Oxidative Stress Is Involved in the Renal Dysfunction Induced by Sinoaortic Denervation in Rats

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The hypothesis that oxidative stress contributes to renal dysfunction in sinoaortically denervated (SAD) rats was investigated. Rats were sinoaortically denervated and received treatment with tempol (0.5 mmol/L in drinking water) for 8 weeks. Although the tempol treatment of the SAD rats had no significant effect on blood pressure or blood pressure variability, it significantly ameliorated the renal dysfunction as indicated by increases in renal blood flow (RBF) and the glomerular filtration rate (GFR) and reductions in plasma creatinine, blood urea nitrogen (BUN), the urine albumin excretion rate (UAE), and the glomerular sclerosis score (GSS). The SAD rats treated with tempol exhibited decreased plasma and renal malondialdehyde (MDA) levels and reduced renal formation of reactive oxygen species (ROS), superoxide (O$_2^-$), peroxynitrite (ONOO$$^-$$) and 3-nitrotyrosine. Treatment with tempol suppressed the nuclear concentration of nuclear factor-kappaB (NF-$\kappa$B) and reduced the renal levels of macrophage chemotactic protein 1 (MCP-1) and interleukin-6 (IL-6). The tempol-treated SAD rats exhibited decreased renal advanced glycation end product (AGE) levels and reduced receptor for advanced glycation end products (RAGE) protein expression. The tempol treatment of the SAD rats restored mitochondrial adenosine triphosphate (ATP) formation, DNA content, membrane integrity and the renal oxygen consumption rate. Additionally, the activities of superoxide dismutase (SOD), glutathione peroxidase (GPx), glutathione S epoxide transferase (GST), and catalase were decreased, and the activities of xanthin oxidase (XO) and reduced nicotinamide adenine dinucleotide phosphate (NADPH) oxidase were enhanced in the kidneys of the SAD rats. In conclusion, our work firstly provided direct evidence that oxidative stress played an important role in the renal dysfunction of SAD rats.

**Key words** oxidative stress; tempol; renal dysfunction; inflammation; mitochondrial function; sinoaortic denervation

Sinoaortic denervation (SAD) is a model that disrupts the baroreceptor reflex system, which leads to increased arterial pressure variability without changes in blood pressure$^1$–$^3$ and results in organ damage that includes cardiac hypertrophy, aortic remodeling, and renal dysfunction.$^4$–$^6$ Chronic renal dysfunction may lead to increased mortality and morbidity, increased costs of medical care, and reduced quality of life.$^7$–$^10$

Previous studies have proposed that the activation of the rennin angiotensin system$^{11}$–$^{13}$ and increases blood pressure variability$^{11}$ help to induce chronic mechanisms underlying the renal dysfunction induced by SAD. Accumulating evidence indicates that redox-sensitive pathways are involved in renal dysfunction, and one of the potential causes of renal disease progression is the accumulation of reactive oxygen species (ROS).$^{14,15}$ Recently, it was reported that ex vivo incubation with tempol (4-hydroxy-2,2,6,6-tetramethyl-piperidine-1-oxyl) restores the vascular relaxation mediated by the endothelium due to the effect of acetylcholine (ACH) in the aortas of SAD rats, which indicates that oxidative stress contributes to the endothelial dysfunction induced by SAD in rats.$^{16}$ Given these findings, we hypothesized that oxidative stress might be involved in the renal dysfunction induced by SAD in rats. Tempol can be used to reduce oxidative injury in animal models, and we therefore tested the effects of treatment with tempol against renal dysfunction induced by SAD in order to investigate the role of oxidative stress in renal injury following disruption of baroreceptor reflex system.

**Experimental**

**Animals** Sinoaortic baroreceptor denervations were performed in male Sprague-Dawley (SD) rats after the rats were anesthetized with 5 mg/kg diazepam and 50 mg/kg ketamine

| Table 1. Hemodynamic Parameters of Rats |
|----------------------------------------|
| **Sham** | **Sham+T** | **SAD** | **SAD+T** |
| Body weight (g) | 336±41 | 345±33 | 329±42 | 331±36 |
| SBP (mmHg) | 139±14 | 141±11 | 143±15 | 140±9 |
| DBP (mmHg) | 94±12 | 92±16 | 96±13 | 96±14 |
| HP (ms) | 138±9.3 | 141±6.9 | 140±7.2 | 142±8.6 |
| SBPV (mmHg) | 8.3±2.6 | 7.9±3.2 | 14.4±4.4$^a$ | 13.2±3.8$^b$ |
| DBPV (mmHg) | 5.4±1.8 | 5.5±2.2 | 9.7±2.0$^a$ | 9.0±1.7$^b$ |
| BRS (ms/mmHg) | 0.66±0.09 | 0.62±0.15 | 0.32±0.08$^a$ | 0.40±0.10$^b$ |

Values are represented as the mean±S.D.; BP, blood pressure; SBP, systolic BP; DBP, diastolic BP; SBPV, systolic BP variability; DBPV, diastolic BP variability; BRS, baroreflex sensitivity; T, tempol; a) p<0.05 versus sham-operated rats. n=10–12 in each group.

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hydrochloride. The neurovascular sheath was exposed through a 1.5-cm incision in the midline of the neck after the sternocleidomastoid muscles were reflected. The superior laryngeal nerves were cut near the vagus, and the aortic depressor fibers were sectioned. The bifurcation region was striped to denervate the sinuses. The rats in the Sham surgery group were incised in the middle of the neck, and the bilateral neck muscles were isolated. All of the experiments were approved by the Bioethics and Investigation Committee, and the institutional animal care guidelines were followed. The rats were randomly

Fig. 1. Effect of Treatment with Tempol on Renal Dysfunction in SAD Rats

Column graphs showed GFR (A), RBF (B), plasma creatinine (C), BUN (D), UAE (E), and GSS (F) in rats. Paraffin-embedded kidney sections were stained with hematoxylin–eosin (G) for evaluation. Concentration-response curves to ACh in isolated afferent arteriole from rats were shown (H). SAD, sinoaortic denervated; GFR, glomerular filtration rate; RBF, renal blood flow; BUN, blood urea nitrogen; UAE, urine albumin excretion rate; GSS, glomerular sclerosis score; T, tempol; Values are the mean±S.D. n=10–12 in each group; * p<0.05 versus Sham-operated rats; # p<0.05 versus SAD rats.
assigned to four groups as follows (n=50–57 per group): group 1, sham-operated rats (Sham); group 2, sham-operated+tempol rats (0.5 mmol/L in the drinking water, Sham+T); group 3, SAD rats (SAD); and group 4, SAD rats+tempol (0.5 mmol/L in drinking water, SAD+T). Eight weeks later, all rats were given normal water for 2 d.

**Western Blotting** The same quantities of prepared samples (20 µg) were electrophoresed by sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE), and Western blotting was performed using the standard protocol. Antibodies against receptor for advanced glycation end products (RAGE), inducible nitric oxide synthase (iNOS), lamin B1, nuclear factor-kappaB (NF-κB) and p65 were obtained from Santa Cruz Biotechnology, Inc. (Santa Cruz, CA, U.S.A.). The antibodies against β-actin and the horseradish peroxidase (HRP)-conjugated secondary antibody were purchased from ZSBIO, Inc. (Beijing, China).

**Quantitative Real-Time PCR Analysis** Total RNA was extracted from the kidneys using TRIzol reagent (Invitrogen) and was reverse-transcribed using oligo(dT) and reverse transcriptase. Quantitative real-time PCR was performed for interleukin (IL-6) (primers: 5’-TCTACCCAACTTCCAATGCTC-3’ and 5’-TTGGATGGCTTTGTCTAGCC3’), macrophage chemotactrant protein 1 (MCP-1) (5’-GCTTCGCTGCTTC3’ and 5’-CTCGCTGCTGTATTCTCTT3’), and β-actin (5’-AAGTCCTCACCCTCCAAG3’ and 5’-AAGCAATTGCTGCTACCTCCC3’). A Sybr Green-based detection system (Tiangen, Shanghai, China) on a LightCycler system. The samples were analyzed in duplicate. After the PCR runs, the melting curves were analyzed to verify the specificities of the PCR products.

**Detection of the Renal Total ROS, Superoxide (O₂⁻), and Peroxynitrite (OONO⁻)** The renal total OONO⁻, ROS and O₂⁻ formation rates were detected as previously reported. Briefly, prior to the ROS measurements, the samples were incubated in 200 µM 1-hydroxy-3-methoxycarbonyl-2,2,5,5-tetramethylpyrroline (CMH) at 37°C for 30 min. Prior to the before O₂⁻ measurements, the samples were incubated in 200 µM CMH for 30 min and then in 50 U/L polyethylene glycol (PEG)-conjugated superoxide dismutase (SOD) for an additional 30 min. Prior to the OONO⁻ measurements, the samples were incubated in 500 µM 1-hydroxy-3-carboxypyroline (CPH) for 30 min.

**Mitochondrial Function of the Kidneys** The mitocon-
dria from the rat kidneys were isolated with differential centrifugation. The mitochondrial ROS formation was estimated according to a chemiluminescence method. To reduce $O_2^-$ generation via redox reactions, the samples contained $5\mu mol/L$ lucigenin. The mitochondrial ATP formation rate was determined with a kit purchased from BioVision (Mountain View). The degree of mitochondrial swelling was measured as previously reported.20)

**Oxygen Consumption Rate in the Kidneys** The extracted kidneys were rinsed several times with unbuffered Dulbecco’s modified Eagle’s medium (DMEM) (pH 7.4). The kidneys were minced, and 10-mg samples were transferred into the wells of a microplate. A screen was added to cover the samples such that free perfusion was permitted, and tissue movement was minimized. Next, 500-$\mu L$ unbuffered DMEM supplemented with 2.5 mM D-glucose, 200-$\mu M$ L-carnitine and 50 $\mu M$ bovine serum albumin (BSA)-conjugated palmitic acid was added to each well. The microplates were transferred to an incubator. After incubation for 1 h at 37°C, an extracellular flux analysis was performed. After three baseline measurements, 1 $\mu M$ rotenone and 10 $\mu M$ antimycin A were added. Next, the oxygen consumption rates were monitored before they stabilized. The assay was repeated twice, and two replicates from each rat were examined.

**Measurements of the Oxidant and Antioxidant Enzyme Activities** The SOD activities in the kidneys were determined with an SOD-525TM kit purchased from OXIS International (Foster, CA, U.S.A.). The measurement of the reduced nicotinamide adenine dinucleotide phosphate (NADPH) oxidase activities in the kidneys were performed as previously reported.21) Renal catalase activity was determined according to a previously described method.22) The measurements of the glutathione S epoxide transferase (GST) activities in the kidneys were performed with a GST Fluorometric Activity Assay Kit purchased from BioVision (Mountain View, CA, U.S.A.). The glutathione peroxidase (GPx) activities in the kidneys were measured with a testing kit obtained from Cayman Chemicals (Ann Arbor, MI, U.S.A.). The xanthine oxidase (XO) activities in the kidneys were measured with a previously reported fluorometric kinetic assay method.23) The results were normalized to the protein concentrations.

**Statistical Analysis** The statistical significances of the differences between pairs of groups were analyzed with Student’s $t$-tests, unless the samples failed a normality test, in which case the Mann–Whitney rank-sum test was used. The significances of differences between three or four groups were examined with two-way ANOVA followed by Bonferroni $t$-tests. Significance was accepted at the level of $p<0.05$.

**Results**

**Effects of Tempol Treatment on the Hemodynamic Parameters (HPs)** Increased diastolic BP variability (DBPV), systolic BP variability (SBPV) and reduced baroreflex sensitivity (BRS) were observed in the SAD rats (Table 1). Similar body weights, SBPs, DBPs, and HPs were observed between the two groups. Thus, the HPs did not significantly
Effects of Tempol Treatment on Renal Dysfunction  Significant reductions in glomerular filtration rate (GFR) (Fig. 1A) and renal blood flow (RBF) (Fig. 1B) were noted in the SAD rats. Additionally, significantly elevated plasma creatinine (Fig. 1C), blood urea nitrogen (BUN) (Fig. 1D), and urine albumin excretion rate (UAE) (Fig. 1E) levels were noted in the SAD rats compared with the sham-operated rats. Morphological analyses revealed an increased level of glomerular sclerosis score (GSS) (Figs. 1F, G) in the SAD rats. Result of concentration–response curves to ACh revealed that renal endothelial function was impaired in SAD rats (Fig. 1H).

All of these renal HPs, GSS, and endothelial function were at least partially restored in the SAD rats by consumption of tempol.

Effect of Tempol on Renal Oxidative Stress in the SAD Rats  The levels of malondialdehyde (MDA) in the plasma (Fig. 2A) and kidneys (Fig. 2B) and the formations of ROS (Fig. 2C), O$_2^-$ (Fig. 2D), OONO$^-$ (Fig. 2E) and 3-nitrotyrosine (Fig. 2F) in the kidneys were increased in the SAD rats compared with the sham-operated rats. Following tempol treatment, the MDA levels in the kidneys and plasma were reduced, and the formations of 3-nitrotyrosine, OONO$^-$, O$_2^-$, and ROS in the kidneys were inhibited.

Effect of Tempol Treatment on Renal Inflammation The expression of nuclear NF-$\kappa$B p65 was increased (Fig. 3A) in the kidneys of the SAD rats. The tempol treatment of the SAD rats elicited decreased expression of nuclear NF-$\kappa$B p65 in the kidneys, which suggested that the tempol inhibited the nuclear translocation of NF-$\kappa$B in the kidneys of the SAD rats. Renal iNOS protein expression (Fig. 3B) and the levels of MCP-1 mRNA (Fig. 3C) and IL-6 mRNA (Fig. 3D) were enhanced in the SAD rats. The tempol treatment of the SAD rats suppressed iNOS protein expression and reduced the mRNA levels of MCP-1 and IL-6 in the kidneys.

Effect of Treatment with Tempol on the Renal Advanced Glycation End Products (AGEs)/RAGE Pathway in the SAD Rats  Increased renal AGE levels (Fig. 4A) and RAGE protein expression (Fig. 4B) were observed in the SAD rats. Tempol treatment of the SAD rats reduced the renal AGE levels and suppressed RAGE protein expression.

Effect of Tempol Treatment on Renal Mitochondrial Function  ROS formation was increased (Fig. 5A), while ATP formation was reduced (Fig. 5B) in the kidneys of the SAD rats. Increased mitochondrial swelling (Fig. 5C) and decreased mitochondrial DNA content (Fig. 5D) were observed in the kidneys of the SAD rats. The tempol treatment of the SAD rats inhibited the formation of mitochondrial ROS and ATP and also reduced the mitochondrial swelling and increased the mitochondrial DNA content. The oxygen consumption rate (OCR) was decreased in the kidneys of the SAD rats compared with those of the young rats, and the tempol treatment of the SAD rats restored the renal OCR. The mitochondrial membrane potential (MMP, Fig. 5F) was reduced in the kidneys of the SAD rats, and this MMP loss was significantly reduced following tempol treatment.

Effects of Tempol Treatment on Rennin Angiotensin System in Kidneys of SAD Rats  The levels of AngII (the central element of rennin angiotensin system, Fig. S1, supplementary data) were higher in kidneys of SAD rats. Treatment with tempol had no significant effect on the levels of AngII in kidneys of both sham-operated rats and SAD rats.

Oxidant/Antioxidant Enzyme Activities in the Kidney Homogenates of the SAD Rats  The activities of SOD (Fig. 6A), GPx (Fig. 6B), GST (Fig. 6C), and catalase (Fig. 6D) were reduced, and the activities of XO (Fig. 6E) and NADPH oxidase (Fig. 6F) were increased in the kidneys of the SAD rats.

Discussion  Oxidative stress describes physiological imbalance between the formation of ROS and the ability of the body to remove them. At normal concentrations, ROS may act as a second-messenger system that is involved in the maintenance of vascular tone and tubular function in the kidney. If this balance is disrupted, excess ROS can induce various types of kidney damage. In the SAD rats, and imbalance resulted from the reductions of the activities SOD, catalase, GPx and GST, the augmentations of the activities of NADPH oxidase and XO, and the impairment of the renal mitochondria.

Some previous small studies have indicated that antioxidants may play an important role in the development of...
chronic renal failure and that treatment with antioxidants can prevent the progression of renal insufficiency.\(^{25}\) In the present study, chronic tempol treatment suppressed ROS formation in the kidneys, as indicated by reduced MDA levels and \(O_2^-\) and \(\text{ONOO}^-\) formation, and preserved renal function in the SAD rats. These findings indicated that oxidative stress contributed to the progression of the renal dysfunction induced by SAD. Previous reports have revealed that chronic or acute tempol treatment can reduce arterial pressure in some rat models of hypertension.\(^{26–30}\) However, our study and others\(^{31}\) have demonstrated that tempol treatment has no effect on blood pressure or blood pressure viability. Therefore, the protective effect of tempol against the renal dysfunction induced by SAD is independent of its modulatory effect on blood pressure.

Apart from its anti-oxidative properties, tempol exhibited an anti-inflammatory property in this study and in previous studies.\(^{32}\) Chronic inflammation and oxidative stress are features that are associated with the activation of NF-κB and have pivotal roles in the pathogenesis of chronic renal failure. NF-κB is activated by cytoplasmic ROS, which not only leads to increased mitochondrial ROS production\(^{33}\) but also induces the expression of pro-inflammatory cytokines, such as MCP-1 and IL-6.\(^{34,35}\) In the present study, tempol treatment suppressed the translocation of NF-κB into the nucleus and reduced renal inflammation in the SAD rats. Therefore, we conclude that oxidative stress emerges earlier than inflammation and is the primary abnormality in the kidneys of SAD rats.

The accumulation of AGEs has been found to play an important role in the development of chronic kidney disease.\(^{36}\) Recently, it was reported that RAGE activation induces the remodeling of the aorta and endothelial dysfunction.\(^{18}\) Early in the Maillard reaction, glycation depends on the glucose concentration. Therefore, due to the availability of high quantities of glucose, glycation can enhance diabetes mellitus.\(^{37}\) However, it had been noted that oxidizing conditions and ROS play pivotal roles in the formation of glycoxidation products, which are an important class of AGEs that accumulate in tissues.\(^{38}\) In the present study, tempol treatment reduced the renal AGE levels and downregulated RAGE protein expression in the SAD rats. Therefore, renal AGEs/RAGE activation in SAD rats might be secondary to oxidative stress.

In the present study, consumption of tempol effectively preserved the renal mitochondrial function of the SAD rats, including ATP formation and membrane integrity. Mitochondria play important roles in tissue oxygen gradients, death signaling, and \(\text{H}_2\text{O}_2\) signaling. Mitochondrial dysfunction plays an important role in the gradual loss of renal energy, which causes the structural characteristics of chronic kidney disease.\(^{38,39}\) Our results indicated that oxidative stress might contribute to the mitochondrial dysfunction in the kidneys of SAD rats.

Finally, limitation of this work should be noted that only...
one antioxidant and only one dose of tempol was investigated in SAD rats. Some additional works about several other antioxidants and several doses of tempol were required in future work.

In conclusion, our work firstly demonstrated that oxidative stress contributed to SAD-induced renal dysfunction in rats, possibly through the activation of the AGEs/RAGE pathway, the induction of inflammation, and the impairment of mitochondrial function. Additionally, our results indicated that tempol could potentially be a novel countermeasure against the renal injury induced by baroreflex dysfunctions in patients.

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Conflict of Interest The authors declare no conflict of interest.

Supplementary Materials The online version of this article contains supplementary materials.

References
1) Schreihofer A. M., Sved A. F., Am. J. Physiol., 266, R1705–R1710 (1994).
2) Jacob H. J., Alper R. H., Brody M. I., Hypertension, 14, 501–510 (1989).
3) Norman R. A. Jr., Coleman T. G., Dent A. C., Hypertension, 3, 119–125 (1981).
4) Su D. F., Miao C. Y., Clin. Exp. Pharmacol. Physiol., 28, 709–715 (2001).
5) Parati G., Pumidossi G., Albini F., Malaspina D., Mancia G., J. Hypertens., 5, 93–98 (1987).
6) Sega R., Corrao G., Bombelli M., Beltrame L., Facchetti R., Grassi G., Ferrario M., Mancia G., Hypertension, 39, 719–724 (2002).
7) Majais S. K., Story K., Brouillette J., Takano T., Soroka S., Franek C., Mendelsson D., Finkelstein F. O., Clin. J. Am. Soc. Nephrol., 4, 1293–1301 (2009).
8) Collins A. J., Foley R. N., Chavers B., Gilbertson D., Herzog C., Johansen K., Kasisk B., Kutner N., Liu J., St. Peter W., Guo H., Gustafson S., Heubner B., Lamb K., Li S., Li S., Peng Y., Qiu Y., Roberts T., Skeans M., Snyder J., Solid C., Thompson B., Wang X., Weinhandel E., Zaun D., Arko C., Chen S-C., Daniels E., Ebben J., Frazier E., Hanzlik C., Johnson R., Sheets D., Wang X., Forrest B., Constantine E., Everson S., Eggers P., Agoda I., Am. J. Kidney Dis., 59 (Suppl. 1), A7 (2012).
9) Matsushita K., van der Velde M., Astor B. C., Woodward M., Levey A. S., de Jong P. E., Coresh J., Garrevoort R. T., Lancet, 375, 2073–2081 (2010).
10) Vanholder R., Massy Z., Argiles A., Spasovski G., Verbeke F., Lameire N., Nephrol. Dial. Transplant., 20, 1048–1056 (2005).
11) Miao C. Y., Xie H. H., Wang J. J., Su D. F., Acta Pharmacol. Sin., 23, 713–720 (2002).
12) Xie H. H., Miao C. Y., Liu J. G., Su D. F., J. Cardiovasc. Pharmacol., 41, 325–331 (2003).
13) Li L., Yi-Ming W., Li Z. Z., Zhao L., Yu Y. S., Li D. J., Xia C. Y.,
Liu J. G., Su D. F., Pharmacol. Res., 58, 196–201 (2008).

14) Zhang C., Chen H., Xie H. H., Shu H. J., Su D. F., J. Hypertens., 21, 2141–2148 (2003).

15) Zhang C., Xie H. H., Lu Z. A., Zhi M. J., Su D. F., J. Cardiovasc. Pharmacol., 43, 663–668 (2004).

16) Small D. M., Coombes J. S., Bennett N., Johnson D. W., Gobe G. C., Nephrology (Carlton), 17, 311–321 (2012).

17) Wilcox C. S., Am. J. Physiol. Regul. Integr. Comp. Physiol., 289, R913–R935 (2005).

18) Wu F., Feng J. Z., Qiu Y. H., Yu F. B., Zhang J. Z., Zhou W., Yu F., Wang H. D., Atherosclerosis, 229, 287–294 (2013).

19) Elks C. M., Mariappan N., Haque M., Guggilam A., Majid D. S., Francis J., Am. J. Physiol. Renal Physiol., 296, F298–F305 (2009).

20) Mariappan N., Soorappan R. N., Haque M., Sriramula S., Francis J., Am. J. Physiol. Heart Circ. Physiol., 293, H2726–H2737 (2007).

21) Li Y. L., Gao L., Zucker I. H., Schultz H. D., Cardiovasc. Res., 75, 546–554 (2007).

22) Beers R. F. Jr., Sizer I. W., J. Biol. Chem., 195, 133–140 (1952).

23) Onody A., Csonka C., Giricz Z., Ferdinandy P., Cardiovasc. Res., 58, 663–670 (2003).

24) Araujo M., Welch W. J., Curr. Opin. Nephrol. Hypertens., 15, 72–77 (2006).

25) Chen J., He J., Ogden L. G., Batuman V., Whelton P. K., Am. J. Kidney Dis., 39, 460–468 (2002).

26) Schnackenberg C. G., Wilcox C. S., Hypertension, 33, 424–428 (1999).

27) Nishiyama A., Yoshizumi M., Hitomi H., Kagami S., Kondo S., Miyatake A., Fukunaga M., Tamaki T., Kiyomoto H., Kohno M., Shokoji T., Kimura S., Abe Y., J. Am. Soc. Nephrol., 15, 306–315 (2004).

28) Rancourt M. E., Rodrigue M. E., Agharazii M., Larivière R., Lebel M., Am. J. Hypertens., 23, 314–320 (2010).

29) Koeners M. P., Braam B., Joles J. A., J. Hypertens., 29, 1160–1166 (2011).

30) MIYATA T., Maeda K., Kurokawa K., Van Ypersele de Strihou C., Nephrol. Dial. Transplant., 12, 255–258 (1997).

31) Manoli I., Syssel J. R., Li L., Houllier P., Garone C., Wang G., Zerfas P. M., Cusmano-Ozog K., Young S., Trivedi N. S., Cheng J., Sloan J. L., Chandler R. J., Abu-Asab M., Tsokos M., Elkahlon A., Rosen S., Enns G. M., Berry G. T., Hoffmann V., DiMauro S., Schnerrmann J., Venditti C. P., Proc. Natl. Acad. Sci. U.S.A., 110, 13552–13557 (2013).