The relationship between activity pattern and muscle adaptation in skeletal muscle

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Muscle is highly plastic in terms of size (maximum force), speed, maximum power and endurance. Well-controlled studies in animals have shown that the adult skeletal muscle fibre has a remarkable ability to modify its gene expression so that with long term substantial changes in the daily activity pattern the contractile phenotype can be modified across the whole spectrum of fibre type found in control muscle. The contractile phenotype in this context includes the isoform content of myosin and therefore the maximum velocity of shortening, the mitochondrial content and therefore the specific force and aerobic capacity (endurance), and the calcium handling proteins and therefore the speed of activation and relaxation. With voluntary exercise in human subjects similar responses are observed, although the degree of transformation is restricted by the practical limitations of exercise dosing to changes in mitochondrial activity and muscle size rather than the more profound changes in contractile protein isoform that can be induced with artificial activation over a substantial proportion of the day.

Physical exercise is widely acknowledged to have a beneficial effect, and conversely a sedentary lifestyle is considered to increase the risk of diabetes, stroke, heart disease and pathology related to obesity. Various attempts have been made to specify what level of activity is required to negate the harmful effects of inactivity, but it is difficult to make universally applicable recommendations. For example the National Health Service in England recommends approximately 150 minutes per week of moderate exercise, defining moderate as the level at which one is able to speak but beginning to find difficulty in singing! Refinement of such recommendations exists on a progressive scale so, for example, sports scientists might suggest exercise at a level that increases the heart rate to a certain proportion of the calculated maximum, or increases oxygen consumption by a certain amount. There is an enormous amount of informal wisdom available for individuals who wish to improve their endurance or strength. For example, in very general terms, strength training programmes are designed to ensure that the participant exercises each target muscle group to the extent that voluntary contraction at that level is no longer achievable. In other words, that the muscle, at least temporarily, is overloaded. The general principle is that such overload induces adaptive mechanisms in the muscle that produce changes that increase the load or volume of contraction at which failure occurs.

But what mechanism or pathway in muscles is triggered by overload, and results in adaptations that can be seen as a homeostatic mechanism to change the properties of the muscle so that the overload is negated? There is now a large literature on potential adaptive mechanisms and it is clear that they comprise hundreds of signalling molecules and molecular pathways. Egan and

Zierath[1]provide a recent review that can be recommended. In terms of the sensed parameter, muscle cells have been shown to be responsive via enzyme activity that is modulated by changes in the stretch of the membrane it which it is located (reviewed in [2], by signalling molecules that are activated by a reduction in the energy supply available to the contracting muscles[3], by signalling molecules that are activated by changes in intracellular calcium (which is also the immediate trigger for contraction)[4] and by ion channel mechanosensors that change their permeability in membranes under load. Some kinases such as focal adhesion kinase are part of structures that connect the sarcomeres to the tension-bearing components of the myofascia and are thus ideally placed to sense changes in the integrated load experienced by muscles [5]. However it is true to say that the relative importance of these various mechanisms is difficult to judge at present, and that there is redundancy and interaction among the adaptive systems. Most of them have been identified by measuring responses to extreme experimental changes of loading, such as unloading by hind limb suspension in rodents or overloading by removing or disabling agonistic muscles. Further insight is gained when a link can be established between mutation of the gene product in question and a recognised human congenital abnormality of muscle function.

It is, however, well established that muscle adaptation involves pathways that have profound adaptive effects within individual muscle fibres. For example, in the transformation of a muscle fibre from the fast type to the slow type that takes place with cross reinnervation of a fast muscle with a slow nerve or by imposing a continuous low frequency electrical activation, the fast isoforms of myosin that constitute the thick filaments of the sarcomere are progressively replaced by slower isoforms over a time course of a few weeks[6]. Such a remarkable change of phenotype in a continuously working muscle requires coordination of gene expression to generate the new proteins, as well as control of the removal and probable recycling of the fast protein isoforms.

One consequence of the discovery of so many intracellular pathways that are potentially involved in sensing and responding to changes in activity in muscle, is that this diversity of mechanisms may well be differentially sensitive among individuals. And therefore exercise programs defined in terms of, for example, exercise at a particular power output per kilogramme of body mass, may produce the anticipated benefits in one individual but not in another. In this context it may be that exercise programs that are designed with some acceptance of individual variability, and based for example on exercise to exhaustion, may have a better chance of effectiveness in a mixed cohort than programmes that ‘dispense’ a fixed exercise dose.

Furthermore, there is some evidence that some effects of exercise may not be continuously variable but may require the crossing of some threshold of activity (or cellular response to activity) in order to trigger a change. Salmons[7, 8] discussed the concept of relatively stable phenotypic types that we observe as muscle fibre types. There is still no comprehensive study of the response space of muscle in terms of the relationship between ‘amount’ or ‘pattern’ of activity as an input and muscle phenotype as an output. However, the discovery of early markers of change (such as changes in key mRNA levels) mean that we can design experiments that look in detail at the initial response of a muscle to a new pattern of activity without the need for long and expensive experiments designed to observe the ultimate outcome of a period of modified activity. Once hypotheses have been generated in short term experiments, then the long term outcomes can be investigated .

What therefore are the determinants of muscle phenotype? The simplest and perhaps the most important hypothesis is that the total amount of activity per day is the primary determinant of muscle fibre type. In other words, that muscles (or more accurately motor units) that receive a large number of action potentials become slow contracting, with a high volume percentage of mitochondria. They are therefore suited for endurance activity, but cannot produce a high power output per unit mass because the myosin ATPase does not hydrolyse ATP quickly and a large proportion of the muscle cell volume is dedicated to oxidative production of ATP rather than generation of force. In contrast, motor units that receive low daily amounts of activation express fast isoforms of myosin and have a higher proportion of the muscle cell volume made up by contractile units (sarcomeres) because they contain a lower volume percentage of mitochondria. Because ATP can be supplied to the contractile units at a high rate for a short time by anaerobic glycolysis, these motor units have a high maximum power output, but their action cannot be sustained and they succumb to fatigue in repeated series of contractions.

Such a control mechanism would thus require the muscle cell to direct its transcriptional activity in response to the integrated number of activations. This could be signalled by an integration of calcium spikes in the cytoplasm, or possibly an integration of substrate consumption (ATP) or product release (AMP). Hennig and Lomo[9] showed that the activity patterns of motor units in a mixed muscle tended to cluster by total active time (number of action potentials per day).

This is not the complete picture however. Hennig and Lomos paper also showed that motor unit firing properties also tend to cluster by burst frequency, although for the motor units that were assumed to be the fast types, the frequency ranges overlap considerably. On this basis, several authors have emphasised the effect of the frequency of activation within an individual contraction, pointing to the fact that fast motor units tend to be activated with a high neural firing rate, whereas slow motor units tend to be activated with a low neural firing rate. Thus there is the beguiling concept that high frequencies tend to make a muscle fast and low frequencies slow. If this were true, then there would need to be a mechanism whereby a motor unit detected the frequency of activation. Of course it is possible to imagine a cellular resonant circuit that discriminated between high and low frequency, but we also have to bear in mind that the contractile consequence of activation at high and low frequency is already different. Because the motor unit acts as an integrator, the force generated within a burst of firing from a motor unit depends on the frequency of firing during the burst. Low frequencies produce oscillatory movements because the process of activation, sarcomere shortening, and relaxation produced by a single impulse is complete before the next pulse arrives at the endplate. On the other hand if the frequency is high enough that the following pulse arrives before relaxation from the prior pulse is complete then the effect of the second pulse produces an additive effect. Thus, the force produced by a motor unit is related to the frequency of activation, the relationship being s-shaped but approximately linear between about 10 Hz and 50 Hz for a typical human muscle.

Westgaard and Lomo [10]in their remarkable series of experiments during the 80s did claim that the effect of high frequency to make a muscle fast was still detectable even when the total amount of stimulation was very large. In denervated rat soleus they delivered 100Hz bursts for 0.6s every 1.2s, an average frequency of 50Hz, and claimed that the muscle was still faster to contract and relax than muscle stimulated with a large amount of low frequency stimulation. This finding has not been adequately followed up and deserves reappraisal in both denervated and innervated muscles.

However, in stimulation experiments that have used bursts of stimulation to produce fused tetani (and therefore relatively high frequencies) over several weeks, the principle that it is necessary to keep the total amount of activity as low as possible to prevent unwanted slowing of the muscle has been supported[11]. It was also the basis of 'demand stimulation' in the clinical application known as cardiomyoplasty, in which trained skeletal muscle was used to assist the heart.[12]

On the other hand, some muscles do have very high levels of habitual activation and appear to be able to maintain this high level without further adaptive change. The diaphragm and the laryngeal muscles are examples.

Sutherland et al [13]investigated whether a muscle would show a differential response between amount of impulses and integrated contractile response by using optimised bursts. They exploited the doublet effect in fast muscle to produce greater integrated force than is elicited by uniform inter pulse intervals. They showed that with patterns of activation that had a daily average frequency of 2.5 Hz, or 21600 pulses per day, the muscle weight and the speed of activation were dependent on the pattern of delivery whereas the speed of shortening, the proportions of the myosin isoforms and resistance to fatigue were not different between the two groups. This demonstrates that we may not consider muscle fibres as having fixed relationships between speed and size and fatigue resistance. To some extent at least these aspects can be differentially regulated.

Strength and conditioning in human sport science.

Such discussion of the precise relationship between pattern of activation and muscle response may seem rather remote from the very well-publicised world of human performance training. Sportsmen and women are familiar with the difference between strength (or resistance' training and 'endurance' training. The traditional pattern is that strength training should involve few contractions at high intensity whereas endurance training should involve longer periods of exercise at lower intensity. However, this dichotomy is over simplified; for example, adaptations to improve general fitness, defined as the ability to use oxygen to generate muscle work, can also be achieved by brief periods of high intensity exercise whose merit may be that such a schedule is more practicable for the modern time-pressured lifestyle.[14]

To begin to understand this additional complexity we must consider that, in normal activity, the motor units within a muscle are not all recruited at the same time and with the same pattern of activation. The previous discussion of muscles responding to patterns and amounts of activity assumes that an experimental muscle can be considered as if it were one large motor unit. This is a useful approximation in experiments in which the whole muscle can be activated synchronously by implanted electrodes. However the claimed benefits in time-saving of short bursts of high intensity exercise (sprint interval training) over longer periods of lower intensity exercise probably have to do with the principle of progressive recruitment. In a mixed muscle, there exist some motor units that are only recruited when maximum force is required. In normal contemporary daily life, it is rare for maximum force to be required because we rarely lift at our maximum capacity, or push a load that is almost too heavy for us to move, or need to jump as high as we are able. However, if we purposely cycle or run at maximum capacity for a brief period than signalling pathways in these rarely-activated motor units may be triggered and a useful systemic response gained.

Therefore, in novel clinical applications of functional electrical stimulation, there are some general principles as already discussed that can help in choosing an appropriate pattern of stimulation. There are a number of examples in which experimental studies have attempted to find a pattern of daily activation that achieved a particular contractile phenotype. Just one example is given here to illustrate the logic involved. Cardiomyoplasty is a term used to refer to the use of skeletal muscle to assist the failing heart[15]. In this case, untrained skeletal muscle was known to be incapable of working continuously at the typical continuous work rate of heart muscle. On the other hand, it was shown that continuous low frequency stimulation of skeletal muscle could produce a transformation so profound that skeletal muscle could not only become so resistant to fatigue that it could work continuously at a similar work rate to that of the heart, but also that too much adaptation could cause such extreme slowing that its assistive potential was compromised[16]. Experiments in rabbits showed that transformation to a stable intermediate state was achievable[17], and subsequent work showed that such worked could be translated to larger animals, but that it was necessary to apply a scaling factor to the timing parameters[11].

The desire to improve muscle properties by exercise or activation by an assistive technology such as electrical or magnetic stimulation has various aspects, illustrated by the following statements:

This muscle is too weak; I want to make it stronger

This muscle fatigues too quickly; I want to make it work for longer

This muscle does not respond at all to electrical stimulation; I want to make it excitable

This muscle hurts too much for me to go training; I want to speed up recovery

My muscles are not what they used to be

These questions and longings are an integral part of contemporary society. There is work to be done in academic science laboratories, and we have better tools now than ever before to investigate the relationship between muscle use and muscle phenotype. In particular we are able to perform transcriptomic, proteomic and metabolomics studies even on very small pieces of trained muscle. We are able to recruit muscle in experimental animals with precisely programmed patterns of exercise using miniature implantable muscle stimulators. And an increasing number of laboratories have the expertise and permission to take muscle samples from human subjects.

Because of the global obsession with sports, fitness and body image, there is a huge amount of informal experimentation based on novel coaching practices in elite sport that are not accessible in the same way as formal experimental studies, and a very lively online debate based almost exclusively on individual case reports . These often discuss complex programmes of exercise, usually with the aim of strengthening and achieving an ideal physique, but also notable for being a lifestyle choice that takes up a great deal of time. One of the main reasons for the large time cost is the desire to exercise each muscle group say three times a week. And thus with a modest estimate of 12 target muscle groups (for example, flexors and extensors of ankle, knee, hip, elbow, shoulder plus

abdominal and spinal groups) this requires at least 3 intensive gym sessions per week. At the other end of the scale there are many persons that would like to exercise more to gain the benefits of physical exercise but are restricted by joint pain, weakness, muscle injuries or cramping. If we wish to keep our population more able and less dependent into old age, we must also concentrate on these problems and find solutions that are evidence-based. These will include the use of physical therapy, and warm up routines to minimize injury but the great majority of work in this area is focused on the young and in particular the young athlete or sports player. A significant problem in the current approach to exercise science is perhaps that almost any experimental manipulation of timing of food intake, timing of exercise, pattern and intensity of exercise can be shown in normal muscle to be influential on protein synthesis or signaling pathways. This is a genuine difficulty because there is the possibility of almost infinite variation for both voluntary and imposed muscle contraction in terms of the combination of frequency, burst length, interburst interval, (duty cycle) activity/rest ratio, load, level of recruitment, sets, reps, sessions per day, sessions per week, warm up, warm down, tapering towards a competition or maximal effort and so on.

We need a period of synthesis of research findings into models that can predict which practical lifestyle interventions are most likely to be beneficial in which part of the human population. A stark example of the potential inapplicability of research findings in healthy humans to groups in need of intervention is the lack of response of obese sedentary individuals to the benefits that might be expected from exercise[18]. Another aspect of applicability to all individuals is that there is increasing evidence that the adaptive process itself can become defective. Indeed, defective protein processing is not only well-established in neurodegenerative disease such as Alzheimer's but also now in cardiac and skeletal muscular disorders[19].

In conclusion, we have a large array of evidence on which to base decisions concerning prescription of exercise and the potential therapeutic use of functional electrical stimulation. We also have a large number of powwerfulr techniques and well-equipped laboratories to provide further evidence. As ever, there is the need to perform disciplined experiments that are designed to discriminate between competing hypotheses. As in many other fields, we need to integrate the information provided by investigations of single pathways into a holistic understanding of muscle adaptation. The new techniques of proteomics, metabolomics and pathway analysis will help us in this task.

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