Original Article

Effects of Isokinetic versus Isotonic Training on Salivary Immunoglobulin A and Alpha Amylase in Advanced Level Adolescent Weightlifters

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Abstract:

The purpose of this study was to examine the effects of isokinetic training versus isotonic training among adolescent state-level weightlifters on salivary immunoglobulin A (SIgA) and alpha (α)-amylase responses immediately post-training and one month after cessation of training. Nineteen young advanced level weightlifters (age: 14.74 $\pm$ 1.37 years old) participated in this study. They were divided into two groups namely isokinetic and isotonic. Participants in the isokinetic group performed 24 sessions (eight sessions per week for eight weeks) of isokinetic training by using an isokinetic dynamometer whereby participants in the isotonic group performed 24 sessions of isotonic training by using a dumbbell. Saliva samples were collected pre- and post-training, and one month after cessation of training and was analyzed for SIgA and α -amylase concentrations and their secretion rates. As a result, SIgA and α -amylase concentrations and secretion rates were not significantly affected by the type of training in advanced level of adolescent weightlifters. Furthermore, there was no significant effect on the saliva parameters measured across time.

Key Words: immune responses, youth, weightlifting, resistance training

Introduction

Immune responses are largely depending on the type, intensity and duration of the exercise, fitness level, environmental factors and the duration of measurement (Nieman & Pedersen, 2000). In general, prolonged and high intensity exercise may induce suppressed immune function and subsequently increase the risk of infection. On the other hand, regular and moderate intensity exercise may enhance immune function and decrease the infection risk below average (Nieman, 1994). However, these findings were proposed based on numerous studies on endurance type of exercise. To date, limited studies have been carried out to investigate the effects of resistance exercise on immune responses (Sclczak et al., 2015).

Resistance exercise is a form of physical exercise which can cause muscle damage and a change in immune response. Resistance training could impose a significant change on the immune system despite its shorter duration of effort (Koch, 2010). Isokinetic exercise causes an alteration in the immune system. There are several studies that highlighted the usage of isokinetic exercise to investigate the immune responses such as inflammation by measuring the level of cytokines. Lee et al., (2011) found significant changes of IL-6 plasma concentration after ten weeks of passive repetitive isokinetic training exercise in high intensity group compared to low intensity and control groups. Another study by Minetto et al., (2006) reported that an acute bout of high intensity isokinetic exercise induced a high concentration of serum IL-6 in both power (e.g., bodybuilder) and endurance (e.g., long-distance cyclist) athletes although greater IL-6 concentration level was observed in power athletes. However, study with regards to salivary immunoglobulin A (SIgA) and alpha (α)-amylase responses are still scarce to date.

SIgA is a type of salivary antimicrobial proteins that is important for defense against pathogens which invade mucosal surfaces of oral cavity. It has been shown that SIgA play roles in oral immunity by preventing adherence of microbial and neutralizing enzymes, toxins and viruses (Marcotte & Lavoie, 1998). A few studies have demonstrated that following prolonged and high intensity exercise, SIgA concentration decreases and in turns may increase the risk of getting infection (Fahlman and Engels, 2005; Gleeson et al., 1999).

Salivary α -amylase is an enzyme that is important to host defense by inhibiting the adherence and growth of certain bacteria (Scannapieco et al., 1994). Ehlert et al. (2006) proposed that increment of α -amylase might reflect the interaction of stress-dependent sympathetic and parasympathetic stimulation via central nervous noradrenergic input. Moreover, its secretion appears to be dependent on the intensity of exercise (Bishop et al., 2000). It has been reported that α -amylase concentration increased following aerobic exercise (Chatterton et al., 1996).

Despite of the listed benefits of isokinetic training, to date, no studies have been carried out to compare the effects of isokinetic training versus isotonic training on immune responses. Therefore, the aim of the present study is to examine the effects of isokinetic training and isotonic training on the SIgA and α -amylase responses. In addition, this study also aims to investigate the cessation effects of training on the measured parameters.

Materials & Methods

Participants

A priori sample size calculation showed that 8 participants per group are sufficient to yield 0.8 power of the study with effect size of 0.6 (large sample size according to Cohen, 1988). Those weightlifters with at least two years of experience in competitive weightlifting and without previous history of shoulder injury were included. Their age was between 13 and 17 years old. Participants were excluded from the study if they were unable to adhere to at least 85% of training loads during the intervention and/or have undergone rehabilitation for any musculoskeletal injury within the last two years.

Participants were provided with detailed explanation regarding the methodology of the study. Upon agreement, their written consent form was collected. Since they were under 18 years old, assent was obtained from their guardians. The participation in the present study was voluntary. All procedures were conducted with compliance to Declaration of Helsinki 1975. Ethical approval was obtained from Human Ethical Committee of Universiti Sains Malaysia (USM/JEPeM/14110457).

Procedure

The entire experiment was conducted during the preparatory phase of the weightlifters' training cycle. All participants were recruited from the same pool of weightlifters with standardized training program provided by their coach. Their program included five strength training sessions and one tactical session per week. The duration of their standardized training session was two hours per session. Meanwhile, the training program prescribed in the current study (e.g., isokinetic and isotonic upper limb resistance training) was an additional training for the athletes.

After obtaining their written informed consent, baseline measurement of weight and height, and saliva samples collection was carried out. Then, participants were grouped into either isokinetic or isotonic groups by matching their weight. After that, participants performed the training program (either isotonic or isokinetic) for eight weeks with three sessions per week. Immediately post-training and one month after training cessation, the body height and weight were measured and saliva samples were collected again.

For the saliva samples collection, participants arrived at the laboratory at 8.30 a.m. after an overnight fasting. Participants then sat quietly for 15 minutes. With an initial swallow to empty the mouth, saliva was collected through four minutes of unstimulated dribbling into the pre-weighed sterile bijoux tube with eyes open, head tilted slightly forward, and without using any mouth and tongue movements. All saliva samples were kept at -80°C until analysis. Saliva samples were analysed for SIgA concentration using an ELISA kit (AssayMax™ ELISA Kits, Assaypro LLC, USA) and α -amylase concentration using an ELISA kit (Human α -amylase-AMS/AMY, QAYEE-BIO, China).

The saliva flow rate and saliva antimicrobial protein secretion rate (SIgA and α -amylase) were calculated as follows:

- Saliva volume (ml) = Difference in weight (g) of bijoux tube before and after the collection of saliva assuming saliva density of 1.0 g/ml
- Saliva flow rate (ml/min) = $\frac{\text{Saliva volume (ml)}}{\text{Collection time (min)}}$
- SIgA/ α -amylase secretion rate ($\mu\text{g/min}$) = Saliva flow rate (ml/min) x SIgA/ α -amylase concentration ($\mu\text{g/ml}$)

Training Program

The training program consisted of 24 sessions of the training with three sessions per week for eight weeks. Participants of the isokinetic training group performed the training at the Sports Science Laboratory, USM by using an isokinetic dynamometer (Multi-joint System Biodex Pro, Shirley, NY, USA). At the beginning, participants perform familiarization whereby they were asked to complete one set of 12 reciprocal internal and external shoulder rotations in concentric mode. The training program commenced following at least three days of recovery. During each training sessions, ten minutes of warm-up emphasizing on the shoulder was performed by each participant followed by a minute of active rest. The dynamometer was calibrated with known weight before each training sessions. A correction for the mass of the limb and lever arm system was made on all torque curves. The preparation of participant on the dynamometer followed the manufacturer's manual closely to ensure safety of the participants.

In terms of participant preparation, for the first until the eighth sessions, participants were trained in the seated position with 45° of shoulder abduction. In this position the scapular was positioned so that equal distribution of external rotators and internal rotators can be trained. The angular velocity set was at $120^{\circ}.\text{s}^{-1}$, with 12-15 repetitions for two sets. For the ninth to the 16th sessions, the seated position with 90° of shoulder abduction was selected. The angular velocity applied was $240^{\circ}.\text{s}^{-1}$ with 10-12 repetitions for three sets. For the 17th to 24th sessions, training was conducted in standing position while diagonally lifting the bar. The angular velocity was set at $360^{\circ}.\text{s}^{-1}$ with 8-10 repetitions for four sets. For all training positions, rest interval between the sets was provided for one minute. Training was conducted for both sides of upper limb. For cool down, participants performed a proper shoulder stretching and ice pack was applied on the shoulder for ten minutes to reduce muscle soreness. Training was progressive in terms of the number of sets, angular velocity applied and

changes of body position to resemble upper limb's movement during weightlifting. The details of the program were summarized in Table 1.

Table 1 Characteristics of prescribed isokinetic and isotonic training

Sessions	Body position	Repetitions	Sets	Rest (min)	Velocity ($^{\circ}\cdot s^{-1}$)
1 to 8	Seated with 45° of shoulder abduction	12-15	2	1	120
9 to 16	Seated with 90° of shoulder abduction	10-12	3	1	240
17 to 24	Standing while lifting the bar diagonally overhead	8-10	4	1	360

Velocity was applied for isokinetic training only

Each training session was completed approximately in one hour including the warming up and stretching sessions. The same lifting positions, duration of rest interval between sets, number of sets and repetitions were applied by the isotonic group however their training was conducted using a constant weight (e.g., dumbbells of 50% of their upper limb's weight). However, the angular velocity was not fixed in isotonic group.

Statistical analysis The saliva volume, saliva flow rate, SIgA and α -amylase concentrations and secretion rates were compared between the isokinetic and isotonic training group for the three time points (e.g., pre- and post-training, and one month after training cessation) using general linear model mixed ANOVA with repeated measure to determine the interaction between types of training program and time period. Statistical significance was accepted at $p < 0.05$. Data were expressed as mean $\pm$ SD.

Results

In this study, a total of 23 participants were recruited from Kelantan state-level weightlifting team with range of age between 13-17 years old. However, only 19 participants successfully completed all the 24 sessions of training and participated in a series of immunological tests conducted at the pre- and post-training and one month after the training cessation. Four participants dropped out due to lack of adherence to the prescribed training program. Data only from the remaining participants' were included in the subsequent analyses.

Descriptive data of physical characteristics of participants were tabulated in the Table 2. The general linear model mixed ANOVA with repeated measure design revealed significant time and group interaction ($df = 2$, $F = 7.137$, $p < 0.05$) and significant main effects of time ($df = 2$, $F = 57.19$, $p < 0.05$) on mean body weight. An increment of body weight was observed in both groups at post-training compared to the pre-training and at one month after training cessation compared to the post-training.

Table 2: Physical characteristics of participants (N = 19)

	Isokinetic Group (n=8)	Isotonic Group (n=11)
Age (years)	14.36 $\pm$ 1.06	14.82 $\pm$ 1.60
Height (m)		
Pre-training	1.59 $\pm$ 0.07	1.58 $\pm$ 0.11
Post-training	1.60 $\pm$ 0.07	1.59 $\pm$ 0.11
1 month after training cessation	1.61 $\pm$ 0.07	1.59 $\pm$ 0.11
Body Weight (kg)		
Pre-training	70.82 $\pm$ 19.63	57.00 $\pm$ 10.79
Post-training	73.24 $\pm$ 19.40*	58.25 $\pm$ 10.88*
1 month after training cessation	74.49 $\pm$ 19.97**	65.00 $\pm$ 25.28**
Body Fat Percentage (%)		
Pre-training	33.87 $\pm$ 11.30	24.93 $\pm$ 5.86
Post-training	36.31 $\pm$ 10.57	27.96 $\pm$ 6.14
1 month after training cessation	36.31 $\pm$ 10.58	26.74 $\pm$ 7.18
Fat Free Mass (kg)		
Pre-training	45.44 $\pm$ 8.64	46.63 $\pm$ 15.47
Post-training	45.54 $\pm$ 9.47	45.46 $\pm$ 13.59
1 month after training cessation	46.27 $\pm$ 9.44	46.67 $\pm$ 14.01

* Significantly different from its respective pre-training value

** Significantly different from its respective post-training value

Table 3 summarizes the descriptive data and results of mean saliva volume and saliva flow rate. After eight weeks of training, the general linear model mixed ANOVA with repeated measure design revealed no significant time and group interaction ($df = 1.255$, $F = 0.513$, $p = 0.524$) and no significant main effects of time ($df = 1.255$, $F = 0.610$, $p = 0.481$) on mean saliva volume between both groups. Similar results were obtained for saliva flow rate whereby no significant time and group interaction ($df = 1.258$, $F = 0.523$, $p = 0.520$) and no significant main effects of time ($df = 1.258$, $F = 0.602$, $p = 0.485$) between the groups.

Table 3: Saliva volume and flow rate following isokinetic and isotonic training

Groups		Pre-training	Post-training	1 month after training cessation
Isokinetic Group	Saliva volume (ml)	2.07 ±0.11	2.06 ±0.31	2.07 ±0.58
	Saliva flow rate (ml/min)	0.51 ±0.03	0.51 ±0.08	0.52 ±0.14
Isotonic Group	Saliva volume (ml)	2.12 ±0.27	1.86 ±0.31	2.16 ±0.68
	Saliva flow rate (ml/min)	0.53 ±0.07	0.47 ±0.08	0.54 ±0.17

Descriptive data and results of mean salivary IgA and its secretion rate were presented in Table 4. After eight weeks of training, the general linear model mixed ANOVA with repeated measure design revealed no significant time and group interaction ($df=2$, $F=0.62$, $p=0.544$) and no significant main effects of time ($df=2$, $F=0.794$, $p=0.46$) on mean salivary IgA volume. Similarly, no significant time and group interaction ($df=2$, $F=0.042$, $p=0.959$) and no significant main effects of time ($df=2$, $F=1.837$, $p=0.177$) were observed the salivary IgA secretion rate.

Table 4: Salivary immunoglobulin A (SIgA) concentration and secretion rate following isokinetic and isotonic training

Groups		Pre-training	Post-training	1 month after training cessation
Isokinetic Group	SIgA concentration (µg/ml)	489.68 ±74.91	426.33 ±63.03	492.53 ±79.38
	SIgA secretion rate (µg/min)	278.98 ±84.48	241.69 ±82.41	278.87 ±73.65
Isotonic Group	SIgA concentration (µg/ml)	496.11 ±190.83	480.18 ±194.67	511.07 ±133.65
	SIgA secretion rate (µg/min)	262.47 ±104.93	228.66 ±117.43	275.57 ±97.56

Table 5 summarizes the descriptive data and results of mean salivary amylase and its secretion rate. After eight weeks of training, the general linear model mixed ANOVA with repeated measure design revealed no significant time and group interaction ($df=2$, $F=0.955$, $p=0.395$) and significant main effects of time ($df=2$, $F=4.486$, $p=0.019$) on mean salivary amylase volume. However, no significant time and group interaction ($df=1.343$, $F=0.282$, $p=0.668$) and no significant main effects of time ($df=1.343$, $F=1.241$, $p=0.292$) were observed for the salivary amylase secretion rate.

Table 5: Salivary α -amylase concentration and secretion rate following isokinetic and isotonic training

Groups		Pre-training	Post-training	1 month after training cessation
Isokinetic Group	α -amylase concentration (µg/ml)	120.70 ±10.03	127.22 ±20.99	118.88 ±23.68
	α -amylase secretion rate (µg/min)	68.60 ±15.00	80.83 ±34.76	71.84 ±31.74
Isotonic Group	α -amylase concentration (µg/ml)	97.69 ±23.57	117.43 ±26.14	96.79 ±19.71
	α -amylase secretion rate (µg/min)	51.94 ±13.09	55.19 ±12.14	60.23 ±29.18

Discussion

In the present study, we found no significant difference of SIgA and α -amylase concentration and their secretion rate in advanced level of adolescent weightlifters following eight weeks of either isotonic or isokinetic training and its cessation. To date, no study has been conducted to compare the effects of isotonic and isokinetic training on SIgA and α -amylase concentration and their secretion rate particularly in advanced skill level of young athletes.

Findings about the effects of exercise on SIgA were inconsistent. For an example, Fricker et al., (1999) reported a decreased in SIgA concentration after more than seven months of training but it returned to the pre-training level after training cessation. Similarly, other study also reported a decrease of SIgA concentration among swimmers in post-training session after three months of training (Tharp & Barnes, 1990). In contrast,

Tharp (1991) reported an increased level of SIgA concentration among the young male basketball players after two months of training. However, Tsai et al., (2012) reported an increase level of SIgA due to prolonged and intensive resistance training among the weightlifters. Nevertheless, to date limited studies have been conducted that involved resistance training, particularly isokinetic exercise.

Theoretically, SIgA is secreted by both acinar and ductal units under the stimulation of α - and β -adrenergic and peptidergic receptors whereby the stimulation of β -adrenoreceptors caused an increase of SIgA secretion rate. However, prolonged β -adrenoreceptors stimulation appeared to reduce the replenishment of SIgA into the glandular pool (Proctor et al., 2003). The inconsistency of the secretory immune response of SIgA concentration and secretion rate may be attributed to the interaction between different types of stimulation and their receptors during exercise. This discrepancy may also be attributed to the differences during the time of the saliva collection (Burns et al., 2004) and due to the different methods of expressing SIgA, nutritional status of the individual, and the exercise protocol employed.

In terms of SIgA secretion rate, we observed that there was no significant difference across the groups but further calculation showed the trend of decrement from pre- to post-test in both groups with more decline was noted in isotonic group. Previous research showed that the SIgA secretion rate was reduced following a triathlon race Steerenberg et al., (1997) and tennis drills (Novas et al., 2003) but it was found to be stable following participation in soccer (Bishop et al., 1999) and cycling (Blannin et al., 1998). To the best of our knowledge, currently there were no studies comparing the effects of isokinetic and isotonic resistance training on SIgA concentration and secretion rate. Therefore, we can only compare with the results from different type of sports rather than different mode of muscle contraction during resistance training.

Nevertheless, majority of findings from a recent review article (Koibuchi & Suzuki, 2014) indicate that salivary α -amylase activity and secretion rate consistently increase after an exercise. Similarly, Allgrove et al., (2008) reported an increase of α -amylase following exercise at 50% $\text{VO}_{2\text{ max}}$ although it was not significant. It has been reported that α -amylase activity was associated to adrenergic activation. Therefore it was expected that exercise would elevate the activity of α -amylase above the baseline values since exercise can escalate the signals to sympathetic nervous system (McKune et al., 2014).

In terms of body fat percentage and fat free mass, there was no interaction between time (e.g., pre-training, post-training and one month after training cessation tests) and training groups (e.g., isokinetic and isotonic). This result suggests the intervention period had no effect on altering the body fat percentage and fat free mass. However, a significant interaction of time and intervention was observed in terms of body weight. The data showed the increment of body weight both in isotonic and isokinetic training groups at post-training and one month after the training cessation compared to their respective pre- and post-training values. Therefore, we suggested that the cause of increased body weight but not in body fat was due to the normal growth among the adolescent participants rather than caused by the prescribed training program.

The studies on the effects of resistance training especially comparison of isokinetic and isotonic training were scarce as most of the previous studies focused more on endurance rather than resistance exercise. Studies that compared the effects of training across three time points (e.g., before training, after training and one month after training cessation) on immune responses are lacking. Furthermore, less studies were involved with advanced level of weightlifters who have different physiological adaptations towards resistance training compared to novice and recreationally active population.

Conclusions

As a conclusion, this study found that SIgA and α -amylase responses towards both isokinetic and isotonic training were not significantly different in advanced level of adolescent weightlifters. More studies should be carried out involving isokinetic exercise and immune function since this research area is scarce to date.

Conflicts of Interest

There are no conflicts of interest to declare.

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