A View of the Clock - Analysis and Synthesis

"In solving a problem, the grand thing is to be able to reason backwards....In the everyday affairs of life it is more useful to reason forward. There are fifty who can reason synthetically for one who can reason analytically."

*Sherlock Holmes in 'A Study in Scarlet'*

"Life without synthesis........it's impossible"

*Maxim Gorky in 'My Universities'
Two basic propositions emerging from a review of the literature on circadian clock resetting, and also implied by the findings of the present study are: 1) the SCN clock shows differential sensitivity to stimuli, and 2) the SCN is a learning clock.

1. Differential sensitivity of the SCN clock:

There are two ways by which SCN regulates temporal sequencing and cycling of body functions: (a) by generating oscillatory output signals, such as neuronal firing rate (Inouye and Kawamura, 1979; 1982; Inouye, 1996; Meijer et al., 1996), and vasopressin secretion (Earnest and Sladek, 1986; Gillette and Reppert, 1987), which relay the information of phase of the clock as the time of day signal; and (b) by gating the clock's own sensitivity to incoming signals which adjust the clock timing (Gillette, 1996).

The incoming signals conveying environmental and/or organismic information can affect the clock timing only if they arrive during those specific phases of the circadian cycle when the clock recognizes them as 'error signals'. These error signals conveying the information of asynchrony between environmental or organismic state and the clock's time prompt resetting of the clock. Thus, the clock does a temporal filtering of signals accessing its time keeping system across the circadian cycle (Gillette, 1996).

There are three temporal domains during which the clock shows differential sensitivity and responses to various stimuli: day time domain, night time domain and dawn/dusk domain; and the stimuli which effectively reset the clock in one temporal domain may not do so in other domains (Gillette, 1996).

During the day, the SCN clock is sensitive to any manipulation of the pathways regulated by cAMP, but not at night since the molecular gate, thought to be opened by serotonin receptors (Medanic and Gillette, 1992; Prosser et al., 1994), through which the cAMP accesses the clock is closed at night (Prosser and Gillette, 1989; Gillette, 1996).
In the night time domain, both cGMP and Ca\(^{2+}\)/NO activated pathways access the clock; the former pathway is primarily gated by M\(_1\) muscarinic ACh receptors activated by carbachol or ACh (Liu and Gillette, 1996), and the latter by NMDA receptors activated by light or glutamate (Ding et al., 1994; Ebling, 1996).

However, in the time domain of (subjective)dawn or (subjective) dusk, the clock is relatively insensitive to manipulations of cAMP, cGMP and Ca\(^{2+}\)/NO pathways (Gillette, 1996). Instead, this domain is characterised by the clock’s sensitivity to melatonin (McArthur et al., 1991; Gillette and McArthur, 1996) and NPY (Shibata and Moore, 1993; Inouye, 1996). Thus, the clock has different “windows of sensitivity” (Medanic and Gillette, 1993).

In the present study, the results of dorsal raphe inactivation effect on the multiple unit activity (MUA) of SCN (Chapter III) and RHT neurotransmitter levels in SCN (Chapter IV) also imply a preferential sensitivity of the clock to serotonergic neurotransmission. Both MUA and ELISA studies produced results which were indirectly consistent with NPY activation of the SCN clock, and not conforming to serotonin depletion (due to dorsal raphe inactivation) as detailed in Ch.III and IV. These results could be due to reduced sensitivity of SCN and increased sensitivity of IGL to serotonin during daytime (Mason, 1986). Thus, these results imply differential sensitivity of the SCN clock to extrinsic stimuli, and complement the earlier findings suggesting preferential opening of the clock’s “windows of sensitivity”.

Another related aspect of the clock’s preferential sensitivity is its differential responses to a given stimulus across a circadian cycle. For instance, light causes a phase advance in the subjective dawn and a phase delay in the subjective dusk (Meijer and Reitveld, 1989; Schwartz, 1993). Phase response of the clock to various agents such as NMDA, substance P, somatostatin, 5-HT/agonist, GABA/agonist and NPY also varies across a circadian cycle (Inouye and Shibata, 1994; Inouye, 1996).

In the present study, the effect of a behavioural stimulus, REM sleep deprivation (a putative antidepressant treatment), was studied on the rhythms of RHT neurotransmitters, and GABA and monoamines of SCN. The results showed
(vide Ch.V) that the rhythms of SCN neurotransmitters had differential responses to REM sleep deprivation, possibly, at least partly, due to changes in the input to the clock (dorsal raphe, for instance). This suggests that the clock partly owes its differential sensitivity to the nature of the incoming inputs (signals).

2. SCN is a learning clock

2.1. SCN and learning

Learning is broadly defined as a modification of response resulting from experience (Thompson, 1967). Since application of light or glutamate (Meijer et al., 1988), 5-HT (Prosser et al., 1992) and NPY (Medanic and Gillette, 1993; Huhman and Albers, 1994) can alter the responsiveness of the SCN to the same stimuli at later times of the same 24 hr period (i.e., modification of response), SCN clock was suggested to have the ability of learning.

Later, the learning ability of the SCN clock was substantiated by the identification in SCN of cellular events normally associated with learning. These cellular correlates of learning identified in SCN are:-

(a) slowly activating and inactivating membrane conductances (Walsh et al., 1995; Bouskila and Dudek, 1995) which can make the cellular response depend on its recent history of activity (Marder et al., 1996); (b) phosphorylation of cAMP Response Element Binding (CREB) protein (Ding et al., 1997), considered as a part of the molecular switch required for consolidation of long term memory (Bailey et al., 1996); (c) induction of long term potentiation - LTP - by NMDA (Nisikawa et al., 1998), and long term depression - LTD - by NPY (van den Pol, 1996), considered as the cellular equivalents and bases of learning and memory; besides the proposed role of LTD as a mechanism of rhythmogenicity (O’ Donovan and Rinzel, 1997); and (d) neurons displaying burst firing (Meijer and Reitveld, 1989; Zhang et al., 1995), considered sufficient to produce LTP and LTD (Lisman, 1997).

In this study, DR inactivation lowered and phase advanced the firing rate (Ch.III), and reduced the glutamate level (Ch.IV) of SCN. This effect was not expected of 5-HT depletion due to DR inactivation (where a phase delay and an
increase in firing and increased glutamate level were anticipated); instead it was
equivalent to that produced by NPY activation (Medanic and Gillette, 1993; van den
Pol et al., 1996; Gribkoff et al., 1998). This preferential activation of NPY could be
due to the increased sensitivity to 5-HT of NPY neurons of IGL - compared to SCN-
during day (the time of DR inactivation in this study) (Mason, 1986). Since NPY is
known to induce LTD - one form of cellular learning - in glutamatergic neurons of
SCN (van den Pol et al., 1996), the observed effects could be the expression of
neuronal learning of SCN (vide Ch.IV and V), a possible advantage of preferential
NPY activation.

2.2. REM sleep and learning

Although REM sleep has been implicated in a variety of functions (vide 5.4.),
its involvement in the modification of learned responses (Crick and Mitchison,
1983; Winson, 1993; Hennevin et al., 1995) is probably the most important in view
of its role in adapting the animal to changes in the environment.

In foetus, young and adult mammals, spontaneous repetitive activation of the
CNS circuitry during REM sleep (hippocampal theta rhythm, for instance) are
considered to enhance and maintain the neural circuitries coding for phylogenetic
-inherited autonomic functions of the species - and ontogenetic - experiential
information such as learned responses of the individual - memories (Kavanau,
1996). This process of repetitive and spontaneous activation of ontogenetic and
phylogenetic circuitry is called “dynamic stabilization” (Kavanau, 1994).

The circadian period of all the individuals of a given species, normally, is
relatively constant - compared to phase - with minor variations (Davis and
Viswanathan, 1996), and hence can be considered as a phylogenetic memory.
However, the phase of the circadian rhythm varies across the population of a
species, depending on the individual’s prior experience, and therefore represents the
ontogenetic memory.

Since REM sleep significantly contributes to the dynamic stabilization of
neural circuits, the ontogenetic memory of circadian phase and the phylogenetic
memory of circadian period are possibly retained and consolidated by the modulatory action of REM sleep on the SCN pacemaker.

In this study, the possible modulatory action of REM sleep on the circadian phase (ontogenetic memory) of the SCN was investigated by estimating REM sleep deprivation induced changes in the rhythms of some neurotransmitters (Ch.V) which are considered to modulate SCN clock functioning.

The results showed that REM sleep deprivation had differential responses on the rhythms of neurotransmitters in the various input and the output pathways of the SCN: a phase advance in the RHT input i.e. Glu, SP, and a phase delay in the monoaminergic (except dopamine which showed a phase advance) and GABAergic inputs; and a phase advance in the VIP output (vide Table5.A). Therefore, one function of the REM sleep could be equilibrating the RHT input with the monoaminergic input, and thus to regulate the output of the SCN clock to produce an appropriately phased signal. This function is possibly modulated to a great extent by the serotonergic raphe nuclei. Thus, this study suggests that the ontogenetic memory of circadian phase is modulated by REM sleep.