**Name of journal: World Journal of Translational Medicine**

**ESPS Manuscript NO: 3809**

**Columns: REVIEW**

**Hypo-activity induced skeletal muscle atrophy and potential nutritional interventions: A review**

Bostock EL *et al.* Disuse associated muscle wasting

Emma L Bostock, Christopher I Morse, Keith Winwood, Islay McEwan, Gladys LOnambélé-Pearson

**Emma L Bostock, Christopher I Morse, Keith Winwood, Islay McEwan, Gladys LOnambélé-Pearson,** Institute for Performance Research, Department of Exercise and Sport Science, Manchester Metropolitan University, Crewe, CW1 5DU, United Kingdom

**Author contributions:** All the authors contributed to this work.

**Correspondence to: Gladys L Onambélé-Pearson, PhD,** Department of Exercise and Sport Science, Manchester Metropolitan University, Crewe Green Road, Valentine Building, Room 2-7, Crewe, CW1 5DU, United Kingdom. g.pearson@mmu.ac.uk

**Telephone:** +44-161-2475594 **Fax:** +44-161-2476386

**Received:** May 24, 2013 **Revised:** September 7, 2013

**Accepted:** November 1, 2013

**Published online:**

**Abstract**

Periods of hypo-activity result in profound changes in skeletal muscle morphology and strength. This review primarily addresses the differential impact of de-training, bed-rest, limb immobilisation and unilateral lower limb suspension on muscle morphology, strength and fatigability. The degree of muscle atrophy differs depending on the hypo-activity model and the muscles in question, with the leg and postural muscles being the most susceptible to atrophy. Hypo-activity also results in the dramatic loss of strength that often surpasses the loss of muscle mass, and consequently, the nervous system and contractile properties adapt to adjust for this excessive loss of strength. In addition, the degree of muscle strength loss is different depending on the hypo-activity model, with immobilisation appearing to have a greater impact on strength than unloaded models. There is a step-wise difference in the magnitude of muscle loss so that, even after accounting for differential durations of interventions immobilisation ≥ unilateral lower limb suspension ≥ bed-rest ≥ de-training. Muscle fatigability varies between hypo-activity models but the results are equivocal and this may be due to task-specific adaptations. This review also addresses potential nutritional interventions for attenuating hypo-activity induced muscle atrophy and strength declines, in the absence of exercise. Essential amino acid supplementation stands as a strong candidate but other supplements are good contenders for attenuating hypo-activity induced atrophy and strength losses. Several potential nutritional supplements are highlighted that could be used to combat muscle atrophy but extensive research is needed to determine the most effective.

© 2013 Baishideng. All rights reserved.

**Key words:** Immobilisation; Disuse; Muscle size; Muscle strength; Nutrition supplementation; Muscle fatigability

**Core tip:** This review summarises and compares the morphological, strength and fatigability changes in response to different models of hypo-activity. The hypo-activity models include de-training, bed-rest, immobilisation and unilateral lower limb suspension. There is a step-wise difference in the magnitude of muscle and somewhat strength losses so that, even after accounting for differential durations of interventions immobilisation ≥ unilateral lower limb suspension ≥ bed-rest ≥ de-training. Muscle fatigability varies between hypo-activity models but the results are equivocal and this may be due to task-specific adaptations. This review also highlights several potential nutritional interventions for attenuating hypo-activity induced changes.

Bostock EL, Morse CI, Winwood K, McEwan I, Onambélé-Pearson GL. Hypo-activity induced skeletal muscle atrophy and potential nutritional interventions: A review

**Available from:**

**DOI:**

**INTRODUCTION**

Skeletal muscle is one of the most adaptable tissues in the body, and as such, it is capable of altering its structure in response to different levels of physical activity. Prolonged reductions in muscle activity and mechanical loading result in many physiological adaptations in skeletal muscle form and function [[1-4](#_ENREF_1)]. Muscle atrophy (decrease in muscle mass) is seen during reduced activity (*e.g.*, sedentary behaviour, de-training) [[5-8](#_ENREF_5)] or disuse models (*e.g.*, immobilisation, head-down tilt bed-rest) [[1](#_ENREF_1), [3](#_ENREF_3), [9](#_ENREF_9), [10](#_ENREF_10)]. It is evident that the degree of muscle atrophy is not constant across muscle groups or hypo-activity models [[2](#_ENREF_2),[3](#_ENREF_3),[11](#_ENREF_11),[12](#_ENREF_12" \o "Yue, 1997 #847)].

Simply reducing normal levels of activity can be classed as the first stage of disuse. Decrements in muscle mass and strength have been documented in trained humans undergoing de-training [[5-8](#_ENREF_5), [13-15](#_ENREF_13)]. Bed-rest conditions result in the removal of normal weight-bearing forces acting on the bones of the lower limbs in the vertical position and a decrease in number and/or magnitude of muscle contractions, particularly in the postural musculature. During bed-rest, muscular contraction is still possible although it is limited and the muscular force required for producing movement is very much diminished once ground reaction forces are removed. A more rigid immobilisation can be achieved by casting a limb, resulting in more rapid decrements in muscle mass than does bed-rest alone. The final method of hypo-activity commonly reported in the literature is that of unilateral lower limb suspension (ULLS), a method of reducing habitual activity whilst causing lesser degree of inconvenience to the participants.

The purpose of this review is to assess the varying impact of different hypo-activity models on the skeletal muscle system. This is broken down into the effects of hypo-activity on muscle morphology, muscle strength and muscle fatigability. In order to provide some homogeneity in the results based on the variable duration of the hypo-activity, values are presented per week and where relevant the duration of the hypo-activity is provided in parenthesis. Exercise prescription is not always a practical prescription, even when it would be recommendable, to individual’s under-going immobilisation or bed-rest after trauma or illness, due to the presence of counter indications for exercise such as pain, immobilisation in a cast, *etc.* Thus, other interventions are required to attenuate losses in muscle mass and function. Therefore, this review will also discuss potential nutritional interventions for preventing the loss of muscle mass/function seen with hypo-activity, where increased physical activity is not combined with the nutritional treatment. Studies were found using search terms “bed-rest and atrophy” and “immobilisation and atrophy” in PubMed. However, this returned over 1400 hits. To focus our search criteria, only data on healthy humans were selected through the inclusion of the “human” and “clinical trial” filters in the PubMed search. This resulted in 86 studies, suitable for inclusion in the present review.

**MUSCLE MORPHOLOGY**

***Muscle anatomical cross sectional area***

Anatomical cross sectional area (ACSA) is the cross-sectional area of the muscle at right angles to its longitudinal axis. Muscle ACSA is a major determinant of maximum voluntary contraction (MVC) torque [[16](#_ENREF_16), [17](#_ENREF_17)] and hypo-activity models have been shown to result in the decrease in this parameter. Figure 1 shows relative change in muscle anatomical cross sectional area (ACSA), physiological cross sectional area (PCSA) and volume in response to hypo-activity models. Values are taken from the references used in the text for de-training (ACSA – 40 d and 24 wk) [[6](#_ENREF_6), [8](#_ENREF_8)], bed-rest (ACSA – 30 d to 17 wk) [[2](#_ENREF_2), [11](#_ENREF_11), [18](#_ENREF_18), [19](#_ENREF_19)] (PCSA – 20 d) [[1](#_ENREF_1), [9](#_ENREF_9), [20](#_ENREF_20)] (Volume – 7 d and 32 d) [[11](#_ENREF_11), [21](#_ENREF_21)], immobilisation (ACSA – 9 d to 4 wk) [[3](#_ENREF_3), [4](#_ENREF_4), [12](#_ENREF_12), [22-26](#_ENREF_22)] (Volume – 2 wk and 4 wk) [[10](#_ENREF_10), [12](#_ENREF_12), [27](#_ENREF_27)] and ULLS (ACSA – 23 d and 4 wk)[[28-31](#_ENREF_28)]. Where there are missing bars, this shows gaps in the literature (*i.e.*, values are not available for a parameter during a specific hypo-activity model). Values are presented as means; error bars denote SD. Periods of detraining (24 wk) have resulted in a decrease in ACSA of the quadriceps [[6](#_ENREF_6)]. Likewise, Narici *et al*[[8](#_ENREF_8)] reported decreases in leg ACSA (approximately 0.7%/wk) in response to 40 d de-training.

Stricter hypo-activity models result in greater decreases in muscle ACSA. Following 30 d bed-rest Convertino *et al*[[11](#_ENREF_11)] reported decreases in ACSA of the calf (approximately 1.1%/wk) and thigh (approximately 1.9%/wk). Similarly, a 2.4%/wkdecrease in plantar flexors was found following 5 wk horizontal bed-rest [[18](#_ENREF_18)]. Muscle group-specific adaptations have been demonstrated in skeletal muscle ACSA of the leg and lumbar musculature after 17 wk of bed-rest [[2](#_ENREF_2)]. The plantar-flexors were more susceptible to atrophy (approximately 1.8%/wk) than the dorsiflexors (approximately 0.9 to 1.2%/wk) [[2](#_ENREF_2)]. The intrinsic lumbar muscles atrophied approximately 0.5%/wkbut there was no significant change in psoas muscle mass[[2](#_ENREF_2)]. Rittweger *et al*[[19](#_ENREF_19)] reported a decrease in calf muscle ACSA (approximately 2.0%/wk), which was greater than the reported decrease in the forearm ACSA (0.5%/wk) in response to 90 d bed-rest.

Immobilisation of the leg through plaster cast has shown to decrease calf ACSA (approximately 3 to 5%/wk) after just 2 wk [[4](#_ENREF_4), [22](#_ENREF_22)]. Changes in quadriceps ACSA (approximately 8.3%/wk) have also been documented with as little as 10 d leg cast immobilisation [[25](#_ENREF_25)]. Similarly, Veldhuizen *et al*[[3](#_ENREF_3)] reported decreases in quadriceps ACSA (approximately 5.3%/wk) with 4 wk leg casting. Immobilisation of the knee using a brace has also resulted in decreases in muscle ACSA [[24](#_ENREF_24), [26](#_ENREF_26)]. Fourteen days of knee-brace immobilisation has resulted in ACSA decreases of the thigh (approximately 3.1%/wk), quadriceps (approximately 2.9 to 3.8%/wk), gastrocnemius (approximately 4.7%/wk) and soleus (approximately 3.3%/wk) muscles [[24](#_ENREF_24), [26](#_ENREF_26)]. Yasuda *et al*[[26](#_ENREF_26)] found no sex-based differences in the quadriceps ACSA response to knee-brace mediated immobilisation. There is considerably less data on immobilisation-induced atrophy of the upper limb muscles. Casting of the arm for as little as 9 d has shown to decrease ACSA of the forearm (approximately 3.2%/wk) [[23](#_ENREF_23)]. Yue *et al* [[12](#_ENREF_12)] investigated the effect of 4 wk elbow joint immobilisation with a fibre glass cast and reported a decrease in elbow flexor ACSA (approximately2.8%/wk).

Tesch *et al*[[32](#_ENREF_32)] developed a model to study the effects of an unloaded limb in humans that allows for freely moveable joints but minimises load bearing. In this ULLS method, a sling suspends one lower leg and the contralateral shoe has an elevated sole to allow for a relaxed position of the unloaded limb. ULLS also results in decreases in muscle ACSA, though to a lesser degree than seen with immobilisation. ULLS of 23 d has been reported to decrease knee extensor (approximately 3%/wk) [[30](#_ENREF_30)] and plantar flexor (approximately 2.7%/wk) [[31](#_ENREF_31)] ACSA. Correspondingly, Clark *et al*[[28](#_ENREF_28),[29](#_ENREF_29)] reported decreases in plantar flexor (approximately 2.0 to 2.3%/wk) and knee extensor (approximately 2.0%/wk) [[29](#_ENREF_29)] ACSA in response to 4 wk ULLS. It would therefore seem that in terms of ACSA at least, the most impactful model of hypo-activity is immobilisation.

***Muscle physiological cross sectional area***

Physiological cross-sectional area (PCSA) is the area of the muscle at right angles to the longitudinal axis of the fibres. Muscle PCSA has been associated with the maximal force generating capacity of a muscle [[33](#_ENREF_33)] and has been shown to decrease with bed-rest [[1](#_ENREF_1), [9](#_ENREF_9), [20](#_ENREF_20)]. Twenty days bed-rest has been shown to decrease PCSA of the thigh (between approximately 2.7 to 3.6%/wk) [[1](#_ENREF_1), [20](#_ENREF_20)]. Akima *et al*[[9](#_ENREF_9)] demonstrated muscle group-specific adaptations, demonstrating a decrease in PCSA of knee extensor (approximately 2.5%/wk), knee flexor (approximately 4.0%/wk) and plantarflexor (approximately 4.5%/wk) muscles in response to 20 d of 6 degrees head-down-tilt bed rest. It is generally accepted that muscle losses are greater in the knee extensors than the knee flexors after unloading in humans[[34](#_ENREF_34)]. Akima *et al*[[9](#_ENREF_9)] demonstrated the opposite to this, which could be due to the methodology used to determine PCSA. In addition, since a muscle placed in a shortened position experiences a greater degree of atrophy than one placed in a lengthened position [[35](#_ENREF_35)], the pattern/magnitude of disuse would therefore be expected to be modulated by both the mode of hypo-activity and the joint angle adopted in the immobilisation. Bed-rest, however, had no effect on the PCSA of the tibialis interior [[9](#_ENREF_9)]. The tibialis anterior experiences lower activation during habitual physical activities than other muscles such as the plantar flexor, and as such may explain the lack of decrease in tibialis anterior muscle PCSA with bed-rest. Comparisons of bed-rest to other hypo-activity models in terms of PCSA changes is not yet possible, as research is lacking with this parameter being measured.

***Muscle volume***

Muscle volumeis a major determinant of joint torque [[36](#_ENREF_36)] and has been shown to decrease in response to bed-rest and immobilisation models [[10-12](#_ENREF_10),[21](#_ENREF_21),[27](#_ENREF_27)]. Muscle volume of the thigh decreases (approximately 3%/wk) with as little as 7 d bed rest [[21](#_ENREF_21)]. Following 30 d bed rest, Convertino *et al*[[11](#_ENREF_11)] reported decreases in calculated leg volumes of the calf (approximately 2.3%/wk) and thigh (approximately 1.1%/wk). Yue *et al*[[12](#_ENREF_12)] investigated the effect of 4 wk elbow joint immobilisation with a fibre glass cast and reported a decrease in elbow flexor volume (approximately 2.9%/wk). A case study of a fracture patient who fractured the fifth metatarsal of the right foot displayed substantial and rapid losses in muscle volumes, both proximally and distally to the immobilisation site after 4 wk subsequent immobilisation [[10](#_ENREF_10)]. The degree of muscle volume decrease varied between the different muscle sites of the triceps surae (approximately 5.5%/wk), quadriceps (approximately 6.0%/wk) and hamstrings (approximately 1.6%/wk) [[10](#_ENREF_10)]. This is in agreement with the general acceptance that muscle volume is lost to a greater extent in the knee extensors compared to the knee flexors [[34](#_ENREF_34)]. Urso et al demonstrated different responses to 2 wk adductor pollicis (AP) immobilisation between younger and older males [[27](#_ENREF_27)]. AP volume decreased approximately 2.1%/wk (not significant) in young males and significantly decreased by approximately 4.8%/wk in older males [[27](#_ENREF_27)].

***Upper vs lower limb***

Immobilisation through casting appears to have a greater effect on the lower limb musculature than the upper body. This is not surprising since the habitual loading of the lower extremities, because of body weight in normal ambulation and even in the absence of intended physical exertion, is far more substantial than that in the upper extremities. Understandably, this thereby affects the required threshold of decrease in muscle activity necessary to negatively impact on muscle metabolism. [Relative change in muscle ACSA, PCSA and volume in response to hypo-activity models. Values are separated into the effect of each hypo-activity model on the upper limb (UL) versus the lower limb (LL). The values are taken from the references used in the text for de-training (ACSA\_LL) [[6](#_ENREF_6), [8](#_ENREF_8)], bed-rest (ACSA\_UL) [[19](#_ENREF_19)] (ACSA\_LL) [[2](#_ENREF_2), [11](#_ENREF_11), [18](#_ENREF_18), [19](#_ENREF_19)] (PCSA\_LL) [[1](#_ENREF_1), [9](#_ENREF_9), [20](#_ENREF_20)] (Volume\_LL) [[11](#_ENREF_11), [21](#_ENREF_21)], immobilisation (ACSA\_UL) [[12](#_ENREF_12), [23](#_ENREF_23)] (ACSA\_LL) [[3](#_ENREF_3), [4](#_ENREF_4), [22](#_ENREF_22), [24-26](#_ENREF_24)] (Volume\_UL) [[12](#_ENREF_12), [27](#_ENREF_27)] (Volume\_LL) [[32](#_ENREF_32)] and ULLS (ACSA\_LL) [[28-31](#_ENREF_28)]. Where there are missing values, this shows gaps in the literature *(i.e.*, values are not available for a parameter during a specific hypo-activity model) (Table 1)]. Forearm muscle ACSA decreased 4.1% with 9 d arm casting [[23](#_ENREF_23)], whereas, a similar period of immobilisation of the lower limb with 10 d casting resulted in an 11.8% decrease in quadriceps ACSA [[25](#_ENREF_25)]. Similarly, with longer periods of immobilisation the effect seems to be greater in the lower limbs. In response to 4 wk elbow joint casting, Yue *et al*[[12](#_ENREF_12)] reported an 11.2% decrease in elbow flexor ACSA, whereas, Veldhuizen *et al*[[3](#_ENREF_3)] reported a 21% decrease in quadriceps ACSA in response to 4 wk leg casting.

***Intramuscular adipose tissue***

Using signal intensity analysis of lower limb magnetic resonance images (MRI) Manini *et al*[[37](#_ENREF_37)] discriminated between the relative changes in adipose and skeletal muscle tissue resulting from a 4 wk period of ULLS. In addition to the characteristic reduction in muscle ACSA, there was a concomitant 15% increase in intermuscular adipose content after 4 wk of lower limb suspension [[37](#_ENREF_37)]. Thus, these findings suggest, that hypo-activity induced alterations in skeletal muscle morphology goes beyond muscle atrophy alone.

***Summary***

Together, these findings show that the extent of muscle atrophy differs depending on the hypo-activity model. Certain factors may modulate the differential responses to hypo-activity models (*e.g.*, age, nutritional status). Indeed, both Kortebein *et al*[[38](#_ENREF_38)] and Yue *et al*[[27](#_ENREF_27)] suggested that older individuals experience greater losses in muscle mass when compared to younger individuals. A change in nutritional status, whether it is due to physiological changes directly caused by hypo-activity or to altered behaviour that is caused by hypo-activity and leads to changes in diet, could affect the physiological systems in question. The above also suggest that the degree of muscle atrophy differs between muscle groups, with the leg and postural muscles being most susceptible to atrophy. This is likely to be due to the comparatively substantial decrease in habitual weight-bearing forces applied to the lower limb during hypo-activity. Hypo-activity not only decreases muscle content, but also impacts on the intrinsic composition of the said skeletal muscle through increased adiposity [[37](#_ENREF_37)] and altered muscle architecture [[39](#_ENREF_39)].

The decrease in muscle mass seen with hypo-activity may be the result of an imbalance between protein synthesis and protein breakdown [[40-42](#_ENREF_40)]. In response to 14 d simulated microgravity, Ferrando *et al*[[40](#_ENREF_40)] reported a loss of lean muscle mass, accompanied with a 14% decrease in protein synthesis and no change in protein breakdown. Similarly, Gibson *et al*[[41](#_ENREF_41)] reported a marked fall in muscle protein synthesis in response to 7 wk leg immobilisation. A shorter period of immobilisation (21 d) provided little evidence of increases in mRNA for catabolic enzymes or increases in enzyme activity during this period [[43](#_ENREF_43)]. However, there is some evidence to suggest that increases in catabolic potential do occur, and that this event happens very quickly (48 h) after immobilisation[[42](#_ENREF_42)]. Nevertheless, collectively the evidence suggests that protein breakdown is unlikely to be a key modulator in the process of muscle atrophy occurring during immobilisation in humans [[44](#_ENREF_44),[45](#_ENREF_45)].

The molecular signalling responses to de-training are only just beginning to be investigated, and to date, only changes in metabolic proteins have been reported in human skeletal muscle [[46](#_ENREF_46), [47](#_ENREF_47)]. With bed-rest, Ogawa *et al*[[48](#_ENREF_48)] reported increased mRNA expression of the E3 ligases, Cbl-b and Atrogin-1 in response to 20 d bed-rest. This was accompanied by a threefold increase in ubiquitinated proteins[[48](#_ENREF_48)]. Investigation into the effects of limb immobilisation on cell signalling in humans is limited. Modest changes in mRNA for many genes in the first 2 d after immobilisation have been reported but these changes do not affect protein levels of most transcripts [[42](#_ENREF_42)]. However, the Akt protein synthesis pathway and extracellular matrix components seem to be affected within 48 hours of immobilisation [[42](#_ENREF_42)]. Chen *et al*[[49](#_ENREF_49)] and Jones *et al*[ [50](#_ENREF_50)] reported increases in the E3 ligases, Atrogin-1 and MuRF-1 in response to 11 to 14 d immobilisation in humans. These changes were not seen with 48 h immobilisation [[42](#_ENREF_42)] and are therefore thought to only occur after long duration (days rather than hours) immobilisation. Increased metallothionein expression in human skeletal muscle fibres has been associated with exposure to physiological stress, which results in elevated levels of reactive oxygen species (ROS) [[51](#_ENREF_51)]. Urso et al reported a more than two-fold increase in metallothioneins in human skeletal muscle with 48 h of immobilisation [[42](#_ENREF_42)]. However, neither Chen *et al*[[49](#_ENREF_49)] nor Jones *et al*[ [50](#_ENREF_50)] identified changes with longer periods of immobilisation. This may suggest that metallothioneins are increased in the first few days of hypo-activity to prevent ROS-mediated DNA or cellular damage. De Boer *et al*[[43](#_ENREF_43)] investigated the effects of ULLS on gene expression and cell signalling. They reported increased expression of mRNA for MuRF-1 by approximately 3 fold after 10 d without changes in MAFbx or tripeptidyl peptidase II mRNA, but all decreased between 10 and 21 d[[43](#_ENREF_43)]. These authors concluded that both myofibrillar and tendon protein synthetic rates show progressive decreases during 21 d of disuse; in muscle this is accompanied by decreased phosphorylation of FAK, with no marked increases in genes for proteolytic enzymes [[43](#_ENREF_43)]. Overall, whilst it is clear that cell signalling responses differ between hypo-activity models; further research is needed to provide a definitive description of the timing, magnitude and nature of these molecular adaptations.

**MUSCLE STRENGTH**

The associated decline in strength through hypo-activity can be best described based on the mode of assessment. Both isometric and dynamic strength have been reported to decline with hypo-activity, the relative magnitude of which appears to largely reflect the patterns of atrophy described above.

***Isometric strength***

Hypo-activity models alter muscular isometric torque. After 40 d de-training, Narici *et al*[[8](#_ENREF_8)] reported a decrease in knee extension isometric MVC (approximately 2.1%/wk). Similarly, maximum isometric quadriceps strength has been reported to decrease with 90 d de-training (approximately 1.3%/wk) [[5](#_ENREF_5)]. More dramatic losses in isometric torque are seen with stricter hypo-activity models. Bed-rest models have been shown to decrease maximum voluntary force of plantar flexion (approximately 7.5%/wk) [[52](#_ENREF_52)] and knee extensor torque (approximately 4.1 to 5.0%/wk) [[53](#_ENREF_53)]. Correspondingly, Kawakami *et al*[[1](#_ENREF_1)] showed a decrease in muscle force for knee extension (approximately 3.8%/wk) with 20 d bed-rest.

Studies using 2 wk of cast immobilisation have reported decreases in triceps surae isometric MVC torque (approximately 8.5 and 12%/wk) [[4](#_ENREF_4), [54](#_ENREF_54)]. A discrepancy between the two studies may be due to the degree of immobilisation. Gondin *et al* [[54](#_ENREF_54)] simply immobilised the ankle joint, whilst, White *et al*[[4](#_ENREF_4)] utilised a full leg cast. Knee-brace mediated immobilisation has resulted in a decrease in knee extensor and plantar flexion isometric strength (approximately 11.2 and 12.7%/wk, respectively) [[24](#_ENREF_24)]. Knee-cast mediated immobilisation resulted in a slightly larger decrease in isometric leg strength (approximately 15.7%/wk) [[55](#_ENREF_55)]. Christensen *et al*[[22](#_ENREF_22)] utilised a knee-to-toe plaster cast and reported a decrease in isometric calf muscle strength (approximately 4.5%/wk). Studies using casting to immobilise the elbow joint have found decreases in isometric MVC of the elbow flexors (approximately 5.3 to 8.8%/wk) [[12](#_ENREF_12), [56](#_ENREF_56), [57](#_ENREF_57)], and a decrease in the maximum load that could be lifted [[12](#_ENREF_12)]. A more dramatic decrease in isometric MVC torque has been reported in the flexors and extensors of the wrist (approximately 22.8 to 25.3%/wk) in response to immobilisation [[23](#_ENREF_23), [58](#_ENREF_58)].

With ULLS, isometric torque appears to be affected to a lesser degree than with immobilisation models. An explanation for the above observation may be that ULLS removes weight-bearing but allows for freely moveable joints (hence a degree of muscular activity) whereas immobilisation is a more rigid model that does not allow joint movement (hence a greater restriction of muscular activity). Studies have reported plantar flexor isometric MVC torque to decrease (approximately 5 to 7 %/wk) with ULLS [[28](#_ENREF_28), [31](#_ENREF_31)]. With ULLS, increased fluctuations in plantar flexion (approximately 3%/wk) and knee extension (approximately 5.5%/wk) isometric force have been demonstrated [[29](#_ENREF_29)].

***Isokinetic strength***

In addition to the established decline in isometric strength (torque and force), hypo-activity models (de-training, bed-rest, immobilisation and ULLS) also result in reductions in dynamic torque outputs. Hypo-activity models also result in changes to dynamic torque outputs. After 14 d de-training isokinetic eccentric and concentric knee extension force has been shown to decrease by approximately 6 and 1.2%/wk, respectively [[7](#_ENREF_7)]. With as little as 14 d bed-rest decrements in knee extensor 1 repetition maximum (approximately 4.5%/wk) are seen along with a fall in MVC (approximately 7.5%/wk) [[59](#_ENREF_59)]. After 6 wk bed-rest maximum voluntary concentric knee extensor torque was shown to decrease uniformly across angular velocities (approximately 4.1 to 5.0%/wk) [[53](#_ENREF_53)]. Muscle-specific adaptations are evident with bed-rest, as shown by Dudley *et al*[[60](#_ENREF_60)] who reported a decrease in concentric and eccentric isokinetic knee extensor peak torque (approximately 4.4%/wk), with no alterations in knee flexors in response to 30 d 6 degrees head-down bed-rest. Again muscle-specific adaptations were demonstrated by Le Blanc *et al*[[18](#_ENREF_18)] who reported a decrease in plantar flexor concentric isokinetic strength (approximately 2.6%/wk) and no change in the isokinetic strength of the dorsiflexors with 5 wk bed-rest. As with the knee extensors *vs* knee flexors difference in sensitivity to hypo-activity alluded to above, the plantar flexor muscles experience a greater level of recruitment during gait than the tibialis anterior. Thus, habitual muscle recruitment prior to hypo-activity would appear to be a large determinant of the relative magnitude of hypo-activity-induced changes.

Results from lower limb immobilisation models indicate that short-term immobilisation is associated not only with atrophy but with a diminished capacity of the muscle to perform both concentric and eccentric strength [[23](#_ENREF_23), [55](#_ENREF_55)]. Lower limb casting results in a dramatic decrease in isokinetic quadriceps strength (approximately 29.1%/wk) [[25](#_ENREF_25)]. There is evidence that the effect of leg cast immobilisation on isokinetic strength of the knee extensors and flexors is greater in the knee extensors, demonstrated by a fall in peak torque of approximately 13.3%/wkfor the knee extensors and approximately 3.3%/wkfor knee flexors [[3](#_ENREF_3)]. Cast immobilisation of the arm also results in decreased concentric (approximately 6.9 to 16.9%/wk) and eccentric (approximately 9.7 to 14.4%/wk) strength for flexion, extension, pronation and supination of the wrist [[23](#_ENREF_23)].

Less dramatic decreases in isokinetic strength are seen with ULLS compared to immobilisation. De Boer *et al*[[30](#_ENREF_30)] found a decrease in isokinetic knee extensor torque in response to 23 d ULLS (approximately 6.4%/wk). Similarly, after 4 wk ULLS mean average peak isokinetic torque is decreased (approximately 4.3%/wk) [[61](#_ENREF_61)]. With as little as 14 d ULLS, a decrease in peak isokinetic torque (approximately 5 to 8.6%/wk) and total work performed (approximately 7.5 to 10.0%/wk) by knee extensors and flexors was reported [[62](#_ENREF_62)].

***Strength vs size changes***

There is evidence to suggest that decreases seen in strength in response to hypo-activity models are greater than the changes seen in muscle size. With de-training the loss in leg muscle ACSA (approximately 0.7%/wk) was not as great as the decrease seen in knee extension MVC (approximately 2.1%/wk) [[8](#_ENREF_8)]. Similarly, in bed-rest Kawakami *et al*[[1](#_ENREF_1)] suggested that the decrease in knee extension mean muscle force (approximately 3.8%/wk) seen after 20 d head down bed-rest was related more to changes in neural activation to those in PCSA (approximately 2.7%/wk). Correspondingly, Berg *et al*[[53](#_ENREF_53)] suggested that the decline seen in strength (approximately 4.1 to 5.0%/wk) could not be entirely accounted for by decreased ACSA (approximately 2.3%/wk), and that the strength loss could also be due to factors resulting in decreased neural input to muscle and/or reduced specific tension of muscle, as evidenced by a decreased torque to EMG ratio. Discrepancies between decreases in muscle size and muscle strength have also been reported in upper and lower immobilisation studies. White *et al* [[4](#_ENREF_4)] reported an approximately 5%/wk decrease in muscle ACSA whilst triceps surae MVC decreased approximately 12%/wk. Additionally, the upper limb decreases in forearm ACSA (approximately 3.2%/wk) were much smaller than those reported in forearm flexor and extensor strength (approximately 22.8 to 25.3 %/wk) [[23](#_ENREF_23)]. Again, in ULLS models muscle torque (approximately 5 to 7%/wk) appears to decrease to a greater degree than muscle ACSA (approximately 2.3 to 2.7%/wk) [[28](#_ENREF_28), [31](#_ENREF_31)].

***Summary***

Bed-rest appears to have varying degrees of impact on the upper and lower body. After 14 d of 6 degrees head down bed-rest maximum voluntary force for plantar flexion was decreased (approximately 7.5%/wk) whilst no effect was observed on maximal voluntary force of hand grip [[52](#_ENREF_52)]. Similar results were demonstrated by LeBlanc *et al*[[2](#_ENREF_2)] who showed after 17 wk of continuous bed-rest that isokinetic muscle strength decreased significantly in the thigh and calf with no loss in the arms. These results further support the idea that the lower limbs are primarily affected by bed-rest, more so than the upper limb. However, Gogia *et al*[[63](#_ENREF_63)] did observe a decrease in elbow flexor torque (approximately 3.8%/wk) and a non-significant decrease in elbow extension torque (approximately 1.4%/wk) after 5 wk of bed-rest. Thus, suggesting that strength in the upper limb is affected by bed-rest but only in specific muscles during specific tasks.

Together, these findings show that in addition to the reduction in muscle mass, hypo-activity also results in a dramatic loss of strength [Figure 2 relative change in isometric and isokinetic strength in response to hypo-activity models. Figure 2A Values taken from references in the text for upper body changes in strength in response to de-training, bed-rest (isokinetic) [[2](#_ENREF_2), [52](#_ENREF_52), [63](#_ENREF_63)], immobilisation (isometric) [[12](#_ENREF_12), [23](#_ENREF_23), [56-58](#_ENREF_56)] (isokinetic) [[23](#_ENREF_23)] and ULLS. Figure 2B values taken from references in the text for lower body changes in strength in response to de-training (isometric) [[5](#_ENREF_5), [8](#_ENREF_8)] (isokinetic) [[7](#_ENREF_7)], bed-rest (isometric) [[1](#_ENREF_1), [52](#_ENREF_52), [53](#_ENREF_53)] (isokinetic) [[18](#_ENREF_18), [53](#_ENREF_53), [59](#_ENREF_59), [60](#_ENREF_60)], immobilisation (isometric) [[4](#_ENREF_4), [22](#_ENREF_22), [24](#_ENREF_24), [54](#_ENREF_54), [55](#_ENREF_55)] (isokinetic) [[3](#_ENREF_3), [25](#_ENREF_25)] and ULLS (isometric) [[28](#_ENREF_28), [29](#_ENREF_29), [31](#_ENREF_31)] (isokinetic) [[30](#_ENREF_30), [61](#_ENREF_61), [62](#_ENREF_62)]. Where there are missing bars, this shows gaps in the literature (*i.e.*, values are not available for that parameter during a specific hypo-activity models)]. Values are presented as means; error bars denote SD. Models in which the joint is immobilised appear to have a greater impact on strength than unloaded models. These changes in muscular strength vary between hypo-activity models. The degree of loss in muscular strength surpasses the loss of muscle mass. Therefore, other alterations in the neuromuscular system, other than the reduction in contractile proteins must contribute to the excessive loss of strength. Voluntary force production is associated with neurological and skeletal muscle properties, thus suggesting these two factors as mechanisms accounting for the loss of strength with hypo-activity models.

**MUSCLE FATIGABILITY**

Studies have also examined the impact of hypo-activity models on the fatigability of skeletal muscle. Kamiya *et al*[[64](#_ENREF_64)] showed no change in time to fatigue after 14 d bed-rest. After a longer period of bed-rest (8 wk), Mulder *et al*[[65](#_ENREF_65)] demonstrated an increase in fatigability (7.2%-10.2%/min decrease in maximum voluntary isometric torque per minute exercise; or approximately 0.9-1.3%/wk fatigability increment). The contrast between the two studies would tend to suggest a delay in the impact of hypo-activity on muscle fatigability.

The effect of immobilising a limb has various different effects on skeletal muscle fatigability. Two weeks of full leg cast immobilisation resulted in no effect on muscle fatigability [[4](#_ENREF_4)]. In contrast, Veldhuizen *et al*[[3](#_ENREF_3)] found a decrease in isokinetic quadriceps endurance work from 9.1 kJ to 5.6 kJ after 4 wk leg cast immobilisation. These results suggest that short periods of lower limb immobilisation (≤ 2 wk) have little effect on muscle fatigability whilst longer periods of immobilisation (≥ 4 wk) increases muscle fatigability. Studies investigating the effects of immobilisation on skeletal muscle fatigability in the upper limbs have found different effects to those in the lower limbs. Similar to lower limbs shorter periods of immobilisation in the upper limbs appear to have minimal effects on muscle fatigability [[23](#_ENREF_23)]. Unlike the lower limb, longer periods of immobilisation of the upper limb show a trend towards increased resistance to fatigability. Following 3 wk of hand-forearm immobilisation time to task failure increased by 21% (approximately 7%/wk) [[66](#_ENREF_66)]. Semmler *et al*[[56](#_ENREF_56)] investigated the effects of fiberglass cast immobilisation of the elbow joint, and reported 7 out of the 12 immobilised participants exhibited an unusual pattern of muscle activity during a fatiguing contraction after immobilisation. In those individuals with this unusual pattern of muscle activity there was an associated increase in the ability to maintain a contraction over an extended period of time in the elbow flexor muscles [[56](#_ENREF_56)]. The physiological basis for the sometimes observed immobilisation-induced decreased fatigability, is not clear but it is likely to be related to neural factors[[56](#_ENREF_56)]. In contrast to this, Miles *et al*[[67](#_ENREF_67)] found an increase in fatigability in response to 3 wk arm suspension in untrained but not trained individuals. Previous research showed that ULLS led to increased fatigability after 4 wk of unloading [[61](#_ENREF_61)]. Results from Deschenes *et al*[[62](#_ENREF_62)] found a contrasting decrease in fatigability after just 2 wk of unloading.

Collectively these results suggest that muscle fatigability varies between different hypo-activity models [Figure 3 relative change in muscle fatigability in response to hypo-activity models (mean ± SD). Positive percentage change depicts an increase in fatigability whilst negative percentage change shows a decrease in fatigability. Values are separated into the effect of each hypo-activity model on the upper limb (UL) versus the lower limb (LL). The values are taken from the references used in the text for bed-rest (LL) [[64](#_ENREF_64), [65](#_ENREF_65)], immobilisation (UL) [[23](#_ENREF_23), [66](#_ENREF_66)] (LL) [[3](#_ENREF_3), [4](#_ENREF_4)] and ULLS (LL) [[61](#_ENREF_61), [62](#_ENREF_62)]. Where there are missing bars, this shows gaps in the literature (*i.e.*, values are not available for a parameter during a specific hypo-activity model)]. Shorter periods of hypo-activity (≤ 2 wk) generally appear to have little impact on fatigability. Muscle fatigability appears to increase in weight-bearing muscles but immobilisation in the upper body suggests an increase in resistance to fatigue. Differences between studies could be due to the duration of unloading or in the method used to test fatigue resistance. The mechanisms that cause fatigue are specific to the task being performed [[68](#_ENREF_68), [69](#_ENREF_69)]. Therefore, variability between fatigue resistance responses to hypo-activity models may be due to task specificity. Studies investigating a comparison of different fatigue tasks before and after hypo-activity are sparse. Yue *et al*[[12](#_ENREF_12)] demonstrated a task-dependent effect on muscle fatigue with substantially increased endurance time (reduced fatigability) at a low force (20% MVC) and no statistical effect at a moderate force (65% MVC) in the elbow flexors. The selective improvement of fatigue resistance for the low-force contraction was accompanied by the absence of a change in the time course of the twitch, suggesting that the immobilisation-induced adaptation included and improved efficacy of some excitation-contraction processes and underscored the major role of these mechanisms in determining the endurance time for low-force, long-duration contractions. It appears that the hypo-activity induced adaptations in muscle fatigability vary with the specifics of the task being performed. More research is needed to investigate these task-specific responses to different models of hypo-activity.

Numerous adaptations in fatigue mechanisms have been hypothesised to explain the observed preservation and decrease in fatigability in response to hypo-activity. As stated previously, hypo-activity results in muscle atrophy and a decrease in muscle strength, have been reported to be accompanied by myofiber transitions from slow to fast [[70](#_ENREF_70)] and a shift in fuel metabolism away from lipid fuels toward glycolysis [[71](#_ENREF_71)]. Typically these changes are associated with increased fatigability. Cardiovascular adaptations with hypo-activity [[72](#_ENREF_72)] reduces oxygen delivery and oxygen utilization which may impair prolonged exercise capacity. Additionally, exercise tolerance may be influenced by impaired muscle activation after hypo-activity [[1](#_ENREF_1), [54](#_ENREF_54)]. In light of this, the reports of decreased fatigability with hypo-activity are puzzling, and the underlying mechanisms remain unclear. It is possible that an atrophy-induced decrease in absolute force production will result in decreased intramuscular pressure. This in turn, will increase blood flow to the muscle and increase supply to match the metabolic demand [[56](#_ENREF_56), [73](#_ENREF_73)]. Other potential mechanisms include adaptations in the neural activation strategy utilised [[56](#_ENREF_56)], adaptations in the basal inorganic phosphate concentration [[74](#_ENREF_74)], and changes in excitation-contraction coupling [[12](#_ENREF_12)].

**NUTRITIONAL SUPPLEMENTATION**

As mentioned above, there is strong evidence that protein synthesis is decreased in response to periods of bed-rest and immobilisation [[40](#_ENREF_40), [41](#_ENREF_41), [43](#_ENREF_43)]. That resistance exercise provides an anabolic stimulus during hypo-activity is undisputed [[9](#_ENREF_9), [59](#_ENREF_59), [75](#_ENREF_75)]. When supplemented with nutritional interventions, the benefits of exercise during bed-rest appear additive [[76](#_ENREF_76)], thereby suggesting different synergistic pathways for counteracting atrophy. It may not always be practical to prescribe exercise to counteract the atrophy brought about by inactivity. In these cases, such as trauma, pharmaceuticals may be used and have been tried with varying degrees of success [[77](#_ENREF_77)]. However, effective long-term medication is not a palatable option (*e.g.*, costs, side effects, repeated injections). Where exercise is not a practical prescription, supplementing the diet with potential/recognised hypertrophic nutrients may be an effective and easily adhered to intervention programme for preventing the loss of muscle mass/function seen with hypo-activity. In this latter therapeutic group, potential candidates include proteins (essential amino acids (EAAs) and Leucine in particular), creatine, omega-3 fatty acids, vitamin-D (Vit-D) and antioxidants, to name but a few [[78](#_ENREF_78), [79](#_ENREF_79)].

***Protein***

Stuart *et al*[[80](#_ENREF_80)]. sought to determine whether the catabolic effects of bed-rest in humans was due to a decrease in protein synthesis, and if so, to assess whether increasing the amount of dietary protein might be beneficial The calculated non-oxidative Leucine disappearance was used as a measure of whole-body-protein synthesis, which was shown to decrease when dietary protein was low. Bed-rest resulted in a 24% decrease in nonoxidative Leucine disappearance in participants assigned to a lower-protein diet (0.6 g protein·kg body wt-1·d-1), whereas Leucine kinetics were unchanged by the same bed-rest protocol in participants who received a higher-protein diet (1.0 g protein·kg body wt-1·d-1) [[80](#_ENREF_80)]. In other words, whereas protein synthesis is suggested here to decrease with bed-rest, dietary supplementation of protein appears to protect against this deleterious response.

***Essential amino acids***

Bolus oral ingestion of EAAs produces a several-fold increase in plasma amino acid levels [[81](#_ENREF_81)] and has been shown to stimulate net protein synthesis to a greater extent than a mixed meal or a solution containing nonessential amino acids [[82](#_ENREF_82)]. Studies have shown that providing a nutritional supplement enriched with EAAs could improve lean body mass, strength and physical function even without exercise [[83](#_ENREF_83)]. Previous studies by Stein et al have shown improved nitrogen balance during both 6 and 14 d of bed-rest when provided with a daily supplementation of 11 g of branch-chain amino acids (BCAA), compared with the same dose of nonessential amino acids [[84](#_ENREF_84), [85](#_ENREF_85)]. It appears that a greater dose of EAAs (49.5 g/d) during 28 d bed-rest prevented any noticeable changes in muscle mass [[86](#_ENREF_86)]. Paddon-Jones *et al*[[86](#_ENREF_86)] however, reported that during this 28 d period that although no changes in muscle mass were observed they did find a decline in muscle strength. Nonetheless, the decrease in muscle strength with EAAs (11%) was still noticeably less than the decrease in strength seen in the control group (23%) [[86](#_ENREF_86)]. These results collectively demonstrate a positive effect of EAAs supplementation during periods of bed-rest ranging from 6 to 28 d on both muscle mass and function [[84-86](#_ENREF_84)].

***Creatine***

Creatine supplementation is another potential supplement that may attenuate hypo-activity induced decreases in muscle size and strength. Johnston et al reported that short-term (29 d) creatine supplementation (20 g/d) attenuates the loss in muscle mass and strength during upper arm immobilisation [[87](#_ENREF_87)]. It is well known that muscle total creatine content can be rapidly raised by a high-dose oral creatine intake [[88](#_ENREF_88)] and that long-term creatine intake can enhance the effects of weight training on muscle size and strength [[89](#_ENREF_89), [90](#_ENREF_90)]. Creatine supplementation during 10 wk of resistance training has been shown to accelerate the rate of muscle hypertrophy in young adults who previously had their knee flexors immobilised for 2 wk [[91](#_ENREF_91)]. Furthermore, 14 d creatine supplementation during hind-limb immobilisation lessened the rate of loss in the plantarflexors in a rodent model [[92](#_ENREF_92)]. Additionally, Op’t Eijinde *et al*[[93](#_ENREF_93)] showed that creatine supplementation prevented the loss of glucose transporter type 4 (GLUT4) during muscle disuse and increased muscle GLUT4 content above normal levels during subsequent rehabilitation. Collectively these studies suggest that creatine supplementation during resistance training and rest may be effective at reversing or maintaining lower-body muscle mass during and after an immobilised state.

***Antioxidants***

Intricate antioxidant defence systems in the body work to continually manage oxidative stress. To counteract ROS, enzymatic and nonenzymatic antioxidants work together [[94](#_ENREF_94)]. Enzymes work to improve or maintain an antioxidant balance and to avert oxidative damage by scavenging or preventing transformation of ROS to intracellular molecules and inhibiting their conversion to more deleterious forms. Endogenous nonenzymatic antioxidants such as vitamins-C and –E, carotenoids and flavonoids play important roles by contributing to the antioxidant system as cofactors for antioxidant enzymes. Results from Zwart *et al*[[95](#_ENREF_95)] provide evidence that increased oxidative stress occurs during bed-rest. These data are also supported by results of several other studies that show evidence for elevated oxidative stress and increased ROS [[96-98](#_ENREF_96)]. It would be interesting to see whether antioxidant supplementation during hypo-activity models will have beneficial effects on these outcome measures and furthermore, see whether this would then result in the attenuation of muscle loss in these models.

***Vitamin-D***

Ceglia proposed Vit-D supplementation as an effective nutritional intervention to attenuate age related sarcopenia [[99](#_ENREF_99)]. Vit-D supplementation (800 IU per day) for periods of 8 to 12 wk has been reported to reduce postural sway and improve the risk of falling in elderly individuals [[100](#_ENREF_100), [101](#_ENREF_101)]. Longer periods (12 mo) of Vit-D supplementation (800 IU per day) in the elderly has been shown to increase strength, decrease body sway and increase physical performance [[102](#_ENREF_102)]. However, in a healthy elderly population with no Vit-D deficiency Vit-D supplementation does not appear to improve muscle strength or function [[103](#_ENREF_103), [104](#_ENREF_104)]. It remains to be seen whether Vit-D supplementation in healthy persons with no Vit-D deficiency, any enhancement in muscle structural or contractile properties can be attained in the presence of hypo-activity.

***Omega-3 (EPA)***

Recent studies by Smith et al supplemented healthy young and elderly individuals with omega-3 fatty fish-oils for 8 wk and found a significant increase in the muscle protein synthetic response to amino acid administration [[105](#_ENREF_105), [106](#_ENREF_106)]. They concluded in the elderly model that omega-3 fatty acids might be useful for the prevention and treatment of sarcopenia [[105](#_ENREF_105)]. Dietary fish oil has also been shown to alleviate soleus muscle atrophy during immobilisation in association with Akt signalling in rats [[107](#_ENREF_107)]. It would therefore seem reasonable to suggest that more investigation is needed in to the potential of omega-3 fatty acids as a nutritional supplement for attenuating muscle atrophy with hypo-activity. In parallel, it is believed that omega-3 fatty acids may impact on lean body mass though decreasing the effectiveness of catabolic cytokines, reduced protein degradation and improving insulin sensitivity [[108](#_ENREF_108)]. There is evidence to suggest that eicosapentaenoic acid (EPA) an omega-3 fatty acid may reduce the pro-inflammatory cytokines associated with inflammation [[109](#_ENREF_109)]. Magee et al demonstrated *in vitro* that EPA inhibits the effects of TNF-α by reducing its apoptotic effects and enabling myogenesis [[109](#_ENREF_109)]. It is however debatable whether this supplement would be useful in combating muscle atrophy where, as seen in human hypo-activity models, there is scant evidence for increased protein breakdown [[40](#_ENREF_40)].

**CONCLUSION**

Hypo-activity models result in profound changes in skeletal muscle morphology and strength. Muscle mass and strength losses vary between different hypo-activity models, with immobilisation causing the most profound decreases, greater than bed-rest and limb suspension. Decrements in muscle size and strength are seen in response to hypo-activity models with the greatest decrements seen in antigravity muscles. The decreases in strength seen with hypo-activity models surpass the losses in muscle mass and as such, the nervous system and contractile properties adapt to adjust for this excessive loss of strength. Nutritional supplementation may stand as a viable intervention to combat muscle atrophy with hypo-activity when exercise is not a practical prescription. There are several potential nutritional supplements that could be used to combat muscle atrophy but extensive research is needed to determine the most affective.

**REFERENCES**

1 **Kawakami Y**, Akima H, Kubo K, Muraoka Y, Hasegawa H, Kouzaki M, Imai M, Suzuki Y, Gunji A, Kanehisa H, Fukunaga T. Changes in muscle size, architecture, and neural activation after 20 days of bed rest with and without resistance exercise. *Eur J Appl Physiol* 2001; **84**: 7-12 [PMID: 11394257 DOI: 10.1007/s004210000330]

2 **LeBlanc AD**, Schneider VS, Evans HJ, Pientok C, Rowe R, Spector E. Regional changes in muscle mass following 17 weeks of bed rest. *J Appl Physiol (1985)* 1992; **73**: 2172-2178 [PMID: 1474100]

3 **Veldhuizen JW**, Verstappen FT, Vroemen JP, Kuipers H, Greep JM. Functional and morphological adaptations following four weeks of knee immobilization. *Int J Sports Med* 1993; **14**: 283-287 [PMID: 8365837 DOI: 10.1055/s-2007-1021178]

4 **White MJ**, Davies CT, Brooksby P. The effects of short-term voluntary immobilization on the contractile properties of the human triceps surae. *Q J Exp Physiol* 1984; **69**: 685-691 [PMID: 6514993]

5 **Andersen JL**, Aagaard P. Myosin heavy chain IIX overshoot in human skeletal muscle. *Muscle Nerve* 2000; **23**: 1095-1104 [PMID: 10883005 DOI: 10.1002/1097-4598(200007)]

6 **Häkkinen K**, Alen M, Kallinen M, Newton RU, Kraemer WJ. Neuromuscular adaptation during prolonged strength training, detraining and re-strength-training in middle-aged and elderly people. *Eur J Appl Physiol* 2000; **83**: 51-62 [PMID: 11072774 DOI: 10.1007/s004210000248]

7 **Hortobágyi T**, Houmard JA, Stevenson JR, Fraser DD, Johns RA, Israel RG. The effects of detraining on power athletes. *Med Sci Sports Exerc* 1993; **25**: 929-935 [PMID: 8371654]

8 **Narici MV**, Roi GS, Landoni L, Minetti AE, Cerretelli P. Changes in force, cross-sectional area and neural activation during strength training and detraining of the human quadriceps. *Eur J Appl Physiol Occup Physiol* 1989; **59**: 310-319 [PMID: 2583179 DOI: 10.1007/BF02388334]

9 **Akima H**, Kubo K, Kanehisa H, Suzuki Y, Gunji A, Fukunaga T. Leg-press resistance training during 20 days of 6 degrees head-down-tilt bed rest prevents muscle deconditioning. *Eur J Appl Physiol* 2000; **82**: 30-38 [PMID: 10879440 DOI: 10.1007/s004210050648]

10 **Grosset JF**, Onambele-Pearson G. Effect of foot and ankle immobilization on leg and thigh muscles' volume and morphology: a case study using magnetic resonance imaging. *Anat Rec (Hoboken)* 2008; **291**: 1673-1683 [PMID: 18951503 DOI: 10.1002/ar.20759]

11 **Convertino VA**, Doerr DF, Mathes KL, Stein SL, Buchanan P. Changes in volume, muscle compartment, and compliance of the lower extremities in man following 30 days of exposure to simulated microgravity. *Aviat Space Environ Med* 1989; **60**: 653-658 [PMID: 2764848]

12 **Yue GH**, Bilodeau M, Hardy PA, Enoka RM. Task-dependent effect of limb immobilization on the fatigability of the elbow flexor muscles in humans. *Exp Physiol* 1997; **82**: 567-592 [PMID: 9179575]

13 **Colliander EB**, Tesch PA. Effects of detraining following short term resistance training on eccentric and concentric muscle strength. *Acta Physiol Scand* 1992; **144**: 23-29 [PMID: 1595350 DOI: DOI: ]

14 **Häkkinen K**, Komi PV. Electromyographic changes during strength training and detraining. *Med Sci Sports Exerc* 1983; **15**: 455-460 [PMID: 6656553 DOI: 10.1249/00005768-198315060-00003]

15 **Houston ME**, Froese EA, Valeriote SP, Green HJ, Ranney DA. Muscle performance, morphology and metabolic capacity during strength training and detraining: a one leg model. *Eur J Appl Physiol Occup Physiol* 1983; **51**: 25-35 [PMID: 6684028 DOI: 10.1007/BF00952534]

16 **Bamman MM**, Newcomer BR, Larson-Meyer DE, Weinsier RL, Hunter GR. Evaluation of the strength-size relationship in vivo using various muscle size indices. *Med Sci Sports Exerc* 2000; **32**: 1307-1313 [PMID: 10912898 DOI: 10.1097/00005768-200007000-00019]

17 **Narici MV**, Landoni L, Minetti AE. Assessment of human knee extensor muscles stress from in vivo physiological cross-sectional area and strength measurements. *Eur J Appl Physiol Occup Physiol* 1992; **65**: 438-444 [PMID: 1425650 DOI: 10.1007/BF00243511]

18 **LeBlanc A**, Gogia P, Schneider V, Krebs J, Schonfeld E, Evans H. Calf muscle area and strength changes after five weeks of horizontal bed rest. *Am J Sports Med* 1988; **16**: 624-629 [PMID: 3239619 DOI: 10.1177/036354658801600612]

19 **Rittweger J**, Frost HM, Schiessl H, Ohshima H, Alkner B, Tesch P, Felsenberg D. Muscle atrophy and bone loss after 90 days' bed rest and the effects of flywheel resistive exercise and pamidronate: results from the LTBR study. *Bone* 2005; **36**: 1019-1029 [PMID: 15811637 DOI: 10.1016/j.bone.2004.11.014]

20 **Kawakami Y**, Muraoka Y, Kubo K, Suzuki Y, Fukunaga T. Changes in muscle size and architecture following 20 days of bed rest. *J Gravit Physiol* 2000; **7**: 53-59 [PMID: 12124185]

21 **Ferrando AA**, Stuart CA, Brunder DG, Hillman GR. Magnetic resonance imaging quantitation of changes in muscle volume during 7 days of strict bed rest. *Aviat Space Environ Med* 1995; **66**: 976-981 [PMID: 8526835]

22 **Christensen B**, Dyrberg E, Aagaard P, Kjaer M, Langberg H. Short-term immobilization and recovery affect skeletal muscle but not collagen tissue turnover in humans. *J Appl Physiol (1985)* 2008; **105**: 1845-1851 [PMID: 18927270 DOI: 10.1152/japplphysiol.90445.2008]

23 **Miles MP**, Clarkson PM, Bean M, Ambach K, Mulroy J, Vincent K. Muscle function at the wrist following 9 d of immobilization and suspension. *Med Sci Sports Exerc* 1994; **26**: 615-623 [PMID: 8007811 DOI: 10.1249/00005768-199405000-00015]

24 **Oates BR**, Glover EI, West DW, Fry JL, Tarnopolsky MA, Phillips SM. Low-volume resistance exercise attenuates the decline in strength and muscle mass associated with immobilization. *Muscle Nerve* 2010; **42**: 539-546 [PMID: 20658567 DOI: 10.1002/mus.21721]

25 **Thom JM**, Thompson MW, Ruell PA, Bryant GJ, Fonda JS, Harmer AR, Janse de Jonge XA, Hunter SK. Effect of 10-day cast immobilization on sarcoplasmic reticulum calcium regulation in humans. *Acta Physiol Scand* 2001; **172**: 141-147 [PMID: 11442454 DOI: 10.1046/j.1365-201X.2001.00853.x]

26 **Yasuda N**, Glover EI, Phillips SM, Isfort RJ, Tarnopolsky MA. Sex-based differences in skeletal muscle function and morphology with short-term limb immobilization. *J Appl Physiol (1985)* 2005; **99**: 1085-1092 [PMID: 15860685 DOI: 10.1152/japplphysiol.00247.2005]

27 **Urso ML**, Clarkson PM, Price TB. Immobilization effects in young and older adults. *Eur J Appl Physiol* 2006; **96**: 564-571 [PMID: 16369818 DOI: 10.1007/s00421-005-0109-1]

28 **Clark BC**, Fernhall B, Ploutz-Snyder LL. Adaptations in human neuromuscular function following prolonged unweighting: I. Skeletal muscle contractile properties and applied ischemia efficacy. *J Appl Physiol (1985)* 2006; **101**: 256-263 [PMID: 16514004 DOI: 10.1152/japplphysiol.01402.2005]

29 **Clark BC**, Pierce JR, Manini TM, Ploutz-Snyder LL. Effect of prolonged unweighting of human skeletal muscle on neuromotor force control. *Eur J Appl Physiol* 2007; **100**: 53-62 [PMID: 17287986 DOI: 10.1007/s00421-007-0399-6]

30 **de Boer MD**, Maganaris CN, Seynnes OR, Rennie MJ, Narici MV. Time course of muscular, neural and tendinous adaptations to 23 day unilateral lower-limb suspension in young men. *J Physiol* 2007; **583**: 1079-1091 [PMID: 17656438 DOI: 10.1113/jphysiol.2007.135392]

31 **Seynnes OR**, Maffiuletti NA, Maganaris CN, de Boer MD, Pensini M, di Prampero PE, Narici MV. Soleus T reflex modulation in response to spinal and tendinous adaptations to unilateral lower limb suspension in humans. *Acta Physiol (Oxf)* 2008; **194**: 239-251 [PMID: 18485122 DOI: 10.1111/j.1748-1716.2008.01874.x]

32 **Tesch PA**, Berg HE, Häggmark T, Ohlsén H, Dudley GA. Muscle strength and endurance following lowerlimb suspension in man. *Physiologist* 1991; **34**: S104-S106 [PMID: 2047402]

33 **Powell PL**, Roy RR, Kanim P, Bello MA, Edgerton VR. Predictability of skeletal muscle tension from architectural determinations in guinea pig hindlimbs. *J Appl Physiol Respir Environ Exerc Physiol* 1984; **57**: 1715-1721 [PMID: 6511546]

34 **Bloomfield SA**. Changes in musculoskeletal structure and function with prolonged bed rest. *Med Sci Sports Exerc* 1997; **29**: 197-206 [PMID: 9044223]

35 **Williams PE**, Goldspink G. The effect of immobilization on the longitudinal growth of striated muscle fibres. *J Anat* 1973; **116**: 45-55 [PMID: 4798240]

36 **Fukunaga T**, Miyatani M, Tachi M, Kouzaki M, Kawakami Y, Kanehisa H. Muscle volume is a major determinant of joint torque in humans. *Acta Physiol Scand* 2001; **172**: 249-255 [PMID: 11531646 DOI: 10.1046/j.1365-201x.2001.00867.x]

37 **Manini TM**, Clark BC, Nalls MA, Goodpaster BH, Ploutz-Snyder LL, Harris TB. Reduced physical activity increases intermuscular adipose tissue in healthy young adults. *Am J Clin Nutr* 2007; **85**: 377-384 [PMID: 17284732]

38 **Kortebein P**, Ferrando A, Lombeida J, Wolfe R, Evans WJ. Effect of 10 days of bed rest on skeletal muscle in healthy older adults. *JAMA* 2007; **297**: 1772-1774 [PMID: 17456818 DOI: 10.1001/jama.297.16.1772-b]

39 **de Boer MD**, Seynnes OR, di Prampero PE, Pisot R, Mekjavić IB, Biolo G, Narici MV. Effect of 5 weeks horizontal bed rest on human muscle thickness and architecture of weight bearing and non-weight bearing muscles. *Eur J Appl Physiol* 2008; **104**: 401-407 [PMID: 18320207 DOI: 10.1007/s00421-008-0703-0]

40 **Ferrando AA**, Lane HW, Stuart CA, Davis-Street J, Wolfe RR. Prolonged bed rest decreases skeletal muscle and whole body protein synthesis. *Am J Physiol* 1996; **270**: E627-E633 [PMID: 8928769]

41 **Gibson JN**, Halliday D, Morrison WL, Stoward PJ, Hornsby GA, Watt PW, Murdoch G, Rennie MJ. Decrease in human quadriceps muscle protein turnover consequent upon leg immobilization. *Clin Sci (Lond)* 1987; **72**: 503-509 [PMID: 2435445]

42 **Urso ML**, Scrimgeour AG, Chen YW, Thompson PD, Clarkson PM. Analysis of human skeletal muscle after 48 h immobilization reveals alterations in mRNA and protein for extracellular matrix components. *J Appl Physiol (1985)* 2006; **101**: 1136-1148 [PMID: 16763108 DOI: 10.1152/japplphysiol.00180.2006]

43 **de Boer MD**, Selby A, Atherton P, Smith K, Seynnes OR, Maganaris CN, Maffulli N, Movin T, Narici MV, Rennie MJ. The temporal responses of protein synthesis, gene expression and cell signalling in human quadriceps muscle and patellar tendon to disuse. *J Physiol* 2007; **585**: 241-251 [PMID: 17901116 DOI: 10.1113/jphysiol.2007.142828]

44 **Rennie MJ**. Anabolic resistance: the effects of aging, sexual dimorphism, and immobilization on human muscle protein turnover. *Appl Physiol Nutr Metab* 2009; **34**: 377-381 [PMID: 19448702 DOI: 10.1139/H09-012]

45 **Rennie MJ**, Selby A, Atherton P, Smith K, Kumar V, Glover EL, Philips SM. Facts, noise and wishful thinking: muscle protein turnover in aging and human disuse atrophy. *Scand J Med Sci Sports* 2010; **20**: 5-9 [PMID: 19558380 DOI: 10.1111/j.1600-0838.2009.00967.x]

46 **Burgomaster KA**, Cermak NM, Phillips SM, Benton CR, Bonen A, Gibala MJ. Divergent response of metabolite transport proteins in human skeletal muscle after sprint interval training and detraining. *Am J Physiol Regul Integr Comp Physiol* 2007; **292**: R1970-R1976 [PMID: 17303684 DOI: 10.1152/ajpregu.00503.2006]

47 **Simsolo RB**, Ong JM, Kern PA. The regulation of adipose tissue and muscle lipoprotein lipase in runners by detraining. *J Clin Invest* 1993; **92**: 2124-2130 [PMID: 8227328 DOI: 10.1172/JCI116813]

48 **Ogawa T**, Furochi H, Mameoka M, Hirasaka K, Onishi Y, Suzue N, Oarada M, Akamatsu M, Akima H, Fukunaga T, Kishi K, Yasui N, Ishidoh K, Fukuoka H, Nikawa T. Ubiquitin ligase gene expression in healthy volunteers with 20-day bedrest. *Muscle Nerve* 2006; **34**: 463-469 [PMID: 16868939 DOI: 10.1002/mus.20611]

49 **Chen YW**, Gregory CM, Scarborough MT, Shi R, Walter GA, Vandenborne K. Transcriptional pathways associated with skeletal muscle disuse atrophy in humans. *Physiol Genomics* 2007; **31**: 510-520 [PMID: 17804603 DOI: 10.1152/physiolgenomics.00115.2006]

50 **Jones SW**, Hill RJ, Krasney PA, O'Conner B, Peirce N, Greenhaff PL. Disuse atrophy and exercise rehabilitation in humans profoundly affects the expression of genes associated with the regulation of skeletal muscle mass. *FASEB J* 2004; **18**: 1025-1027 [PMID: 15084522 DOI: 10.1096/fj.03-1228fje]

51 **Penkowa M**, Keller P, Keller C, Hidalgo J, Giralt M, Pedersen BK. Exercise-induced metallothionein expression in human skeletal muscle fibres. *Exp Physiol* 2005; **90**: 477-486 [PMID: 15640275 DOI: 10.1113/expphysiol.2004.029371]

52 **Kamiya A**, Michikami D, Shiozawa T, Iwase S, Hayano J, Kawada T, Sunagawa K, Mano T. Bed rest attenuates sympathetic and pressor responses to isometric exercise in antigravity leg muscles in humans. *Am J Physiol Regul Integr Comp Physiol* 2004; **286**: R844-R850 [PMID: 14701716 DOI: 10.1152/ajpregu.00497.2003]

53 **Berg HE**, Larsson L, Tesch PA. Lower limb skeletal muscle function after 6 wk of bed rest. *J Appl Physiol (1985)* 1997; **82**: 182-188 [PMID: 9029214]

54 **Gondin J**, Guette M, Maffiuletti NA, Martin A. Neural activation of the triceps surae is impaired following 2 weeks of immobilization. *Eur J Appl Physiol* 2004; **93**: 359-365 [PMID: 15490220 DOI: 10.1007/s00421-004-1225-z]

55 **Hortobágyi T**, Dempsey L, Fraser D, Zheng D, Hamilton G, Lambert J, Dohm L. Changes in muscle strength, muscle fibre size and myofibrillar gene expression after immobilization and retraining in humans. *J Physiol* 2000; **524 Pt 1**: 293-304 [PMID: 10747199 DOI: 10.1111/j.1469-7793.2000.00293.x]

56 **Semmler JG**, Kutzscher DV, Enoka RM. Limb immobilization alters muscle activation patterns during a fatiguing isometric contraction. *Muscle Nerve* 2000; **23**: 1381-1392 [PMID: 10951441 DOI: 10.1002/1097-4598(200009)23: 9<1381: : AID-MUS9>3.0.CO; 2-5]

57 **Vaughan VG**. Effects of upper limb immobilization on isometric muscle strength, movement time, and triphasic electromyographic characteristics. *Phys Ther* 1989; **69**: 119-129 [PMID: 2913580]

58 **Lundbye-Jensen J**, Nielsen JB. Central nervous adaptations following 1 wk of wrist and hand immobilization. *J Appl Physiol (1985)* 2008; **105**: 139-151 [PMID: 18450985 DOI: 10.1152/japplphysiol.00687.2007]

59 **Bamman MM**, Clarke MS, Feeback DL, Talmadge RJ, Stevens BR, Lieberman SA, Greenisen MC. Impact of resistance exercise during bed rest on skeletal muscle sarcopenia and myosin isoform distribution. *J Appl Physiol (1985)* 1998; **84**: 157-163 [PMID: 9451630]

60 **Dudley GA**, Duvoisin MR, Convertino VA, Buchanan P. Alterations of the in vivo torque-velocity relationship of human skeletal muscle following 30 days exposure to simulated microgravity. *Aviat Space Environ Med* 1989; **60**: 659-663 [PMID: 2764849]

61 **Berg HE**, Dudley GA, Hather B, Tesch PA. Work capacity and metabolic and morphologic characteristics of the human quadriceps muscle in response to unloading. *Clin Physiol* 1993; **13**: 337-347 [PMID: 8370234 DOI: 10.1111/j.1475-097X.1993.tb00334.x]

62 **Deschenes MR**, Giles JA, McCoy RW, Volek JS, Gomez AL, Kraemer WJ. Neural factors account for strength decrements observed after short-term muscle unloading. *Am J Physiol Regul Integr Comp Physiol* 2002; **282**: R578-R583 [PMID: 11792669 DOI: 10.1152/ajpregu.00386.2001]

63 **Gogia P**, Schneider VS, LeBlanc AD, Krebs J, Kasson C, Pientok C. Bed rest effect on extremity muscle torque in healthy men. *Arch Phys Med Rehabil* 1988; **69**: 1030-1032 [PMID: 3214261]

64 **Kamiya A**, Iwase S, Michikamia D, Fua Q, Mano T. Muscle sympathetic nerve activity during handgrip and post-handgrip muscle ischemia after exposure to simulated microgravity in humans. *Neurosci Lett* 2000; **280**: 49-52 [PMID: 10696809 DOI: 10.1016/S0304-3940(99)00995-7]

65 **Mulder ER**, Kuebler WM, Gerrits KH, Rittweger J, Felsenberg D, Stegeman DF, de Haan A. Knee extensor fatigability after bedrest for 8 weeks with and without countermeasure. *Muscle Nerve* 2007; **36**: 798-806 [PMID: 17661376 DOI: 10.1002/mus.20870]

66 **Clark BC**, Hoffman RL, Russ DW. Immobilization-induced increase in fatigue resistance is not explained by changes in the muscle metaboreflex. *Muscle Nerve* 2008; **38**: 1466-1473 [PMID: 18932206 DOI: 10.1002/mus.21127]

67 **Miles MP**, Heil DP, Larson KR, Conant SB, Schneider SM. Prior resistance training and sex influence muscle responses to arm suspension. *Med Sci Sports Exerc* 2005; **37**: 1983-1989 [PMID: 16286870 DOI: 10.1249/01.mss.0000176302.99185.be]

68 **Enoka RM**, Duchateau J. Muscle fatigue: what, why and how it influences muscle function. *J Physiol* 2008; **586**: 11-23 [PMID: 17702815 DOI: 10.1113/jphysiol.2007.139477]

69 **Hunter SK**, Duchateau J, Enoka RM. Muscle fatigue and the mechanisms of task failure. *Exerc Sport Sci Rev* 2004; **32**: 44-49 [PMID: 15064647 DOI: 10.1097/0003677-200404000-00002]

70 **Fitts RH**, Riley DR, Widrick JJ. Physiology of a microgravity environment invited review: microgravity and skeletal muscle. *J Appl Physiol (1985)* 2000; **89**: 823-839 [PMID: 10926670]

71 **Stein TP**, Wade CE. Metabolic consequences of muscle disuse atrophy. *J Nutr* 2005; **135**: 1824S-1828S [PMID: 15987873]

72 **Convertino VA**. Cardiovascular consequences of bed rest: effect on maximal oxygen uptake. *Med Sci Sports Exerc* 1997; **29**: 191-196 [PMID: 9044222]

73 **Bodor M**. Muscle endurance after limb immobilization. *Muscle Nerve* 2002; **25**: 923-94; author reply 923-94; [PMID: 12115986 DOI: 10.1002/mus.10135]

74 **Shaffer MA**, Okereke E, Esterhai JL, Elliott MA, Walker GA, Yim SH, Vandenborne K. Effects of immobilization on plantar-flexion torque, fatigue resistance, and functional ability following an ankle fracture. *Phys Ther* 2000; **80**: 769-780 [PMID: 10911415]

75 **Ferrando AA**, Tipton KD, Bamman MM, Wolfe RR. Resistance exercise maintains skeletal muscle protein synthesis during bed rest. *J Appl Physiol (1985)* 1997; **82**: 807-810 [PMID: 9074967]

76 **Brooks N**, Cloutier GJ, Cadena SM, Layne JE, Nelsen CA, Freed AM, Roubenoff R, Castaneda-Sceppa C. Resistance training and timed essential amino acids protect against the loss of muscle mass and strength during 28 days of bed rest and energy deficit. *J Appl Physiol (1985)* 2008; **105**: 241-248 [PMID: 18483167 DOI: 10.1152/japplphysiol.01346.2007]

77 **Jones TE**, Stephenson KW, King JG, Knight KR, Marshall TL, Scott WB. Sarcopenia--mechanisms and treatments. *J Geriatr Phys Ther* 2009; **32**: 83-89 [PMID: 20039588 DOI: 10.1519/00139143-200932020-00008]

78 **Kim JS**, Wilson JM, Lee SR. Dietary implications on mechanisms of sarcopenia: roles of protein, amino acids and antioxidants. *J Nutr Biochem* 2010; **21**: 1-13 [PMID: 19800212 DOI: 10.1016/j.jnutbio.2009.06.014]

79 **Sakuma K**, Yamaguchi A. Novel intriguing strategies attenuating to sarcopenia. *J Aging Res* 2012; **2012**: 251217 [PMID: 22500226 DOI: 10.1155/2012/251217]

80 **Stuart CA**, Shangraw RE, Peters EJ, Wolfe RR. Effect of dietary protein on bed-rest-related changes in whole-body-protein synthesis. *Am J Clin Nutr* 1990; **52**: 509-514 [PMID: 2203254]

81 **Paddon-Jones D**, Sheffield-Moore M, Zhang XJ, Volpi E, Wolf SE, Aarsland A, Ferrando AA, Wolfe RR. Amino acid ingestion improves muscle protein synthesis in the young and elderly. *Am J Physiol Endocrinol Metab* 2004; **286**: E321-E328 [PMID: 14583440 DOI: 10.1152/ajpendo.00368.2003]

82 **Tipton KD**, Gurkin BE, Matin S, Wolfe RR. Nonessential amino acids are not necessary to stimulate net muscle protein synthesis in healthy volunteers. *J Nutr Biochem* 1999; **10**: 89-95 [PMID: 15539275 DOI: 10.1016/S0955-2863(98)00087-4]

83 **Katsanos CS**, Kobayashi H, Sheffield-Moore M, Aarsland A, Wolfe RR. A high proportion of leucine is required for optimal stimulation of the rate of muscle protein synthesis by essential amino acids in the elderly. *Am J Physiol Endocrinol Metab* 2006; **291**: E381-E387 [PMID: 16507602 DOI: 10.1152/ajpendo.00488.2005]

84 **Stein TP**, Donaldson MR, Leskiw MJ, Schluter MD, Baggett DW, Boden G. Branched-chain amino acid supplementation during bed rest: effect on recovery. *J Appl Physiol (1985)* 2003; **94**: 1345-1352 [PMID: 12471043 DOI: 10.1152/japplphysiol.00481.2002]

85 **Stein TP**, Schluter MD, Leskiw MJ, Boden G. Attenuation of the protein wasting associated with bed rest by branched-chain amino acids. *Nutrition* 1999; **15**: 656-660 [PMID: 10467608 DOI: 10.1016/S0899-9007(99)00120-3]

86 **Paddon-Jones D**, Sheffield-Moore M, Urban RJ, Sanford AP, Aarsland A, Wolfe RR, Ferrando AA. Essential amino acid and carbohydrate supplementation ameliorates muscle protein loss in humans during 28 days bedrest. *J Clin Endocrinol Metab* 2004; **89**: 4351-4358 [PMID: 15356032 DOI: 10.1210/jc.2003-032159]

87 **Johnston AP**, Burke DG, MacNeil LG, Candow DG. Effect of creatine supplementation during cast-induced immobilization on the preservation of muscle mass, strength, and endurance. *J Strength Cond Res* 2009; **23**: 116-120 [PMID: 19130643 DOI: 10.1519/JSC.0b013e31818efbcc]

88 **Harris RC**, Söderlund K, Hultman E. Elevation of creatine in resting and exercised muscle of normal subjects by creatine supplementation. *Clin Sci (Lond)* 1992; **83**: 367-374 [PMID: 1327657]

89 **Terjung RL**, Clarkson P, Eichner ER, Greenhaff PL, Hespel PJ, Israel RG, Kraemer WJ, Meyer RA, Spriet LL, Tarnopolsky MA, Wagenmakers AJ, Williams MH. American College of Sports Medicine roundtable. The physiological and health effects of oral creatine supplementation. *Med Sci Sports Exerc* 2000; **32**: 706-717 [PMID: 10731017 DOI: 10.1097/00005768-200003000-00024]

90 **Vandenberghe K**, Goris M, Van Hecke P, Van Leemputte M, Vangerven L, Hespel P. Long-term creatine intake is beneficial to muscle performance during resistance training. *J Appl Physiol (1985)* 1997; **83**: 2055-2063 [PMID: 9390981]

91 **Hespel P**, Op't Eijnde B, Van Leemputte M, Ursø B, Greenhaff PL, Labarque V, Dymarkowski S, Van Hecke P, Richter EA. Oral creatine supplementation facilitates the rehabilitation of disuse atrophy and alters the expression of muscle myogenic factors in humans. *J Physiol* 2001; **536**: 625-633 [PMID: 11600695 DOI: 10.1111/j.1469-7793.2001.0625c.xd]

92 **Aoki MS**, Lima WP, Miyabara EH, Gouveia CH, Moriscot AS. Deleteriuos effects of immobilization upon rat skeletal muscle: role of creatine supplementation. *Clin Nutr* 2004; **23**: 1176-1183 [PMID: 15380911 DOI: 10.1016/j.clnu.2004.03.004]

93 **Op 't Eijnde B**, Ursø B, Richter EA, Greenhaff PL, Hespel P. Effect of oral creatine supplementation on human muscle GLUT4 protein content after immobilization. *Diabetes* 2001; **50**: 18-23 [PMID: 11147785 DOI: 10.2337/diabetes.50.1.18]

94 **Matés JM**, Pérez-Gómez C, Núñez de Castro I. Antioxidant enzymes and human diseases. *Clin Biochem* 1999; **32**: 595-603 [PMID: 10638941 DOI: 10.1016/S0009-9120(99)00075-2]

95 **Zwart SR**, Oliver SA, Fesperman JV, Kala G, Krauhs J, Ericson K, Smith SM. Nutritional status assessment before, during, and after long-duration head-down bed rest. *Aviat Space Environ Med* 2009; **80**: A15-A22 [PMID: 19476165 DOI: 10.3357/ASEM.BR07.2009]

96 **Pawlak W**, Kedziora J, Zolynski K, Kedziora-Kornatowska K, Blaszczyk J, Witkowski P. Free radicals generation by granulocytes from men during bed rest. *J Gravit Physiol* 1998; **5**: P131-P132 [PMID: 11542322]

97 **Pawlak W**, Kedziora J, Zolynski K, Kedziora-Kornatowska K, Blaszczyk J, Witkowski P, Zieleniewski J. Effect of long term bed rest in men on enzymatic antioxidative defence and lipid peroxidation in erythrocytes. *J Gravit Physiol* 1998; **5**: P163-P164 [PMID: 11542339]

98 **Zezerov AE**, Ivanova SM, Morukov BV, Ushakov AS. [Lipid peroxidation in the human blood during a 120-day period of anti-orthostatic hypokinesia]. *Kosm Biol Aviakosm Med* 1989; **23**: 28-33 [PMID: 2716266]

99 **Ceglia L**. Vitamin D and its role in skeletal muscle. *Curr Opin Clin Nutr Metab Care* 2009; **12**: 628-633 [PMID: 19770647 DOI: 10.1097/MCO.0b013e328331c707]

100 **Bischoff HA**, Stähelin HB, Dick W, Akos R, Knecht M, Salis C, Nebiker M, Theiler R, Pfeifer M, Begerow B, Lew RA, Conzelmann M. Effects of vitamin D and calcium supplementation on falls: a randomized controlled trial. *J Bone Miner Res* 2003; **18**: 343-351 [PMID: 12568412 DOI: 10.1359/jbmr.2003.18.2.343]

101 **Pfeifer M**, Begerow B, Minne HW, Abrams C, Nachtigall D, Hansen C. Effects of a short-term vitamin D and calcium supplementation on body sway and secondary hyperparathyroidism in elderly women. *J Bone Miner Res* 2000; **15**: 1113-1118 [PMID: 10841179 DOI: 10.1359/jbmr.2000.15.6.1113]

102 **Pfeifer M**, Begerow B, Minne HW, Suppan K, Fahrleitner-Pammer A, Dobnig H. Effects of a long-term vitamin D and calcium supplementation on falls and parameters of muscle function in community-dwelling older individuals. *Osteoporos Int* 2009; **20**: 315-322 [PMID: 18629569 DOI: 10.1007/s00198-008-0662-7]

103 **Grady D**, Halloran B, Cummings S, Leveille S, Wells L, Black D, Byl N. 1,25-Dihydroxyvitamin D3 and muscle strength in the elderly: a randomized controlled trial. *J Clin Endocrinol Metab* 1991; **73**: 1111-1117 [PMID: 1939527 DOI: 10.1210/jcem-73-5-1111]

104 **Johnson KR**, Jobber J, Stonawski BJ. Prophylactic vitamin D in the elderly. *Age Ageing* 1980; **9**: 121-127 [PMID: 7395656 DOI: 10.1093/ageing/9.2.121]

105 **Smith GI**, Atherton P, Reeds DN, Mohammed BS, Rankin D, Rennie MJ, Mittendorfer B. Dietary omega-3 fatty acid supplementation increases the rate of muscle protein synthesis in older adults: a randomized controlled trial. *Am J Clin Nutr* 2011; **93**: 402-412 [PMID: 21159787 DOI: 10.3945/ajcn.110.005611]

106 **Smith GI**, Atherton P, Reeds DN, Mohammed BS, Rankin D, Rennie MJ, Mittendorfer B. Omega-3 polyunsaturated fatty acids augment the muscle protein anabolic response to hyperinsulinaemia-hyperaminoacidaemia in healthy young and middle-aged men and women. *Clin Sci (Lond)* 2011; **121**: 267-278 [PMID: 21501117 DOI: 10.1042/CS20100597]

107 **You JS**, Park MN, Song W, Lee YS. Dietary fish oil alleviates soleus atrophy during immobilization in association with Akt signaling to p70s6k and E3 ubiquitin ligases in rats. *Appl Physiol Nutr Metab* 2010; **35**: 310-318 [PMID: 20555375 DOI: 10.1139/H10-022]

108 **Siddiqui RA**, Shaikh SR, Sech LA, Yount HR, Stillwell W, Zaloga GP. Omega 3-fatty acids: health benefits and cellular mechanisms of action. *Mini Rev Med Chem* 2004; **4**: 859-871 [PMID: 15544547 DOI: 10.2174/1389557043403431]

109 **Magee P**, Pearson S, Allen J. The omega-3 fatty acid, eicosapentaenoic acid (EPA), prevents the damaging effects of tumour necrosis factor (TNF)-alpha during murine skeletal muscle cell differentiation. *Lipids Health Dis* 2008; **7**: 24 [PMID: 18638380 DOI: 10.1186/1476-511X-7-24]

**P-Reviewer:** Gorgey AS **S-Editor:** Song XX **L-Editor:** **E-Editor:**

**Figure 1 Relative change in muscle anatomical cross sectional area, physiological cross sectional area and volume.**

**Figure 2 Relative change in isometric and isokinetic strength.** A: Upper limb; B: Lower limb.

**Figure 3 Relative change in muscle fatigability.**

**Table 1 Relative change in upper and lower limb muscle anatomical cross sectional area, physiological cross sectional area and volume**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
|  | ACSA\_UL  (%) | ACSA\_LL  (%) | PCSA\_UL  (%) | PCSA\_LL  (%) | Volume\_UL  (%) | Volume\_LL  (%) |
| De-training | - | -0.6 | - | - | - | - |
| Bed-rest | -0.5 | -1.5 | - | -3.5 | - | -2.1 |
| Immobilisation | -3.0 | -4.4 | - | - | -3.3 | -4.4 |
| ULLS | - | -2.4 | - | - | - | - |
| Mean (SD) of  4 models | -1.8 (1.8) | -2.2 (1.6) |  | -3.5 (0.01) | -3.3 (0.01) | -3.3 (1.6) |

ACSA:Anatomical cross sectional area; PCSA:Physiological cross sectional area; ULLS: Unilateral lower limb suspension.