actively feeding, approximately same sized individuals were chosen for analyses. Commercially available progesterone (supplied by the Patel Chest Institute, Delhi), was injected (200 µg hormone in 0.1 ml refined peanut oil) into experimental animals in the vicinity of the brain. The controls were injected with 0.1 ml refined peanut oil alone. Injections were given in the early hours of the morning. Brain tissue, pooled from 16 to 18 animals at 0 °C (weighing 8 to 10 mg), was weighed in ice-cold Ringer[10], and analyzed 2 h after injection, for the estimations of total protein following the micro Biuret method[11].

RNA was extracted by the method of SCHMIDT-THANNHAUSSER-SCHNEIDER[12] and estimated by orcinol colour reaction following the colorimetric procedure described by GLICK[13].

Acetylcholinesterase (AchE, E.C.3.1.1.7). The tissues were homogenized in ice-cold 0.02 *M* phosphate buffer, pH 7.0. They were centrifuged for 30 min at 6,000 rpm and the supernatant was used for assay. AchE activity was determined spectrophotometrically by the method of HESTRIN[14].

Results and discussion. It is obvious from the data presented in the Table that the protein content decreased ($p < 0.01$) in the brain on in vivo administration of progesterone. Paralleling the decrease in proteins, RNA levels also decreased significantly as a function of progesterone injection (Table). The decrease in the level of proteins of the CNS of progesterone administered tadpoles appears to be the direct consequence of the protein destructive nature of progesterone[15,16]. It is also possible that such a decrease may be due to a change in protein synthesis. Significant decrease in RNA level observed in the brain of tadpoles on in vivo administration of progesterone, also points to deceleration in the activity of the protein-synthetic machinery in progesterone administered animals.

The significant decrease in the activity levels of acetylcholinesterase in the developing amphibian (Table) on injection of progesterone may be the direct consequence of proteine destructive action of progesterone as stated earlier[16]. It is therefore possible that the decrease in the activity levels of acetylcholinesterase is a reflection of the decrease in the enzyme synthetic processes caused by the progesterone administration.

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The Tension/Length Relationship of an Insect (*Calliphora erythrocephala*) Supercontracting Muscle

J. HARDIE[1,2]

Department of Zoology and Comparative Physiology, The University of Birmingham, Birmingham B15 2TT (England),
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Summary. The tension/length curves of an insect supercontracting striated muscle are described. Both vertebrate and invertebrate smooth (non-striated) muscles show a close similarity to these curves. Thus, although insects possess only striated muscle, some of these muscles can perform the function of smooth muscle of other animals.

It has often been stated that insects posses only striated muscle[3,4]. Thus these animals do not appear to have any muscles which are structurally equivalent to the smooth (non-striated) muscles of other invertebrates and vertebrates. This lack of smooth muscle might be expected to impose limitations on the range of physiological type of muscle in insects. In particular, the functioning of the isotonic visceral muscles would be restricted. However, it was the isotonic body-wall muscles of the blowfly larva that provided an explanation of how certain striated muscles of insects could perform the type of activity more usually associated with smooth muscle[5,6]. These muscles possess perforated Z discs; at short sarcomere lengths the thick and thin myofilaments penetrate the Z perforations and enter adjacent sarcomeres (Figure 1). Thus it is possible for this type of sarcomere to contract down to below A band length with no concomitant change in myofilament length. This phenomenon of 'supercontraction' of striated muscle was first observed in molluscs[8], and has also been reported in chelicerates[9,10] and in vertebrates[11]. Reversible contraction has been observed down to lengths much shorter than those possible for 'classical' striated muscles with solid Z discs (22% of initial length in blowfly muscle[5]; < 30% of initial length in barnacle muscle[8]). However, there are no detailed reports in the literature of the tension/length relationship of this type of muscle.

Figure 2 shows the tension/length curves of a supercontracting bodywall muscle fibre from a blowfly larva. Active tension is produced during a change in fibre length from 0.47 mm to 2.19 mm (a 79% length change). Optimum length (muscle length where maximum active tension is developed) is 1.25 mm while passive tension is first recorded at approximately 0.8 mm and increases at greater lengths. It was not possible to measure a meaningful, in vivo resting length due to the usual difficulties

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[2] Present address: Agricultural Research Council Insect Physiology Group, Department of Zoology and Applied Entomology, Imperial College, London, England.
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encountered with very extensible muscles[14]. Maximum tetanic tension was approximately 0.17 kg/cm², a figure much lower than is usual for insect muscle (1.5–2.0 kg/cm² [15–18]) but is comparable with certain other muscles[19].

Perhaps the best way to discuss the significance of the tension/length relationship demonstrated is by reference to other muscles (Figure 3). The most striking feature of the active tension/length curves (Figure 3A) is the difference in working range. For the classical striated muscle (frog sartorius and locust flight muscle) the range is narrow. In this case the working range of the sarcomere is limited to where the myofilaments just overlap and where the thick filaments collide with and buckle against the Z disc[22]. It is interesting that the active tension/length curves which most closely match that of the blowfly muscle are those of *Mytilus* A.B.R.M. and guinea-pig taenia coli muscle. The former is a non-striated, paramyosin containing muscle with no Z discs per se. The corresponding structures are blocks of dense material with thin filaments attached at each end[23, 24]. These bodies do not restrict contraction since they allow the paramyosin filaments to slide past them. Guinea-pig taenia coli muscle is a classical vertebrate smooth muscle, again possessing dense bodies which are presumably analogous to Z material[25]. The working ranges of these 2 muscles are remarkably wide, with guinea-pig muscle

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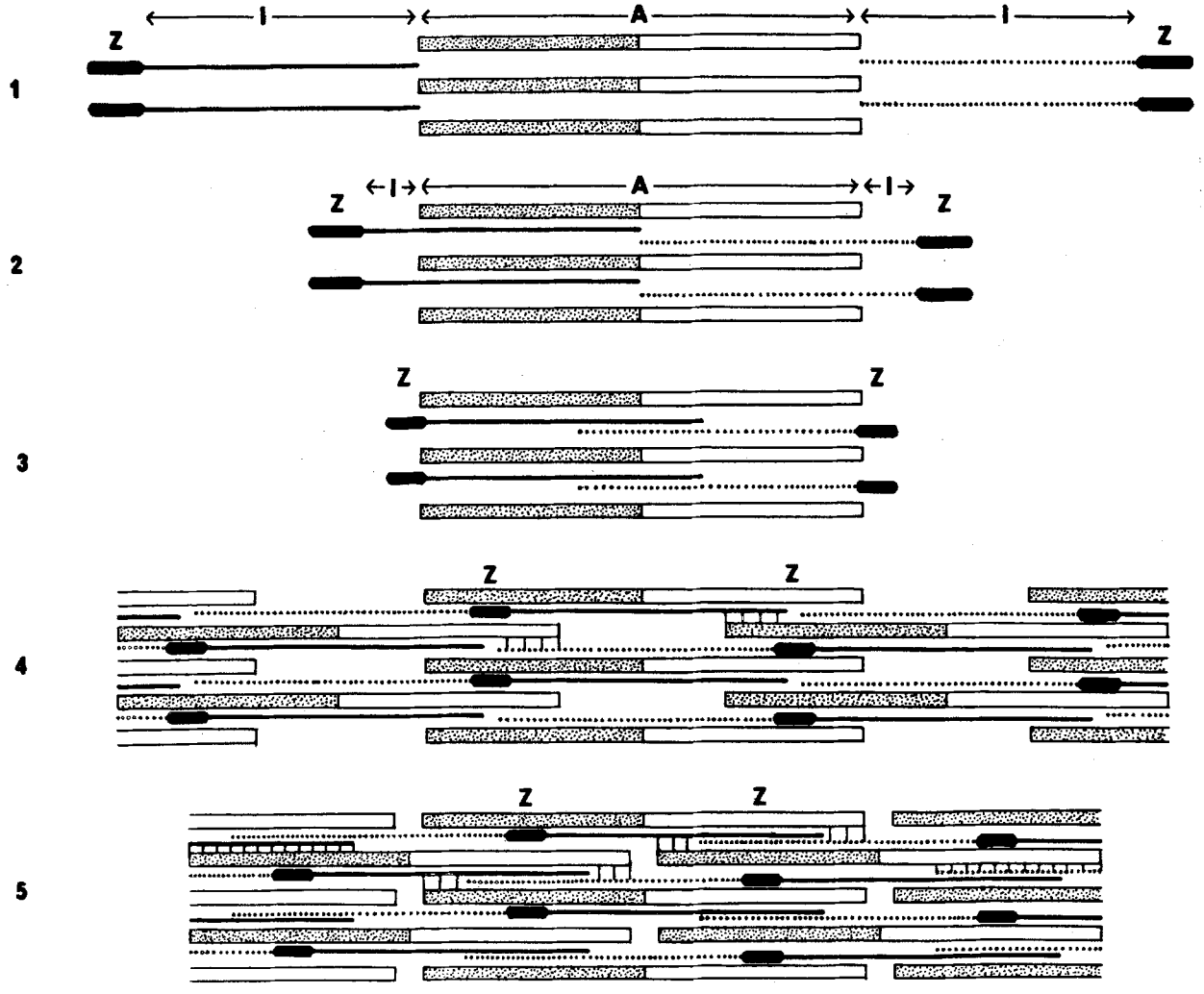

Fig. 1. Diagram to show the hypothetical myofilament array in a supercontracting body-wall muscle of the larval blowfly during contraction, modified from Osborne[5]. For ease of presentation the myofilaments are drawn in register, although electron microscopy demonstrates a more staggered alignment[5, 7]. It is assumed that the thin filaments on either side of the Z line have opposite polarities, and that each half of the thick filament has opposite polarity. The polarities are indicated in the diagram such that the thick filament marked ▩ will form cross bridges with the thin filament —. Thick filament □ will form cross bridges with the thin filament · · ·. Possible cross bridges formed between thick and thin filaments of adjacent sarcomeres are shown in stages 4 and 5. Myofilament lengths are those of *Calliphora* muscle[7]. Thick filament length = 3.2 μm; thin filament length = 2.0 μm. Z band thickness decreases from 0.4 μm in relaxed muscle to 0.3 μm in the contracted state. Stage. 1, sarcomere fully stretched 7.6 μm; 2, sarcomere length 4.4 μm; 3, sarcomere length 3.3 μm. Thick filaments begin to penetrate the Z disc; 4, sarcomere length 2.2 μm. Thin filaments are penetrating the Z disc; 5, sarcomere length 1.7 μm.

capable of maintaining active tension over a 90% length change. More recently MILLER[26] has described the tension/length curves of the leech dorsal longitudinal muscle, an obliquely striated muscle possessing Z rods. The working range afforded by this structure lies between classical striated muscle and blowfly supercontracting muscle.

At sarcomere lengths in supercontracting muscle, where the thick filaments begin to penetrate the Z disc (Figure 1, stage 3), it might be expected that there would be a drop in tension as the possible cross-bridge sites are reduced[5]. However, OSBORNE[5] predicted that the staggered arrangement of the myofilaments within a supercontracting sarcomere may account for smooth contraction as penetration of the thick filaments is asynchronous. The smooth active tension curve seen with a whole fibre at shorter lengths (Figure 2) may to some extent reflect this prediction but probably also reflects heterogeniety of sarcomere length.

Comparison of the passive tension/length curves (Figure 3B) also shows the similarity of blowfly larval muscle to nonstriated muscle. The striated muscles show a great resistance to stretch, due to the parallel elastic elements. However, the non-striated muscles and blowfly muscle show much less resistance. Indeed, high resistance to stretch and a wide working range are incompatible.

NAGAI and GRAHAM[27] claim that cockroach proctodeal muscles are supercontracting, but a previously published tension/length curve[18] does not reflect this, as active tension was not recorded during tetanus. *Limnophilus* (Trichoptera) abdominal muscles do, however, show a wide working range[17] but there are apparently no reports on the ultrastructure of this muscle. It has long been known that slight variation in striated muscle structure gives rise to muscles with widely varying physiological properties[28]. The presence of perforated Z discs endows supercontracting muscles with certain properties more usually associated with smooth muscles. Thus the functions of non-striated muscles in other animals may be performed in insects by a modified striated muscle. At present ultrastructural evidence has demonstrated this type of muscle in the body-wall and viscera of dipterans[5,6,29–32] and lepidopterans[33–35]. There is much less evidence for its presence in other orders, although it has been found in a dictyopteran[27].

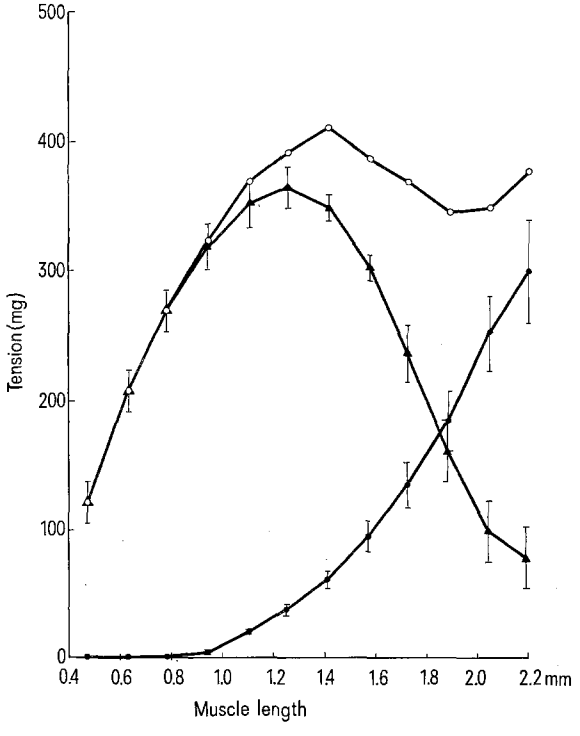

Fig. 2. The tension/length relationship of an isolated longitudinal ventrolateral muscle fibre, 13B[12] of *Calliphora erythrocephala* larva (3rd stage). The figures are a mean of 8 measurements recorded at 20 °C. ○, total tension; ●, passive tension; ▲, active tension; I, standard error limit. Tension was recorded with a transducer manufactured from silicone strain gauges. The maximum series compliance of the recording set up was < 6% of the optimum muscle length. Stimulation was indirect via the segmental nerve, and was supramaximal at 77/sec; this was not maximum tetanus but was chosen to prevent fatigue. Tension in the muscle rose sharply to a plateau, which was measured. The preparation was submerged in a physiological saline[13]. All measurements were made as the preparation length was increased. With this indirect type of stimulation it is possible that stretching the muscle has an effect on activation. This would not however, detract from the wide working range, although it may affect the shape of the active and total tension/length curves.

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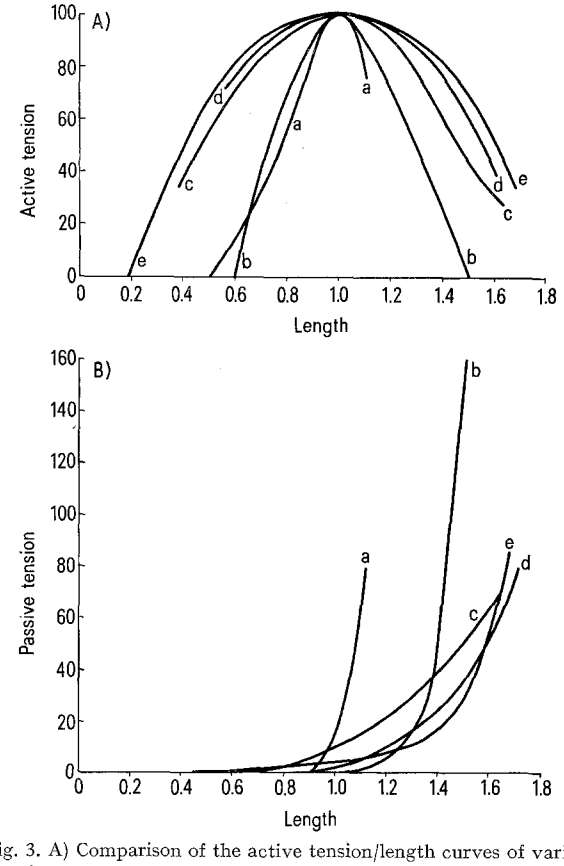

Fig. 3. A) Comparison of the active tension/length curves of various muscles.
B) Comparison of the passive tension/length curves of various muscles. a) Locust flight muscle[16]. b) Frog or toad sartorius muscle[20]. c) *Calliphora* muscle 13B. d) *Mytilus* anterior byssal retractor muscle (ABRM)[21]. e) Guinea-pig taenia coli muscle[14]. Tension is expressed as a percentage of maximum active tension, and length relative to the optimum length (where maximum active tension is produced).