Water quality improvement agent(new type CAC-717) also acts as a prion disinfectant

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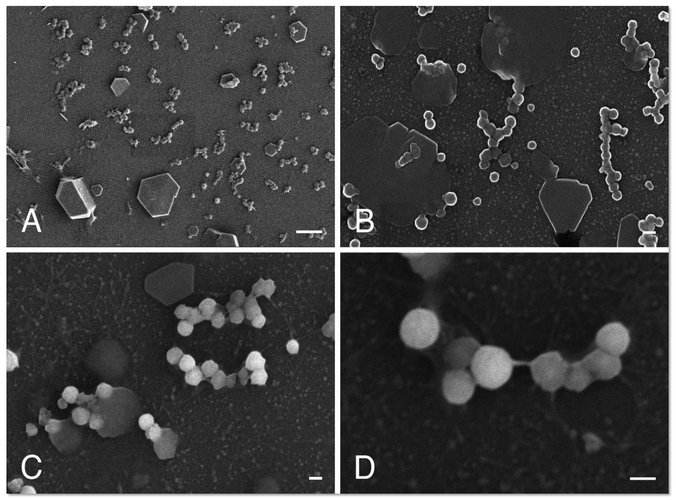
Introduction

In Prion2018 we reported electrically charged material (CAC-717) by continuously applying an electric field to mineral water containing calcium hydrogen carbonate. A Teflon insulation-coated electrostatic field electrode (N-800N, Mineral Activation Technical Research Center; Japan Patent No. 5864010) was used to create the electric field at a voltage of 2×10^4 V for 48 h. CAC-717 solution in distilled water (Japan Patent No. 5778328, FDA/USA Regulation No. 880.6890 Class 1 disinfectant) had a pH of 12.39 ± 0.03 and contained calcium hydrogen carbonate particles (1,120 mg/L) and carbonate complex microparticles (50–500 nm) with a mesoscopic structure. CAC-717 works as a soil amendment was evaluated by growth of green tea leaves in Japanese farm lands. In Prion2019 we report that CAC-717 absorbed by ceramics (Fig. 1) were applied to fresh or sea water to work as water quality improvement agents.



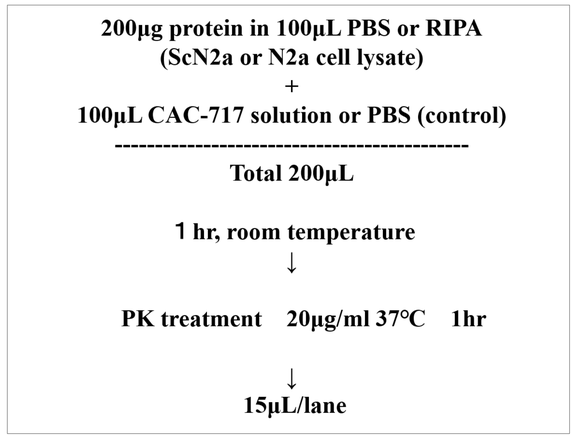
(Fig. 1)

(A)Scanning electron microscope image of the mesoscopic structure of calcium hydrogen carbonate. Spheroid structures were created after high-voltage treatment of aqueous solutions. Scale bar 1000 nm.

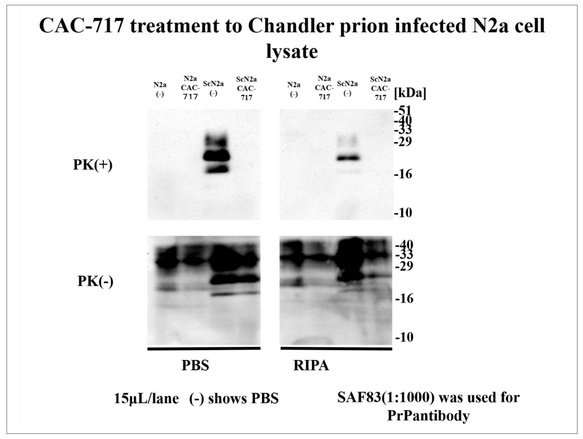
(B) Higher-magnification scanning electron microscope image of the mesoscopic structure of calcium hydrogen carbonate. Scale bar 100 nm.

(C) Higher-magnification scanning electron microscopic image of calcium hydrogen carbonate. Chain-like spheroid structures were observed with small hexagonal formations. Picture is taken with stronger electric voltage. Scale bar 100 nm.

(D) Small fiber structures connecting the spheroid structures to form the chain-like structure. Scale bar100 nm.



(Fig. 2)



(Fig. 3)

CAC-717 as a prion disinfectant

Mouse prion (Chandler) infected N2a cell (ScN2a) lysate was subsequently mixed with equal volume of CAC-717 solution. As a negative control, uninfected N2a cell lysate was diluted 2-fold with PBS or RIPA buffer. 200 µg lysate protein was diluted in 100 µg of buffer (PBS or RIPA) and mixed with 100 µℓ of PBS. Mixtures were incubated at room temperature for 1 h then immediately diluted in PBS before determining the amount of prion in each sample for Western blotting with SAF83 monoclonal antibody. Prion was not detected in the ScN2a cell line lysate mixed with CAC-717 solution after testing with Western Blotting. However, PrP^C levels was intact in both uninfected N2a cell and infected ScN2a cells, detected by Western Blotting (Fig. 2-3).

Water quality improvement agent

CAC-717 ceramics placed inside concrete blocks were sprayed into sea water by the seashore of Shimane Prefecture, Japan. Then alteration of water environment was observed for 6 months. Number of sea urchin, sea cucumber, abalones, and Sazae (Fig. 4-6) were calculated inside the concrete blocks with CAC-717 ceramics. Yield, as calculated by the number of these marine animals, was then determined. No animal was observed in the blocks with ceramics without CAC-717. While large amounts of animals were attracted and growing inside the blocks with CAC-717 ceramics.

(Fig. 4)

(Fig. 5)

(Fig. 6)

Sterilizing action

Ongoing experiment shows that CAC-717 coated ceramics are effective in sterilization against many kinds of bacteria and viruses (Kirisawa et al, personal communication). Salked lime became hardened in the shape of board, but CAC-717-coated ceramics remained as soft powder until the end of experiment (6 months post-scattering). This sterilizing action was not decreased by coexisting proteins as reported previously for influenza viruses (1). The alkalinity of feces was measured after mixing with CAC-717 solution. Initially, CAC-717 in dishes had a pH of 12.1 ± 0.3 (n=4) tested with a pH meter. CAC-717 and the bovine feces mixture (0.1 mL) were placed in Petri dishes. After 10 min, the solution was measured with a pH meter. The pH reading on the meter was 12.0 ± 0.3 (n=4). There was no significant difference in pH reading between the CAC-717 solution and CAC-717-feces mixture after 10 min.

Evaluation

Evaluation of water environment, nutrient content, composition, and microbiological activity tend to increase following use of water quality improvement agents. Numerous studies have therefore to be examined for the effect of water quality improvement agent, beside animal yield and quality. Our future aims are to determine the microbiological activity of CAC-717 ceramics after spraying with CAC-717.

Reference
1. Nakashima R, Kawamoto M, Miyazaki S, Onishi R, Furusaki K, Sakudo A, Onodera T. (2017) Evaluation of calcium hydrogec carbonate mesoscopic crystals as a disinfectant for influenza A viruses. J Vet Med Sci 79:939-942.