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diauxic with GGE + 5 mM glucose (Figure 2b). Addition of glucose to GGE significantly suppressed growth of Alcaligenes sp. (Figure 2c).

Growth of B. subtilis (i) on GGE was high in presence of GGE + sucrose (Figure 3d) and was diauxic in GGE + 2.5 mM sucrose. In GGE + 5 mM sucrose Alcaligenes sp. showed diauxic growth (Figure 3c). C. albicans did not utilize GGE even in the presence of glucose or sucrose (Figure 2d, 3d).

It is of interest to record that two strains of B. subtilis from the same forest soil had contrasting characteristics with respect to utilization of GGE. The inability of B. subtilis (ii) and C. albicans to utilize GGE suggests that they lack the capacity to produce the enzymes required for utilization of the compound.

Attempts have been made to enhance the degradation of lignin model substances by using additives such as succinate, glucose and cellulose. Munakata et al. reported that glyceral increased the degradation of 3-(2-methoxy-4-formylphenox)-1,2-propanediol by Flavobacterium feruginium and F. ringense. In contrast, cell yield of Alcaligenes sp. and B. subtilis (i) was higher when grown on GGE alone than when grown on glucose.

Low levels of glucose and high levels of nitrogen allowed rapid release of [14C]-CO2 from methoxy groups of vanillic and syringic acids. Pseudomonas tabaci, P. putida and Rhodotorula mucilaginosa cells grown on glyceral or glucose oxidized aromatic substances only after a prolonged lag because of enzyme repression. The diauxic pattern we have observed must be due to utilization of the easily metabolizable substrate glyceral or sucrose initially, followed by utilization of GGE.


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Ethanol injection into the suprachiasmatic nuclei disrupts the day night feeding rhythm in the Wistar rat

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Effect of ethanol injection on the circadian rhythm of food intake in Wistar rats was investigated. Ethanol was injected into the bilateral suprachiasmatic nuclei (SCN) through a chronically implanted cannula. The circadian rhythm of food intake was disrupted by injections during the beginning of light and dark (LD) phases of the LD cycle, with no significant change in total daily food intake. Increase or decrease in concentration of ethanol did not produce any change in the extent of disruption of the circadian rhythm of food intake. It is suggested that, by inhibiting vasopressin function, ethanol altered the circadian rhythm of food intake.

Under normal environmental conditions the light-dark (LD) cycle acts as a zeitgeber. Difficulties in the entrainment of various circadian rhythms. In the absence of the environmental cues these endogenously generated circadian rhythms free run with a period of approximately 24 h (ref. 1). Entrainment by periodic stimuli in addition to the LD cycle is also possible. Certain chemicals can be a source of such periodic stimuli and act as a non-photic zeitgeber, though their ability to modify the circadian rhythms varies, from mimicking the effect of light by carbachol to slowing or dissociating of circadian rhythms.

Ethanol, which is one of the inhibitors of vasopressin (VP) function, has been found to lengthen the endogenous tidal rhythms in isopods and to lengthen the period of locomotor activity rhythm in hamsters. VP has been identified as a putative neurotransmitter at the suprachiasmatic nuclei (SCN) and it is likely to be part of the control of various circadian rhythms. The objective of the present study was to investigate the effects of ethanol injection into SCN on the circadian rhythm of food intake in the rat.

Wistar rats weighing 200-300 g were housed in individual cages. The temperature of the animal room was kept within 22 ± 2°C. The lighting regime consisted of 12:12 LD cycle with lights on from 0900 h to 2100 h. All animals had a minimum of 7 days prior to the start of the experiment. Food and water were given ad libitum.

Under nembutal anaesthesia (50 mg/kg), intraperitoneally 22-gauge stainless steel guide cannulae were chronically implanted in the midline to deliver injections into SCN bilaterally. The stereotaxic co-

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ordinates were AP on bregma, L = 0 and V = 8.6 ± 1 (ref. 7). To deliver injection into the area near SCN, cannulae were implanted with the same co-ordinates except that AP was 0.5 mm posterior to bregma.

One group (group I) of 9 rats received injections of iso-osmolal ethanol (1.6%) at the beginning of light phase (9 am local time, 0 h circadian time) of the LD cycle, while another group (group II, n = 5) received the injections at the beginning of dark phase (9 pm local time, 12 h circadian time). A third (group III, n = 6) and fourth (group IV, n = 5) groups received injections of 1 and 2% ethanol respectively at the beginning of light phase. Two other groups served as controls: one (group V, n = 8) received 0.9% saline (1 μl) and the other (group VI, n = 5) ethanol (1.6%) into the area near SCN, also at the beginning of light phase.

Food consumption measurement started 48 h after surgery and was continued till the end of the experiment. A measured quantity of food was presented to the animals at the beginning of each light (9 am local time, 0 h circadian time) and dark (9 pm local time, 12 h circadian time) phase. Food remaining at the end of each phase was weighed and consumption was calculated. On day 7 post-operation 1 μl of ethanol was injected into SCN at the beginning of light or dark phase, depending on the group, at the rate of 0.1 μl/min; rate of injection of saline was the same. To avoid ambiguity of results due to chemical and morphological changes at the site of injection each animal had only one injection6. The site of injection was stained using ferric chloride. Details of the procedure adopted for injection and histological sectioning are mentioned elsewhere9–11.

Statistical analysis was performed using Student’s paired t test.

Table 1 shows changes in food consumption pattern as a result of ethanol injection into SCN. After ethanol injection into SCN at the beginning of light phase (group I) the ratio of food consumed in light phase to that in dark phase increased (columns 2, 3 and 4, 5 in Table 1). Injection at the beginning of dark phase (group II) also had a similar effect. The change in the LD ratio was statistically significant (P < 0.001) only for the light and dark phase immediately following ethanol injection (see columns 6 and 7). Saline injection into SCN (group V) and injection of ethanol into the area near SCN, (group VI) had no effect on the pattern of food intake. Injection of 1% and 2% ethanol (groups III and IV) also disrupted the pattern of food intake to almost the same extent as that observed with 1.6% ethanol. Total daily food intake remained fairly constant throughout the experiment in spite of the change in the LD ratio.

Lesions of SCN, the neural generator of circadian rhythms, are known to produce changes in many circadian rhythms12, including feeding rhythms13. Partial lesions of rostral and dorsomedial regions of SCN have been reported to alter the circadian rhythm of food intake14. These regions of SCN contain exclusively vasopressinergic neurons, which happen to be the largest among the peptide-containing neurons of SCN15.

Ethanol is a known central suppressant of arginine vasopressin (AVP)16 and inhibits VP function4. It has been reported by Eisenhofer et al.17 that acute ethanol ingestion impairs memory, probably through ethanol-induced inhibition of VP release. There are also other reports18,19, which have given rise to the concept that VP secretion is suppressed by ethanol.

The results of this study indicate inhibition of VP function by ethanol as evidenced by disruption of food intake. This disruption is quite likely since VP is essential at SCN for the integration and expression of normal circadian rhythms20. Moreover it was recently reported5 that the V1 receptor antagonist d(CH2)5-

<table>
<thead>
<tr>
<th>Group</th>
<th>Before injection</th>
<th>24 h after injection</th>
<th>48 h after injection</th>
</tr>
</thead>
<tbody>
<tr>
<td>L</td>
<td>D</td>
<td>L</td>
<td>D</td>
</tr>
<tr>
<td>I</td>
<td>5.1 ± 1.1</td>
<td>14.0 ± 1.5</td>
<td>9.5 ± 1.0</td>
</tr>
<tr>
<td></td>
<td>(36.0 ± 0.96)</td>
<td>(64 ± 0.56)</td>
<td>(47.4 ± 0.54)</td>
</tr>
<tr>
<td>II</td>
<td>3.4 ± 0.2</td>
<td>8.5 ± 0.9</td>
<td>5.2 ± 0.5</td>
</tr>
<tr>
<td></td>
<td>(26.2 ± 1.92)</td>
<td>(71.7 ± 1.92)</td>
<td>(45.0 ± 0.77)</td>
</tr>
<tr>
<td>III</td>
<td>2.9 ± 0.2</td>
<td>8.1 ± 0.5</td>
<td>5.4 ± 0.2</td>
</tr>
<tr>
<td></td>
<td>(27.3 ± 2.6)</td>
<td>(72.7 ± 2.6)</td>
<td>(44.5 ± 0.91)</td>
</tr>
<tr>
<td>IV</td>
<td>3.8 ± 0.3</td>
<td>8.3 ± 0.6</td>
<td>5.5 ± 0.04</td>
</tr>
<tr>
<td></td>
<td>(31.5 ± 1.4)</td>
<td>(68.3 ± 1.4)</td>
<td>(46.7 ± 0.99)</td>
</tr>
<tr>
<td>V</td>
<td>5.9 ± 0.63</td>
<td>13.3 ± 1.42</td>
<td>5.6 ± 0.5</td>
</tr>
<tr>
<td></td>
<td>(30.7 ± 0.68)</td>
<td>(69.3 ± 0.7)</td>
<td>(30.2 ± 1.5)</td>
</tr>
<tr>
<td>VI</td>
<td>3.9 ± 0.2</td>
<td>8.1 ± 0.3</td>
<td>5.4 ± 0.2</td>
</tr>
<tr>
<td></td>
<td>(32.0 ± 0.75)</td>
<td>(68.0 ± 0.7)</td>
<td>(30.8 ± 1.4)</td>
</tr>
</tbody>
</table>

Values are means ± SE.
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Tyr (Me)AVP disrupts the circadian rhythm of food intake when injected into SCN. The disruption seen in this study was transient and was neither photoperiodic nor dose-dependent.

VP has been implicated in many of the central integrative processes in addition to its classical role in water and electrolyte metabolism. Recently it has been suggested that VP may have a role in stress-induced feeding as well. The understanding of its role in the control of circadian rhythms has come far from studies in brattlebore rats only, which are deficient in VP synthesis. However, the presence of a separate neuroanatomical system responsible for the circadian cerebrospinal fluid VP, as suggested by Schwartz and Reppert, and its effective insulation from osmotic regulation of blood VP makes this peptide important in circadian time-keeping.

In view of the report that ethanol can alter the electrical activity of some brain areas, in this study it was found that ethanol injection may also alter the activity of SCN neurons and thereby disrupt the circadian rhythm.


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Embryotoxicity of RU 486 in English albino rabbit, Oryctolagus cuniculus

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The synthetic steroid RU 486, when administered orally at daily doses of 6.4 mg, 32.0 mg, and 160.0 mg rabbit (low, high and toxic dose respectively) during the period of organogenesis to CDRI colony-bred ad female rabbits, caused 100% resorptions in all test groups. Control animals had no resorptions.

RU 486, a synthetic progestosterone-receptor blocker, was observed to be effective for termination of pregnancy. It is advisable that a distinction be made between extended pharmacological effect and its teratogenic effect on the embryo. This communication reports the results of an embryotoxic evaluation of RU 486.

Colony-bred adult nulliparous female rabbits were mated to bucks of proven fertility. Copulation was confirmed by the presence of sperm in the vagina and the day when sperm was noticed was designated day zero of pregnancy. Mated rabbits were divided into 4 groups of 5 animals each and the compound was administered orally on day 6 to day 15 post-coitus as follows: group I, control; group II, low-dose group, contraceptive dose (CD), 6.4 mg/rabbit/day; group III, high dose group, CD × 5; group IV, toxic-dose group, CD × 25, 1600 mg/rabbit/day.

Body weight of all animals was recorded on days 1, 14, 21, 28 and 30 post-mating. On day 30 post-coitus caesarean sections were performed on all animals at the number of corpora lutea; number of implantation sites, number of resorptions, number of live/dead foetuses; size, weight and gross abnormality of each foetus; viability, growth deformities and newborns were recorded.

Half of the foetuses were fixed in Bouin's solution and were examined for visible abnormalities by sectioning method. The remaining foetuses were cleared in 1% KOH solution and stained by Dawson Alizarin Red technique for visualization of ossification defects.

None of the mothers showed any noticeable deviation in food intake throughout the experimental period. There was no mortality in any of the groups. There was steady gain in body weight of all animals of all groups.

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