

zer-Direktschreiber'. The ATP-relaxing solution contained (in mM): ATP, 17.5; $MgCl_2$, 17.5; imidazole, 10; EGTA, 4; Na-azide, 4. The (ATP-free) standard rigor solution contained (in mM): KCl, 100; imidazole, 10; EGTA, 4; Na-azide, 4. The other rigor solutions contained (in mM): KCl, 10; $MgCl_2$, 30 (or $CaCl_2$, $BaCl_2$, $SrCl_2$, K_2SO_4 or $KHPO_4$); imidazole, 10; EGTA, 4; Na-Azide, 4. Rigor solutions with lower concentrations of divalent ions (5–30 mM) were obtained by mixing these solutions in the appropriate relation. The pH was adjusted at room temperature to 6.5 by addition of HCl or NaOH. The experiments were carried out at room temperature (ca. 20°C).

Results. In order to produce rigor tension skeletal muscle fibre bundles relaxed in the ATP-relaxing solution were incubated in the ATP-free standard rigor solution¹. Herein the isometric tension rose (Figure 2a) to about 0.9 kg/cm² (standard deviation 0.3 kg/cm², $n = 10$). The following immersion in a Mg-containing rigor solution produced a tension fall to about 85% of the tension level in standard rigor solution (Figure 2a, Table). The 100% tension level was reached again after reimmersion in standard rigor solution. Such tension cycles could be repeated up to 20 times without any notable decrease in the extent. In experiments with glycerol-extracted rabbit psoas fibres (unpublished experiments) and DLM-fibres (Figure 2b) similar findings were obtained. Rigor tension was also produced by transferring the fibres from ATP-relaxing solution directly into Mg-containing rigor solution. The subsequent immersion in standard rigor solution induced a further tension increase of about 15% to the 100% level followed by a tension decrease in Mg-rigor solution.

The tension decreased by raising the Mg^{++} concentration from 5–30 mM. Reversible tension cycles were also induced by other divalent cations (Table): at a concentration of 30 mM the tension fall relative to the 100% level, was similar in Mg-, Ca-, Ba- and Sr-containing rigor solutions. The isometric rigor tension was reversibly decreased by lowering the pH of the standard rigor solution (Table). The reverse effect was obtained in rigor solutions containing SO_4 and by raising the pH (Table), whereas rigor

solutions with PO_4^{--} did not seem to affect the isometric tension.

Discussion. Provided that the crossbridges do not rotate on the actin filament while in rigor⁸ the elastic force between actin and myosin filaments produced by the crossbridges, and therefore the rigor tension, should increase with wider spacing of actin and myosin filaments.

We assume that this consequence of the crossbridge models (Figure 1)^{3,9} is verified by the present experiments, as raising the pH not only increases the rigor tension, but also the interfilamentar distances, to about the same extent of 10%^{4,5}, whereas the reverse effect on isometric tension and spacing of actin and myosin filaments is induced by divalent cations or lowering the pH. According to ROME^{4,5} the variation of the interfilamentar distance might be produced by electrostatic effects between the filaments. Apart from the interpretation discussed, the correlation between rigor tension and myofilamentar distances might also be explained on the basis of the electrostatic model proposed by ELLIOT et al.¹⁰ or KOMINZ's¹¹ osmotic theory.

Zusammenfassung. Die Rigorspannung glycerinextrahierter Skelettmuskelfasern (*Caiman crocodilus*) und fibrillärer Insektenflugmuskelfasern (DLM) (*L. maximus*) konnte durch Zugabe von zweiwertigen Kationen und pH-Erniedrigung reversibel erniedrigt werden sowie durch pH-Erhöhung und Zugabe von Sulfationen erhöht werden. Da unter ähnlichen Bedingungen im Rigor Veränderungen des Abstandes von Aktin- und Myosinfilamenten nachgewiesen wurden^{4,5}, wurden die beobachteten Spannungsänderungen im Rigor auf Grund neuerer Crossbridge-modelle^{3,9} diskutiert.

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Effect of divalent ions and pH on the isometric rigor tension; [glycerol-extracted fibre bundles from skeletal muscle (*Caiman crocodilus*)]

Solution	pH	Tension ^a	n
10 mM KCl + 30 mM $MgCl_2$	6.5	80.6 ± 10.2	66
10 mM KCl + 30 mM $CaCl_2$	6.5	82.6 ± 11.3	16
10 mM KCl + 30 mM $BaCl_2$	6.5	79.2 ± 15.1	5
10 mM KCl + 30 mM $SrCl_2$	6.5	77.0 ± 13.2	5
10 mM KCl + 30 mM K_2SO_4	6.5	118.2 ± 10.2	10
100 mM KCl	6.0	87.9 ± 5.3	5
100 mM KCl	7.0	112.8 ± 6.3	5

^a Tensions (mean values and standard deviations from the mean) given in percentage of the values obtained in standard rigor solution (100 mM KCl, pH 6.5, 20°C). All solutions contained 10 mM imidazole, 4mM Na-azide and 4mM EGTA.

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Overload and Training, Compensatory Hypertrophy and Contraction Characteristics of M. plantaris in Female and Male Rats

In a series of experiments concerning the effect of overload and training on the fast and pennate-fibred M. plantaris (MP)¹⁻³ we also compared the effect in male and female rats. A recent report on compensatory hyper-

trophy (CH) of leg muscles in the rat of both sexes⁴ produced findings essentially similar to ours, i.e., that there are no differences between the sexes. These authors said: 'This finding is not really surprising, if one considers that

Table I. Female rats. Weight and contraction characteristics of *M. plantaris* (mean \pm standard deviations)

Female rats	CNT N = 5	DNT N = 5	DT N = 5	Analysis of variance	Scheffé test CNT-DNT	CNT-DT	DNT-DT
Rat weight (g)	182 \pm 12.1	193 \pm 10.8	185 \pm 8.7	—	—	—	—
Muscle weight/ Rat weight (%)	0.119 \pm 0.0043	0.152 \pm 0.0120	0.178 \pm 0.0035	$\times \times \times$	$\times \times \times$	$\times \times \times$	$\times \times \times$
Twitch/Tetanus	0.148 \pm 0.0087	0.142 \pm 0.0150	0.135 \pm 0.0119	—	—	—	—
Tetan. isom. force/ Cross sectional area (kgf/cm ²)	7.3 \pm 0.30	7.9 \pm 0.53	7.6 \pm 0.63	—	—	—	—
Twitch contract. time (msec)	15 \pm 1.5	15 \pm 0.7	14 \pm 0.4	—	—	—	—
V _o /l _o (mm/sec/cm)	67 \pm 3.9	60 \pm 3.1	65 \pm 4.6	\times	\times	—	—
a/P _o (%)	30 \pm 3.9	30 \pm 3.3	29 \pm 2.0	—	—	—	—

CNT, control not trained; DNT, denervated not trained; DT, denervated trained; Statistical analysis: —, not significant; \times , $0.01 \leq P \leq 0.05$; $\times \times$, $0.005 \leq P \leq 0.01$; $\times \times \times$, $P \leq 0.005$.

limb muscles as a whole are not dependent upon sexual hormones ...'. Since we furthermore measured the characteristics of isometric and isotonic contraction of the MP, applying a different operation to evoke excessive use, and included training as well, the data might be of interest to supplement the work of the authors mentioned above.

Methods. 15 male and 15 female Wistar rats, 7 weeks old, were used. The male and the female group was divided into 3 subgroups of 5 rats. 1 subgroup served as a control (CNT-control not trained). The rest was operated according to the procedure of denervation plus transplantation². In short, *M. gastrocnemius* and *M. soleus* were denervated and distal tendon of MP was transplanted to that of *M. gastrocnemius*. After the operation, these rats were divided into the subgroups DNT (denervated not trained) and DT (denervated trained). The DT rats were trained on a motor-driven endless belt at 30° at a speed of 300 m/h. The training started the day after the operation. A time schedule of 2 h per day 5 days per week was followed until 48 h before the contractions were measured. The mean age of the rats was 92 days range 82–107 days (females) and 93 days range 81–105 days (males) on the day of the measurements. Contraction characteristics of the MP were established in a nerve-muscle preparation (*N. tibialis* – *M. plantaris*) in situ. The

MP was freed from the surrounding tissue, blood vessels and the nerve to the MP were kept intact. The *N. tibialis* was transected at ca 3 cm proximal from the MP. The distal tendon of MP was connected to either the force or displacement transducer. The skinflaps of the wound were drawn up and aside and a paraffin pool was made which was kept at 35–36°C. All measurements occurred at optimal muscle length (*l*_o)³: i.e., muscle length at which twitch contraction time (time to peak) is minimal and twitch contraction force (peak force) is maximal. Tetanic stimulation frequency was 185 Hz. Isotonic contractions are elicited with afterloads between 1% and 60% of maximal isometric tetanic force (*P*_o). Maximal velocity (*V*_o) was obtained by calculation using Hill's formula.

After the contraction studies, the muscle at optimal length was kept connected to the transducer. The rat was killed with an overdose of pentobarbital (Nembutal) and 2 h later when the rat was in rigor – mortis the muscle

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Table II. Male rats. Weight and contraction characteristics of *M. plantaris* (mean \pm standard deviations)

Male rats	CNT N = 5	DNT N = 5	DT N = 5	Analysis of variance	Scheffé test CNT-DNT	CNT-DT	DNT-DT
Rat weight (g)	294 \pm 22.5	266 \pm 16.6	235 \pm 9.6	$\times \times \times$	—	$\times \times \times$	$\times \times$
Muscle weight/ Rat weight (%)	0.110 \pm 0.0091	0.135 \pm 0.0133	0.167 \pm 0.0100	$\times \times \times$	$\times \times$	$\times \times \times$	$\times \times \times$
Twitch/Tetanus	0.165 \pm 0.0188	0.145 \pm 0.0168	0.155 \pm 0.0122	—	—	—	—
Tetan. isom. force/ Cross sectional area (kgf/cm ²)	7.1 \pm 0.54	7.7 \pm 0.41	7.5 \pm 0.28	—	—	—	—
Twitch contract. time (msec)	15 \pm 1.3	15 \pm 1.4	15 \pm 1.1	—	—	—	—
V _o /l _o (mm/sec/cm)	71 \pm 3.0	61 \pm 4.9	67 \pm 3.9	$\times \times$	$\times \times$	—	—
a/P _o (%)	31 \pm 6.6	28 \pm 3.3	26 \pm 2.1	—	—	—	—

See legend to Table I.

was dissected. Lastly muscle length and weight were determined. More details of the method may be found in the papers by BINKHORST^{1,3}.

Results and discussion. There are significant differences between the absolute values of muscle weight and length of male and female rats of the same age and the contraction parameters closely related to these values such as force and velocity. The main reason for these differences is that the male rats are larger and heavier than the females. To study the effect of sex mainly relative values will be used.

In Table I and II the data of female and male rats respectively are presented. The results of the statistical analyses are also included in the Tables. The analysis of variance and the SCHEFFÉ⁶ multiple comparison test were done on the subgroups.

Rat weight in the male groups differed significantly: the trained rats have the lowest weight which is in agreement with a recent report⁷. In female rats this difference is not found. This finding might be important for studies of physical activity, caloric intake and obesity, in so far as there are differences in reaction depending on the sex of the rat. Muscle hypertrophy (from muscle weight/rat weight) can be seen in the operated rats; hypertrophy is larger in the operated and trained rats (DT rats).

Maximal speed of contraction per unit muscle length (V_0/l_0) is significantly different only in the CNT-DNT comparison in female as well as male rats: the CNT rats show the highest speed. It could be shown³ that there is a significant positive correlation between V_0/l_0 and the cosine of the fibre angle, which is significantly increased in the hypertrophic pennate fibred MP. Is it assumed that the increased angle at least partly offers an explanation for the lower speed in the hypertrophic muscles.

The parameters twitch/tetanus, twitch contraction time, and a/P_0 (curve parameter for the force-velocity relation according to Hill's formula) to not show any

significant differences. It is concluded that the overloaded MP remains fast, which is in agreement with previous findings¹.

The results as presented in the Tables indicate that for the MP there are no differences in reaction to overload and training between the sexes.

Lastly Student's *t*-test was applied to the data of the CNT, DNT and DT rats to compare the female and male groups. Significant difference were found in the rat weight of all equal subgroups (all $P < 0.005$). For the relative parameters only 2 significant differences were found which may be meaningless taking in account the large number of tests. Generally speaking, the female and male rats do not react differently to the applied procedure, except for body weight.

Zusammenfassung. Bei der Ratte wurde durch Denervation des M. gastrocnemius und des M. soleus eine kompensatorische Hypertrophie des M. plantaris bewirkt und die distale Sehne des M. plantaris auf diejenige des M. gastrocnemius transplantiert. Ein Teil der operierten Tiere wurde trainiert, die Muskelgewichte bestimmt und isotonische sowie isometrische Kontraktionen untersucht. Die Muskelhypertrophie war bei den trainierten Ratten am stärksten ausgeprägt.

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Effects of Prostaglandins PGE_1 and $PGF_{2\alpha}$ on Oxygen Consumption, Sodium and Potassium Content of Renal Tissue

Prostaglandin E_1 , a hypotensive substance, elicits natriuresis when infused in the kidney. This natriuretic effect is coincident with an increase in renal plasma flow, a decrease in the para-amino hippuric extraction rates, but produces no observable change in glomerular filtration rate and systemic blood pressure¹⁻³. Since the natriuretic effect of PGE_1 cannot be explained by an

increase in sodium tubular load, the other explanation is that sodium reabsorption is reduced either by the increase in blood flow per se^{4,5} or by the action of PGE_1 on tubular cells.

The natural occurrence of prostaglandins in the kidney suggest that they may have a physiological role in the regulation of sodium and water excretion. This possibility led us to study the effect of PGE_1 on oxygen consumption, sodium and potassium content of renal cortical slices of rats to find out whether PGE_1 has a direct effect on the kidney.

Material and methods. Male Wistar rats (150–200 g) were used. The animals were decapitated and both kidneys and the liver removed and cut into slices of 0.3 mm thickness⁶. The slices were kept in Petri dishes with

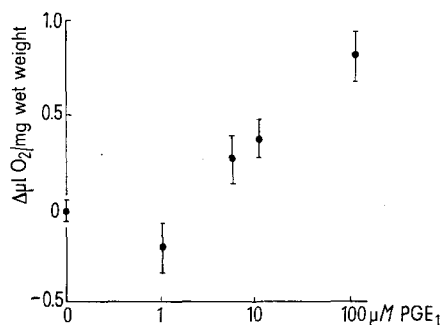


Fig. 1. Relationship between the increment of oxygen consumption ($\Delta \mu l$) and concentration of prostaglandin E_1 (PGE_1) in the medium. Each point represent the mean \pm SD.

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