Association of mitochondrial DNA polymorphisms and/or haplogroups with elite Japanese athlete status

Noriyuki Fuku1*, Eri Mikami1-3 and Masashi Tanaka1

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A number of familial and twin studies have assessed the relative contribution of genetic and environmental factors to physical performance or its-related traits and have estimated that there is a significant genetic component to their phenotypes. In addition, aerobic capacity has been found to have stronger maternal inheritance than paternal. This finding implies that functional differences in maternally inherited mtDNA-encoded proteins involved in oxidative phosphorylation affect aerobic performance. In this article, therefore, we focus on associations between mtDNA polymorphisms/haplogroups and elite Japanese athlete status. From sequencing analysis of the control region in the mtDNA, certain mtDNA polymorphisms and haplogroups were shown to be associated with elite Japanese endurance athlete status, probably due to enhanced ATP production by mitochondria in the cardiac and skeletal muscles or both. This phenomenon is in agreement with several previous reports on Caucasian and African populations. It should be noted that certain mtDNA polymorphisms or haplogroups are also associated with elite Japanese sprint athlete/power status, probably due to enhanced calcium dynamics in the skeletal muscle. Thus, mtDNA polymorphisms/haplogroups influence not only aerobic performance but also anaerobic performance.

Keywords: mitochondrial DNA, haplogroup, polymorphism, athlete status, physical performance, endurance, sprint

Introduction

Background. Human physical performance is essentially multifactorial and is determined by a range of environmental (i.e., physical training, nutrition, and technological aids) and genetic factors. De Moor et al.1 studied 4488 British adult monozygotic and dizygotic female twins and estimated that the heritability of athletic status is 66%. It has also been reported that genetic factors are major determinants of physical performance-related traits such as maximal oxygen uptake (VO\text{2}max)2,3 and muscle strength4,5. The strong genetic contributions to physical performance and/or its-related traits indicate the possibility of using genetic approaches to individualize training approaches for enhancing competitive abilities in sports or even to help select appropriate athletic events through the use of genetic screening for extending the personal limit of athletes (i.e., talent identification). These possibilities provide the rationale and motivation for genetic studies of physical performance or its-related traits. To date, numerous studies have attempted to identify genetic polymorphisms that relate to physical performance and response to physical training. In 2009, over 200 genes in both nuclear DNA and mitochondrial DNA (mtDNA) have been reported to be associated with physical performance and health-related fitness6,7. The number of genes associated with physical performance-related phenotypes is expected to increase dramatically with the application of genome-wide methods to elite athlete cohorts; albeit such studies are only in their infancy8.

Mitochondria are essential to all higher organisms for sustaining life, and are extremely important in energy metabolism, providing 36 molecules of ATP per glucose molecule in contrast to the 2 ATP molecules produced by glycolysis. It is reasonable to hypothesize that mitochondria play an important role in setting aerobic performance, because mitochondria supply the majority of cellular ATP by the process of oxidative phosphorylation (OXPHOS), which is the main source of energy for endurance exercise. Although most DNA is packaged in chromosomes within the nucleus, mitochondria also possess their own circular DNA, designated as mtDNA. The 16,569-base pair (bp) human mtDNA contains 13 genes for mitochondrial OXPHOS, as well as 2 rRNA and 22 tRNA genes.

*Correspondence: nfuku@tmig.or.jp

1 Department of Genomics for Longevity and Health, Tokyo Metropolitan Institute of Gerontology, 35-2 Sakae-cho, Itabashi-ku, Tokyo 173-0015, Japan
2 Graduate School of Sport Sciences, Waseda University, 2-579-15 Mikajima, Tokorozawa, Saitama 359-1192, Japan
3 Japan Society for the Promotion of Science, 5-3-1 Kojimachi, Chiyoda-ku, Tokyo 102-0083, Japan

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Abstract A number of familial and twin studies have assessed the relative contribution of genetic and environmental factors to physical performance or its-related traits and have estimated that there is a significant genetic component to their phenotypes. In addition, aerobic capacity has been found to have stronger maternal inheritance than paternal. This finding implies that functional differences in maternally inherited mtDNA-encoded proteins involved in oxidative phosphorylation affect aerobic performance. In this article, therefore, we focus on associations between mtDNA polymorphisms/haplogroups and elite Japanese athlete status. From sequencing analysis of the control region in the mtDNA, certain mtDNA polymorphisms and haplogroups were shown to be associated with elite Japanese endurance athlete status, probably due to enhanced ATP production by mitochondria in the cardiac and skeletal muscles or both. This phenomenon is in agreement with several previous reports on Caucasian and African populations. It should be noted that certain mtDNA polymorphisms or haplogroups are also associated with elite Japanese sprint athlete/power status, probably due to enhanced calcium dynamics in the skeletal muscle. Thus, mtDNA polymorphisms/haplogroups influence not only aerobic performance but also anaerobic performance.

Keywords: mitochondrial DNA, haplogroup, polymorphism, athlete status, physical performance, endurance, sprint
that are necessary for protein synthesis within mitochondria\(^9,10\). Therefore, mtDNA diversity is likely to be involved in determining elite endurance athlete status or its-related phenotype such as VO\(_2\)max.

In this article, we discuss the polymorphisms and/or haplogroups of the mtDNA reported to date in association with elite athlete status and its-related traits, especially, in the Japanese population.

**Mitochondrial structure and mitochondrial genetics.** Mitochondria are membrane-enclosed organelles that are found in most eukaryotic cells. These organelles range from 0.5 to 1.0 μm in diameter. Mitochondria are sometimes described as “cellular power plants”, because they provide 36 molecules of adenosine triphosphate (ATP) per glucose molecule in contrast to the 2 ATP molecules produced by glycolysis. Mitochondria are also involved in other functions such as cell growth, cell differentiation, cell death, and so on. A mitochondrion contains inner and outer membranes composed of phospholipid bilayers (Fig. 1). These membranes create two compartments, i.e., matrix (large space) and inter-membrane space (small space) within the organelle, and they have different functions and structures. The inner membrane of the mitochondrion has numerous folds called cristae, which expand the surface area of the inner membrane and thus enhance the ability of the organelle to produce ATP. The matrix contains the enzymes that are associated with the citric acid cycle and beta-oxidation systems. The inner membrane is the site of all of the complexes of the OXPHOS system (i.e., electron transport chain and ATP synthase).

Although most DNA is packaged in chromosomes within the nucleus, mitochondria also have a small amount of their own circular DNA, which is called mtDNA. Each mitochondrion is estimated to contain 2-10 mtDNA copies. The mode of inheritance of the mtDNA is unique, because it is inherited maternally. The entire sequence of human mtDNA has been determined\(^9,10\), and the length of this mtDNA is 16,569 bp, sufficient for 13 messenger RNA (mRNA)-producing genes of the OXPHOS system, 2 ribosomal RNA (rRNA) genes, and 22 transfer RNA (tRNA) genes (Fig. 2-A). mtDNA and its encoded products are important regulators of aerobic ATP production via the mitochondrial OXPHOS system (Fig. 2-B). The mitochondrial electron-transfer chain is composed of 4 enzyme complexes (Complexes I-IV). Three of them (I, III, and IV) contain subunits encoded by mtDNA (Table 1). The other one, namely, Complex II, contains only nuclear DNA-encoded subunits. Mitochondrial ATP synthase (Complex V) comprises 2 subunits encoded by mtDNA (ATP6 and 8), as well as 10-16 subunits encoded by nuclear DNA. Mitochondrial cytochrome b (Cytb) is the only mtDNA-encoded subunit of respiratory Complex III (ubiquinol: ferrocytochrome c oxidoreductase or cytochrome bc1 complex).

mtDNA also contains major and minor non-coding regions\(^11\). The major non-coding region is located between the genes for tRNA-Phe and tRNA-Pro, and is called the control region. This region is about 1.1 kb long (m.16024-16569 plus m.1-576 = 1,122 bp) in humans and contains the main regulatory elements for mtDNA transcription and replication. This is also the region that is most variable in sequence and size among different species, although it contains several conserved elements with possible regulatory functions.

**Mitochondrial haplogroup.** The term ‘haplotype’ is a contraction of the term ‘haploid genotype’. In genetics, a haplotype is a combination of alleles at multiple loci that are transmitted together on the same chromosome.
Therefore, the hereditary mode of mtDNA is haplotypic because mtDNA is inherited maternally. A mitochondrial haplogroup is defined by the presence of a characteristic cluster of tightly linked mtDNA polymorphisms. In modern humans, maternally inherited substitutions in the mtDNA have resulted from the sequential addition of new mutations during the expansion of *Homo sapiens* from Africa to Asia and Europe over the last 200,000 years. Because the mutational rate of the mtDNA is 10 times higher than that of the nuclear DNA, the ancient mitochondrial polymorphisms that have accumulated during the relatively short history of *Homo sapiens* may have contributed to the metabolic characteristics of modern humans. Each of the African haplogroups (mainly, L0-L3), determined by mtDNA sequence analysis, has deeper genetic roots than those haplogroups in European and Asian populations. Mitochondrial haplogroup L3 is proposed to be the ancestor of all non-African populations. European haplogroups (H, I, J, K, S, T, U, V, W, etc.) belong to macrohaplogroup N, whereas Asian haplogroups belong to both macrohaplogroups N and M (haplogroups A, B, F, and N9a to macrohaplogroup N; and haplogroups M7a, M7b, M8, D, and G to macrohaplogroup M). Both macrohaplogroups N and M have haplogroup L3 as a common root (Fig. 3).

**Table 1.** Respiratory chain subunits encoded by mtDNA

<table>
<thead>
<tr>
<th>Complex</th>
<th>Enzyme</th>
<th>No. of subunits</th>
<th>mtDNA-encoded subunits</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>NADH reductase</td>
<td>43</td>
<td>7 (ND1, 2, 3, 4, 4L, 5, and 6)</td>
</tr>
<tr>
<td>II</td>
<td>Succinate reductase</td>
<td>4</td>
<td>-</td>
</tr>
<tr>
<td>III</td>
<td>Cytochrome c reductase</td>
<td>11</td>
<td>1 (Cytb)</td>
</tr>
<tr>
<td>IV</td>
<td>Cytochrome c oxidase</td>
<td>13</td>
<td>3 (COI, 2, and 3)</td>
</tr>
<tr>
<td>V</td>
<td>ATP synthase</td>
<td>17</td>
<td>2 (ATP6, and 8)</td>
</tr>
</tbody>
</table>

**Fig. 2** The human mitochondrial DNA (A) and mitochondrial oxidative phosphorylation system (B). Human mtDNA encodes 13 polypeptides: 7 subunits shown in orange (ND1, ND2, ND3, ND4, ND4L, ND5, and ND6) of Complex I; 1 subunit shown in green (cytochrome b) of Complex III, 3 subunits shown in pink (COI, COII, and COIII) of Complex IV, and 2 subunits (ATP6 and ATP8) of Complex V. mtDNA: mitochondrial DNA; ND: NADH dehydrogenase; ND1: NADH dehydrogenase subunit 1; CO: Cytochrome c oxidase; ATP: ATP synthase; 12S: 12S rRNA; 16S: 16S rRNA.
Heritability of physical performance

Heritability is defined as the additive proportion of observed total phenotypic variation in a particular trait (e.g., physical performance) that can be attributed to inherited genetic factors in contrast to environmental ones. In genetics, it is the relative importance of genetics versus the environment that determines the phenotype. The values of heritability estimates can range from 0 (0%) to 1 (100%). This heritability is a function of genetic and environmental factors, and the interaction between them; e.g., monozygotic traits usually have a high heritability estimate (almost 100%), whereas only a relatively smaller proportion of multifactorial disorders can be explained by genetic factors alone.

The topic of the role of genetic and environmental factors in the manifestation of physical performance-related phenotypes, such as $V\cdot O_2$max, was first addressed by Klissouras in 1971. He showed that the $V\cdot O_2$max of monozygotic twins is more alike than that of dizygotic twins, and concluded a heritability estimate of 91% of the total phenotype diversity, although the sample size of his study was small. Subsequent examination of moderate or large sample sizes has indicated a smaller effect of heritability estimates on $V\cdot O_2$max. Among large-scale studies, Sundet et al. reported that the heritability estimate accounts for approximately 60% of maximum aerobic power based on data obtained from 436 monozygotic and 622 dizygotic male twin pairs registered with the Norwegian Army Draft Board. Similarly, Bouchard et al. reported that the heritability estimate of $V\cdot O_2$max in the sedentary state, adjusted for age, sex, and body mass, is approximately 50% based on data from 429 individuals from 86 nuclear families in the HERITAGE Family Study. Relatively recently, De Moor et al. provided indirect support for these observations in their study on 4488 adult female twins from the TwinsUK Adult Registry, which employed the first genome-wide linkage scan for sports. They estimated the heritability of athlete status at 66%. This proportion of genetic factors regarding athlete status seems to be reasonable in terms of the heritability estimates for aerobic performance from large cohorts as mentioned above (approximately 50-60%).

Interestingly, the data on $V\cdot O_2$max in the HERITAGE Family Study, mentioned above, also indicated that maternal influence, i.e., mitochondrial inheritance, accounts for as much as 30% of familial transmission, in which inheritance constitutes well more than half of the genetic factors. Lesage et al. also reported stronger maternal inheritance (than paternal) for $V\cdot O_2$max in their familial study. These results suggest a possible mitochondrial genetic component active in athletic performance.

Association study on physical performance

Restriction fragment length polymorphisms. RFLP (restriction fragment length polymorphism) is an old technique (but still useful) for detecting variation in DNA sequences. A restriction enzyme cuts double-stranded DNA at a specific sequence, which is usually from 4 to 6 base pairs in length. The restricted DNA fragments, i.e., polymorphisms, are usually separated by agarose gel electrophoresis. Analysis of DNA variation by RFLP has been an important tool for genome mapping, identification of disease-related genes, genetic fingerprinting, and so on.

The first report to address the association between mtDNA variations and a physical performance-related trait ($V\cdot O_2$max) was made by Dionne et al. They assessed the relationship between mtDNA RFLPs and baseline $V\cdot O_2$max, and its response to 20-week endurance training, by examining 15 mtDNA RFLPs generated by 22 restriction enzymes in 46 sedentary North Americans of Quebec City. These 22 RFLP sites had been generated by base substitutions located in protein-coding regions as well as in non-coding regions. They found that carriers of 3 mtDNA RFLPs, 2 in the ND5 gene and 1 in the tRNA-Thr...
gene, have a VO₂max in the untrained state significantly higher than the non-carriers; whereas carriers of 1 mtDNA RFLP in the ND2 gene have a lower initial VO₂max. Carriers of 3 RFLPs in the ND5 gene are also associated with a lower VO₂max. Subsequently, Rivera et al. 24) examined 125 Caucasian male elite endurance athletes and 65 sedentary controls for a possible association between elite endurance athlete status and 4 mtDNA RFLPs (3 in the ND5 and 1 in the control region), which were previously reported to be associated with VO₂max as mentioned above 25). They found no significant associations between mtDNA RFLPs and elite endurance athlete status. In the 1990’s, RFLP technology was generally considered the gold standard for genotyping, etc. Although it has been the most accurate method for gene typing, it has some drawbacks, namely, sample size, sample quality, specific restriction enzyme (cannot be used for all sequence variations), and the length of time for analysis. Currently, the RFLP method is becoming less used for genotyping due to the rise of inexpensive DNA sequencing technologies.

Mitochondrial haplogroups. As mentioned above, a mitochondrial haplogroup is defined by the presence of a characteristic cluster of tightly linked mtDNA polymorphisms, namely, a set of mtDNA polymorphisms (See section 1-3). The matrilineal inheritance of mtDNA and linear accumulation of polymorphisms have allowed the construction of detailed mtDNA phylogenies 26). These phylogenies display the variation and diversity of human mtDNA and allow haplogroup identification through the analysis of a small number of haplogroup-specific polymorphisms, i.e., polymorphisms detected in the hypervariable segment 1 (HVS1: approximately 350 base pairs) of the mtDNA control region, which is a mutational hotspot 27). This matrilineal pattern of descent means that individual haplotypes share linked complexes of polymorphisms common to all sequences in a haplogroup.

We sequenced HVS1 and the C>A polymorphism at m.5178 within the ND2 gene of the mtDNA by use of an automated DNA sequencing machine from Applied Biosystems, and compared the percent frequency of mitochondrial haplogroups found in 141 Japanese Olympians, from various sports, with 672 Japanese controls 27). These athletes were classified as endurance/middle-power athletes (EMA=81) or sprint/power athletes (SPA=60). Subjects were classified into 12 major Japanese mitochondrial haplogroups (i.e., F, B, A, N9a, N9b, M7a, M7b, M*, G2, G1, D5 or D4) on the basis of the presence of HVS1 polymorphisms and several protein-coding region polymorphisms. The distribution of mitochondrial haplogroups in EMA and SPA, relative to that in the controls, is shown in Fig. 4. When the frequency of each haplogroup versus the sum of all others was compared between EMA or SPA and controls, we found that EMA displays an excess of haplogroup G1 (OR 2.52 [95% CI 1.05-6.02], P=0.032; Fig. 4), with 8.9% in the EMA group compared with 3.7% in the control. On the other hand, SPA shows a greater proportion of haplogroup F (OR 2.79 [95% CI 1.28-6.07], P=0.007; Fig. 4), with 15.0% in the SPA group relative to 6.0% in the control.

Our previous reports also suggested that certain mitochondrial haplogroups are associated with elite Kenyan endurance athlete status 28) and Jamaican/African-American sprint athlete status 29). Elite Kenyan endurance athletes display an excess of haplogroup L0 and a dearth of haplogroup L3 relative to the general Kenyan popula-

![Fig. 4](image-url)
Obesity.

We have proposed a hypothesis to explain this inconsistency (Fig. 5). The main function of mitochondria is to produce ATP by OXPHOS; and while the uncoupling of mitochondrial OXPHOS generates heat, it concomitantly reduces the production of ATP. Conversely, more tightly coupled OXPHOS would be expected to decrease heat production and result in higher efficiency of ATP production. This improved efficiency in ATP production could explain, in part at least, the association between haplogroup G1 and endurance performance. However, this energy conservation by mitochondrial OXPHOS may predispose to obesity in sedentary individuals later on in life - a phenomenon commonly referred to as the “thrifty” genotype and/or “thrifty” phenotype. These hypotheses require further investigation.

2. Sprint/power performance-related haplogroup F:

Although numerous studies have reported associations between aerobic performance phenotypes and mitochondrial haplogroups, studies on associations between “anaerobic” performance phenotypes and mitochondrial haplogroups are lacking. Because “anaerobic” capacity relies more heavily upon glycolysis than upon mitochondrial OXPHOS, it is not commonly believed that certain mtDNA polymorphisms and/or mitochondrial haplogroups are related to anaerobic capacity. However, it should be noted that we found a positive association between mitochondrial haplogroup F and elite Japanese SPA status. We also examined the effect of mitochondrial haplogroups on anaerobic performance phenotypes such as muscle power, muscle strength, and muscle mass in 480 healthy Japanese non-athlete adults, and found that macrohaplogroup N is significantly associated with stronger leg extension power and higher vertical jump performance compared

1. Endurance performance-related haplogroup G1:

Among the mtDNA polymorphisms characteristic of haplogroup G1, the m.15497G>A transition, causing Gly251Ser replacement in the Cytb gene, seems to be a functional polymorphism. A previous survey, conducted as part of the National Institute for Longevity Longitudinal Study on Aging, revealed that this m.15497G>A transition is associated with obesity in the middle aged-to-elderly Japanese population. This observation may indicate increased efficiency of mitochondrial energy conservation at the cytochrome bc1 complex, resulting in decreased energy consumption. As mentioned above, we recently reported that this obesity-associated mitochondrial haplogroup, namely, haplogroup G1, is also associated with elite Japanese EMA status. This finding seems to be inconsistent with the association between G1 and obesity.

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with macrohaplogroup M for these Japanese subjects. Mitochondrial haplogroup F, which is associated with elite Japanese SPA status, is one of the major components of macrohaplogroup N, which is associated with muscle power in non-athletic Japanese individuals. Although we could not demonstrate a significant association between mitochondrial haplogroup F and anaerobic performance phenotypes in the 480 non-athletic individuals, both leg extension power (18.3 ± 1 vs. 17.3 ± 0.3 watts/kg body weight) and vertical jump performance (38.2 ± 1.9 vs. 36.5 ± 0.5 cm) tended to be higher in subjects with mitochondrial haplogroup F than in those with other haplogroups.

Previous studies also focused on the roles of mitochondrial haplogroup F and anaerobic performance phenotypes in the 480 non-athletic individuals, both leg extension power (18.3 ± 1 vs. 17.3 ± 0.3 watts/kg body weight) and vertical jump performance (38.2 ± 1.9 vs. 36.5 ± 0.5 cm) tended to be higher in subjects with mitochondrial haplogroup F than in those with other haplogroups.

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For SPA status m.152T>C T 95.1 (639) 98.0 (98) 0.192 0.40 (0.09 to 1.67) 88.2 (75) 0.010 2.58 (1.22 to 5.45) m.16278C>T C 91.7 (616) 90.0 (90) 0.578 1.22 (0.60 to 2.48) 100.0 (85) 0.006 0.00 - T 8.3 (56) 10.0 (10) 0.0 0.0 0.0 0.0 (0)

CON: Controls; EMA: endurance/middle-power athletes; SPA: sprint/power athlete; OR: odds ratio; CI: confidence intervals.

**Polymorphisms in the control region.** We have already stated that mtDNA contains 37 genes, i.e., 13 mRNAs, 22 tRNAs, and 2 rRNAs. The mtDNA also contains major non-coding regions, which include the control region or D-loop. The control region of mtDNA contains the heavy strand origin of replication, the promoters for heavy (H) and light (L) strand transcription, and the binding site of mitochondrial transcriptional factor A (TFAM). Therefore, it is likely that the polymorphisms in these functional regions of mtDNA cause a difference in the sensitivity of this TFAM binding site. Thus, the control region may contain important polymorphisms influencing physical performance.

To examine this hypothesis, we sequenced the entire control region of mtDNA by use of an automated DNA sequencing machine from Applied Biosystems, and compared the percentage frequencies of polymorphisms found in 185 Japanese international athletes from various sports, mainly track & field, with those found in 672 Japanese controls. The subjects were comprised of 185 elite Japanese athletes who had represented Japan at international competitions (i.e., 100 EMA and 85 SPA) and 672 Japanese controls. Table 2 displays the results of all polymorphisms that reached statistical significance (P<0.05) when the frequencies of polymorphisms were compared between the athletes and controls. The EMA group displayed an excess of three polymorphisms [m.152T>C, m.514(CA)n repeat (n≥5), and poly-C stretch at m.568-573 (C≥7)] compared to the controls. On the other hand, the SPA group showed a greater frequency of the m.204T>C polymorphism compared to the controls. In addition, none of the SPA individuals had m.16278C>T polymorphism, whereas the frequencies of this polymorphism in controls and EMA were 8.3% and 10.0%, respectively. These findings imply that several polymorphisms detected in the control region of mtDNA may influence physical performance, probably in a functional manner.

**Table 2.** Polymorphisms in the mtDNA control region associated with SPA or EMA status

<table>
<thead>
<tr>
<th>Polymorphism</th>
<th>CON (n=672)</th>
<th>EMA (n=100)</th>
<th>SPA (n=85)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>% (n)</td>
<td>% (n)</td>
<td>P value</td>
</tr>
<tr>
<td>For EMA status m.152T&gt;C</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>T</td>
<td>81.5 (548)</td>
<td>72.0 (72)</td>
<td><strong>0.025</strong></td>
</tr>
<tr>
<td>C</td>
<td>18.5 (124)</td>
<td>28.0 (28)</td>
<td></td>
</tr>
<tr>
<td>m.514(CA)n (CA)3 or (CA)5</td>
<td>40.6 (273)</td>
<td>27.0 (27)</td>
<td><strong>0.009</strong></td>
</tr>
<tr>
<td>Poly-C stretch at m.568-573 (C≥7)</td>
<td>96.0 (645)</td>
<td>89.0 (89)</td>
<td><strong>0.003</strong></td>
</tr>
<tr>
<td>(C)6</td>
<td>4.0 (27)</td>
<td>11.0 (11)</td>
<td></td>
</tr>
<tr>
<td>For SPA status m.204T&gt;C</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>T</td>
<td>95.1 (639)</td>
<td>98.0 (98)</td>
<td>0.192</td>
</tr>
<tr>
<td>C</td>
<td>4.9 (33)</td>
<td>2.0 (2)</td>
<td></td>
</tr>
<tr>
<td>m.16278C&gt;T</td>
<td>91.7 (616)</td>
<td>90.0 (90)</td>
<td>0.578</td>
</tr>
<tr>
<td>T</td>
<td>8.3 (56)</td>
<td>10.0 (10)</td>
<td></td>
</tr>
</tbody>
</table>
1. Endurance performance-related m.152T>C polymorphism

The m.152T>C polymorphism is located next to the second heavy-strand replication origin (m.146-151). Although there is no direct evidence explaining an association between endurance performance and m.152T>C polymorphism of the mtDNA control region, some interesting findings have been reported to explain this association. Fish et al. suggested that the nucleotide position m.151 in the mtDNA is important for accelerating mtDNA replication in response to physiological demands. Another polymorphism, the m.150C>T polymorphism adjacent to m.151, is reportedly associated with longevity and remodeling of the replication-origin site. We previously reported that mitochondrial haplogroups N9a, G1, and D5 are associated with resistance against the metabolic syndrome in Japanese women, and that haplogroup N9a is also associated with resistance against type 2 diabetes mellitus in an Asian population. Interestingly, all of these haplogroups are accompanied by the 150C>T polymorphism. These findings suggest the importance of the region around the heavy-strand replication-origin site, and consequently allow the prediction that polymorphisms near this site are functional to some extent. The m.152T>C polymorphism, which is associated with endurance performance, seems to be one of the potentially functional polymorphisms influencing mtDNA replication and/or mitochondria biogenesis.

2. Endurance performance-related m.514(CA)n length polymorphism

Another EMA status-related polymorphism, i.e., the m.514(CA)n repeat, displays variation in length. The frequency of m.514(CA)≤4 is significantly lower in EMA individuals than in controls, whereas that of m.514(CA)≥5 is higher in the former than in the latter. It should be noted that Murakami et al. reported that the mtDNA content in the vastus lateralis muscle is higher in healthy sedentary Japanese males with m.514(CA)≥5 than in those with m.514(CA)≤4 (Fig. 6). mtDNA content is significantly associated with citrate synthase activity and VO2peak. This apparent link between the long m.514(CA)n repeat (n≥5) in the mtDNA control region and enhanced aerobic capacity, due to an increase in mtDNA content in the skeletal muscle (or cardiac muscle), may explain why this polymorphism is associated with elite EMA status.

3. Sprint/power performance-related m.204T>C and m.16278C>T polymorphisms

The nucleotide position at m.204 is located 9 bp upstream of the conserved sequence block 1 (CSB1: m.213-235), a region associated with RNA priming of mtDNA replication. On the other hand, the nucleotide position at m.16278 is located in the 7S DNA (m.16106-191), which is also called the short single-stranded DNA fragment, a region associated with mtDNA copy number. As shown in Table 2, the frequencies of m.204T>C and m.16278C>T polymorphisms in SPA and EMA deviated in opposite directions from the controls (P=0.013 and P=0.003, respectively). Opposite effects on elite SPA and EMA status of different genotypes at a single locus have also been observed in studies on the genes encoding angiotensin-converting enzyme (ACE), α-actinin-3 (ACTN3), and peroxisome proliferator-activated receptor-alpha (PPARA). These genes and/or genotype regulate, at least in part, muscle properties such as fiber-type composition and metabolic dynamics in the skeletal muscle.

Sprint/power athletes obviously require a proliferation of fast-twitch muscle fibers, and 70-80% of the skeletal muscle fibers in elite-level SPA individuals are fast-twitch muscle fibers. Sprint performance also relies more on anaerobic ATP production by glycolysis than on aerobic ATP production by mitochondrial OXPHOS. Possible mechanisms underlying the results presented here explain why the mtDNA control region polymorphisms are associated with elite-status Japanese athletes whether they are of EMA or SPA status. Mitochondria play an important role in determining muscle fiber-type composition, because it was previously reported that PPAR gamma coactivator 1-alpha (PPARGC1A) and/or PPAR-delta (PPARD), which are regulators of mitochondrial biogenesis, drive the ratio of slow/fast-twitch muscle fibers. If certain control-region polymorphisms, e.g., m.204T>C or m.16278C>T, in mtDNA down- or up-regulate mtDNA replication and/or transcription by PPARs or a variation, muscle composition can be changed to fast- or slow-twitch muscle fiber. In addition, decreased ATP generation from mitochondrial OXPHOS, due to mitochondrial dysfunction by mtDNA mutation, induces compensatory up-regulation of the cytoplasmic glycolysis process, which is called the Warburg effect. Therefore, decreased mitochondrial function contributes to enhanced capacity.
for generation of ATP by glycolysis. This effect can, at least in part, explain why athletes with certain polymorphisms in the mtDNA control region associated with mitochondrial function, such as mtDNA replication and/or transcription, have an advantage or disadvantage in terms of sprint/power performance.

Conclusions and future directions

In this article, we mainly discussed the potential roles of mtDNA polymorphisms and haplogroups for determining elite Japanese athlete status. It is well known that certain mtDNA polymorphisms and/or haplogroups are associated with elite endurance athlete status, as we and other researchers previously reported, because proteins of the mitochondrial OXPHOS system, some of which are encoded by mtDNA, play an important role in aerobic ATP production, especially in the skeletal muscle. On the other hand, it has not been commonly believed that certain mtDNA polymorphisms and/or haplogroups are related to “anaerobic” capacity, because “anaerobic” capacity relies more heavily upon glycolysis than upon mitochondrial OXPHOS. However, we have provided the first evidence that certain mtDNA polymorphisms and/or haplogroups are not only associated with endurance performance but also with sprint/power performance. This finding, namely, an association between mitochondrial haplogroup and sprint/power performance, has also been replicated in African-American sprinters. In addition, we reported an association between mitochondrial haplogroup and muscle power in non-athletic individuals. This may sound strange, but it appears reasonable to accept this phenomenon because mitochondria are involved in the regulation of energy metabolism as well as intracellular Ca²⁺ concentration, which is a trigger of muscle contraction. Further extensive studies are necessary to understand the functional link between mtDNA polymorphisms/haplogroups and sprint performance.

While it is a well-known fact that environmental factors such as training and diet affect athletic performance, genetic factors seem to play an important role in athletic performance as well. Hopkins⁴⁰, in 2001, pointed out that “If you want your kids to be great athletes, marry a great athlete”. Although he had a radical view of sports at that time, his opinion is not incorrect now. For instance, De Moor et al.¹ reported the heritability estimate of athletic status to be 66%. However, there is not enough evidence to explain, in detail, the relationship between genetic factors and elite athlete status. Current studies on athletic performance-associated polymorphisms (PAPs), which were previously mostly published from candidate gene-association studies⁴⁵-⁶⁰, have elucidated the polygenic profile for determining athletic performance; but there is still not enough evidence to explain the genetic component. Further investigation in the area of sports science will focus on more detailed analysis of genetic polymorphisms detected in both mtDNA and nuclear DNA, e.g., whole genome sequencing for mtDNA and genome-wide association studies (GWAS) for nuclear DNA⁷⁰, even though GWAS may not be able to find genetic polymorphisms with a large effect that explain the overall genetic factors predicted by heritability estimates⁷¹.

References


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