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# **ACTN3 R577X POLYMORPHISM AND BODY COMPOSITION PROFILE OF INDONESIAN KARATE ATHLETES**

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## **Abstract**

An elite athletes is a complex trait that influenced by environmental factors and genetic predisposition. Karate is a part of combat sports that requires a high level of physical, physiological, complex technical skill and tactical excellent for success. One of candidate gene for excellent performance is  $\alpha$ -actinin 3 (*ACTN3*). This research was to investigated *ACTN3* R577X polymorphism and body composition profile of Indonesian Karate athletes. The study involved 29 Indonesian elite Karate athletes. Genotyping was performed by polymerase chain reaction and restriction fragment length polymorphism analysis using DdeI enzyme restriction. Body composition (height, weight, body mass index and percent of body fat) were measured. The genotype distribution was RR = 3 (10,3%), RX = 24 (82,8%), and XX = 2 (6,9%). No significant differences in body composition were found among genotypes. In conclusion, R allele was an advantage for success in Karate.

**Key words:** *ACTN3* polymorphism, genetic predisposition, Indonesian karate athletes.

## **INTRODUCTION**

Physical performance is a complex phenotype influenced by environmental (training, diet), behavioral and genetics factors (Gineviciene, *et al.*, 2011). The changes, at both cellular and molecular levels, that occur during physical exercise involve gene expression. For example, the skeletal muscles can vary in efficiency and the cardiovascular system can be more or less subject to fatigue, depending on how genes are expressed. The general hypothesis is that there is an inheritance component affecting physical and athletic fitness that is able to interact with environmental factors, particularly with training. Therefore, to understand the biological aspects of performance it is essential to understand the roles played by genes (Calo *et al.*, 2008).  $\alpha$ -actinin 3 (*ACTN3*) is one gene that indentified as candidate gene for explaining the inter-individual variability.

$\alpha$ -actinin are an family of actin-binding protein that play structural and regulatory roles in cytoskeletal organization. In human skeletal muscles, *ACTN2* and *ACTN3* are an important structural component of the Z disc and anchoring actin thin filaments (Mills *et al.*, 2001). The *ACTN2* gene is expressed in all fibers, while *ACTN3* is restricted to fast twitch fiber (Clarkson *et al.*, 2005). A common human polymorphism of the *ACTN3* gene was identified as a loss of function nonsense mutation replacing an arginine codon 577 (577R) with the premature stop codon (577X). This allele is unable to encode  $\alpha$ -actinin 3 protein, but because the *ACTN2* gene is expressed in both type I and II fibers, it can compensate for the loss of the *ACTN3* protein in type II fibers in individuals who are 577X homozygotes.

The nonsense allele is found in every human population, with a wide variation, implying that balancing selection may have been involved in maintaining the polymorphism. This suggest that the *ACTN3* genotype may be one of the factors that influence normal variation in muscle function (Yang *et al.*, 2003).

Several studies have investigated the relationship between *ACTN3* gene polymorphism and sport performance. Some reports indicated the positive association between the presence of the 577R allele and the capacity to perform high power muscle contractions (Clarkson *et al.*,

2005). The XX genotype is markedly under-represented among elite sprint, strength and power athletes. These previous studies suggested that  $\alpha$ -actinin 3 deficiency has a detrimental effect on the function of fast skeletal muscle fibers (Yang *et al.*, 2003; Druzhevskaya *et al.*, 2008). Karate is a sport that requires a high level of physical, physiological, complex technical skill and tactical excellent for success.

Modern karate consists of numerous repetitions of high intensity actions per fight lasting 1–3 s each, separated by low-intensity hopping-stepping movements ( $18\pm 6$  s) and short referees' breaks ( $9\pm 6$  s). Single defensive or offensive technique last  $0.3\pm 0.1$  to  $1.8\pm 0.4$  s for the shortest and longest duration actions, respectively (Tabben *et al.*, 2013). Physical conditioning is important for Karate athletes. Because competition is the focal point of athletic training, any training program should mimic the competition and reflect the desired adaptation.

Properly designed plyometric exercises and strength and ballistic training may increase punching and kicking speed, or power. Much of the power in various techniques, not only kicking techniques but also even hand techniques in karate, is generated through the hip rotation and related leg actions. To optimize power generated through the hip rotation, twist, crunches and other variations of rotary movements should be used, and also power exercises such as cleans and snatches should be used to increase power generated through the legs. It has been suggested that flexibility training may be needed to increase the range of motion (Iide, K *et al.*, 2008). Performing resistance training with short rest intervals, traditional cardiovascular interval training, and punching and kicking as quickly as possible with short rest intervals are recommended to increase the ability to buffer acid muscle and blood concentrations. The competitors may perform cardiovascular conditioning 3 days per week for short period of time to assist anaerobic recovery.

Karate is full body contact sport that injury can be happened. The common injuries in karate are sprains and bruises of the fingers, toes, and limbs. Most of these injuries could be prevented by hand, foot, and shin protectors. The significant injury sites are the head, neck, shoulder, and lower back. The strength training program in karate would include the neck, rotator cuff, and core stability and flexibility exercises. In addition to these exercises, the ballistic muscle contractions essential to various karate techniques necessitate development of agonist/antagonist muscle balance.

Overall karate fighting is ranked as a high intensity event. Lehmann (1998) has suggested that anaerobic metabolism is considered as the predominant source of energy in this sport. On the other hand, Beneke *et al.* (2004) have investigated the energetics of karate kumite based on measures of aerobic and anaerobic metabolism during simulated fighting, and concluded that the aerobic metabolism is predominant during this activity, although the decisive actions (*i.e.*, attacks and defense techniques) are maintained mainly via the anaerobic alactic system activation. Determinant actions in karate kumite are based on activities that require a high metabolic rate. Owing to the needs of aerobic and anaerobic demands of karate competition, elite class karate athletes are usually prescribed a mixed training regime that elicits adaptation in both energy pathways. The physiological profile of elite competitors has been presented recently showing the needs of developing both metabolisms in karate athletes (Tabben *et al.*, 2013).

Beside physical conditioning, specific anthropometric characteristics are needed to be successful in this sport. Athletes who have optimal physique are more likely to succeed than those who lack the general characteristics. In athletes, body composition measure are widely used to prescribe desirable body weights, to optimize competitive performance, and to assess the effects of training. It is generally accepted that a lower relative body fat is desirable for successful competition in most of the sports. This is because additional body fat adds to the weight of the body without contributing to its force production or energy producing capabilities. (Shafeeq *et al.*, 2010).

Based on knowledge about the role of *ACTN3* R577X polymorphism in skeletal muscle performance, the aim of this study was to investigated *ACTN3* R577X polymorphism and body composition in Indonesian Karate athletes.

## RESEARCH METHOD

### Subjects

Twenty nine Indonesian Karate athletes of elite and sub-elite level were participated in this study (14 male and 15 female). Subjects gave written consent to participate after being informed of the study protocol and procedures. The Ethics Committee of the Faculty of Medicine of Gadjah Mada University approved the study protocol.

### Body Composition Measurement

The height, weight, body mass index (BMI), and percent of body fat were measured. Body height was measured to the nearest 0,1 cm with the subjects standing with their back to stature meter. Weight was measured to the nearest 0,1 kg. Body mass index (BMI, in  $\text{kg}/\text{m}^2$ ) was calculated as weight (in kg) divided by height (in  $\text{m}^2$ ). Percent of body fat was determined by measuring the chest, abdominal, and quadriceps skinfolds using a caliper.

### Genotyping

DNA was extracted from peripheral venous blood by standard procedures. Genotyping of the *ACTN3* polymorphism was performed using polymerase chain reaction (PCR). The resulting PCR products were genotyped by restriction fragment length polymorphism (RFLP). The amplified fragment subsequently underwent digestion by *DdeI* restriction enzyme. Digested PCR fragments were separated by 3% agarose gel electrophoresis, staining with ethidium bromide, and visualized by ultraviolet.

## RESEARCH RESULTS AND DISCUSSION

Genotype distribution of RR, RX and XX genotypes in this study was 10,3%, 82,8%, and 6,9%, respectively. The allele frequencies are 51,7 % and 48,3 % for the R allele and X allele, respectively. The distribution of the *ACTN3* genotypes is given in table 1.

Table 1. *ACTN3* genotype distribution of the athletes

Sex	n	Genotypes, %		
		RR	RX	XX
Male	14	48,3	85,8	7,1
Female	15	13,3	80	6,7
Total	29	10,3	82,8	6,9

Body composition characteristics were given in table 2.

Table 2. Body Composition characteristics in total sample

Characteristics	Mean $\pm$ SD
Height, cm	163,0 $\pm$ 7,6
Weight, kg	63,1 $\pm$ 10,2
BMI, $\text{kg}/\text{m}^2$	23,6 $\pm$ 2,5
Percent of body fat, %	15,7 $\pm$ 8,9

Table 3. the *ACTN3* genotype distribution of Kata and Kumite

Characteristics	n (%)	Genotypes, n (%)		
		RR	RX	XX
Kata	13 (44,8)	2 (7,1)	9 (85,8)	2 (7,1)
Kumite	16 (55,2)	1 (6,3)	15 (93,7)	0 (0)
Total	29	3 (10,3)	24 (82,8)	2 (6,9)

Our data about physical characteristics of Indonesian Karate Athletes are different to the ones observed by Nezhad and Farhadi (2012) in Iran National Karateka. The height and body mass in Indonesian Karate athletes are lower compared to the above-mentioned study. The difference may be due to the race, diet and effect of training. BMI ( $23,6 \pm 2,5 \text{ kg/m}^2$ ) and percentage of body fat ( $15,7 \pm 8,9 \%$ ) have been an ideal body composition. It is generally accepted that a lower relative body fat is desirable for successful competition in most of the sports, even in Karate.

Previous studies have shown highly significant association between *ACTN3* genotype and sprint/power performance, while the nonfunctional allele (577X) was believed to provide an advantage for endurance performance. The finding of complete absence of XX genotype in our study was low (6,9%). In Australian Caucasians and Asian (Japanese), XX genotype frequencies from 18 to 25% were established (Yang et al., 2007). The frequencies of XX genotype were observed very low (1 to 2%) in Kenyan, Jamaican and American sprinters (Scott et al., 2010). Low frequencies (11%) were reported in Ethiopian athletes (Yang et al., 2005).

*ACTN3* genotype is associated with human athletic performance (Yang et al., 2003). The RR genotype was predominant in the group of individuals practicing speed and power oriented sports (Holdys, 2011). In our research, RR and RX genotype of *ACTN3* gene were found in 27 athletes (93,1%). The R allele of *ACTN3* gene is considered an advantage for speed and power sports.

## CONCLUSIONS

The genotype distribution of *ACTN3* R577X gene were RR = 3 (10,3%), RX = 24 (82,8%), and XX = 2 (6,9%). No significant differences in body composition were found among genotypes. In conclusions, the data indicated that R allele was an advantage for success in Karate.

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