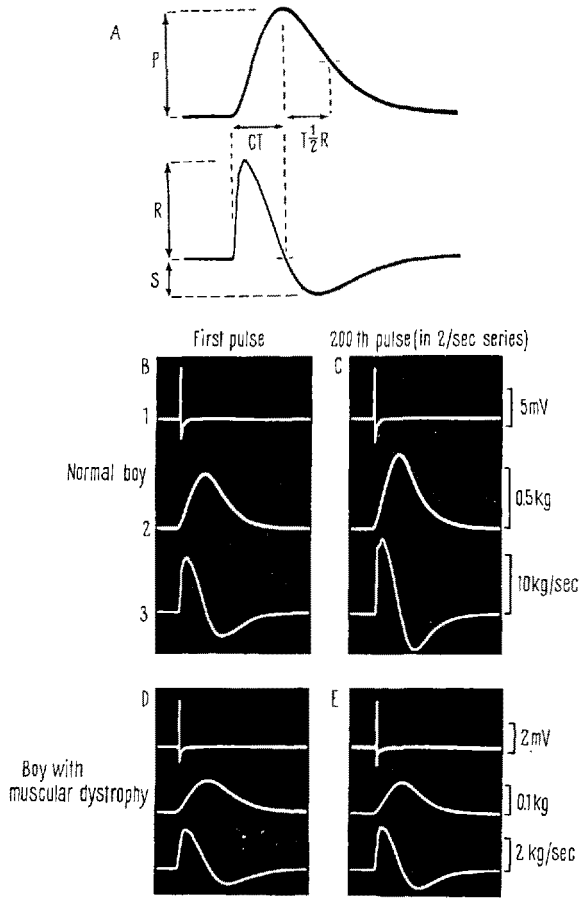
Human Sex-Linked Muscular Dystrophy: Kinetics of the Isometric Twitch

Muscular dystrophy refers to a group of inherited degenerative diseases which involve primarily the skeletal muscle fibres. The genetic defect which initiates the muscle fibre disorder is unknown but research progress can be anticipated since the molecular processes involved in the activation and contraction of the myofibrils are being uncovered[1,2] while methods for the direct investigation of human muscles are steadily improving. The present report is concerned with the Duchenne type of human muscular dystrophy, which is the commonest and most severe form of the disease and which is generally inherited as an X-linked recessive trait. Physiological studies of muscle contraction in situ were carried out in 15 patients aged 5–14. Six normal boys aged 9–14 served as controls, besides a more extensive series of adults. Supramaximal electric pulses of 50 μsec duration are delivered to the ulnar nerve at the wrist. The belly-tendon electrical response and the isometric myogram of the adductor pollicis muscle are recorded on cathode-ray oscilloscopes, along with the first derivative of the myogram[3,4]. The i.m. temperature measured with a thermistor needle is 37–38 °C. Figure A shows the measurements performed on the enlarged photographic records of the twitch (upper trace) and its derivative (lower trace). The mean force P is 0.415 kg in the normal boys and 0.120 kg in the patients. Corresponding figures for the maximum tetanic force P_0 are 3.75 and 1.30 kg. The force decreases with the progress of dystrophic involvement, as evaluated by electromyographic analysis of the voluntary motor unit potentials in the same muscle[5].

Repetitive stimulation of the motor nerve at 2/sec for 2 or 3 min elicits a characteristic shortening of the twitch time-course in normal muscle (Figure)[4,6,7]. This phenomenon is also present and indeed prominent in the dystrophic muscles. In our study of twitch kinetics we therefore compare the features of the 1st and the 200th twitches belonging to an uninterrupted series at 2/sec. The simultaneously recorded electrical response of the muscle discloses only slight changes of duration and amplitude of its second phase probably related to a slight increase in conduction velocity of the muscle spike[3]. There is no evidence of variation in neuro-muscular excitation during the 2/sec series and the prominent changes of the twitch are obviously related to contractile processes. In normal boys, as in adults[6], the twitch presents a considerable staircase potentiation during the series and the 200th twitch has a larger force P and rate of tension development R (Figure B, C). In the dystrophic boys the potentiation of twitch force and rate is either reduced or absent, depending on the severity of dystrophic involvement of the muscle, although the characteristic shortening of relaxation time still occurs (Figure D, E).

The mean contraction time CT of the first twitch elicited in the rested muscle is 76 msec in normals and 85 msec in patients, a barely significant difference. For the 200th twitches in the 2-/sec series, the mean CT are 69 and 73 msec respectively (Table). The isometric CT is related to the speed of activation of the myofilaments which in turn depends on the intrinsic activity of myosin ATPase[8], provided the amount of Ca^{++} released from the sarcoplasmic reticulum into the myoplasm is adequate[9]. The finding of normal CT in dystrophic muscle thus allows us to predict normal activity for the myosin ATPase of the still excitable muscle fibres[10]. The mean relaxation time $T^1/_2R$ (see Figure A) for the first twitch is 61 msec in the normal boys and 75 msec in the patients. This difference of + 23% is significant, but it is much smaller than the threefold increase reported for gastrocnemius of mice with genetic muscular dystrophy[11]. The difference vanishes when the 200th twitches in the 2/sec series are compared in the 2 groups (Table). The differen-



Cathode ray-oscillograms of the responses of the adductor pollicis muscle to a supramaximal electric pulse delivered to the ulnar nerve at the wrist. Intramuscular temperature 37–38 °C. (A) measurements performed on the isometric twitch (upper trace) and on its first derivative (see text). (B, C) normal boy, simultaneous record of belly-tendon electrical response (1), isometric twitch (2) and the latter's first derivative (3). (D, E) same for boy with Duchenne muscular dystrophy. Actual involvement of the adductor pollicis muscle is indicated by the reduced mean duration of motor unit potentials (from 9.17 to 6.4 msec) and by difficulties for voluntary graduation.

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tial record allows us to estimate the maximum rate of relaxation which we designate S. Its mean value is 3.4 kg/sec in normal boys and 0.72 kg/sec in the patients. Since S is no doubt influenced by the absolute force developed by the corresponding contraction, we rather consider the relaxation speed/kg as given by the ratio S:P. The mean ratios are 10 in normals and 6.5 in the dystrophics. They become equal at 8.4 for the 200th twitch (Table).

In conclusion the dystrophic disorder produces characteristic changes in the capability for tension development and for staircase potentiation in skeletal muscle. These contractile anomalies appear at a preclinical stage when the electrical responses of the muscle still appear normal[5]. In the present paper we emphasize that the twitch kinetics undergoes rather little changes in human dystrophic muscle. Our study refers to the rested muscle studied in situ, at 37–38 °C, under optimum initial tension and with supramaximal stimulation of the motor nerve. Under these conditions the twitch relaxation time is slightly prolonged (+ 23%) and the relaxation speed/kg is reduced (− 35%). However the 2/sec stimulation eliminates these differences as the relaxation accelerates more in dystrophic muscle than in normal muscle (Table). These findings for human muscle are at variance with those reported for dystrophic mouse muscle in vitro in which relaxation was 3 times slower than in controls[11]. Our results suggest that the Ca^{++} sequestration process governing relaxation[12] in the sarcomeres is not irreversibly damaged by Duchenne muscular dystrophy. The observations on contraction times also suggest that the myosin ATPase has normal activity in the still excitable dystrophic muscle fibres[13].

	Normal boys (6)	Duchenne dystrophy (15)	*t*-test
CT_1 (msec)	76 ± 4	85 ± 11	$P = 0.06$
CT_{200}	69 ± 5	73 ± 8	$P = 0.25$
$T^1/_2R_1$ (msec)	61 ± 7	75 ± 9	$P < 0.01$
$T^1/_2R_{200}$	58 ± 11	61 ± 8	$P = 0.5$
$S_1:P_1$	10 ± 1.2	6.5 ± 1.4	$P < 0.01$
$S_{200}:P_{200}$	8.4 ± 2.3	8.4 ± 1.4	$P = 0.9$

Résumé. Les propriétés mécaniques du muscle adducteur du pouce atteint de dystrophie musculaire de type Duchenne ont été étudiées in situ chez 15 malades. La force produite par la secousse isométrique et le tétanos isométrique est réduite dans tous les cas. Le temps de contraction de la secousse isométrique et son temps de relaxation sont légèrement supérieurs aux valeurs de contrôle. Cette différence disparait au cours de la stimulation répétée du nerf à 2/sec. Ces observations montrent que l'activité ATPase de la myosine (déterminant la vitesse de contraction) et le processus de séquestration du Ca^{++} myoplasmique dans le réticulum sarcoplasmique (déterminant la vitesse de relaxation) ne sont pas altérés de façon irréversible dans la dystrophie musculaire de Duchenne.

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The Effect of Partial Hepatectomy upon Circadian Distribution of Mitosis in the Cornea of Rats

The stimulation of mitosis which occurs in the cornea of rats following partial hepatectomy has been intensively investigated as to its relation to physiological mechanisms which control cell division[1].

The stimuli are apparently related to adrenal gland function because, while it is absent in adrenalectomized rats, it is restored in dexamethasone treated adrenalectomized rats[2]. Whether this stimulating action results from direct or indirect glucosteroid effect upon the metabolism of corneal epithelial cells is not known at the present time[3].

In previous work, the occurrence of cells in mitosis in the cornea of normal and in partially hepatectomized rats was determined in only 2 time periods of the day i.e. 10.00 and 23.00, and thus afforded a partial view only of the diurnal distribution of mitotic activity[3]. Therefore, additional data were sought in order to obtain: (1) a truly circadian distribution of mitosis: (2) a more complete picture of the first 24 h period post partial hepatectomy, in which, according to previous data, increased mitotic activity was not present and the absence of which was thought to be related to either post surgery fast, stress and/or displacement of the morning mitotic peak to some other time period of the day. Thus the different groups of partially hepatectomized rats were sacrificed at 03.00, 07.00, 10.00, 15.00, 19.00 and 23.00 of the day. The control levels of mitosis for the different periods of the day were obtained in 2 separate experiments carried out with 1 month interval between them. The possibility that significant physiological variations in the levels of mitosis could occur from day to day, in spite of the apparent maintenance of environmental conditions, was thus verified.

Female rats of the Wistar strain, with weight ranging between 120–140 g, were used in all experiments. Partial hepatectomy was performed according to the technique of Higgins and Anderson[4]. The animals were sacrificed

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