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The Influence of ACTN3 Gene Polymorphism on VO2max and Sprint Speed Based on Sprint Interval Training Intervention

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Abstract

It is known that one of the physical fitness factors is a non-modifiable genetic factor, one of which is the ACTN3 gene. The influence of this gene polymorphism on physical fitness response, both aerobic and anaerobic, to intervention is still limited. Sprint Interval Training (SIT) as one intervention factor enhances physical fitness, both aerobic and anaerobic. The research aims at observing whether ACTN3 gene polymorphism influences VO2max and Sprint Speed with SIT intervention. VO2max is a parameter of aerobic physical fitness, while sprint speed is a parameter of anaerobic physical fitness.

Twenty-eight male students of 18-25 years old were taken as the research's subjects from the Student Activity Unit of Sports of Jenderal Soedirman University using the consecutive sampling method. The subjects were divided by genotypes identified using a PCR-RFLP method into three groups, namely RR, RX, and XX. All samples undertake three sessions of SIT per week for five weeks with work to rest ratio (W:R) = (1:8). Before and after undertaking SIT regimen, the subjects were examined for VO2max using a Multi-Stage Fitness Test method and for speed using a Sprint 30 m method. The analysis was conducted using a One-Way ANOVA test at a significance level of 0.05. There was a significant difference at mean value VO2MAX ($p=0.033$) and speed ($p=0.048$) with each genotype group, and the highest change takes place with the genotype group RR. Study concluded that ACTN3 gene polymorphism influences a change in VO2MAX and speed after Sprint Interval Training intervention.

Keywords: *ACTN3 gene, Vo2max, Sprint speed, Sprint interval training.*

Introduction

Physical fitness profoundly influences an athlete's performance. There are two components of physical fitness, namely aerobic fitness, and anaerobic fitness. Each sport requires different physical fitness with each other. The peculiarity of each sport makes the basis for sport practitioners to adjust their physical training and nutrition. The genetic factor is something extraordinary, which are often forgotten. Genetic factor determines about 20-80% of the individual's performance concerning physical fitness dominance [1]. Some individuals are born with aerobic physical fitness dominance and vice versa. ACTN3 gene is a gene which encodes the production of protein α-actinin-3 released by muscle fiber type 2[2]. Protein α-actinin-3 is responsible for the formation of fast and robust movement like a sprint[3]. ACTN3 gene has three polymorphisms, with polymorphism XX, the production of protein α-actinin-3 is less than with polymorphism RR. Previous researches on ACTN3 have proven that RR polymorphism has better anaerobic fitness. However, polymorphism's influence on the outcome of physical exercise has not been studied much, whether specific polymorphism tends to result in better physical fitness with a physical training intervention.

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Therefore, we were interested in observing how ACTN3 gene polymorphism influences physical fitness, both aerobic and anaerobic, with particular physical training intervention[3].

One physical training optimally enhances physical fitness, both aerobic and anaerobic, is Sprint Interval Training (SIT). Sprint Interval Training (SIT) is an intermittent physical training with training period (high intensity) followed with break period (low intensity)[4]. SIT is conducted with training period using short, continuous sprint with maximum workload interspersed with break period with light activities like jogging[5]. Constant training with the SIT method may enhance durability performance[6]. Such training shows a change to better muscle performance and adaptation than regular exercise with shorter training time. Study showed that high-intensity training like SIT with individual ration may progressively enhance recruitment of active type muscle fiber[7]. Therefore, SIT is deemed appropriate to be an intervention in observing how ACTN3 gene polymorphism influences physical fitness, both aerobic and anaerobic. Aerobic fitness parameter shall take VO2max, while anaerobic fitness parameter shall take sprint speed parameter.

Method

Research Design: This quasi-experimental research employed pre- and post-test design approach without a control group. The research subjects were classified based on the results of the ACTN3 gene polymorphism examination into three groups, namely RR, RX, and XX groups.

Research Subject: The 28 male of healthy students, aged between 20 to 25 years old, were divided into three groups, namely 10 students with RR polymorphism, 9 students with RX polymorphism and 9 students with XX polymorphism. The subjects were selected using a consecutive sampling technique. They regularly performed physical training with medium to high intensity at least 2-3 times per week for minimum the last three weeks. Subjects had to be a healthy man and pass the physical fitness examination; Physical Activity Readiness Questionnaire/PAR-Q.

Research Intervention: Sprint Interval Training (SIT) was performed for five weeks with a frequency of 3times/week at interval 1-2 days. SIT regimen employs a ratio of work to rest at 1:8, constituting sprint with an active period of 30 seconds, and the rest period of 4 minutes intermittently for four repetitions. The total duration for one session was 28 minutes, consisting of core training of 18 minutes, warming-up of 5 minutes, and cooling-down of 5 minutes. The consistent active period intensity took Borg's Scale of 14-18, regular active rest period brings Borg's Scale 10-13, and consistent warming-up and cooling-down intensity take Borg's Scale 10-13 [8].

ACTN3 gene polymorphism examination: The PCR-RFLP method examined the ACTN3 gene polymorphisms as previous study[9]. PCR was conducted using forward primer 5' CTG TTG CCT GTG GTA AGT GGG 3', and reverse 5' TGG TCA CAG TAT GCA GGA GGG 3'. The DdeI enzyme was used to digest PCR product 60 minutesin 37^0 C. An electrophoresis was conducted with agarose gel 2.5% at 100 volt for 50 minutes.

VO2 max examination: A change to VO_{2MAX} was examined using a Multi-Stage Fitness Test method with a run from one sign to another sign at a distance of 20 meters and in harmony with previously recorded sound. Subjects were required to follow the rhythm as long as possible and the test was ceased when subjects fail to reach the end of the 20m track for 2 stages or get exhausted[9,10].

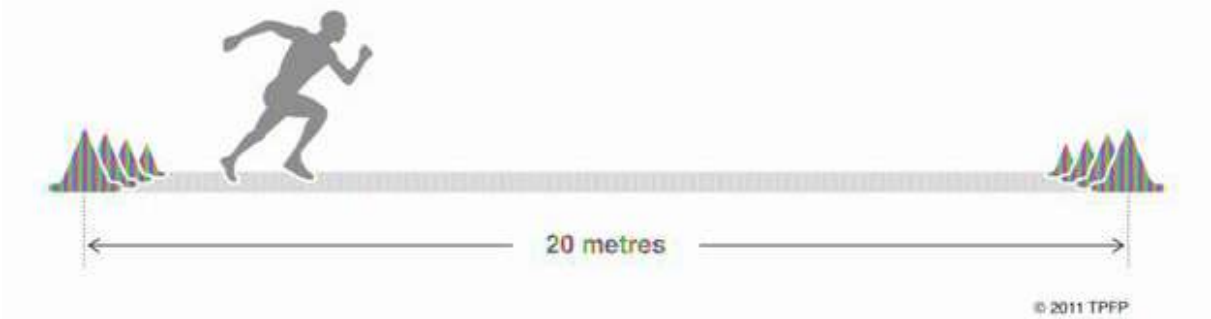

Figure 1. Multi-stage Fitness Test

Sprint Speed Examination:

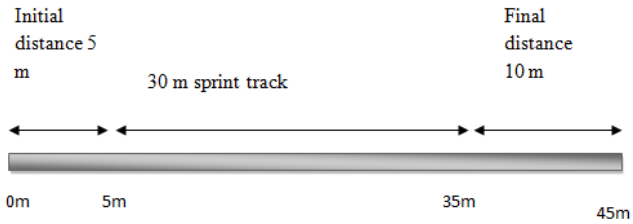

Figure 2. Sprint Speed Measurement Track

A change to sprint speed was examined with a sprint on a 30 meters track[11]. Additional 5 meters of initial distance was given to minimize any reaction time effect at the start of sprint on the travel time of 30 meters sprint. A final distance of 10 meters was given so that subjects may reduce their speed conveniently in prevention of injury risk.

Statistical Analysis: A change to VO_{2MAX} after SIT with all groups of ACTN3 gene was tested with a paired T-Test. The difference in the change to VO_{2MAX} after SIT between the ACTN3 gene groups was tested using One Way ANOVA. A Post Hoc test was employed as an advanced test to One Way ANOVA. The result of $p \leq 0.05$ was deemed significant.

Results

The baseline data of research subjects' age and body mass index (BMI) with each group are presented in Table 1. Table 1 shows that the age and BMI of subjects between groups are relatively equal, with the highest mean age is of RR group (21.10 ± 1.28 years old) and the highest mean BMI is of RR group (23.60 ± 1.24 kg/m^2).

Table 1. Characteristics of Age and Body Mass Index (BMI) among Subjects

Variable Characteristics	RR	RX	XX
Mean Age (years) ± SD	21.10 ± 1.28	20.50 ± 1.58	20.70 ± 1.15
Mean BMI (kg/m^2) ± SD	23.60 ± 1.24	22.44 ± 2.03	23.16 ± 1.95

The Paired T-test results show that there is significant difference in both speed variable and VO2 max before and after SIT intervention variable (Table 2).

Table 2. Paired T-Test results between Speed and VO2max Correlation

Variables	Pre SIT(mean±SD)	Post SIT (mean ± SD)	P Value
Speed	7.24 ± 0.50	7.51 ± 0.59	0.000
VO2max	35.29 ± 4.60	43.39 ± 5.63	0.000

The one way ANOVA test results (Table 3) show that the change to sprint speed and VO2max after SIT is statistically different between the ACTN3 gene groups with p = 0.048 for speed and p = 0.033 for VO2max ($p < 0.05$).

Table 3. One Way ANOVA Test Results comparing the Genotypes and Interventions

Variables	Pre SIT (mean ± SD)	Post SIT (mean ± SD)	Variable Change	P Value
Speed				
RR	7.55 ± 0.35	8 ± 0.37	0.45	
RX	7.12 ± 0.53	7.51 ± 0.56	0.39	0.048
XX	7.04 ± 0.50	7.26 ± 0.47	0.22	
VO2max				
RR	37.43 ± 6.01	47.45 ± 6.46	10.02	
RX	35.37 ± 4.40	34.28 ± 4.15	1.09	0.033
XX	34.28 ± 4.15	42.32 ± 5.32	8.04	

The Post-Hoc analysis to observe polymorphism group with significant difference revealed that significant difference takes place with RR and XX groups for speed (p=0.017) and RR and RX groups for VO2max (p=0.010). The highest improvement of speed and VO2max takes place with RR polymorphism group.

Discussion

This research proves that Sprint Interval Training intervention may enhance speed and VO2max. SIT training is one method of HIIT (High Intensity Interval Training), in which HIIT is defined as a high intensity training consisting of aerobic and anaerobic combination. High intensity training for individual will give benefit of enhancement of muscle mass, while low intensity training will enhance mitochondrial mass and oxidative enzyme activity[3]. A Study reported that HIIT training is correlated with recruitment of type IIa muscle fiber[12]. This theory reflect the informationthat high intensity training will change the composition of muscle fiber, from type I and type IIx muscle fiber to type IIa muscle fiber. Type IIa muscle fiber is muscle fiber with components almost equal to type I muscle fiber, in which the components include number of mitochondria, capacity of oxidative phosphorylation and capillary content[13,14]. The components may support muscle metabolism in a training which requires durability or VO2max. Previous researches also prove that SIT training performed for about 5 weeks shows a change to skeletal muscle metabolism, that it increases creatine kinase enzyme (creatine phosphate catalysis) and myokinase enzyme (resynthesis of ATP from ADP) with phosphate system[15,16]. Such improvement of creatine kinase and myokinase enzymes gives certain benefit to sport which requires speed like sprint. Therefore, performing SIT training will obtain double benefits in the form of enhancement of aerobic performance, in this case with enhancement of VO2max, and also enhancement of anaerobic performance, as shown with enhanced performance of sprint speed.

Furthermore, it proves that ACTN3 gene polymorphism influences speed and VO2max enhancement response with SIT intervention. The best enhancement of speed and VO2max is with RR gene polymorphism. Genotype RR with ACTN3 will encode the formation of protein α-actinin 3 the most. Skeletal muscle with protein α-actinin 3 has type 2 muscle fiber which works faster and is able to make more maximal contraction than muscle fiber without protein α-actinin 3 (XX)[17]. The reason is that protein α-actinin 3 interacts with calcium and calmodulin-dependent protein phophatase calcineurinso that protein α-actinin 3 may make more distribution of type II muscle fiber[18]. More type II muscle fiber causes better speed. In addition, mass enhancement of type IIa muscle fiber causes enhancement of durability in training.

Previous study on the rats to examine the difference in muscle biochemical characteristics found that the activities of Citrate synthase is found higher with type IIa muscle fiber than with any other type of muscle fiber[19]. Itcatalyze acetyl-CoA in krebs cycle of muscle cell oxidative metabolism track. Such enhanced activity of citrate synthase makes type IIa muscle fiber able to have stronger durability than any other type of muscle fiber[20].

It has beenproposed that m TOR and p70S6k phosphorylation are higher with ACTN3 gene R allele after physical sprint training[21]. mTOR and p70S6k phosphorylation serve to regulate skeletal muscle hypertrophy[22]. Therefore, we may conclude that hypertrophy and strength enhancement of skeletal muscle will be higher with individual with R allele after certain period of physical training. Moreover, another study also propose that testosterone level in male and female athletes is higher with individual with ACTN3 gene R allele[23]. This allows individual with R allele to have higher enhancement of muscle strength with physical training intervention, particularly resistance training. Similarly, the same occurs with the influence of ACTN3 on VO2max enhancement during SIT training. VO2 max is higher with genotype XX. However, after endurance training, the highest VO2 max enhancement takes place with individual with RR allele[24].

The influence of ACTN3 gene, particularly RR polymorphism, on the performance makes ACTN3 gene called “a gene for speed”. Researches show that ACTN 3 gene influences training adaptation, which in this case is speed and VO2max enhancement response during SIT. Moreover, ACTN3 evidently influences not only training adaptation, but also recovery speed during training and reduces injury risk [25].

This research is limited that it does not recall respondents’ nutrition, while it is possible that respondents’ energy during measurement may influence the result. Nutrition like high carbohydrate and protein may influence training adaptation.

Conclusion

The ACTN3 gene polymorphism influences a change to speed and VO_{2MAX} after Sprint Interval Training (SIT) intervention. The best response take place with genotype RR group.

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Conflict of Interest: Authors declare that there are no conflict of interest in submitting this manuscript. All authors are responsible for developing and completing this manuscript submission.

Ethical Clearance: The research has been reviewed by Ethical Committee of Health Research, Faculty of Medicine, Universitas Jenderal Soedirman.

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