

USE OF SELF-POWERED TENG ELECTRICAL STIMULATION DEVICE BASED ON CHITOSAN THIN FILM AND SILVER NANOPARTICLES FOR ANTIBACTERIAL APPLICATIONS AND PARKINSON'S DISEASE TREATMENT

ABSTRACT

In this study, a TENG (Triboelectric Nanogenerator) power generation device was successfully designed based on the principle of converting mechanical energy into electrical energy, where a chitosan membrane serves as a triboelectric positive material and silver nanoparticles (AgNPs) are incorporated. The CHI-AgNP (CA1) nanocomposite membrane was successfully synthesized using a casting method, demonstrating its effectiveness as an electrode material. The TENG device utilizing this membrane exhibited promising output performance (up to 40V and 2 $\mu\text{A}/\text{cm}^2$) along with enhanced antibacterial activity due to electrical stimulation (ES).

Furthermore, experiments on *Caenorhabditis elegans* confirmed the safety of S-TENG stimulation. The device significantly improved symptoms associated with Parkinson's disease and showed that electrical stimulation did not significantly affect the lifespan, body size, or reproductive capacity of *C. elegans*. The dopamine synthesis process, starting from tyrosine, involved the *cat-2* and *bas-1* genes, indicating that electrical stimulation improved the slowing response rate of the nematodes to 66.87%, compared to 18.78% in the non-stimulated control group. Additionally, the stimulated group exhibited significant recovery in dopaminergic neuronal fluorescence compared to the control group. Electrical stimulation also increased the expression of the *cat-2* and *dat-1* genes.

Keywords: TENG power generation device, CHI-AgNP (CA1) nanocomposite membrane, Parkinson's disease

I. INTRODUCTION

With the rapid development of renewable energy technology, especially the triboelectric effect (triboelectric nanogenerators), the conversion of mechanical energy into electrical energy has attracted significant attention from the research community. Devices based on this effect can generate a continuous, environmentally friendly power source while minimizing negative environmental impacts. However, a major challenge remains: the use of non-biodegradable synthetic materials, which limit their applicability in biomedical fields.

Chitosan, a natural biopolymer with biodegradability, non-toxicity, and excellent charge-generation properties, has shown potential as an environmentally friendly triboelectric material. Chitosan can serve as a triboelectric positive electrode material in power generation devices, paving the way for research into green energy harvesting from naturally available materials.

One crucial application of this technology is in treating neurodegenerative diseases, particularly Parkinson's disease (PD). PD is a chronic degenerative disorder affecting millions of people annually, and current treatment methods, including medications and surgery, remain limited. Recent research has indicated that electrical stimulation from TENG devices can positively impact PD treatment by enhancing dopaminergic behavior and restoring damaged dopamine neurons. This finding opens up the potential for using triboelectric generators in biomedicine as a non-invasive treatment for Parkinson's disease.

Based on theoretical and practical foundations, our research project titled **“Use of Self-Powered TENG Electrical Stimulation Device Based on Chitosan Thin Film and Silver Nanoparticles for Antibacterial Applications and Parkinson's Disease Treatment”** aims not only to develop clean and sustainable energy sources but also to create biomedical applications that improve the quality of life for Parkinson's patients and individuals with other neurological disorders.

2.1. Theoretical Background

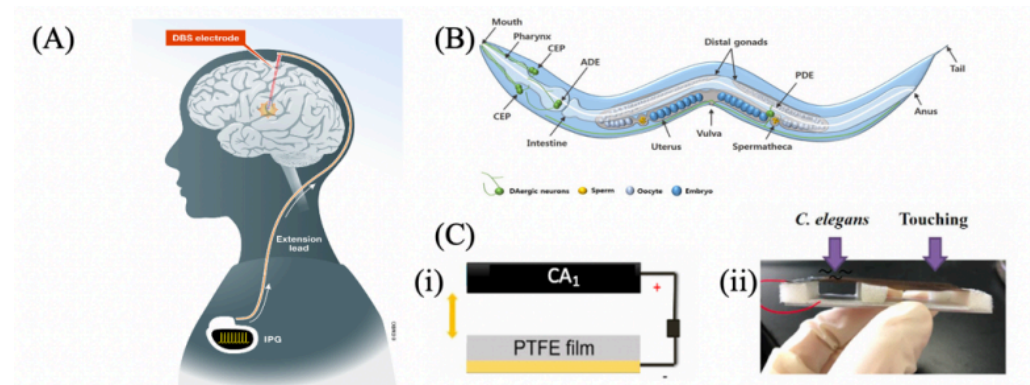


Figure 2.1: (A) Deep Brain Stimulation (DBS) for Parkinson's treatment. (B) Dopaminergic neurons in *C. elegans*. (C) TENG device design: (i) schematic model and (ii) experimental setup.

Based on the demonstrated electrical stimulation potential of TENG devices, our study expands its application in Parkinson's disease treatment using *C. elegans* as a model organism. DBS is a conventional treatment for PD motor symptoms, but its

invasiveness and surgical risks limit its application. Therefore, we propose a TENG device as a non-invasive alternative. *C. elegans* is advantageous due to its transparency, short lifespan, genetic similarity to humans, simple nervous system, and mutant strains relevant to PD research.

II. EXPERIMENTAL METHODS

2.2. Experimental Equipment and Chemicals

- Chemicals: Chitosan, AgNO_3 , distilled water.
- Equipment: Magnetic stirrer, mold, drying oven, aluminum sheet, PolyTetraFluoroEthylene (PTFE) membrane.

2.3. Experimental Procedure

2.3.1. Casting Process for CHI–AgNPs Membrane

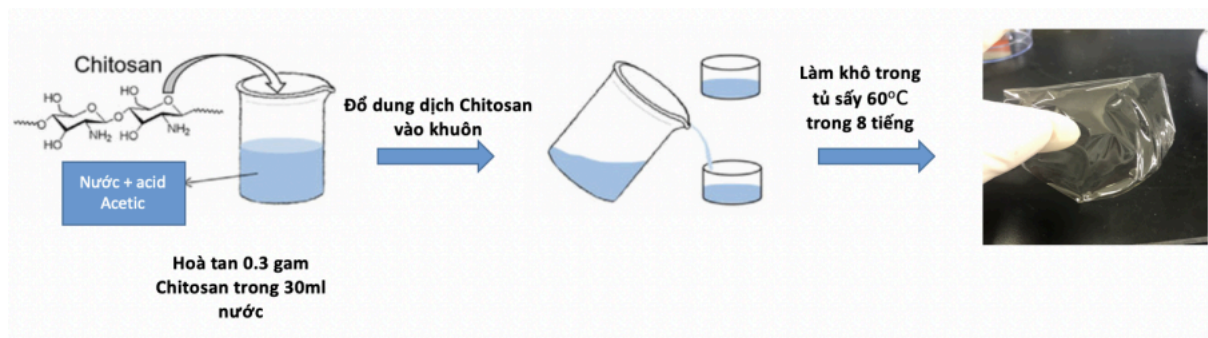


Figure 2.2. Schematic diagram of Chitosan membrane.

- Dissolve 0.3g of Chitosan in 30ml of H_2O (1% CH_3COOH) using a magnetic stirrer.
- When the Chitosan solution is completely dissolved, pour 30ml of the solution into a mold. Next, place the mold in an oven at 60°C for 8 hours, then remove it to cool. After that, separate the Chitosan membrane from the mold. The Chitosan membrane is cut into small pieces for IR, UV-Vis, SEM spectroscopy, and for designing the electricity generation device.

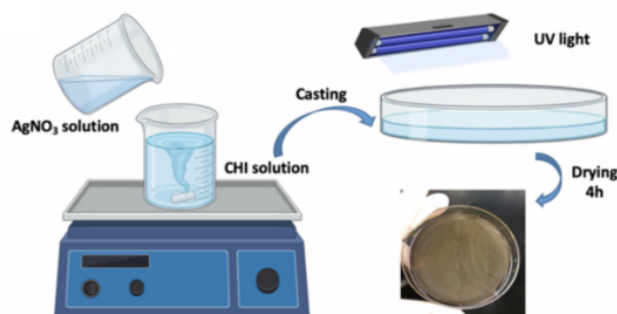


Figure 2.3. Schematic diagram for the preparation of CHI–AgNPs cast membrane.

- Chitosan is used as a stabilizing and reducing agent to synthesize silver nanoparticles from silver nitrate. Therefore, first, dissolve the AgNO₃ solution in 1% Chitosan (CH₃COOH) solution using a magnetic stirrer. When the silver salt is dissolved in the acidified chitosan solution, the silver ions bind to the polymer chain through amino groups, resulting in the formation of silver particles attached to chitosan.
- When the two solutions are completely dissolved, pour them into a mold and use a UV lamp to dry for 4 hours.

2.3.2. Design process for the electricity generation device

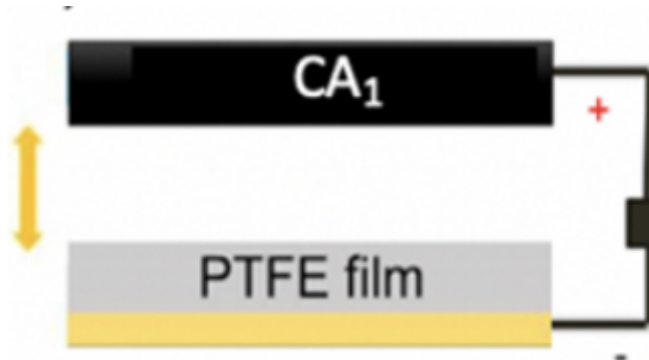


Figure 2.4. Layout design of TENG.

An electricity generation device consists of a Chitosan layer acting as the positive triboelectric layer and an aluminum layer attached to a PolyTetraFluoroEthylene (PTFE) layer as the negative triboelectric layer. Wires are connected to the two aluminum electrode layers. The thin layers are of the same size, 2*2cm². The electricity generation device is connected to a digital oscilloscope (MDO3052, Tektronix) to measure voltage and current intensity.

2.3.3. Study on the functionality of the electricity generation device using Chitosan membrane in the treatment of Parkinson's disease.

Study on the biocompatibility of the TENG device: Experiments were conducted to evaluate the biocompatibility and safety based on three aspects: Lifespan of *C. elegans*, body size, and reproductive ability. Statistical analysis of lifespan was performed using Kaplan-Meier survival analysis. Body size images of N2 *C. elegans* strains with and without ES on day 1 of adulthood. Body size was assessed by the computer image analysis program, Image J of the parent individual size of Wild-type N2 on day 1 of adulthood (**p < 0.001).

Study on the ability to improve Parkinson's related symptoms: Previous studies have shown that dopaminergic neurons have the ability to detect mechanical stimuli from the external environment, thereby modulating behavior through the interaction between neurons and motor neurons, leading to a decrease in *C. elegans*' movement speed. Specifically, when *C. elegans* moves on a smooth surface in a food-free environment and comes into contact with a food-containing bacterial surface, dopamine neurons detect changes in the environment and slow their movement. Based on this principle, an experiment called the basic slowing response was performed to assess the effect of electrical stimulation (ES) on dopamine neurons.

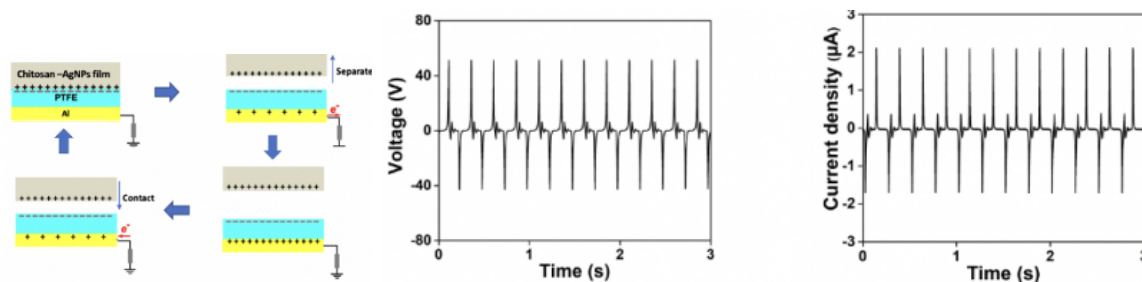
In the experiment, two *C. elegans* strains were used: wild-type N2 and CB1112, in which the *cat-2* gene (coding for the enzyme producing levodopa, the precursor of dopamine from tyrosine) has been knocked out. Each worm was treated with ES (** $p < 0.001$, $n = 25 \sim 30$). ES was performed at the L4 stage, and the movement speed was verified 24 hours later, with the results expressed as a percentage.

Study on the ability to activate dopamine transport: 6-Hydroxydopamine (6-OHDA) is a known neurotoxin that causes Parkinson's disease in *C. elegans*. In this experiment, *C. elegans* was treated with 6-OHDA at a concentration of 25 mM. The transgenic strain Pdat-1::GFP BZ555 was used to quantify the expression of the gene encoding the dopamine transporter (DA) *dat-1*, expressed in two pairs of ADE and CEP cells as well as one pair of PDE DA neurons. The fluorescence intensity was quantified from the DA transporter and assessed via image analysis program.

Study on the expression of genes related to neuroprotective mechanisms in *C. elegans*: 6-Hydroxydopamine (6-OHDA) was used to induce damage to dopamine neurons, followed by ES treatment. Worms were treated with ES (* $p < 0.05$, ** $p < 0.01$, $n = 20 \sim 30$). After 24 hours, the expression levels of the *cat-2* gene, a gene related to dopamine synthesis by encoding the enzyme tyrosine hydroxylase (TH), and the *dat-1* gene, a gene related to the reuptake of dopamine into presynaptic neurons, were analyzed using quantitative real-time polymerase chain reaction (RT-qPCR).

III. RESULTS AND DISCUSSION

3.1. Material characteristics



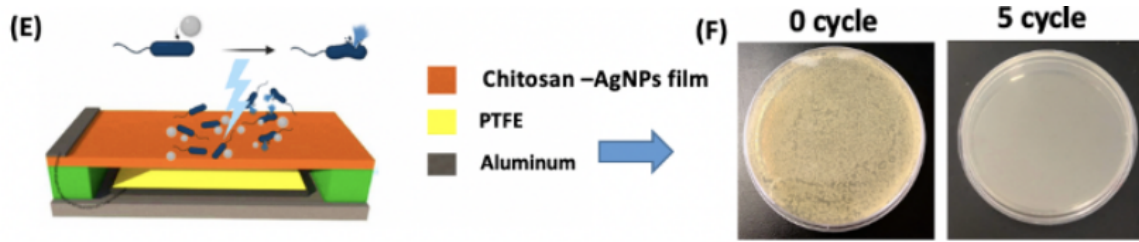


Figure 3.1: Electrical Output Performance of CHI-AgNPs Cast S-TENG-Based Film. (A) TENG design layout and the triboelectric power generation mechanism using CHI-AgNPs membrane. (B-C) Voltage and current density of CHI-AgNPs membrane. (D) Proposed mechanism to enhance antibacterial performance. (E) Antibacterial results after each cycle against *Escherichia coli* strains.

Figure 3.1A describes the power generation mechanism of the S-TENG electrical stimulation device, which combines contact electrification and electrostatic induction. When the CHI-AgNPs film comes into contact with the PTFE membrane, the electron displacement creates a current flow when the two membranes separate. The output performance results of the TENG are shown in Figure 3.1B and 3.1C, with an output voltage of 40V and a current density of $2.22 \mu\text{A}/\text{cm}^2$. This value demonstrates the potential application of the device as an electrical stimulation (ES) source.

We also examined the sterilization capability of the TENG device. Figure 3.1D illustrates the antibacterial mechanism: the CHI-AgNPs membrane acts as an electrode in contact with bacteria, increasing antibacterial efficiency. The results in Figure 3.1E show that after 5 TENG cycles, bacteria were completely eliminated, proving that antibacterial effectiveness was significantly improved due to electrical stimulation.

3.2. Biocompatibility Study of the TENG Device

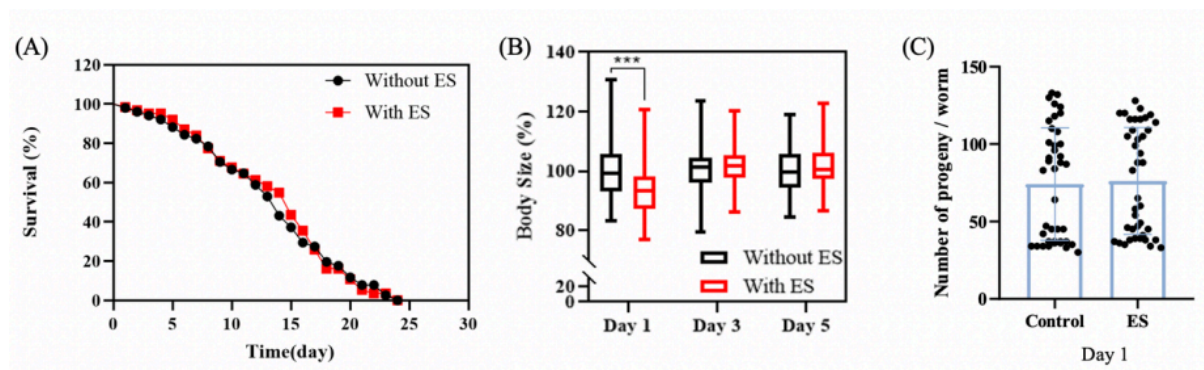


Figure 3.2: Biocompatibility of ES in *C. elegans*. (A) Lifespan analysis. (B) Body size measurement. (C) Parental body size of wild-type N2 worms on day 1 of adulthood ($p < 0.001$).

Since our treatment method relies on electrical stimulation from the TENG device, ensuring biocompatibility is crucial. Experiments were conducted to evaluate biocompatibility and safety. Electrical stimulation was applied to the wild-type N2 strain at the L4 stage, and there was no significant difference in lifespan between the stimulated nematodes and the control group (Figure 3.2A). The average lifespan of the stimulated nematodes was 13.33 ± 5.46 days, slightly higher than the control group (12.92 ± 5.68 days), showing that ES is safe. Body size changes were evaluated on days 1, 3, and 5 after stimulation; although body length decreased on day 1 ($94.02 \pm 9.06\%$), it recovered on day 3 ($102.04 \pm 7.08\%$) and day 5 ($101.20 \pm 7.38\%$) (Figure 3.2B). This result shows that the ES method does not affect *C. elegans* growth. Regarding reproductive ability, ES-treated nematodes laid an average of 76.23 ± 34.58 eggs per day, higher than the control group (74.2 ± 36.35 eggs) (Figure 3.2C), with no significant difference, confirming the safety of ES through the TENG device.

3.3. Investigation of the TENG Electrical Stimulation Device's Effectiveness in Improving Parkinson's-Related Symptoms

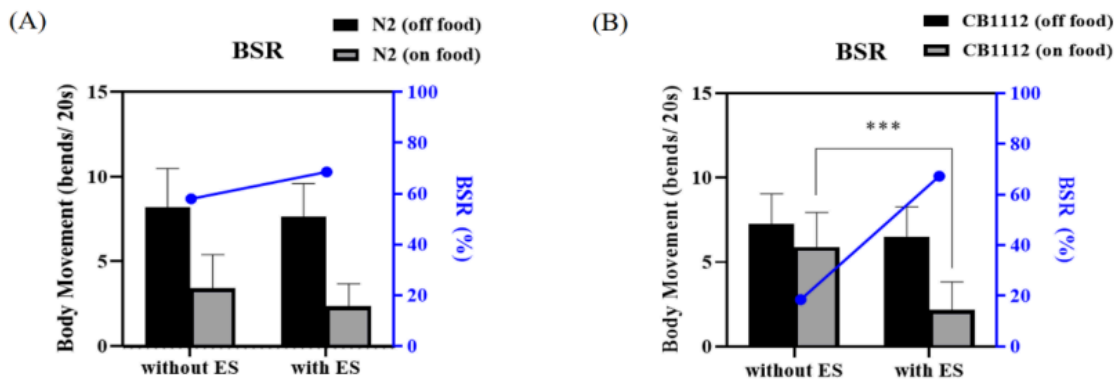


Figure 3.3: Body Movement and Baseline Slowing Response (BSR) Percentage. Wild-type N2 (A) and CB1112 (B) worms were used in this experiment. Each worm was treated with ES ($p < 0.001$, $n = 25 \sim 30$).

The results from the experiment showed that in the wild-type N2 strain, both the ES-treated and non-ES groups exhibited reduced movement speed when transitioning to a bacterial environment. Specifically, the percentage decrease in movement speed was 69.11% for the ES-treated group and 58.53% for the non-ES

group. Although there was a reduction in movement speed in the ES-treated group, the difference between the two groups was not statistically significant (Figure 3.3A).

For the CB1112 strain, due to the impaired ability to produce dopamine, the non-ES group did not exhibit a significant reduction in movement speed compared to the wild-type N2 strain when transitioning to the bacterial environment. This result may be explained by dopamine neuron damage in the CB1112 mutant strain. However, in the ES-treated group, the reduction in movement speed in the bacterial environment was similar to the wild-type group. When calculated as a percentage, the ES-treated group showed a 66.87% decrease in movement speed, whereas the non-ES group only showed an 18.78% decrease (Figure 3.3B). This suggests that behavioral symptoms related to dopamine neuron damage were effectively improved by the ES method.

However, since no significant increase was observed in the experiments with the wild-type N2 strain, where no dopamine neuron damage occurred, it may be assumed that this method is more effective on damaged dopamine neurons than on normal neurons. Therefore, it can be concluded that the ES method using our new TENG device is more effective for treating Parkinson's disease rather than preventing it.

3.4. Investigation of Dopamine Transport Activation Using the S-TENG Electrical Stimulation Device

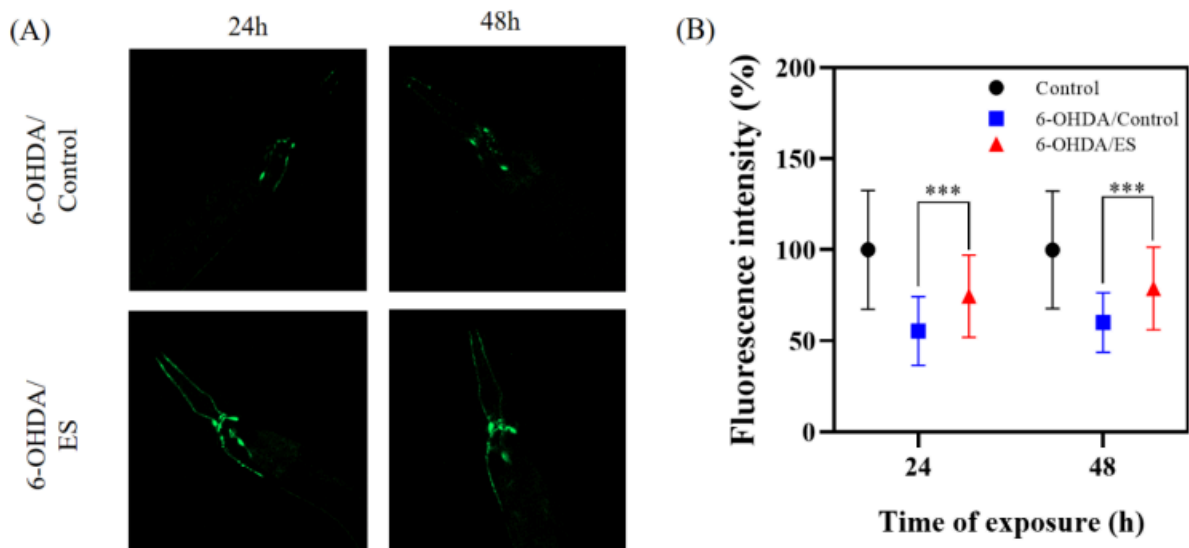


Figure 3.4: Fluorescence Imaging of Dopaminergic (DA) Neurons in BZ555 (A) and DA Transport Enhancement by ES (B). Results are presented with statistical significance levels (** $p < 0.05$, $p < 0.01$, $n = 20 \sim 30$).

Compared to the control samples at days 1 and 2 that were not treated with electrical stimulation (ES), the expression level of *dat-1* in ES-treated nematodes was significantly higher (Figure 3.4A). When taking the control group (100%) that was not treated with 6-OHDA and did not receive ES as the standard, nematodes treated with 6-OHDA showed expression levels of only $55.38 \pm 18.82\%$ after 24 hours and $60.15 \pm 16.35\%$ after 48 hours, indicating a decline in fluorescence expression.

In contrast, in the ES-treated group, the expression level tended to improve significantly, reaching $74.57 \pm 22.58\%$ after 24 hours and $78.75 \pm 22.63\%$ after 48 hours (Figure 3.4B). These results suggest that the ES method has the potential to protect DA neurons from damage caused by 6-OHDA.

3.5. Investigation of Gene Expression in the Dopamine Metabolic Pathway Using the TENG Electrical Stimulation Device

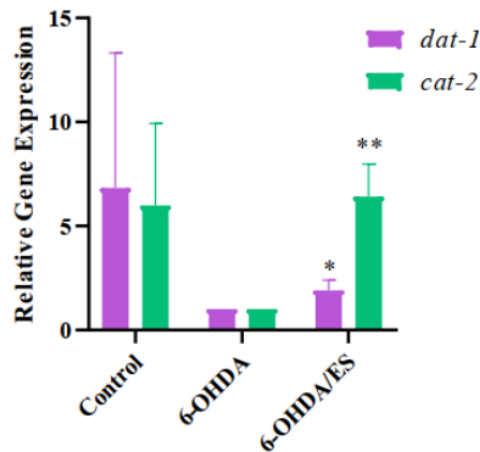


Figure 3.5: Increased relative gene expression in dopamine pathway due to ES. Nematodes were treated with ES (* $p < 0.05$, ** $p < 0.01$, $n = 20 \sim 30$). Increased relative gene expression in dopamine pathway genes (* $p < 0.05$, ** $p < 0.01$).

Through previous experiments, we obtained results showing that the electrical stimulation (ES) method can restore damaged dopamine neurons and effectively improve behavioral symptoms. To investigate the neuroprotective mechanism of *C. elegans* through ES, we conducted additional experiments to analyze the expression of genes related to this mechanism.

The analysis results showed that ES treatment led to a significant increase in the expression level of the *cat-2* gene and the *dat-1* gene, from 1 to 6.42 ± 1.28 and 1.92 ± 0.4 , respectively, in nematodes damaged by 6-OHDA (Figure 3.5). These results suggest that ES has a therapeutic effect on Parkinson's disease by enhancing the expression of these two genes, which are involved in the levodopa conversion

process in the dopamine metabolic pathway and dopamine reuptake pathway. Furthermore, with increased *cat-2* gene expression, it may be considered a key factor in activating the levodopa conversion process.

IV. CONCLUSION AND FUTURE DIRECTIONS

1. Conclusion

We successfully designed a TENG power generation device based on the principle of converting mechanical energy into electrical energy, utilizing a chitosan membrane as the triboelectric positive material. The CHI-AgNP (CA1) nanocomposite membrane was successfully synthesized using a casting method, demonstrating its effectiveness as an electrode material. The TENG device utilizing this membrane exhibited promising output performance (up to 40V and 2 $\mu\text{A}/\text{cm}^2$) along with enhanced antibacterial activity due to electrical stimulation (ES).

Experiments on *Caenorhabditis elegans* confirmed the safety of S-TENG stimulation, with subsequent tests indicating significant improvements in Parkinson's-related symptoms. The study also demonstrated that electrical stimulation did not significantly affect the lifespan, body size, or reproductive capacity of *C. elegans*. The dopamine synthesis process, involving the *cat-2* and *bas-1* genes, suggested that electrical stimulation improved the slowing response rate of the nematodes to 66.87%, compared to 18.78% in the control group. Additionally, the stimulated group exhibited significant recovery in dopaminergic neuronal fluorescence compared to the control group. Electrical stimulation also increased the expression of *cat-2* and *dat-1*, demonstrating the potential of TENG electrical stimulation devices for non-invasive Parkinson's disease treatment.

2. Future Directions

- Expand the research to a mouse model to evaluate the potential application in animals with more complex nervous systems.
- If the results in higher-order animals are promising, proceed to testing on neurons differentiated from patient-derived stem cells in Parkinson's disease.
- Investigate the antibacterial potential of the self-powered TENG device in any conditions.

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