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### Sparse SCN Projections to the PVN Indicate Paracrine Signaling

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# SPARSE SCN VIP PROJECTIONS TO THE PVN INDICATE PARACRINE SIGNALING

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The suprachiasmatic nucleus (SCN) is the body's master circadian pacemaker. How the SCN communicates time-of-day information to the rest of the brain and body remains poorly characterized. My aim was to determine how the SCN communicates time-of-day information to the paraventricular nucleus of the hypothalamus (PVN), an upstream regulator corticosterone release, which has a 24-hour rhythm in the blood. Vasoactive intestinal polypeptide (VIP)-expressing neurons in the SCN have previously been shown to be crucial for coordinating rhythms both within the SCN and body but little is known about their projections to the PVN. I injected a Cre-dependent Brainbow virus unilaterally into the SCN of two VIP-Cre/+ mice and traced the projections within the PVN and sub-PVN. I found that a typical SCN VIP neuron sends divergent projections within the PVN and sub-PVN, but that the large majority of neurites (~75%) bifurcate only 0 to 1 times and that 85% of processes had two or fewer terminals in the PVN. Using a nearest neighbor analysis, I found that most terminals were within 40  $\mu\text{m}$  of each other. By tracing the projections to the ipsilateral and contralateral side of the brain, I also found that VIP neurons have high bilateral connectivity. To determine what cell types receive SCN VIP neuronal input, I double-labeled for SCN VIP neurons and PVN corticotrophin-releasing factor (CRH) neurons and found that SCN VIP neurons sent sparse projections to PVN CRH neurons. These results indicate that individual SCN VIP neurons each target a small population of cells in the PVN or sub-PVN. I concluded that VIP neurons communicate to the PVN through paracrine signaling.