A Mechanistic Approach to Blood Flow Occlusion

Authors
J. P. Loenneke 1, G. J. Wilson 2, J. M. Wilson 3

Affiliations
1 Southeast Missouri State University, Health, Human Performance, and Recreation, Cape Girardeau, United States
2 University of Illinois, Division of Nutritional Sciences, Champaign-Urbana, United States
3 Florida State University, Department of Nutrition, Food, and Exercise Science, Tallahassee, United States

Abstract

Low-Intensity occlusion training provides a unique beneficial training mode for promoting muscle hypertrophy. Training at intensities as low as 20% 1RM with moderate vascular occlusion results in muscle hypertrophy in as little as three weeks. The primary mechanisms by which occlusion training is thought to stimulate growth include, metabolic accumulation, which stimulates a subsequent increase in anabolic growth factors, fast-twitch fiber recruitment (FT), and increased protein synthesis through the mammalian target of rapamycin (mTOR) pathway. Heat shock proteins, Nitric oxide synthase-1 (NOS-1) and Myostatin have also been shown to be affected by an occlusion stimulus. In conclusion, low-intensity occlusion training appears to work through a variety of mechanisms. The research behind these mechanisms is incomplete thus far, and requires further examination, primarily to identify the actual metabolite responsible for the increase in GH with occlusion, and determine which mechanisms are associated to a greater degree with the hypertrophic/anti-catabolic changes seen with blood flow restriction.

Introduction

The American College of Sports Medicine (ACSM) recommends lifting a weight of at least 65% of one’s one repetition maximum (1RM) to achieve muscular hypertrophy under normal conditions. It is believed that anything below this intensity rarely produces substantial muscle hypertrophy or strength gains [17]. However, some individuals are unable to withstand the high mechanical stress placed upon the joints during heavy resistance training. Therefore, scientists have sought lower intensity alternatives such as blood occlusion training, also known as KAATSU training. Blood occlusion training, as the name implies, involves decreasing blood flow to a muscle, by application of a wrapping device, such as a blood pressure cuff. Evidence indicates that this style of training can provide a unique, beneficial mode of exercise in clinical settings, as it produces positive training adaptations at the equivalent to physical activity of daily life (10–30% of maximal work capacity) [1]. Muscle hypertrophy has recently been shown to occur during exercise as low as 20% of 1RM with moderate vascular occlusion (~100mmHg) [32], which could be quite beneficial to athletes [34], patients in post operation rehabilitation, specifically ACL injuries, cardiac rehabilitation patients, the elderly [33,36] and even astronauts [12]. Although muscle hypertrophy would likely benefit those special populations, more research should be done to further our understanding of the proposed benefits to each. The primary mechanisms by which occlusion training stimulates growth include: metabolic accumulation which stimulates a subsequent increase in anabolic growth factors, fast-twitch fiber recruitment (FT), and increased protein synthesis through the mammalian target of rapamycin (mTOR) pathway. Increases in heat shock proteins (HSP), Nitric oxide synthase-1 (NOS-1), and decreased expression of Myostatin have also been observed [15]. The purpose of this manuscript is to describe the physiologic mechanisms by which vascular occlusion leads to skeletal muscle hypertrophy.

Metabolic Accumulation and Growth Hormone

Whole blood lactate [8,34], plasma lactate [7,28,33] and muscle cell lactate [14,15] are all increased in response to exercise with blood flow restriction.
restriction. This is significant, as growth hormone (GH) has shown to be stimulated by an acidic intramuscular environment [34]. Evidence indicates that a low pH stimulates sympathetic nerve activity through a chemoreceptormediated reflex mediated by intramuscular metaboreceptors and group III and IV afferent fibers [38]. Consequently, this same pathway has recently been shown to play an important role in the regulation of hypophy-seal secretion of GH [9, 38].

However, changes in blood lactate are not always predictive of changes in GH. To illustrate, Reeves et al. [28] showed that while occlusion training resulted in a greater GH response than a non-ocluded control, there were no significant differences in blood lactate concentrations between groups. One possibility for the disparity is that occluding blood flow resulted in a slower diffusion of lactate out of muscle tissue, resulting in a more pronounced intramuscular acidic environment and therefore, a greater local stimulation of group IV afferents prior to its diffusion out of the cell. It is also possible that additional intramuscular metabolites stimulated changes in GH as group III and IV afferents are sensitive to changes in adenosine, K+, H+, hypoxia, and AMP. Increases in these metabolites during exercise is thought to drive the pressor reflex leading to increased heart rate and blood pressure, and it is postulated that this may also facilitate increases in GH following occlusion training [27].

Although there is no evidence that GH enhances muscle protein synthesis when combined with traditional resistance exercise in humans [40], occlusion training may be different. Occlusion training elevates GH to levels over that seen with traditional resistance training [18, 19]. One study showed an increase in GH ~290 times greater than baseline measurements [34]. Research on the effects of supraphysiologic dosing of GH with traditional resistance training in humans is limited. And while this research has not yet demonstrated increased hypertrophy, it does appear to indicate that GH administration elevates both the liver iso-form of IGF-1 (Ea) in muscle as well as mechano-growth factor [6]. More recently Ehrenborg and Rosen [6] have in an extensive analysis of the literature on GH concluded that the majority of the improvement with GH is due to the stimulation of collagen synthesis which could provide a protective effect in transferring force from skeletal muscle externally and thus protect against ruptures.

It is unclear if IGF-1 activity is increased in response to occlusion training. More specifically, Takano et al. [33] found a significant increase, whereas two other studies found no increase [1, 15]. Possible reasons as to why there was no increase could be related to the low intensity of the exercise. Kawada and Ishii [15] postulate that IGF-1 may not be necessary for muscle hypertrophy when other factors such as Myostatin, heat shock protein 72 (HSP-72), and nitric oxide synthase-1 (NOS-1) are changed in favor of muscle growth.

**Fiber Type Recruitment**

The size principle suggests that under normal conditions slow twitch fibers (ST) are recruited first and as the intensity increases, fast twitch fibers (FT) are recruited as needed. The novel aspect of occlusion training is that FT are recruited even though the training intensity is low. Moritani et al. [25] postulated that since the availability of oxygen is severely reduced during occlusion, that a progressive recruitment of additional motor units (MU) may take place to compensate for the deficit in force develop-ment. Previous studies have shown significant increases in MU firing rate and MU spike amplitude associated with arterial occlusion suggesting that the recruitment of high threshold MU is not only affected by the force and speed of contraction but also the availability of oxygen [11, 13, 24]. Results from the use of Integrated electromyography (iEMG) are consistent with these findings, demonstrating no practical difference in iEMG activity between low intensity occlusion and high intensity non occlusion training suggesting that a greater number of FT fibers are activated at low intensities [34–36].

**mTOR Pathway**

Increased rates of protein synthesis help to drive the skeletal muscle hypertrophy response [39]. S6K1 phosphorylation – a critical regulator of exercise-induced muscle protein synthesis – has been demonstrated to increase with occlusion training. Phosphorylation of S6K1 at Thr389 was increased by three-fold immediately post exercise with occlusion training, and remained elevated relative to control at three hours post exercise [7]. Moreover research demonstrates that REDD1 (regulated in development and DNA damage responses), which is normally expressed in states of hypoxia, is not increased in response to occlusion training even though hypoxia-inducible factor-1 alpha (HIF-1α) is elevated. Normally HIF-1α mRNA expression correlates with a corresponding elevation in REDD1 [5]. The lack of increases in REDD1 mRNA expression may prove to be important as REDD1 works to reduce protein synthesis through inhibition of the mammalian target of rapamycin (mTOR), responsible for the regulation of translation initiation [5].

Currently there is no clear explanation for this paradox. However it is conceivable that an unknown factor is increased with occlusion training, which influences the transcription of HIF-1α and REDD1.

**Heat Shock Proteins**

HSPs are induced by stressors such as heat, ischemia, hypoxia, free radicals, and act as chaperones to prevent misfolding or aggregation of proteins. HSPs also appears useful to slowing atrophy [15], as HSP-72 plays a protective role in preventing protein degradation during periods of reduced contractile activity [26], by inhibiting key atrophy signaling pathways [4, 31]. The primary pathway involved in mediating protein degradation is the ubiquitin proteasome pathway. Recent in vivo data, demonstrate that increased levels of HSP-70 is sufficient to prevent skeletal muscle disuse atrophy by inhibiting the promoter activation of atrogin-1/muscle atrophy F-box (MAFbx) and muscle-specific RING finger 1 (MuRF1) as well as the transcription factors which regulate their expression, forkhead box O (Foxo) and nuclear factor of P+ NF-P+. Senf et al. [4] also observed that Foxo3a, a member of the Foxo family upregulated during atrophy, not NF-P+ is necessary for the increase in MAFbx and MuRF1 promoter activities during disuse. Regardless both transcriptional factor activities, Foxo and NF-P+ are inhibited with elevated HSP-70 levels. Incidentally, occlusion training has been shown to increase HSP-72 in a rat model [15], and Kawada and Ishii [15] postulated that the increase in HSP-72 could be a potential mechanism by which occlusion increases skeletal mus-
cle hypertrophy and attenuates atrophy [36], likely by inhibiting the mediating pathways of the ubiquitin proteasome pathway.

**NOS-1**

Nitric oxide synthase is an enzyme responsible for converting L-arginine into nitric oxide (NO), a small and electrically neutral molecule capable of moving with ease through tissues [3]. Neuronal NOS (nNOS) is found in the transmembrane/dystrophin protein complex of skeletal muscle [29]. At rest, nNOS continually produces low levels of NO which appear to maintain satellite cell quiescence. During exercise-induced contraction nNOS is thought to be activated by mechanical shear forces, as well as increased intracellular Ca$^{2+}$ concentrations [37]. nNOS is increased in conjunction with occlusion training, possibly mediated by the increased flux of Ca$^{2+}$, as well as reperfusion [15]. According to Anderson et al., [2, 3] a spike in NO production triggers the release of hepatocyte growth factor (HGF) from its binding to the muscle extracellular matrix followed by co-localization with its c-MET receptor on satellite cells leading to their activation. This model is supported by a number of findings demonstrating the inhibition of satellite cells in response to short-term L-arginine methyl ester (L-NAME) treatment following injury or mechanical stretch [3]. Interestingly, Kawada and Ishii [15] did not show an increase in NO, only nNOS, which could be due to the short life span of NO. In this study, NO concentration was measured indirectly by its oxidation products, therefore the obtained values might have resulted from the production and breakdown of NO, both of which might be influenced by the occlusion of blood flow.

**Myostatin**

Myostatin is a negative regulator of muscle growth and mutations of this gene result in overgrowth of musculature in mice, cattle, and humans [21, 22, 30]. Myostatin appears to inhibit satellite cell proliferation because Myostatin-null mice display muscle hypertrophy and increased postnatal muscle growth, which have been linked to increased satellite cell activity [10, 20, 23]. McCroskery et al. [20] conclude that Myostatin is expressed in adult satellite cells and that Myostatin regulates satellite cell quiescence and self-renewal, showing it does play a role in adult myogenesis.

Muscle Myostatin gene expression has been shown to decrease as a result of mechanical overloading [16], as well as in low intensity exercise with occlusion [15]. Occlusion may cause favorable hypertrophic changes in Myostatin as a result of hypoxia and/or the accumulation of metabolic subproducts.

**Conclusion**

In conclusion low-intensity occlusion training works through a variety of mechanisms, with the most prominent being metabolic accumulation, fiber type activation, and mTOR signaling. The research behind these mechanisms is incomplete thus far, and more studies should be included to elucidate the actual metabolite(s) responsible for the increase in GH with occlusion. Furthermore, research should be directed towards determining which particular mechanism(s) is associated to a greater degree with the hypertrophic/anti-catabolic changes seen with blood flow restriction. While we have a base foundation of possibilities, controlled studies addressing each proposed mechanism
would provide a better understanding of each. For example, the paradoxe of REDD1 and HIF-1α should be examined to determine if there is another factor that is increased in response to blood flow restriction. As postulated earlier, perhaps there is an unknown factor that influences the transcription of HIF-1α and REDD1 leading to the paradoxical increase in HIF-1α with a decrease in REDD1 expression. HSPs may also play an important role, specifically in attenuating skeletal muscle atrophy. While animal studies show promise, human studies should be performed to try and confirm the initial findings of Kawada and Ishii.

The mechanisms described in this paper have all been shown to potentially induce skeletal muscle hypertrophy in response to blood-flow restriction. Although some mechanisms may be more prominent than others, all the mechanisms described likely play at least some part in the enhanced skeletal muscle hypertrophy response associated with occlusion training. Fig. 1 summarizes the mechanisms by which blood occlusion training may stimulate muscular hypertrophy.

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