Regulation of skeletal muscle ATP catabolism by AMPD1 genotype during sprint exercise in asymptomatic subjects

BARBARA NORMAN,1 RICHARD L. SABINA,2 AND EVA JANSSON1
1Karolinska Institute, Department of Medical Laboratory Sciences and Technology, Division of Clinical Physiology, Huddinge University Hospital, 141 86 Stockholm, Sweden; and 2The Medical College of Wisconsin, Department of Biochemistry, Milwaukee, Wisconsin 53226

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Norman, Barbara, Richard L. Sabina, and Eva Jansson. Regulation of skeletal muscle ATP catabolism by AMPD1 genotype during sprint exercise in asymptomatic subjects. J Appl Physiol 91: 258–264, 2001.—Deficiency of myoadenylate deaminase, the muscle isoform of AMP deaminase encoded by the AMPD1 gene, is a common myopathic condition associated with alterations in skeletal muscle energy metabolism. However, recent studies have demonstrated that most individuals harboring this genetic abnormality are asymptomatic. Therefore, 18 healthy subjects with different AMPD1 genotypes were studied during a 30-s Wingate test in order to evaluate the influence of this inherited defect in AMPD1 expression on skeletal muscle energy metabolism and exercise performance in the asymptomatic population. Exercise performances were similar across the AMPD1 genotypes, whereas significant differences in several descriptors of energy metabolism were observed. Normal homozygotes (NN) exhibited the highest levels of AMP deaminase activities, net ATP catabolism, and IMP accumulation, whereas intermediate values were observed in heterozygotes (MN). Conversely, mutant homozygotes (MM) had very low AMP deaminase activities and showed no significant net catabolism of ATP or IMP accumulation. Accordingly, MM also did not show any postexercise increase in plasma ammonia. Unexpectedly, MN consistently exhibited greater increases in plasma ammonia compared with NN despite the relatively lower accumulation of IMP in skeletal muscle. Moreover, time course profiles of postexercise plasma ammonia and blood lactate accumulation also differed across AMPD1 genotypes. Finally, analysis of adenosine in leftover biopsy material revealed a modest twofold increase in MN and a dramatic 25-fold increase in MM.

myoadenylate deaminase; adenine nucleotides; inosine monophosphate; adenosine; ammonia

Hydrolysis of ATP provides the energy that drives skeletal muscle contraction and represents a well-studied focal point that links exercise physiology and biochemistry. For example, several studies have shown that ATP content of exercising muscle can be depleted by ~40% during short-term high-intensity exercise (5, 17, 37). Net breakdown of ATP results in an equimolar formation of inosine monophosphate (IMP) (5, 17, 37), the latter produced by deamination of AMP in a reaction catalyzed by AMP deaminase (E.C. 3.5.4.6). The skeletal muscle isoform of AMP deaminase (myoadenylate deaminase) is activated during short-term, high-intensity exercise when the rate of ATP utilization exceeds the potential of the cell to resynthesize ATP.

Approximately 2% of the general Caucasian population is homozygous, and nearly 20% heterozygous, for a prevalent mutation in the AMPD1 gene that encodes myoadenylate deaminase (24, 25, 41). The encoded polypeptide product of the mutant sequence is severely truncated and catalytically inactive (24). Consequently, homozygotes for the mutant AMPD1 allele have extremely low skeletal muscle AMP deaminase activity whereas heterozygotes and normal homozygotes have intermediate and high enzyme activities, respectively (28). Reflecting its prevalence in the general population, several investigators have also reported myoadenylate deaminase deficiency in ~2% of skeletal muscle biopsies submitted for pathological evaluation (for review, see Ref. 30). Approximately one-half of this subset of the myoadenylate deaminase deficient population has associated exercise-related symptoms such as muscle cramps, pain, and early fatigue (9, 19, 20). Nevertheless, the combination of high incidence and associated clinical variability has prompted some investigators to refer to myoadenylate deaminase deficiency simply as a “harmless genetic variant” (41) and “the bane of clinical specialists” (42).

Clinical heterogeneity notwithstanding, a rationale for reduced exercise capacity in connection with myoadenylate deaminase deficiency may be developed based on the premise of accentuated ADP (and AMP) accumulation during fatiguing exercise (for an extended discussion, see Ref. 31). Normally, the AMP deaminase reaction should minimize ADP accumulation in skeletal myocytes during short-term high-intensity exercise by removing AMP and displacing equilibrium of the myokinase reaction (2 ADP ↔ ATP + AMP) in the direction of ATP formation. This process is believed to prevent the inhibitory effect exerted by a decrease in the ATP-to-ADP ratio on the contraction

Address for reprint requests and other correspondence: B. Norman, Clinical Physiology, Huddinge Univ. Hospital, 141 86 Stockholm, Sweden (E-mail: barbara.norman@labtek.ki.se).

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These individuals exhibit no symptoms in everyday life. Furthermore, increases in ADP have been shown to reduce maximal shortening velocity of contracting skeletal muscle (6, 44), which is one characteristic component of muscle fatigue.

We have used this combined information to reason that myoadenylate deaminase deficiency could lead to an earlier decline in exercise performance during conditions of high ATP turnover rates. Therefore, in the present study we applied short-term high-intensity exercise (30-s Wingate test), which has been shown to lead to a pronounced activation of AMP deaminase (4, 8, 12), to investigate the effect of AMPD1 genotype on adenylate catabolism and performance in healthy subjects. The hypotheses to be tested were that 1) a mutation in the AMPD1 gene would lead to attenuated net breakdown of ATP, and corresponding IMP accumulation, and 2) greatly diminished net adenylate catabolism, and thereby greater ADP accumulation, in individuals who are homozygotes for the mutation might result in reduced anaerobic performance, even though these individuals exhibit no symptoms in everyday life.

MATERIALS AND METHODS

Subjects. Fresh whole blood samples were collected from 175 healthy, physically active subjects for extraction of genomic DNA (15). Genotype analysis was performed as described previously (28) and was based on the previously identified AMPD1 mutant allele involving a C34T transition in the AMPD1 open reading frame (24). Eighteen subjects with different AMPD1 genotypes were selected and asked to participate in the present study. Seven (one man and six women) were homozygotes for the normal AMPD1 allele (NN), seven (one man and six women) were heterozygotes and three (one man and two women) were homozygotes for the mutant allele (MM). One female subject with extremely low AMP deaminase activity was found to be heterozygous for the common C34T transition. This individual was included in the mutant homozygote group after she was subsequently shown to be a compound heterozygote harboring a second rare mutation in the AMPD1 gene (WN fishbein, unpublished observation).

The age, height, and body weight (mean and range) of the men were 23 (20–27) yr, 181 (173–194) cm, and 81 (70–89) kg, respectively, and those of the women were 24 (20–32) yr, 168 (158–182) cm and 66 (54–81) kg, respectively. The subjects completed a questionnaire regarding their health and physical activity habits. All considered themselves healthy and none expressed complaints of exercise-induced muscle cramps or myalgias. Physical activity level was assessed by a physical activity index, peak power, and mean power data as dependent variables to examine dependent variable differences among the three different genotype groups.

Experimental protocol. Subjects were instructed not to perform any exhaustive exercise on the preceding day and to abstain from caffeine and nicotine for at least 2 h before reporting to the laboratory after a light breakfast on the day of exercise. The subjects assumed a supine position in the laboratory for 15–20 min, after which blood samples were collected from an antecubital vein, and a percutaneous muscle biopsy sample (3) was taken from the m. quadriceps femoris vastus lateralis (preexercise biopsy). Subjects then moved to a cycle ergometer and performed a 30-s sprint at maximal speed against a resistance of 7.5% of the subjects body weight (Wingate test) (1). Average power output for 5-s periods was automatically registered during exercise. Peak power (the highest 5-s power output) and mean power (the average power output during the 30-s duration of the sprint) were calculated and related to body mass. Immediately after exercise, a second muscle biopsy (postexercise biopsy) was taken from the same incision as the preexercise biopsy. The postexercise biopsies were taken within ~10 s from the cessation of exercise and frozen within a further 5 s. Blood samples were obtained at 3, 6, and 9 min after exercise with the subjects resting in supine position.

Muscle and blood treatment and analysis. Muscle biopsy samples were frozen in isopentane precooled with liquid nitrogen immediately on sampling and stored at −70°C until analyses. Transverse 10-μm sections were cut from the frozen biopsy samples obtained at rest (preexercise), and type I and II myofibers were identified by myofibrillar ATPase stain (36). The remaining part of the preexercise and postexercise biopsy samples were freeze-dried and dissected free from visible connective tissue and blood under a dissection microscope. One portion was homogenized in 0.1 M phosphate buffer, pH 7.7, and used for the analysis of AMP deaminase activity by HPLC (27). A second portion was extracted in 0.4 M perchloric acid, neutralized, and used for the analysis of ATP, ADP, AMP, IMP, inosine, creatine (Cr), and creatine phosphate (CP) by HPLC (26), and lactate by a fluorometric technique (22). Muscle adenosine content was analyzed by HPLC (40) in leftover biopsy material from four heterozygotes and three mutant homozygotes.

Blood samples for determination of plasma ammonia were centrifuged immediately, and the supernatants were frozen in liquid nitrogen and stored at −70°C until analysis by a flow injection technique (38). Blood lactate was analyzed in neutralized perchloric acid extracts of whole blood by a fluorometric enzymatic method (22).

Statistics. Factorial analysis of variance (ANOVA) was performed with AMPD1 genotype as an independent variable and AMP deaminase activity, fiber type composition, training index, peak power, and mean power data as dependent variables to examine dependent variable differences among the three different genotype groups.

Repeated measures analysis of covariance (ANCOVA) with AMPD1 genotype as an independent variable and AMP deaminase activity, fiber type composition, training index, peak power, and mean power data as dependent variables to examine dependent variable differences among the three different genotype groups. The percentage of type I fibers was included as a covariate because fiber type composition of the muscle can have a pronounced effect on metabolism depending on the specific type, intensity and duration of exercise. The same ANCOVA design was used to analyze power decrease during exercise with registrations of power output at 5, 10, 15, 20, 25, and 30 s of cycling. When a significant interaction term was found in the ANCOVA, Scheffe’s post hoc test was used to evaluate differences between the groups.

Repeated-measures ANOVA with AMPD1 genotype as an independent variable and time (before and after exercise) as a dependent variable was used to analyze exercise induced changes in metabolites in the three different genotype groups. The percentage of type I fibers was included as a covariate because fiber type composition of the muscle can have a pronounced effect on metabolism depending on the specific type, intensity and duration of exercise. The same ANCOVA design was used to analyze power decrease during exercise with registrations of power output at 5, 10, 15, 20, 25, and 30 s of cycling. When a significant interaction term was found in the ANCOVA, Scheffe’s post hoc test was used to evaluate differences between the groups.

Repeated-measures ANOVA with AMPD1 genotype as an independent variable and time (before and after exercise) as a dependent variable was applied to analyze the time course for accumulation of blood lactate and plasma ammonia in the different genotypes. When significant interaction term was found in the ANOVA the changes, in blood lactate and plasma ammonia concentrations, at selected points of time were analyzed using Student paired t-test and compared for the different genotypes. P values
were corrected for multiple statistical comparisons according to Holm (16).
Statistical significance was accepted at \( P < 0.05 \). Values are presented as means ± SD.

RESULTS

AMP deaminase activity, fiber-type composition, training index, and power data for different AMPD1 genotype groups are presented in Table 1. There is a pronounced difference in AMP deaminase activities, with ranges for normal homozygotes 1,010–2,169 mmol/kg dry muscle·min\(^{-1}\), for heterozygotes 337–632 mmol·kg\(^{-1}\)·dry muscle·min\(^{-1}\), and for mutant homozygotes 4 –14 mmol·kg\(^{-1}\)·dry muscle·min\(^{-1}\). One female subject who is a compound heterozygote for the common C34T transition and a second rare mutation (see MATERIALS AND METHODS) also has a low AMP deaminase activity (4 mmol·kg\(^{-1}\)·dry muscle·min\(^{-1}\)) and is included in the mutant homozygote subset of individuals. Conversely, there is no significant difference in fiber-type distribution, training index, peak power or mean power during the 30-s Wingate test between the three genotype groups. Power output profiles for the three different genotype groups during the 30-s Wingate test are presented in Fig. 1.

Skeletal muscle metabolite pools before and after exercise and the statistical evaluation of these results are presented in Table 2. Sprint exercise decreases the muscle content of ATP by 40% (decrease of 8.5 mmol/kg dry muscle), in normal homozygotes and somewhat less, 28% (decrease of 6 mmol/kg dry muscle), in heterozygotes. No significant ATP decrease is detected in the mutant homozygotes (genotype group). Consequently, no inosine is detected in the muscle sample. The decrease in ATP is closely matched by a rise in IMP, which is 9.4 mmol/kg dry muscle in normal homozygotes and 6.3 mmol/kg dry muscle in heterozygotes. As expected, there is no significant accumulation of IMP in the mutant homozygotes (genotype × time, \( P < 0.001 \)). An exercise-induced decline in ADP is observed in all three genotype groups (\( P = 0.001 \)). AMP does not change significantly with exercise, but tendencies for higher AMP levels are observed in both pre- and postexercise biopsies of mutant homozygotes (\( P = 0.056 \)). However, the approximate 15-s lapse from the end of exercise until freezing of the muscle sample is sufficient to allow transient changes in AMP and ADP to reequilibrate. Furthermore, the free, active fractions of ADP and AMP are not distinguishable from total amounts of these metabolites measured in muscle extracts. Consequently, the use of muscle biopsy extracts for quantitation of ADP and AMP pools may not accurately reflect the in vivo levels of these metabolites. Future studies involving myoadenylate deaminase-deficient subjects could employ \(^{31}\)P magnetic resonance spectroscopy in an attempt to resolve the technical limitations in measuring ADP pools inherent to the biopsy method.

Total adenine nucleotide content (TAN) plus IMP does not change significantly with exercise in any genotype group. Consequently, no inosine is detected in pre- or postexercise biopsies (detection limit for inosine is 0.05 mmol/kg dry muscle).

Muscle lactate content increases and CP content decreases during exercise, with no difference between the genotypes. The decrease in CP is matched by a corresponding increase in creatine, and the total creatine pool does not change during exercise.

Because of lack of biopsy material, muscle adenosine content was analyzed only in a subset of subjects. Figure 2 shows increased muscle adenosine levels after exercise in two of four heterozygotes (ranges are 0–13 and 8–18 \( \mu \)mol/kg dry muscle before and after exercise, respectively) and in all three mutant homozygotes (ranges are 0–19 and 55–308 \( \mu \)mol/kg dry muscle before and after exercise, respectively) examined, with much higher accumulations in the latter group.

Venous blood lactate and plasma ammonia concentrations before and after 3, 6, and 9 min of sprint

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Table 1. AMP deaminase activity, fiber type composition, training index, and performance data for the Wingate cycling; peak and mean power in NN, MN, and MM and statistical evaluation of differences between genotypes

<table>
<thead>
<tr>
<th></th>
<th>NN (n = 7)</th>
<th>NM (n = 7)</th>
<th>MM (n = 4)</th>
<th>ANOVA P Values</th>
</tr>
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<tbody>
<tr>
<td>AMP deaminase</td>
<td>1,341 ± 416</td>
<td>522 ± 102</td>
<td>9 ± 5</td>
<td>&lt;0.001‡‡</td>
</tr>
<tr>
<td>activity,</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>mmol·kg(^{-1})·dry muscle·min(^{-1})</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Fiber type</td>
<td>51 ± 12</td>
<td>48 ± 11</td>
<td>62 ± 5</td>
<td>0.118</td>
</tr>
<tr>
<td>composition,</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>type I %</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Training index</td>
<td>3 (2–3)</td>
<td>3 (1–3)</td>
<td>3 (2–3)</td>
<td>0.622</td>
</tr>
<tr>
<td>Peak power,</td>
<td>9.2 ± 1</td>
<td>9.0 ± 2</td>
<td>9.2 ± 1</td>
<td>0.962</td>
</tr>
<tr>
<td>W·kg body wt(^{-1})</td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Mean power,</td>
<td>6.7 ± 1</td>
<td>6.9 ± 2</td>
<td>6.8 ± 1</td>
<td>0.935</td>
</tr>
<tr>
<td>W·kg body wt(^{-1})</td>
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</table>

Mutant allele based on a C → T transition at nucleotide +34 (Ref. 24). Values are means ± SD except the values for training index, which are presented as median and range. NN, normal homozygotes; MN, heterozygotes; MM, mutant homozygotes. Post hoc tests: ‡difference between NN and MM; †difference between NM and MM; ‡difference between NN and NM.
exercise are presented in Fig. 3. The time course for venous blood lactate accumulation differs between genotypes (genotype \times time = 0.001). Peak values are observed at 3 min after exercise in mutant homozygotes (n = 4), at 6 min in heterozygotes (n = 7), and at 9 min in normal homozygotes (n = 7). Venous blood lactate levels continue to rise between 3 and 9 min after exercise in normal homozygotes (P = 0.001) and tend to increase in heterozygotes (P = 0.114), whereas mutant homozygotes exhibited no further accumulation (P = 0.592).

Mutant homozygotes do not show any significant increase in venous plasma ammonia after exercise, which is consistent with the absence of detectable IMP accumulation in their muscles during exercise. Unexpectedly, heterozygotes have a greater accumulation of this diffusible metabolite in venous plasma compared with normal homozygotes, despite homoyzgotes, despite a relatively smaller accumulation of IMP in their muscles. Moreover, heterozygotes and normal homozygotes display different time courses for venous plasma ammonia accumulation (genotype \times time = 0.002). Peak ammonia values are observed at 6 min after exercise for both groups, but heterozygotes exhibit a greater increase up to 6 min (P = 0.022) and also a greater decrease between 6 and 9 min compared with normal homozygotes (P = 0.031).

**DISCUSSION**

The aim of the present study was to investigate the effect of AMPD1 genotype, and thus inherently different levels of myoadenylate deaminase expression, on adenine nucleotide metabolism and performance during short-term high-intensity exercise in healthy subjects. Changes in muscle energy metabolites that are proposed to be involved in the development of muscle fatigue (10, 33, 43) were measured in subjects with different AMPD1 genotypes. Power outputs were also measured by using a 30-s Wingate test. These combined analyses reveal genotype-specific metabolic patterns and enable us to evaluate the potential influence of a common AMPD1 mutation on sprint exercise performance.

Short-term, high-intensity exercise results in a dramatic increase in ATP turnover rates in contracting muscle. High rates of ATP turnover are associated with a pronounced activation of AMP deaminase and enhanced accumulation of IMP in fatigued muscle (8, 12, 18, 32). Data presented in this study demonstrate that inherent differences in catalytic potential for AMP deamination conferred by AMPD1 genotype dramatically influence net breakdown of ATP during short-term high-intensity exercise. Homozygotes for the AMPD1 mutant allele have very low skeletal muscle AMP deaminase activities and do not deplete ATP pools or show any significant accumulation of IMP and ammonia during sprint exercise. In contrast, all other subjects investigated in this study exhibit ATP depletion and stoichiometric accumulation of IMP of comparable magnitude to values reported by others using similar exercise protocols (7, 37). However, when analyzed according to AMPD1 genotype, normal homozygotes tended to show greater depletion of ATP and significantly greater accumulation of IMP compared with heterozygotes. Tullson et al. (39) have reported relatively similar accumulations of IMP + inosine across subjects exhibiting more than a threefold range in AMP deaminase activities. However, closer inspection of their data reveals two distinct groups of indi-
Fig. 3. Venous plasma ammonia and blood lactate (means ± SE) at rest and after 30-s sprint exercise in NN, NM, and MM. P values show significance level of the interaction term in the ANOVA in a comparison of NN and NM for ammonia and of NN, NM, and MM for lactate. Mutant allele based on a C→T transition at nucleotide +34 of the AMPD1 gene (24).

In agreement with the absence of IMP accumulation, homozygotes for the AMPD1 mutation also do not exhibit a significant accumulation of venous plasma ammonia after exercise. This strongly supports the concept that ammonia production in normal subjects after short-term high-intensity exercise originates mainly from deamination of AMP to IMP during net adenine nucleotide catabolism in skeletal myocytes (21). Although ammonia and IMP are produced in equimolar amounts in the AMP deaminase reaction, the former is diffusible. Consequently, measurements of intracellular or extracellular ammonia may not accurately reflect the extent of IMP accumulation in muscle. Esbjornsson-Liljedahl and Jansson (7) reported smaller accumulations of plasma ammonia in women compared with men after short-term high-intensity exercise despite no difference in muscle IMP accumulation. Furthermore, Stathis et al. (37) showed that higher plasma ammonia levels up to 10 min after exercise accompanied the attenuation of IMP accumulation in muscle after sprint training. The observed discordance between the accumulation of ammonia and IMP may be due to differences in release of ammonia from muscle to blood.

An unexpected result of this study was the observation that despite smaller accumulations of IMP, heterozygotes consistently exhibit a greater increase in venous plasma ammonia compared with normal homozygotes after exercise. Moreover, heterozygotes show a different time course of accumulation, with a decrease between 6 and 9 min after exercise, whereas normal homozygotes exhibit a continued accumulation. In addition, time courses for the increase of lactate in systemic venous blood differed across the three AMPD1 genotypes. Peak venous blood lactate values are detected at 3, 6, and 9 min after exercise in mutant homozygotes, heterozygotes, and normal homozygotes, respectively.

These combined venous metabolite data may indicate a common mechanism for the release of diffusible metabolites from exercising skeletal muscle into blood. This proposal is based on variable capacities for adenosine production that consequently affect blood flow responses across AMPD1 genotypes. Cytosolic 5' nucleotidase isoform cN-I is relatively abundant in heart and skeletal muscle and produces adenosine during net ATP breakdown (34). Thus cN-I should be relatively more competitive for available myocyte AMP in individuals with decreased levels of myoadenylate deaminase. It is well known that adenosine causes vasodilation and plays an important role in the regulation of cardiac blood flow. Although this phenomenon is less established in skeletal muscle, recent studies in humans have demonstrated a positive correlation between increased interstitial adenosine levels during exercise and blood flow (13, 23). Although data are limited to four heterozygote and three mutant homozygote samples, measurements of muscle adenosine content in leftover biopsy material performed in this study support this hypothesis. A pronounced 25-fold increase in muscle adenosine content after exercise was revealed in the mutant homozygotes, whereas a modest twofold increase was observed in heterozygotes. Similar increases in muscle adenosine content with exercise were previously reported in symptomatic patients with myoadenylate deaminase deficiency, whereas no detectable change in this purine nucleotide was observed in controls (31). Enhanced localized blood flow, to-
gether with more pronounced activation of oxidative phosphorylation by augmented ADP accumulation, in turn, could result in a switch to greater dependence on oxidative energy metabolism in skeletal muscles of heterozygotes and mutant homozygous individuals, thus leading to a diminished net breakdown of ATP. In this fashion, adenosine may act as a link between muscle contraction, metabolic rate, and vasodilatation.

An alternative explanation for greater plasma ammonia accumulation in heterozygotes relative to normal homozygotes may be related to differential flux through the purine nucleotide cycle (PNC). There is evidence for purine nucleotide cycling in human skeletal muscle during exhaustive exercise as suggested by excessive ammonia production relative to IMP accumulation (2). Furthermore, adenylsuccinate synthetase, the enzyme catalyzing the rate-limiting step in the reamination arm of the PNC, is inhibited by high concentrations of IMP (11). These combined data may be used to speculate that purine nucleotide cycling during a 30-s Wingate test is low in normal homozygotes due to the larger accumulation of IMP. Consequently, heterozygous individuals produce more ammonia than normal homozygotes owing to a higher rate of cycling in response to the lower accumulation of IMP during this form of short-term high-intensity exercise.

Despite the dramatic influence of AMPD1 genotype on net breakdown of ATP and accumulation of IMP, and thereby presumably augmented ADP accumulation, no significant difference in performance during a 30-s Wingate test could be detected across the three different genotypes. Thus our hypothesis that greatly diminished adenylate catabolism in mutant homozygous individuals would result in reduced anaerobic performance could not be confirmed. However, it is possible that the 30-s Wingate test, as applied in the present study with a braking force of 7.5% of body weight for all subjects that was not individually optimized, is not sensitive enough to detect small differences in anaerobic performance across the three different genotypes. Regardless, the proposal for a more rapid onset of oxidative energy metabolism in mutant homozygous individuals could be an important compensatory mechanism in myoadenylate deaminase deficiency. This adaptation would lead to a greater replenishment of ATP via oxidative phosphorylation and result in removal of some of the accumulated ADP, thereby preventing its inhibitory action on the velocity of shortening. Support for this hypothesis might be gained in future studies by employing $^{31}$P magnetic resonance spectroscopy to determine whether there is an abnormal accumulation of ADP in myoadenylate deaminase-deficient skeletal muscle during intense, short-term exercise.

To sum up, the high incidence of myoadenylate deaminase deficiency in the general population (24, 28, 41), taken together with an inability to detect lower exercise performance in these individuals (25, 28, this study), implies that this inherited metabolic disorder of skeletal muscle does not typically lead to exercise intolerance. This is also consistent with the opinion that, when an individual exhibiting severe exercise intolerance is found to have an isolated deficiency of myoadenylate deaminase, the disorder may not be due to this enzyme defect alone (42). However, one cannot dismiss the possibility that myoadenylate deaminase deficiency, in combination with another yet-to-be-identified abnormality, may contribute to clinical symptoms observed in these individuals. This viewpoint would also be consistent with the growing number of identified cases of “double trouble” involving myoadenylate deaminase deficiency, in which the presence of a second inherited defect in energy metabolism can produce clinical symptoms more severe than either deficiency alone (42).

Purine metabolism is frequently monitored to assess skeletal muscle energy status in a variety of physiological settings. However, recent studies have demonstrated that more than one of every five individuals in the general population is either heterozygous or homozygous for a common AMPD1 mutation (24, 28, 41). The results of the present study show that individuals with these two genotypes have reduced capacities to deplete ATP pools and accumulate IMP during short-term high-intensity exercise. Moreover, our findings are consistent with the hypothesis that release of diffusible metabolites from skeletal muscle is also affected by AMPD1 genotype. Therefore, it is apparent that genotype should be evaluated as a prerequisite component of studies in which biochemical measurements during maximal exercise in different subjects are planned. Failure to do so may result in misleading data, whereas consideration of this parameter would be expected to enhance the precision of metabolic analyses.

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