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Theresa Rakiewicz

The College at Brockport, pignager@gmail.com

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Determination of the effectiveness of the ZetrOZ wearable ultrasound device (SAM) for the
post-exercise clearance of lactic acid

A Senior Honors Thesis

Submitted in Partial Fulfillment of the Requirements
for Graduation in the Honors College

By
Theresa Rakiewicz
Exercise Physiology Major

The College at Brockport
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Thesis Director: Dr. Craig Mattern, Associate Professor, Exercise Science

Abstract:

Athletes performing in multiple round competitions seek rapid recovery from exercise and restoration of optimal muscle function. The increase in blood lactic acid concentration while performing intense exercise has been well documented; however interventions have not demonstrated the ability to significantly improve lactate removal beyond that of passive recovery. Low intensity therapeutic ultrasound has been previously shown to increase blood flow, fluid circulation, vasodilation, and cellular metabolism. **PURPOSE:** To determine if wearing a long-duration ultrasound therapy device on muscles can accelerate lactic acid kinetics to promote clearance from blood. **METHODS:** A randomized, double blind cross-over study utilizing a lactate-inducing circuit weight training protocol was performed by 10 male participants (22.1 ± 0.46 years). During the first visit to the lab, the 1-repetition maximum (1-RM) for each participant was estimated for the following exercises 1) lunge, 2) squat, 3) leg press, 4) leg extension, and 5) leg curl. During the next two visits participants performed two circuits of the five aforementioned lower body exercises at 70% of their 1-RM. The exercises were performed while wearing functional ultrasound devices during one visit and placebo devices during the other visit. The ultrasound devices were attached to both the quadricep and hamstring muscle groups and worn for 1 hour. Blood lactic acid was measured in duplicate at baseline as well as at 0, 2, 5, 10, 20, 40, and 60 min post-exercise. The area under the curve (AUC) of lactic acid concentration versus post-exercise time was calculated. **RESULTS:** AUC resulting from functional ultrasound devices (344.31 ± 41.12) was significantly lower than placebo devices (396.09 ± 35.24). Lactic acid values from functional devices at time 0, 2, 5, 20, and 40 min post-exercise (10.83 ± 0.8 , 10.61 ± 0.83 , 10.15 ± 0.96 , 6.97 ± 1.04 , 3.60 ± 0.55 mmol/L) were significantly lower than placebo respectively (12.71 ± 1.07 , 11.86 ± 0.61 , 11.69 ± 0.62 , $7.94 \pm .909$, $4.48 \pm .55$ mmol/L). There were no significant differences at time-points 10

and 60 min post-exercise. **CONCLUSION:** These results suggest that a wearable therapeutic ultrasound device promotes blood lactic acid clearance immediately following high intensity exercise, but may offer reduced benefit later in the recovery process.

Introduction:

Resistance training is strengthening muscles by using force to strain against an opposing force like body weight, free weights, strength machines, or bands (4). There are many benefits that go along with resistance training or strength training. Some benefits include a lower risk of mortality and the events of cardiovascular disease, managing chronic diseases especially osteoporosis and Type 2 diabetes mellitus, improving body composition, helping with blood glucose stability, increasing or maintaining resting metabolic rate, increasing muscle mass and strength, facilitating activities of daily living, and even just for appearance (8,10). Since there are so many benefits that are associated with strength training, it is a very popular type of activity for all ages. Athletes weight train to excel at their sport, adults weight train to keep their muscle mass from diminishing or to keep up with their children's activities, elderly people weight train to keep their independence, and anyone may weight train to maintain an overall healthy lifestyle with the advantages that go along with the activity like disease prevention. To strength train, anaerobic glycolysis is needed to create energy.

Anaerobic glycolysis is the breakdown of glucose through steps to either form a product called pyruvate or lactate. Anaerobic glycolysis is the process that occurs during strenuous exercise when the energy demand exceeds oxygen rate of use or supply. If oxygen is present and sufficient then aerobic glycolysis is utilized and pyruvate continues through the citric acid cycle and electron transport chain to produce the final outcome of ATP which is an energy source for the body to do every action it needs to do from moving proteins around the body to contracting

muscles to take a step or bite an apple (4, 10). Glycolysis occurs in the mitochondrion of a cell and through many steps of glucose breaking down with the help of enzymes and electron transporters to carry the hydrogens; the product of pyruvate is formed. Lactate is very similar to pyruvate, the only difference is an extra hydrogen bonded to an oxygen in lactate. An electron transporter, NAD^+ binds with hydrogen and through lactate dehydrogenase, lactate is formed.

Lactate is formed through other processes also such as energy formation from red blood cells that don't have mitochondria and from high glycolytic muscle fibers. These lactate producing processes are balanced with slow twitch muscle fibers that oxidize the lactate for energy. This way lactate is not accumulating in the blood to prevent a lack of homeostasis/normalcy (4, 10).

Even in anaerobic glycolysis, lactate doesn't become a problem right away. Lactate can be diffused into the blood or interstitial space to leave the original site or it can be used as a substrate for producing glycogen, a stored form of glucose, which in a way means lactate is supplying anaerobic glycolysis to keep occurring. The problem arises when ATP, the energy used in glycolysis, isn't formed as quickly as it is being used which creates an increase in lactate levels. Increasing lactate levels will cause fatigue and exercise performance will decline. This occurs because lactate is acidic and certain enzymes can't function in acidic environments causing a diminishing effect on muscle cell's contraction properties (4, 10).

Resistance training uses anaerobic glycolysis as its primary form of energy metabolism. Also, resistance training uses fast twitch muscle fibers for the most part which are the high glycolytic muscle fibers. These fibers cause quick high force bursts of energy but fatigue quickly. Doing activities that use mostly fast twitch muscle fibers can't last long because of the characteristics of these fibers and it is more difficult to recruit fast twitch fibers (4). These two

facts lead to the accumulation of lactate in large amounts especially if there isn't enough recovery time between sets of exercises.

Recovery is essential for people who exercise so that they can minimize soreness and muscle fatigue while restoring energy. Without proper recovery, people can get to a point in exercising where they cannot continue, start to fatigue too quickly, or they can hurt themselves/overtrain. Different recovery methods and times are needed for all different modalities, intensities, and durations of exercise (4, 10). Someone who just ran a marathon needs a different recovery than someone who just lifting weights for their upper body at 50% of their 1-Repitition Maximum.

Recovery comes into play when the muscle cells start to heal and muscles build up adaptively to resist damage from the same activity just done previously (4, 10). Also, lactate levels that were building up during the exercise will start to lower usually within an hour to starting levels. This occurs because the lactate accumulation stops once the exercise stops because there isn't as much lactate produced as there was during the exercise so lactate can be used for all those different reasons and slowly the lactate levels will dwindle as time goes by and the exercise continues to be stopped. This will help with the muscle fatigue feeling also.

Many recovery methods have been studied to test lactic acid kinetics to determine what methods can be effective in post-exercise recovery. Superficial and deep massaging were found to not be effective with post-exercise lactic acid clearance as well as passive static stretching (1). Active recovery was found to be more effective than passive and massage therapy, but combined therapy of active recovery with massage was also a very effective modality based on the study (11).

Ultrasound is the recovery method used in this study. Ultrasound is a heating therapy that can be applied to very small areas of the body. It heats up below the very superficial muscular layer to get to the deeper muscle to increase its temperature temporarily. Ultrasound is typically used to help heal injuries or display images of body parts to see possible injuries on the body. Some common injuries ultrasound helps with are arthritis and impingements (13). Recently, it has been used sometimes for recovery from muscle soreness. Mainly, ultrasound is effective because it increases skeletal muscle temperature which causes vasodilation. Increased vasodilation increases blood flow which in turn transports lactic acid from the muscle to the body's bloodstream where it will be consumed by 1.) the liver for oxidation or conversion into glycogen, 2.) the heart muscle to be used as fuel or 3.) working muscles by oxidation (6, 7, 8, and 14).

Ultrasound can speed up recovery and rehabilitate also by increasing metabolism, minimizing inflammatory response, decreasing joint stiffness, increasing the flexibility of collagen fibers (5). With ultrasound being able to help with all these bodily responses, recovery can be sped up. Increasing the inflammatory response and metabolism means that the healing process is sped up and less metabolic products are formed leading to a shorter time of inflammatory responses and less pain and discomfort overall (13). Since the recovery period is thought to shorten with ultrasound, lactate removal should increase since blood flow is increased speeding up the healing process overall.

ZetrOZ, a portable ultrasound device company, has used its device in the past to help relieve pain and muscle spasms and now it is being tested for lactate removal and recovery (6, 7, 8). These devices give localized low intensity ultrasound to the body parts that they are connected to for a longer duration of time than with higher intensity ultrasound (7, 8). With this

device, it gives all the advantages that ultrasounds can give for a longer period of time and the devices can be connected to the muscle of choice and multiple sites can be connected to increase the area receiving the heat treatment. Therefore my thesis is that ZetrOZ low intensity ultrasound devices speed up muscle recovery from resistance training through an enhanced lactate blood clearance.

Methods:

Subjects

Ten male participants between the ages of 18-40 years old participated in the study. See table 1. Inclusion criteria included 1.) no lower body orthopedic injury history in the past year and 2.) the performance of strength training at least three times a week for the past three months.

Table 1. Participant Characteristics

(n = 10)	Mean \pm SEM
Age (yr)	22.1 \pm 0.45
Height (cm)	178.44 \pm 2.11
Weight (kg)	86.55 \pm 2.62

Study

Each participant came to the laboratory on 4 occasions. The first visit was mainly introductory. Each participant was informed in more detail about the study, signed an informed consent, and filled out a health history questionnaire.

1-Repetition Maximum

The second visit was to gather baseline measurements of 1-repetition maximums. The 1-repetition maximum was tested on 5 lower body exercises which included lunges, leg extensions, smith squat, leg curls, and leg press. To estimate the repetition maximum, they were asked to complete approximately 8-12 repetitions of each exercise using a weight that was expected to produce failure on the 13th repetition. Once the weights and repetitions were collected for each exercise, the formula to estimate the maximum was used: $1\text{-RM (lbs)} = \text{weight lifted (lbs)} / [1.00 - (\text{number of reps} \times 0.02)]$. The 1-RM was recorded for each exercise for every individual.

Circuit Training

The circuit training consisted of the 5 lower body exercises: weighted lunges, leg extensions, smith squat, leg curls, and leg press, using of 70% of their 1-repetition maximum that was calculated for each exercise. Each exercise was completed for 30 seconds at a metronome rate of 44 beats/min. There was a 15 second break between the exercises that gave the participant just enough time to get to the next machine for the next exercise. After the first set of the circuit, the participant was given a 30 second standing break before completing the circuit for a second time.

Blood Lactate Measurements

Lactate concentrations were measured using the YSI 1500 Sport lactate analyzer. The analyzer was calibrated before measuring participant's blood samples. This is a 2 point calibration in which 5 mmol and 15 mmol of standard were analyzed by the machine. Starting with the 5 mmol solution, 2 capillary tubes were filled up to the black line on the capillary tube with the solution and then the tube was placed into the injector after wiping the outside of the tube off to make sure no liquids were on the outside. After the tube is twisted until it is tight in the injector, the injector is placed into the lactate analyzer with the tube inserted into the analyzer

and the plunger is depressed until there is pressure felt which means the cotton ball is at the end of the capillary tube and with the thumb still pressed down, the injector is lifted out of the machine. The same process is repeated with the second capillary tube after the first reading is recorded from the first tube. The values should be between 4.9 mmol-5.1 mmol. For the 15 mmol standard, this solution is run as a blood sample instead of a standard because this machine doesn't calibrate with a solution that high. The same process as the 5 mmol solution is run and the values should be between 14.3 mmol- 15.7 mmol.

Each time lactate concentrations were needed; two capillary tubes were filled of blood and then injected into the analyzer and the two concentrations were averaged.

Ultrasound Devices

Four ultrasound devices were applied to each participant. Each ultrasound device had four bandages connected that were applied to the lower body. On the both quadriceps, one bandage was applied 3-4 inches above the knee and the other bandage was applied 4 inches above that one. On both hamstrings, one bandage was applied 3-4 inches above the knee, similar to the quadriceps placement, and the other bandage was applied where the hamstring and gluteus maximus connect. Most participants needed to shave to allow the bandages to adhere properly. In total 8 bandages were attached to the upper leg. Some of ultrasound devices were the functional devices that applied ultrasound to the muscles while the other devices were placebos. The ultrasound devices were color coded; half were yellow while half were green to distinguish the two types of devices.

Experimental Procedure

The third and fourth visit consisted of going through the circuit resistance training program twice with minimal rest time between exercises at 70% of their 1-RM. Upon arriving,

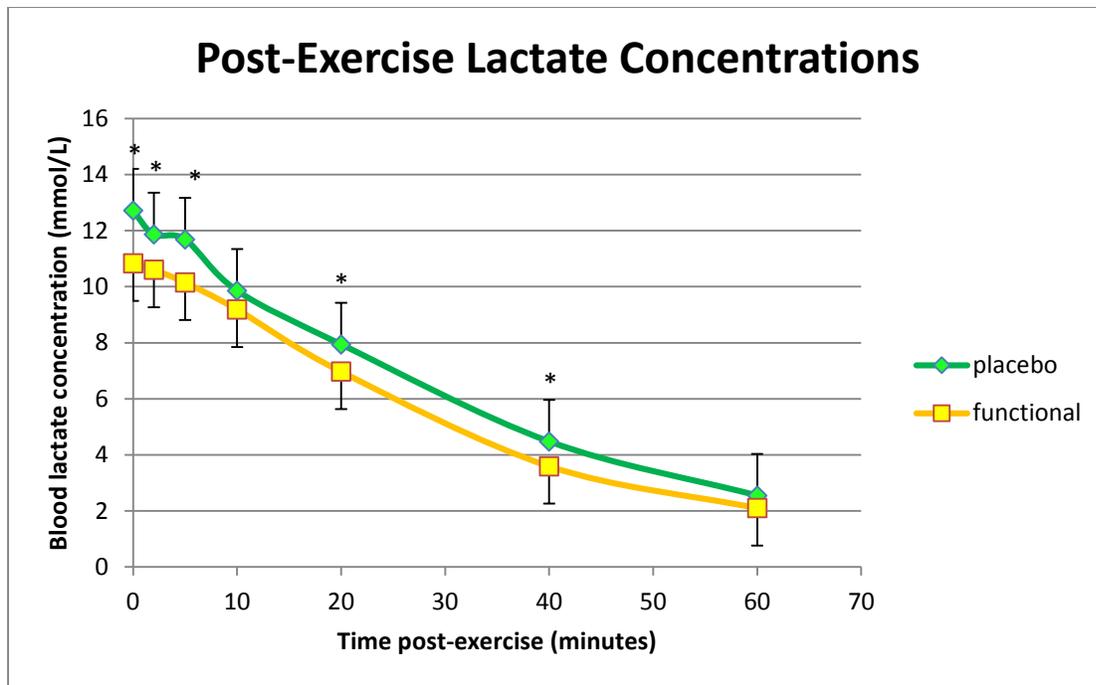
the participant was weighed and then an average baseline blood lactate measurement was taken and recorded. One of the two final visits, the participant was attached to the functional device and the other visit they were attached to the placebo. The study was a double-blind randomized crossover design so the participants and investigators had no knowledge of which device they were applying and wearing. The participants sat leisurely for an hour with the devices attached and at 60 minutes of resting, another lactate measurement was taken and then they began the circuit training. Once the participant finished the circuit the second time, blood lactate concentrations were measured for an hour in timed intervals: immediately post exercise, 2, 5, 10, 20, 40, and 60 minutes post exercise. Blood lactate measurements was recorded for data collection to run through statistical testing for the results.

Statistical Analyses

Time points were compared using one-tailed paired t-testing between the functional and placebo conditions using Microsoft Excel 2010 program. AUC was calculated for each blood lactate measurement and the same one-tailed paired t-test was completed to test for statistical significance. A level of $p < .05$ was used to test for statistical significance. Values were presented as mean \pm SEM.

Results:

At time points 0, 2, 5, 20, and 40 minutes post-exercise, blood lactate concentrations in the ultrasound condition were significantly lower ($p < 0.05$) compared to the placebo condition ($P = 0.02, 0.02, 0.02, 0.03, 0.001$). Time points of 10 and 60 were not significantly different between conditions ($P = 0.14, 0.06$) but it is hypothesized that with more participants time points 10 and 60 will likely become significant. Area under the curve of the entire time the data was collected was also calculated. Area under the curve of lactate in the ultrasound condition (344.31 ± 41.12) was significantly lower compared to the placebo devices (396.09 ± 35.24) ($P = 0.009$).



Graph 1. Blood lactate values were taken at times 0, 2, 5, 10, 20, 40, and 60 minutes after circuit weight training using both the placebo and functional ultrasound devices. For each time point two samples of blood were drawn then averaged. *At time points 0, 2, 5, 20, and 40 minutes post-exercise, values in the ultrasound condition were significantly lower than in the placebo condition ($p < 0.05$). Values are presented as means \pm SEM.

Discussion:

Based on the results from the study, wearing a long-duration ultrasound therapy device on muscles can accelerate lactic acid kinetics to promote clearance from blood. The most critical finding was that most of the time points post-exercise of lactic acid concentrations were significantly lower for the participants while they wore the ultrasound devices. Lactate clearance was accelerated when the participants were wearing the ultrasound devices since their lactate concentration levels were significantly lower. Based on how ultrasound works as a recovery device, it is likely that the heated muscles caused vasodilation which allowed the lactic acid to leave the muscles and go to one or more of the lactate usage locations: liver, heart of slow twitch

muscle fibers. Time point 10 minutes and 60 minutes post exercise were not significantly lower but that is hypothesized only to be because of the small participant pool. With more subjects in the next couple of months, it is likely that at the 10 minute time point the functional ultrasound will also be significantly lower than the placebo like the surrounding time points. At 60 minutes, it is somewhat unclear if the functional device will cause a lowering in lactate concentrations compared to the placebo because the body naturally clears lactate at a certain rate and usually around 60 minutes, the body will clear out the excess lactic acid by itself (1). With more participants, it will be clearer as to if the ultrasound will cause lower lactate concentrations at 60 minutes or if the body will catch up to the ultrasound by 60 minutes.

The other finding that was significant was the area under the curve for total lactic acid concentrations post-exercise was significantly lower using the functional device, which is a finding that supports the hypothesis of the study. If the area under the curve is lower, this means that the total area under all the time points using the functional device is significantly less than the placebo showing that the ultrasound overall had a significant lowering on the lactate concentrations. With each time point looked at as well as the overall post-exercise time, it gives a better overall picture that ultrasound devices do have a significant lactate clearance mechanism to them based on this study.

Limitations to the study

During the study, there were only minor challenges faced. Timing for the participants as well as the data collectors was hard to schedule so participants had different time periods between each visit. Also, some participants did not bleed as easily so it was difficult to get some blood samples, and the bandages that devices connected to the participant's legs fell off or overheated

at times so bandages needed to be replaced and devices needed to cool off to allow for continuing usage.

Practical application

With this study supporting the hypothesis that ultrasound can accelerate lactic acid kinetics, ultrasound can help athletes and active people recover quicker and easier than normally. People everywhere go out to buy recovery drinks, get massages, foam rollers, compression socks and many other items and techniques to help their body feel better quickly to exercise as frequently as they desire. With the portable ultrasound device, strength trainers can lift weights at a high intensity and put on the ultrasound devices to clear the lactic acid quicker so that they will have less muscle damage the next time they want to go strength train again. This can be applied to runners or cyclist or swimmers also. For competitions where multiple bouts of exercise are needed like track and field or wrestling, using the ultrasound device, athletes will be able to compete their first event then recover quicker and more fully to compete again to their highest capability. An advantage of this device is that the person wearing the ultrasound does not need to actively do anything to recover quickly besides put on the bandages and turn on the ultrasound. There is no active recovery of jogging or biking or stretching or rolling out unless they would like to combine the recovery methods to have a more advantageous recovery.

Research

While there has not been much research in the field of ultrasound and lactate, there have been some. One study tested to determine how ultrasound helps with lactate removal and tested the question by swelling up muscle cells by increasing the saline solution the cells were bathed in and applying ultrasound. They hypothesized from the testing that ultrasound does not accelerate lactate leaving the cell, but it accelerates lactate circulating once out of the cell (3). More

research needs to be done to test how ultrasound accelerates lactate kinetics. While ultrasound is still a relatively new topic of interest for exercising recovery, lactic acid removal and recovery methods are very well researched. Active recovery has been supported to have the highest lactate clearance compared to stretching or massage. A study measured blood lactate concentrations on 18 trained male cyclists during two 5k maximal effort cycling tests with a 20 minute recovery between consisting of either passive, active (50% VO_2max), massage, or combined massage and active. Active recovery removed the greatest amount of lactate at minutes 9 and 12, but combined recovery removed the greatest amount at minute 15. Massage is hypothesized to be too superficial to have any real effects on lactate concentrations and the mechanisms of stretching don't seem to effect lactate concentrations either. Nine participants were involved in a study where they exercised at 90% of their VO_2max for 8 minutes followed by a 10 minute recovery intervention of passive, active, stretching, deep or superficial massage. Active recovery had lowered blood lactate concentrations after the intervention while stretching and both massages had similar concentrations to passive recovery (1, 11).

Further Research

There are many different topics within ultrasound and lactate removal that could be further researched. Looking in more detail into how lactate is removed from cells and how ultrasound actually helps accelerate this process is one direction. Another direction could be to try different intensities of ultrasound to find the optimal intensity to maximize recovery. The most important direction should be to compare ultrasound devices to other active forms of recovery like massage, active stretching, jogging or walking to see what the best recovery methods are. Adding portable ultrasound devices to the groupings of recovery methods can have a big effect on active people and athletes everywhere.

Conclusion

Recovery is a very crucial part of any active person's life. Poor recovery can cause unnecessary days off and delays in personal goals and achievements. Proper or even above average recovery can make all the difference in a competition or an accomplishment. There are many already researched recovery methods out there for athletes and everyone to use or do to make their recovery optimal, but portable low intensity ultrasound is a new topic because in the past it has been used mainly for healing in therapy. Ultrasound has the potential to become one of the best recovery techniques out there for the world to use.

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