mimicked in various heart muscle preparations by the administration of cyclic AMP derivatives. In guinea-pig atria, 8-bromo-cyclic AMP in particular exerts a strong positive inotropic effect[9], whereas dibutyryl cyclic AMP is ineffective[19]. Although the effect of catecholamines on the myocardial contractile force is mimicked by 8-bromo-cyclic AMP, the efflux rate of ^{42}K was not enhanced but significantly reduced in the presence of the cyclic nucleotide (τ changed from 37.6 min under control conditions to 43.1 min in the presence of 8-bromo-cyclic AMP; figure 3). The reduction of ^{42}K efflux by 8-bromo-cyclic AMP was unexpected, but this result is in line with the observation that the action potential duration is longer in the presence of dibutyryl cyclic AMP than in the presence of noradrenaline[20]. It is also interesting that in preparations from reserpinized animals dibutyryl cyclic AMP caused inhibition of ^{42}K uptake while noradrenaline had a stimulatory effect[21].

One of us (H.N.) has found that the phosphodiesterase inhibitor papaverine decreases the ^{42}K efflux under the same conditions as described for 8-bromo-cyclic AMP. It remains to be established whether or not these effects are related to the intracellular accumulation of cyclic AMP.

The results show that β-adrenoceptor stimulation by isoprenaline enhances an activity-dependent potassium conductance in the heart. The augmented potassium efflux probably reflects an increase in the current called i_x during the cardiac action potential and may help to provide a shorter systole in the presence of catecholamines. This effect of catecholamines and especially of isoprenaline is not mimicked by 8-bromo-cyclic AMP. It seems possible, therefore that some of the effects of β-adrenoceptor stimulation in the heart are not mediated by the intracellular accumulation of cyclic AMP and that further pathways for the action of catecholamines must be sought.

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Circadian rhythm of β-glucuronidase[1]

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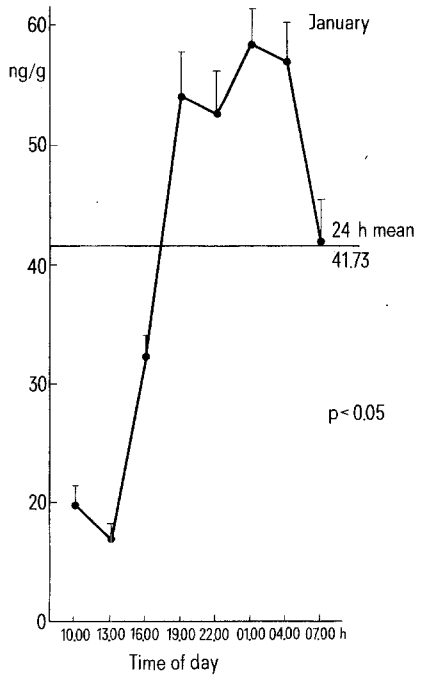
Summary. The circadian rhythm of rat liver β-glucuronidase has been studied in animals kept under highly standardized laboratory condition. A clear 24 h rhythm has been observed for this enzyme with a peak activity at 01.00 h and a trough at 13.00 h.

The circadian rhythm for the lysosomal acid phosphatase has been reported by Bhattacharya and Mayersbach[2]. Furthermore, histochemical studies using the lysosomal marker enzymes (acid phosphatase and β-glucuronidase) have established a rhythmic pattern in the shape, size and distribution of liver lysosomes[3]. These observations suggested a quantitative study of the β-glucuronidase activity of rat liver. The present communication confirms the presence of a circadian rhythm in liver β-glucuronidase activity and discusses the possible functional correlation with the body's detoxicating mechanism.

Materials and methods. Male Wistar rats of HAN breed (Institut für Versuchstierzucht, Hannover, Federal Republic of Germany) were used. For 6 weeks prior to the study, the animals were housed in a room maintained at 22 °C with 55% relative humidity. The room was illuminated artificially from 06.00 to 18.00 h and dark for the next 12-h period. Food and water were supplied ad libitum. The experiment was carried out in the month of January.

The animals were sacrificed at 3-h intervals during an uninterrupted 24-h period. The livers were removed from the animals and weighed immediately. 1 g liver tissue from each animal was homogenized in 0.9% NaCl with an 'ultra turrax' homonizer. The samples were centrifuged at 10,000 × g for 30 min and the β-glucuronidase was estimated according to the method of Fishman[4]. The unit of glucuronidase activity was expressed as nm/g liver tissue.

Results and discussion. As shown in the figure, there is a circadian rhythm in the activity of liver β-glucuronidase ($F = 0.01$). The enzyme activity remains high during the dark phase of the day and goes down after the onset of light i.e. at 07.00 h. The maximum activity ($58.66 + 6.76$) occurred at 01.00 h and the minimum ($16.84 + 2.57$) at 13.00 h. When the lowest and highest means were compared they were statistically different (0.05, t-test). The 24 h mean value was $41.73 + 17.08$. The daily rhythmic variation in the activity of acid phosphatase has been noted in several organs[5] and also in mice and rat liver respectively[2,6]. The peak activity for acid phosphatase in rat liver was observed at 19.00 h and the nadir at 13.00 h[2]. No report has so far been noted on the circadian rhythm of β-glucuronidase activity. When this enzyme activity is compared with acid phosphatase activity, it can be seen that the lowest activity for both the enzymes occurred at the same time (13.00 h) but the peak differed. The liver lysosomes are reported as heterogenous in nature[7]. It has also been reported that the



Circadian variation in β-glucuronidase activity in male rat liver. The activity is expressed in ng/g liver tissue. Each point represents the data from 6 animals with $\pm$ SD.

distribution patterns of all lysosomal enzymes are not identical[7]. This also holds good when the histochemical distribution patterns of these enzymes are compared[3]. Though both acid phosphatase and β-glucuronidase are accepted as the marker enzymes for the lysosomal study, it can be seen from the present study that their activity pattern during a 24-h period is quite different. This confirms once more the heterogenous nature of lysosomes. Therefore the study of lysosomal function through one of the lysosomal enzymes might not give a clear picture of their functional role. Moreover, as the main function of the lysosomal enzymes is to detoxicate toxic materials, the circadian variation in lysosomal enzyme activity is of considerable importance from this point of view. The temporal oscillation of the enzyme activities might influence the body's detoxicating mechanism.

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Modifications of female sex chromatin (Barr body) in rat neurons after reserpine administration[1]

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Summary. 3 groups of rats were sacrificed 30 min, 4 h and 24 h after reserpine (10 mg/kg, i.p.) injection. Toluidine blue stained sections showed that in the motor neurons of the spinal cord the drug, at 4 h, had induced a migration of the Barr body from the nucleolus to the nuclear membrane and an increase in its size and RNA concentration. From our findings we suggest that reserpine may have an activating role on X-linked genes.

Previous studies on the effect of psychoactive drugs at the cellular level have shown changes in nucleic acid histochemistry in rat sympathetic neurons after acute administration of reserpine, which involved the emergence of new RNA both in the nucleus and cytoplasm[5]. Furthermore, changes in nucleoprotein ultrastructure have also been observed in peripheral blood cells of schizophrenic patients after the administration of neuroleptics[6]. Alterations in metaphase chromosome structure have also been induced in vitro by chlorpromazine[7]. The purpose of the present study was to investigate whether the increase in RNA found in sympathetic neurons after reserpine administration[5] was a result of an initial interaction of this drug with the chromatin. To avoid a possible indirect effect through biogenic amines, we chose to study the motor neurons of the spinal cord.

Materials and methods. 18 adult female albino rats, weighing about 200 g each, were divided into 6 groups. 3 groups of rats, given reserpine (10 mg/kg, i.p.), were sacrificed at 30 min, 4 h and 24 h after injection. The remaining 3 groups, given saline and sacrificed at the same time intervals, served as controls. Segments of spinal cord were fixed in 10% formalin. Paraffin sections were stained with Toluidine blue for RNA and with the Feulgen method for DNA.

Results. Observation of the animals indicated that sedation was already evident at 30 min and maximal at 4 h. The effect of the drug had almost been dissipated by 24 h. In Feulgen stained preparations, the sex chromatin (Barr body) of the motor neurons in the controls appeared as a small red granule adjacent to the nucleolus. At 30 min reserpine had induced a partial euchromatization of the Barr body; this decondensation persisted at 4 h. In addition, a migration of this body was observed from its typical position on the nucleolus to the nuclear membrane at 4 h, while at 24 h it was found again, in most neurons, next to the nucleolus. In the Toluidine blue stained sections the motor horn neurons in the control (figure 1) were characterized by clear nuclei, displaying a faintly stained sex chromatin next to the nucleolus, and distinct large Nissl bodies in the perikarya and the dendrites. At 30 min after reserpine administration (figure 2) diffuse RNA occupied the entire nuclear area and was also found in increased concentration in the cytoplasm and on the Nissl bodies. The Barr body, still faintly stained, was located at a distance from the nucleolus. At 4 h (figure 3), when maximal sedation of the animals was observed, the Barr body was intensely stained, noticeably large in size and adjacent to the nuclear membrane. Other striking changes