Reply to Sharifpour et al. light response measurement of the human SCN by 7T fMRI

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PNAS LETTER

Reply to Sharifpour et al.: Light response measurement of the human SCN by 7T fMRI

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We appreciate that Sharifpour et al. (1) took the effort to start a scientific discussion and hope we can clarify issues. In their paper, it is argued that light response measurements of the SCN, as published by us (2), could have mainly originated from nuclei surrounding the SCN. The main reasoning was twofold: 1) The volume of interest (VOI, also referred to as 'mask') contained light-responsive hypothalamic areas that overruled the light response of the SCN, explaining the observed inhibition. 2) The SCN light response should have been an excitation, not an inhibition.

To start with the latter, the light response of suprachiasmatic nuclei (SCN) neurons is present in a subpopulation of all SCN neurons. In nocturnal rodents, the predominant response is an excitation while only few SCN cells respond with inhibition to light (Fig. 1). These responses appear to be driven by glutamate and Gamma-aminobutyric acid (GABA), respectively (3, 4). In contrast, the SCN of diurnal (day active) rodents exhibits relatively less excitations and more inhibitions, as shown by single-unit recordings (Fig. 1). Hence, the expectation of finding an excitatory response in the human SCN is not self-evident and we started without expectation in either direction. Of note, the BOLD response is an 'ensemble tissue level response' of the SCN as opposed to the single-unit recordings.

The choice for the anatomical location, defining our "Volume of interest", was correct according to Sharifpour et al. but large. Indeed, the VOI was 'large', compared to the SCN, and recording a light response from such a small nucleus was a challenge for which we had to adopt dedicated techniques (2). Most readily explainable is that we grouped the light response of all subjects after aligning them on the basis of the hypothalamic contours of each subject, rather than by routine grouping them on the basis of brain contour (Fig. 2). By doing so, we could perform a robust statistical analysis of the SCN, diluting the contribution to the response of possible adjacent areas included in the VOI (see Fig. 1, 3 E-I, Schoonderwoerd et al. (2)).

To what extent are areas immediately surrounding the SCN light responsive? Single-unit recordings in the hamster revealed that from a total of 50 individual SCN neurons, 28% was light responsive, while from 169 individual neurons outside the SCN, it was only 3% (5). Anatomical tracing studies in humans show that the SCN receives dense retinal input, while outside the SCN only few projections are found (8). Of note, the area of the SCN that was targeted by us was localized 1 mm more rostral than the area indicated by Sharifpour et al. resulting in fewer adjacent gray matter areas contributing to the response. On the basis of current studies, we strongly believe that the conclusion from our paper was the most likely one. We are confident that our recommendation to apply all colors of light (daylight) during the day and to refrain from all colors during the night is justified by the data.

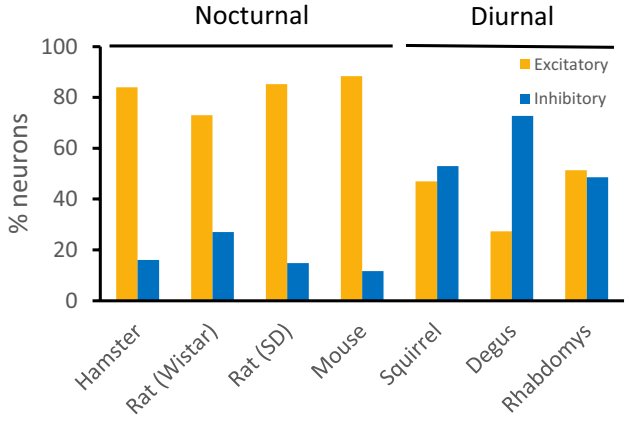

Fig. 1. Adapted from Schoonderwoerd et al. (3) Proportion of light-excited and light-inhibited cells in the SCN of nocturnal and diurnal species. Hamster and rat (Wistar) are from Meijer et al. (5); rat (Sprague–Dawley) and degus data are from Jiao et al. (6); squirrel data are from Meijer et al. (7); and mouse and Rhabdomys data are from Schoonderwoerd et al. (3). Hamsters, rats, and mice are nocturnal species. Squirrels, degus, and Rhabdomys are diurnal species.

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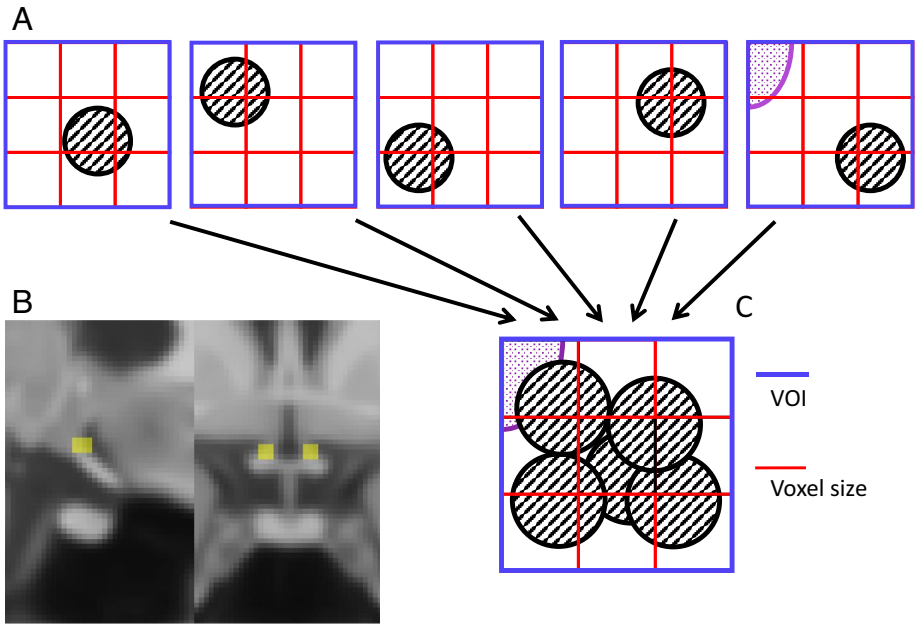

Fig. 2. Schematic drawing of the SCN (black-shaded circle) location (coronal view) within the VOI (*A*), based on the anatomical location of the yellow VOI containing the SCN (*B*) as shown in Schoonderwoerd et al. (2). The schematic drawing shows the between-subject variation in the location of the SCN. Five individual VOIs are schematically drawn in *A*, and in one of those, a small piece of a neighboring hypothalamic nucleus was included for illustration purposes (purple-shaded area). The contribution of this area, if present, will become diluted in the VOI. (*C*) Illustrates the result of grouping the light responses in the VOIs. The VOI size is 3 × 3 mm (in-plane). Our results were registered to the MNI152 template with a 1 × 1 × 1 mm resolution; therefore, the voxel-size in this schematic drawing is 1 × 1 mm. The estimated size of the SCN is 1.1 × 1.1 mm.

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