# Background

Human brain is the biggest energy consuming tissue in human body. Although it only represents 2% of the body weight, it receives 20% of total body oxygen consumption and 25% of total body glucose utilization. For that reason, brain is considered to be the most vulnerable part of human body against the reactive oxygen species (ROS), a by-product of aerobic respiration. Oxidative stress is directly related to a series of brain dysfunctional disease such as Alzheimer's disease, Parkinson's disease etc. Electrolyzed reduced water (ERW) is a functional drinking water containing a lot of molecular hydrogen and a small amount of platinum nanoparticles (Pt NPs, Table Table1).1). ERW is known to scavenge ROS and protect DNA from oxidative damage [1]. We previously showed that ERW was capable of extending lifespan of Caenorhabditis elegans by scavenging ROS [2]. Molecular hydrogen could scavenge ROS and protected brain from oxidative stress [3]. Pt NPs are also a new type of multi-functional ROS scavenger [4].

## **Results**

ERW significantly reduced the cell death induced by H2O2 pretreatment (Figure (Figure 1).1). ERW also scavenged the intracellular ROS and prevented the decrease of mitochondrial membrane potential and ATP production induced by ROS. We also examined the neuroprotective effects of molecular hydrogen and Pt NPs and showed that both molecular hydrogen and Pt NPs contributed to the neuroprotective effects of ERW.

# Conclusion

The results suggest that ERW is beneficial for the prevention and alleviation of oxidative stress-induced human neurodegenerative diseases.

## https://www.nihon-trim.co.jp/common/editor/files/en\_health\_%20benefit\_of\_reduced\_water.pdf

Anti-cancer effects ERW causes telomere shortening in cancer cells (Shirahata et al., 1999). It suppresses tumor angiogenesis by scavenging intracellular ROS and suppressing the gene expression and secretion of vascular endothelial growth factor (Ye et al., 2008). ERW suppresses the growth of cancer cells and microorganisms (Hamasaki et al., 2005; Komatsu et al., 2001) and induces apoptosis together with glutathione in human leukemia HL60 cells (Tsai, Hsu, Chen, Ho, & Lu, 2009). ERW induces differentiation of K562 cells to megakaryocytes (Komatsu et al., 2003), and when supplemented with Pt nanoparticles, it strongly suppresses the two step transformation of NIH3T3 cells by a carcinogen (Nishikawa et al., 2005).

Anti-arteriosclerosis effects ERW suppresses the Cu2 $\flat$ -catalyzed oxidation of human LDL and suppresses triglyceride levels in mice fed high fat foods (Abe et al., 2010). Hydrogen-supplemented water also suppresses arteriosclerosis (see the supplemental information).

Anti-neurodegenerative effects ERW suppresses neural cell death by oxidative stress (Kashiwagi et al., 2005). Hydrogen-supplemented water also exhibits various anti-neurodegenerative disease effects (see the supplemental information).

Other effects Naito et al. (2002) reported that chronic administration with electrolyzed alkaline water inhibits aspirin-induced gastric mucosal injury in rats. Hydrogen-enriched electrolyzed water has been demonstrated to be safe in mutagenicity, genotoxicity and subchronic oral toxicity (Saitoh, Harata, Mizuhashi, Nakajima, & Miwa, 2010). ERW exhibits hepatoprotective effects against CC14-induced liver damage in mice (Tsai et al., 2009). Ionized alkaline water improves the symptoms of metabolic acidosis in experimental animals (Abo-Enein, Gheith, Barakat, Nour, & Sharaf, 2009). ERW also exhibits an anti-hangover effect (Park et al., 2009). Silica hydride found in Funza water in Pakistan suppressed carbon tetrachloride-induced hepatotoxicity in mice (Tsu et al., 2010). Recently, many papers have been published on the suppressive effects of hydrogen molecules contained in ERW on oxidative stress-related diseases (see the supplementary information). The major functions of reduced water are summarized in Fig. 4.

## Conclusions and perspective

Accumulating evidence has shown that reduced waters are health beneficial and they suppress oxidative stressrelated diseases such as diabetes, cancer, arteriosclerosis, neurodegenerative diseases, and the side effects of hemodialysis. The mechanisms of action of reduced water for scavenging ROS are considered to be complicated. ERW contains hydrogen molecules and mineral nanoparticles. Hydrogen molecules and active hydrogen may be new redox regulation factors that can induce the gene expression of antioxidative enzymes. Hydrogen molecules may be converted to active hydrogen by catalyst action of metal nanoparticles to exhibit more potent reducibility. Mineral nanoparticles themselves are new types of multifunctional antioxidants. Mineral hydride nanoparticles, which are H-donors as well as organic antioxidants like ascorbic acid, are also candidates of active agents in reduced waters. NRW may have one or some of the active agents described above. Further investigation on activation methods of water by electricity, magnetic fields or light are likely to contribute to the development of energy-rich waters, which will be beneficial for human health. Reduced water may suppress harmful effects of environmental pollution on the embryo in pregnant women by purifying amniotic fluid and blood. Reduced water might also contribute to the food industry by improving the taste, texture and preservation of foods. In industries, the usage of ERW is Fig. 4. Variety of functions of reduced water. 128 S. Shirahata et al. / Trends in Food Science & Technology 23 (2012) 124e131 expected as washing water to prevent the rust of semiconductors. In the field of environmental remediation, reduced water will prevent the rotting of river and lake water, because the proliferation of bacteria or organisms causing the rotting will be suppressed in a reduced circumstance. Further research on water itself may ultimately reveal the secret of the origin of life.

# https://www.science.gov/topicpages/e/electrolyzed+reduced+water

Antibacterial effect of electrolyzed water on oral bacteria.

PubMed

Lee, Sung-Hoon; Choi, Bong-Kyu

#### 2006-08-01

This study investigated the antibacterial effect of electrolyzed water on oral bacteria both in vitro and in vivo. Tap water was electrolyzed in a water vessel using platinum cell technology. The electrolyzed tap water (called Puri-water) was put in contact with five major periodontopathogens or toothbrushes contaminated with these bacteria for 30 sec. In addition, Puri-water was used as a mouthwash for 30 sec in 16 subjects and the antibacterial effect on salivary bacteria was evaluated. Puri-water significantly reduced the growth of all periodontopathogens in culture and on toothbrushes, and that of aerobic and anaerobic bacteria in saliva, when compared to the effect of tap water. It also significantly reduced mutans streptococci growing on mitis salivarius-bacitracin agar. Our results demonstrate that the electrolyzed tap water is effective as a mouthwash and for toothbrush disinfection.

Preservative effect of electrolyzed reduced water on pancreatic beta-cell mass in diabetic db/db mice.

#### PubMed

Kim, Mi-Ja; Jung, Kyung Hee; Uhm, Yoon Kyung; Leem, Kang-Hyun; Kim, Hye Kyung

#### 2007-02-01

Oxidative stress is produced under diabetic conditions and involved in progression of pancreatic beta-cell dysfunction. Both an increase in reactive oxygen free radical species (ROS) and a decrease in the antioxidant defense mechanism lead to the increase in oxidative stress in diabetes. Electrolyzed reduced water (ERW) with ROS scavenging ability may have a potential effect on diabetic animals, a model for high oxidative stress. Therefore, the present study examined the possible anti-diabetic effect of ERW in genetically diabetic mouse strain C57BL/6J-db/db (db/db). ERW with ROS scavenging ability reduced the blood glucose concentration, increased blood insulin level, improved glucose tolerance and preserved beta-cell mass in db/db mice. The present data suggest that ERW may protects beta-cell damage and would be useful for antidiabetic agent.

Development of device producing electrolyzed water for home care

NASA Astrophysics Data System (ADS)

Umimoto, K.; Nagata, S.; Yanagida, J.

2013-06-01

When water containing ionic substances is electrolyzed, electrolyzed water with strong bactericidal ability due to the available chlorine(AC) is generated on the anode side. Slightly acidic to neutral electrolyzed water (pH 6.5 to 7.5) is physiological pH and is suitable for biological applications. For producing slightly acidic to neutral electrolyzed water simply, a vertical-type electrolytic tank with an asymmetric structure was made. As a result, a small amount of strongly alkaline water was generated in the upper cathodic small chamber, and a large amount of weakly acidic water generated in the lower anodic large chamber. The pH and AC concentration in solutin mixed with both electrolyzed water and had strong bactericidal activity. This solution was slightly acidic to neutral electrolyzed water and had strong bactericidal activity. This device is useful for producing slightly acidic to neutral electrolyzed water as a disinfectant to employ at home care, when considering economic and environmental factors, since it returns to ordinary water after use.

Histological study on the effect of electrolyzed reduced water-bathing on UVB radiation-induced skin injury in hairless mice.

PubMed

Yoon, Kyung Su; Huang, Xue Zhu; Yoon, Yang Suk; Kim, Soo-Ki; Song, Soon Bong; Chang, Byung Soo; Kim, Dong Heui; Lee, Kyu Jae

#### 2011-01-01

Electrolyzed reduced water (ERW), functional water, has various beneficial effects via antioxidant mechanism in vivo and in vitro. However there is no study about beneficial effects of ERW bathing. This study aimed to determine the effect of ERW bathing on the UVB-induced skin injury in hairless mice. For this purpose, mice were irradiated with UVB to cause skin injury, followed by individually taken a bath in ERW (ERW-bathing) and tap water (TW-bathing) for 21 d. We examined cytokines profile in acute period, and histological and ultrastructural observation of skin in chronic period. We found that UVB-mediated skin injury of ERW-bathing group was significantly low compared to TW control group in the early stage of experiment. Consistently, epidermal thickening as well as the number of dermal mast cell was significantly lowered in ERW-bathing group. Defection of corneocytes under the scanning electron microscope was less observed in ERW-bathing group than in TW-bathing group. Further, the level of interleukin (IL)-11<sup>2</sup>, tumor necrosis factor (TNF)-1<sup>1</sup>± and IL-12p70 in ERW group decreased whereas those of IL-10 increased. Collectively, our data indicate that ERW-bathing significantly reduces UVB-induced skin damage through influencing pro-/anti-inflammatory cytokine balance in hairless mice. This suggests that ERW-bathing has a positive effect on acute UVB-mediated skin disorders. This is the first report on bathing effects of ERW in UVB-induced skin injury.

Electrolyzed-reduced water increases resistance to oxidative stress, fertility, and lifespan via insulin/IGF-1-like signal in C. elegans.

### PubMed

Park, Seul-Ki; Park, Sang-Kyu

#### 2013-01-01

Electrolyzed-reduced water (ERW) scavenges reactive oxygen species and is a powerful anti-oxidant. A positive correlation between oxidative stress and aging has been proved in many model organisms. In Caenorhabditis elegans, many long-lived mutants showed reduced fertility as a trade off against longevity phenotype. We aimed to study the effect of ERW on oxidative stress, fertility and lifespan of C. elegans. We also investigated the genetic pathway involved in the effect of ERW on resistance to oxidative stress and lifespan. We compared lifespan and fertility of worms in media prepared with distilled water and ERW. ERW significantly extended lifespan and increased the number of progeny produced. Then the effect of ERW on resistance to oxidative stress and lifespan of eat-2, a genetic model of dietary restriction, but had no effect on those of age-1, which is involved in insulin/insulin-like growth factor (IGF)-1-like signal. In addition, knockdown of daf-16, the downstream mediator of insulin/IGF-1-like signal, completely prevented the effect of ERW on lifespan via insulin/IGF-1-like signal in C. elegans.

New Approach in Translational Medicine: Effects of Electrolyzed Reduced Water (ERW) on NF-κB/iNOS Pathway in U937 Cell Line under Altered Redox State

#### PubMed Central

Franceschelli, Sara; Gatta, Daniela Maria Pia; Pesce, Mirko; Ferrone, Alessio; Patruno, Antonia; de Lutiis, Maria Anna; Grilli, Alfredo; Felaco, Mario; Croce, Fausto; Speranza, Lorenza

#### 2016-01-01

It is known that increased levels of reactive oxygen species (ROS) and reactive nitrogen species (RNS) can exert harmful effects, altering the cellular redox state. Electrolyzed Reduced Water (ERW) produced near the cathode during water electrolysis exhibits high pH, high concentration of dissolved hydrogen and an extremely negative redox

potential. Several findings indicate that ERW had the ability of a scavenger free radical, which results from hydrogen molecules with a high reducing ability and may participate in the redox regulation of cellular function. We investigated the effect of ERW on H2O2-induced U937 damage by evaluating the modulation of redox cellular state. Western blotting and spectrophotometrical analysis showed that ERW inhibited oxidative stress by restoring the antioxidant capacity of superoxide dismutase, catalase and glutathione peroxidase. Consequently, ERW restores the ability of the glutathione reductase to supply the cell of an important endogenous antioxidant, such as GSH, reversing the inhibitory effect of H2O2 on redox balance of U937 cells. Therefore, this means a reduction of cytotoxicity induced by peroxynitrite via a downregulation of the NF-Î<sup>o</sup>B/iNOS pathway and could be used as an antioxidant for preventive and therapeutic application. In conclusion, ERW can protect the cellular redox balance, reducing the risk of several diseases with altered cellular homeostasis such as inflammation. PMID:27598129

Effect of several electrolyzed waters on the skin permeation of lidocaine, benzoic Acid, and isosorbide mononitrate.

#### PubMed

Kitamura, Toshihiko; Todo, Hiroaki; Sugibayashi, Kenji

#### 2009-02-01

The effects of several electrolyzed waters were evaluated on the permeation of model base, acid and non-ionized compounds, lidocaine (LC), benzoic acid (BA), and isosorbide mononitrate (ISMN), respectively, through excised hairless rat skin. Strong alkaline-electrolyzed reducing water (ERW) enhanced and suppressed the skin permeation of LC and BA, respectively, and it also increased the skin permeation of ISMN, a non-ionized compound. On the contrary, strong acidic electrolyzed oxidizing water (EOW) enhanced BA permeation, whereas suppressing LC permeation. Only a marginal effect was observed on the skin permeation of ISMN by EOW. These marked enhancing effects of ERW on the skin permeation of LC and BMN were explained by pH partition hypothesis as well as a decrease in skin impedance. The present results strongly support that electrolyzed waters, ERW and EOW, can be used as a new vehicle in topical pharmaceuticals or cosmetics to modify the skin permeation of drugs without severe skin damage.

#### Biological effects of electrolyzed water in hemodialysis.

#### PubMed

Nakayama, Masaaki; Kabayama, Shigeru; Nakano, Hirofumi; Zhu, Wan-Jun; Terawaki, Hiroyuki; Nakayama, Keisuke; Katoh, Kiyoshi; Satoh, Toshinobu; Ito, Sadayoshi

#### 2009-01-01

The application of electrolyzed water (EW) at the cathode side to manufacture reverse osmosis (RO) water and hemodialysis (HD) solution can actually lead to less oxidative capacity in chemical terms. The present study examined the biological actions of this water on human polymorphonuclear leukocytes (PMNs), and the clinical feasibility of applying this technology to HD treatment. RO water using EW (e-RO) exhibited less chemiluminescence in luminol-hydrogen peroxide and higher dissolved hydrogen levels (-99.0 ppb) compared with control RO water. The effects of e-RO on PMN viability were tested. HD using e-RO was performed for 12 consecutive sessions in 8 patients for the feasibility test. Basal cellular viability and function to generate superoxide radicals of PMNs were better preserved by e-RO application. In the clinical trial, reductions of blood pressure were noted, but no adverse events were observed. There were no changes in the blood dialysis parameters, although methylguanidine levels were significantly decreased at the end of study. The present study demonstrated the capacity of e-RO to preserve the viability of PMNs, and the clinical feasibility of applying this water for HD treatment. The clinical application of this technology may improve the bio-compatibility of HD treatment. Copyright 2009 S. Karger AG, Basel.

Application of slightly acidic electrolyzed water for decontamination of stainless steel surfaces in animal transport vehicles.

#### PubMed

Ni, Li; Zheng, Weichao; Zhang, Qiang; Cao, Wei; Li, Baoming

#### 2016-10-01

The effectiveness of slightly acidic electrolyzed water (SAEW) in reducing Escherichia coli, Salmonella typhimurim, Staphylococcus aureus or bacterial mixtures on stainless steel surfaces was evaluated and compared its efficacy with composite phenol solution for reducing total aerobic bacteria in animal transport vehicles. Stainless steel surfaces were inoculated with these strains individually or in a mixture, and sprayed with SAEW, composite phenol, or alkaline electrolyzed water for 0.5, 1, 1.5 and 2min. The bactericidal activity of SAEW increased with increasing available chlorine concentration and spraying duration. The SAEW solution of 50mgl -1 of available chlorine concentration showed significantly higher effectiveness than composite phenol in reducing the pathogens on stainless steel surfaces (P<0.05). Complete inactivation of pathogens on stainless steel surfaces were observed after treatment with alkaline electrolyzed water followed by SAEW at 50mgl -1 of available chlorine concentration for 2min or alkaline electrolyzed water treatment followed by SAEW treatment at 90mgl -1 of available chlorine concentration for 0.5min. The efficacy of SAEW in reducing total aerobic bacteria in animal transport vehicles was also determined. Vehicles in the disinfection booth were sprayed with the same SAEW, alkaline electrolyzed water and composite phenol solutions using the automatic disinfection system. Samples from vehicle surfaces were collected with sterile cotton swabs before and after each treatment. No significant differences in bactericidal efficiency were observed between SAEW and composite phenol for reducing total aerobic bacteria in the vehicles (P>0.05). SAEW was also found to be more effective when used in conjunction with alkaline electrolyzed water. Results suggest that the bactericidal efficiency of SAEW was higher than or equivalent to that of composite phenol and SAEW may be used as effective alternative for reducing microbial contamination

# https://www.sciencedirect.com/science/article/pii/S1726490117302617

#### Results

After administration of ERW, the reduced urinary osmolality was increased and elevated  $Na_+$ ,  $K_+$ ,  $Mg_{2+}$  and  $Ca_{2+}$  levels in urine were significantly decreased in cisplatin-induced renal injury mice. Besides, the results demonstrated that significantly decreased elevated serum levels of creatinine and blood urea nitrogen (BUN) and the levels of TBARS in the kidneys that were induced by cisplatin. Moreover, ERW treatment was also found to markedly increase (p < 0.05) the activities of GPx, GR, CAT and SOD, and to increase GSH content in the kidneys. Histopathology showed that ERW protects against cisplatin-induced renal injury to both the proximal and distal tubules.

#### Conclusion

ERW exhibits potent nephroprotective effects on cisplatin-induced kidney damage in mice, likely due to both the increase in antioxidant-defense system activity and the inhibition of lipid peroxidation.

https://www.sciencedirect.com/science/article/abs/pii/S0024320506005923

#### Abstract

Oxidative stress is produced under diabetic conditions and is likely involved in progression of pancreatic  $\beta$ -cell dysfunction found in diabetes. Both an increase in reactive oxygen free radical species (ROS) and a decrease in the antioxidant defense mechanism lead to the increase in oxidative stress in diabetes. Electrolyzed reduced water (ERW) with ROS scavenging ability may have a potential effect on diabetic animals, a model for high oxidative stress. Therefore, the present study examined the possible anti-diabetic effect of ERW in two different diabetic animal models. The genetically diabetic mouse strain  $C_57BL/6J$ -db/db (db/db) and streptozotocin (STZ)-induced diabetic mouse were used as insulin deficient type 1 and insulin resistant type 2 animal model, respectively. ERW, provided as a drinking water, significantly reduced the blood glucose concentration and improved glucose tolerance in both animal models. However, ERW fail to affect blood insulin levels in STZ-diabetic mice whereas blood insulin level was markedly increased in genetically diabetic db/db mice. This improved blood glucose control could result from enhanced insulin sensitivity, as well as increased insulin release. The present data suggest that ERW may function as an orally effective anti-diabetic agent and merit further studies on its precise mechanism.

# https://www.haptra.com/assets/research/Effect%20ARW%20SCI%20Haptra.pd f

Previous studies have demonstrated that AIW has the protective effect against the accumulation of lipid and cholesterol in the body(10,11,15). Lee et al. (unpublished data) designed a study to confirm further the effect of AIW on diseases related with fat accumulation such as obesity. Mice model of obesity induced by feeding high-fat diet was used. In the study, supplementation of AIW showed protection from quickly gaining body weight of mice. This is further supported with the lipid profile, wherein oil red 0 staining revealed that mice fed with AIW controls the accumulation of body fats in liver. Moreover, molecular data showed that supplementation of AIW induced the expression of the gene CYP7A1 which encodes cholesterol 7a-hydroxylase, the first and rate limiting step in the bile acid synthetic pathway, the major site of regulation and primary mechanism for the removal of cholesterol from the body(19). Based on these results, AIW suggests an antiobesity effect via inducing the CYP7A1 that plays critical

role in cholesterol homeostasis in the body. Further alternative mechanism on obesity provides a rationale to explore the supplementation of AIW as a novel therapeutic in humans.

Studies revealed that supplementation of AIW reduced abnormal intestinal fermentation, chronic diarrhea, and gastric hyperacidity (9, 21). AIW exhibits a high pH compared to the tap water, which plays an important role on its known efficacy. Alkaline pH neutralizes acids in the stomach. Since AIW has a high pH, it is justifiable that one mechanism of AIW against hyperacidity and other related accumulated acid and toxic caused diseases is via neutralization of the acids. Another beneficial effect of AIW drinking would be influence on blood pH. Secretion of acidic components in stomach to neutralize AIW increase blood pH to compensate acid base balance, thus help maintaining physiologic homeostasis in human body

## https://link.springer.com/chapter/10.1007/978-94-017-0728-2\_5

Although water is a very stable substance, it is easily electrolyzed to produce hydrogen and oxygen molecules. Weak current produces only active hydrogen and hydrogen molecule without production of oxygen molecule. Active hydrogen is so small that almost all minerals adsorbs or absorbs it. These facts prompted us to propose "active hydrogen theory of reduced water". We have demonstrated that electrolyzed reduced water (ERW) and ground waters such as Hita Tenryosui water in Japan, Nordenau water in Germany, Tracote water in Mexico, which are so called miracle water because of their improving activity on various diseases, are all antioxidative water containing active hydrogen and can scavenge intracellular reactive oxygen species (ROS). Reduced water stimulated glucose uptake into muscle and adipocyte cells as well as insulin. It also stimulated the secretion of insulin from pancreatic beta cells and improved the sugar tolerance damage in type 2 diabetes model mice. Reduced water impaired the tumor phenotypes such as rapid growth, anchorage independent growth in a soft agar, morphology, telomere maintenance, and abilities of invasion, metastasis, and angiogenesis. It activated the cancer immune systems, suppressing the tumor growth *in vivo*. Reduced water is expected for utilization for prevention and therapy of various diseases.

https://www.researchgate.net/publication/307621639\_New\_Approach\_in\_Tr anslational Medicine\_Effects\_of\_Electrolyzed\_Reduced\_Water\_ERW\_on\_NFkBiNOS\_Pathway\_in\_U937\_Cell\_Line\_under\_Altered\_Redox\_State

. Conclusions In summary, the H

2

dissolved in ERW acts as an antioxidant, neutralizing the free radical. This is very important if we consider that oxidative stress contributes to the development of many inflammatory diseases. To this end, further investigations on the benefits of ERW on several diseases with altered cellular redox balance are needed.

Abstract:

It is known that increased levels of reactive oxygen species (ROS) and reactive nitrogen species (RNS) can exert harmful effects, altering the cellular redox state. Electrolyzed Reduced Water (ERW) produced near the cathode during water electrolysis exhibits high pH, high concentration of dissolved hydrogen and an extremely negative redox potential. Several findings indicate that ERW had the ability of a scavenger free radical, which results from hydrogen molecules with a high reducing ability and may participate in the redox regulation of cellular function. We investigated the effect of ERW on H2O2-induced U937 damage by evaluating the modulation of redox cellular state. Western blotting and spectrophotometrical analysis showed that ERW inhibited oxidative stress by restoring the antioxidant capacity of superoxide dismutase, catalase and glutathione peroxidase. Consequently, ERW restores the ability of the glutathione reductase to supply the cell of an important endogenous antioxidant, such as GSH, reversing the inhibitory effect of H2O2 on redox balanceof U937 cells. Therefore, this means a reduction of cytotoxicity induced by peroxynitrite via a downregulation of the NF-xB/iNOS pathway and could be used as an antioxidant for preventive and therapeutic application. In conclusion, ERW can protect the cellular redox balance, reducing the risk of several diseases with altered cellular homeostasis such as inflammation