目的　观察糖尿病肾病（DKD）动物模型Goto-Kakizaki（GK）大鼠骨骼肌蛋白消耗相关线粒体损伤情况，及低蛋白联合α酮酸对其作用。 方法　45只24周龄雄性GK大鼠随机分入正常蛋白组（NPD组）、低蛋白组（LPD组）和低蛋白联合α酮酸组（Keto组）；性别、周龄相同的Wistar大鼠15只为对照组（CTL组），予以正常蛋白饮食。每周称重，同时第24、32、40、48周时检测大鼠尿总蛋白及尿白蛋白、血清葡萄糖、Scr、BUN等变化。光镜下观察48周龄大鼠比目鱼肌琥珀酸脱氢酶（SDH）、烟酰胺腺嘌呤二核苷酸四唑氧化还原酶（NADH）染色后的形态、酶活性，计算肌纤维横截面积和Ⅰ、Ⅱ型肌纤维比例。透射电镜下观察组织超微结构，用分光光度计法检测比目鱼肌柠檬酸合成酶活性，定量PCR法检测线粒体DNA表达。 结果　与CTL组相比，NPD、LPD及Keto组体质量显著下降，尿白蛋白排泄增多，Scr和BUN水平也显著增高；肌纤维横截面积减少（均P＜0.05），Ⅱ型肌纤维比例增加[（37.01±1.85）%、（38.72±1.67）%、（26.77±2.23）%比（18.65±2.37）%，均P＜0.05]；电镜下肌丝部分断裂，线粒体肿胀变形明显；NPD及LPD组肌肉组织柠檬酸合成酶活性降低[（19 260.83±3522.13）、（21 313.11±2266.89） U·min-1·g-1比（24 787.47±1833.76） U·min-1·g-1，均P＜0.05]，线粒体DNA数量减少。与NPD、LPD组相比，Keto组GK大鼠体质量增加显著，血清Scr、BUN及尿蛋白水平均显著降低；比目鱼肌湿重和横截面积轻度增加，Ⅱ型肌纤维比例降低，电镜下肌丝完整，线粒体形态趋于正常，柠檬酸合成酶活性及线粒体DNA表达显著增加（均P＜0.05）。LPD组和NPD组组间差异无统计学意义。 结论　DKD可引起骨骼肌蛋白质消耗，伴线粒体肿胀变形，DNA表达减少及氧化磷酸化功能障碍。低蛋白联合α酮酸饮食则可改善DKD骨骼肌线粒体损伤，缓解蛋白质消耗状况，减轻骨骼肌萎缩程度。

"/> Objective　To observe the mitochondrial damage associated with protein-energy wasting of skeletal muscle in diabetic kidney disease (DKD) model of Goto-Kakizaki(GK) rats and evaluate the effects of low-protein diet supplemented with α-keto acids on muscle wasting.　Methods　Forty-five male 24-week-age GK rats were randomly divided into three groups, normal protein diet group (NPD), low-protein diet group (LPD) and LPD +α-keto group (Keto). Fifteen gender and age matched Wistar rats were served as control group (CTL). The living condition of GK rats was observed and the weight was measured once a week. Urine albumin, serum glucose, creatinine and urea nitrogen were measured at 24, 32, 40, 48 week age. Soleus muscle was observed to calculate the muscle size and the percentage of Ⅰ and Ⅱ type muscle fiber with software after SDH and NADH staining at 48-week-age. Tissue ultrastructure was observed under the transmission electron microscopy. The activity of citrate synthase was detected by spectrophotometer. Expression of mitochondrial DNA was examined by Q-PCR. Results　Compared with the CTL group, NPD, LPD and Keto groups had lower body weight, higher urine albumin, higher serum creatinine and urea nitrogen (P ＜0.05). The cross-sectional area of muscle fibers was larger in CTL group. Compared with CTL group, the muscle fiber was partly broken, the mitochondrial morphology was obviously changed, the percentage of type Ⅱ muscle fiber was increased significantly (P＜0.05), and the activity of citrate synthase and the number of mitochondrial DNA were decreased significantly in NPD, LPD and Keto groups (P＜0.05). In Keto group, muscle wasting was improved compared with NPD and LPD group (P＜0.05), the cross-sectional area of soleus muscle increased and the percentage of type Ⅱ muscle fiber decreased, levels of urine albumin, serum creatinine and urea nitrogen decreased (P＜0.05). Under transmission electron microscopy, the muscle fiber of keto group was intact and mitochondiral morphology was close to that of CTL group. The activity of citrate synthase and number of mitochondiral DNA were higher as compared to CTL group (P＜0.05). There were no significant differences between NPD and LPD group. Conclusions　In DKD condition, protein degradation in the skeletal muscle is accelerated, mitochondrion is swelling, the number of mitochondrial DNA is decreased and mitochondrial function is impaired. Low-protein diet supplemented with α-keto acids can improve mitochondrial damage and muscle wasting induced by DKD.

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糖尿病肾病大鼠骨骼肌蛋白能量消耗相关线粒体损伤及低蛋白联合α酮酸的作用

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Mitochondrial damage in proetin-energy wasting of skeletal muscle in rats with d effect of low-protein diet combined with α-keto acids

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