a more detailed investigation into the mechanism by which Arc1 functions in the insulin producing cells to regulate sleep.

References:

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Acknowledgements:

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