Acid hydrolysis under vigorous condition ($2N \cdot HCl$, reflux) of XX gave an aminosugar, mycaminose[6] (XXII), m.p. 113–115°, which was identical with an authentic sample obtained from leucomycin A_3.

Maridomycin II (II) was oxidized with CrO_3-pyridine complex or MnO_2 to dehydromaridomycin II (XXVI), m.p. 206–207 °C (decomp.), which was identical with authentic sample of carbomycin[7] in UV-, IR-, MS-, NMR-spectra, specific rotation and Rf values on TLC.

The structure of carbomycin has been elucidated by WOODWARD et al.[8].

From these findings, the structure of maridomycin II was determined to be II. Its absolute configuration, except for the C_9-hydroxyl group, was also clarified. Further treatment of XXVI with KI in AcOH yielded dehydrodeepoxymaridomycin II which was identical with carbomycin B[7] in all respects.

Zusammenfassung. Das aus *Streptomyces hygroscopicus* isolierte neue Makrolid Maridomycin II lässt sich mit Säure Mycarcose und Mycaminose spalten. Auf Grund der Oxidation ins Carbomycin sowie der spektroskopischen Daten wurde die Struktur als II erklärt.

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Effect of Diurnal Rhythm and Food Withdrawal on Serum Lipid Levels in the Rat

Occasionally, we have observed from one day to the next marked differences in the levels of serum triglycerides of normal rats. Since fasting can affect serum and liver lipid levels[1,2] and thus may obscure any changes caused by a given treatment, our animals had been allowed access to food until they were killed. When rats of similar age, weight and body weight gain were used, the within group variation of serum triglycerides in both treated and untreated animals rarely exceeded 15%.

BARRETT[3] has found that plasma free fatty acids of rats exhibit a marked diurnal rhythm. This is in accord with the finding of extremely high turnover rates of free fatty acids in the plasma[4]. As far as we are aware, no studies have been conducted on the effect of diurnal rhythm on serum triglycerides in the fed and fasted rat. Therefore, experiments were carried out to determine the effect of fasting and killing time on rat serum triglycerides (as well as on cholesterol and phospholipids).

Methods. Since hemolyzed blood was sometimes obtained in our studies, we first determined the effect of hemolysis on serum triglycerides. Unhemolyzed blood was collected from untreated fed albino rats (Charles River) at 09.00 h and each sample was immediately divided into 2 tubes. The blood from one group was allowed to stand for 3 h and clear serum was obtained after centrifugation. The blood from the second group was hemolyzed with the aid of wooden applicators and centrifuged. Serum triglyceride levels were measured by the semi-automated method of KRAML and COSYNS[5].

For the studies on diurnal rhythm, male albino rats, weighing 180–190 g, were fed Purina lab chow ad libitum. Lighting was automatically regulated to provide 12 daily h of light from 08.00 to 20.00 h. Animals were kept under observation for 3 days and only those with normal weight gain and food intake were used. On day of killing, food (but not water) was withdrawn from half the animals at 08.00 h and the rats were decapitated at various times during the next 24 h. Serum triglycerides[5], cholesterol[6], and phospholipids[7] were measured according to previously described techniques.

Results and discussion. The results of the hemolysis study are presented in the Table. It was found that hemolysis had no effect on serum triglyceride levels in both male and female rats.

The effect of fasting and diurnal rhythm on serum lipids is presented in the accompanying Figure. It was found that both fasting and killing time had a profound effect on serum triglyceride levels. In fed rats, triglyceride glycerol levels at 08.00 h were 14 mg/100 ml and declined to a minimum of 7–8 mg/100 ml in the early evening. Fasting levels were markedly lower than those of fed rats, reaching a minimum of 2–3 mg/100 ml in the evening and

Effect of hemolysis on rat serum trigliyceride levels

Serum	Triglyceride glycerol (mg/100 ml) Male rats	Female rats
Unhemolyzed	15.5 ± 1.43	10.6 ± 1.10
Hemolyzed	16.0 ± 1.64	10.0 ± 0.82
Mean difference	0.5 ± 0.92	0.6 ± 1.12

Blood from Charles River albino rats, weighing 180–190 g was used. Mean difference refers to difference in glycerol levels for each animal as a result of hemolysis. Results are presented as mean ± standard error for 10 rats/group.

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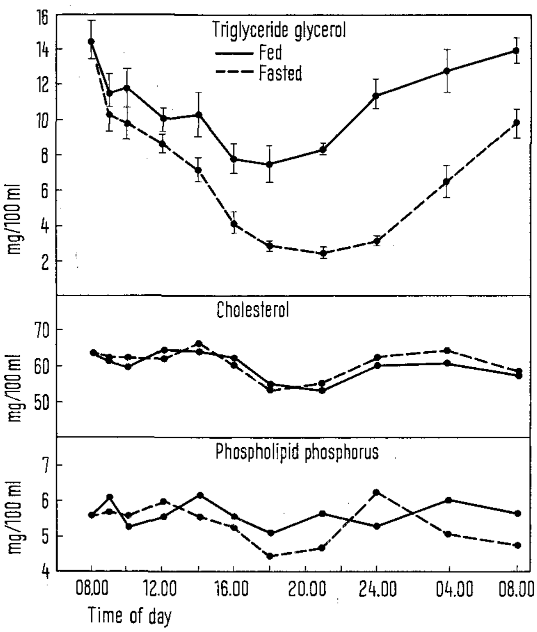
recovering to 9–10 mg/100 ml by 08.00 the following day. The difference between fed and fasting triglyceride levels became statistically significant as early as 14.00 h. Although rats consume most of their daily food intake at night, they do tend to 'nibble' during the day[8]. Comparison of our results with those on the diurnal rhythm of free fatty acids[3] indicates an inverse relationship between rat serum triglycerides and free fatty acids throughout the day.



Effect of fasting and time of day on serum lipids in male rats. Food (but not water) was withdrawn from the fasted animals at 08.00 h. Standard errors are given for serum triglycerides only. Each point represents the mean of 10 rats.

Serum cholesterol and phospholipid levels in both fed and fasted rats remained virtually unchanged during the 24 h test period. No standard errors are presented in the Figure for cholesterol and phospholipids because no statistical difference was found between the fed and fasted rats. As discussed previously[9], much has been reported on the effects of fasting on lipid levels (especially sterols), but literature comparisons are difficult due to such variations as species, strain, sex, age and duration of fast.

The results show that, in contrast to cholesterol and phospholipids, serum triglycerides in both fed and fasted rats exhibit a marked diurnal rhythm and that the effect is more pronounced in fasted animals. Thus, the time of killing is a factor of particular importance in studies of agents affecting serum triglyceride levels.

Résumé. Chez le rat albinos, le niveau des triglycérides sériques varie au cours de la journée, avec un maximum à 08.00 h et un minimum au début de la soirée. Cette variation est plus marquée chez les rats à jeun. Par contre, aucune variation n'a été observée dans les niveaux de cholestérol et de phospholipides.

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The Cholinesterase Activity of Myoneural Junctions from Frog Twitch and Tonic Muscles

At least two different types of extrafusal muscle fibre have been described in the skeletal muscles of frogs. The fibres have been classified as twitch or tonic on the basis of their physiological[1,2], structural[3–5], and biochemical properties[6]. Also the myoneural junctions on these fibres differ in their form and distribution[7]. Günther[8] has shown by staining nerve terminals with silver that 'en-grappe' motor nerve endings occur on the muscle fibres of the tonus bundle of the frog iliofibularis muscle (described by Sommerkamp[3]) and not on fibres in other parts of the iliofibularis or in the sartorius. Other authors[4,9] have demonstrated that the twitch muscles, stained for cholinesterase, have myoneural junctions, 'Endbüschel', which are relatively extensive and which run lengthwise along the muscle fibre with a few oblique branches connecting the main longitudinal terminations. This type of ending occurs in a band of innervation so that nerve terminals on adjacent fibres are more or less at the same level. The en-grappe endings appear irregularly scattered along the muscle fibre and do not occur on the same level on adjacent tonic fibres.

The question posed in this investigation is – can differences in cholinesterase (ChE) activity of myoneural junctions be related to the morphological and functional differences of frog muscles? In order to answer this question a radiometric method[10] was used to give a direct measure of the ChE activities of populations of single myoneural junctions from the different muscles.

The muscles investigated were the iliofibularis (twitch) and its tonus bundle (tonic), sartorius (twitch) and rectus abdominus (mixed) of *Rana tempororia*. The muscles were pinned at their resting length on hard paraffin and incubated in a thiocholine medium[10] for 30 min to make the

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