

Variation of nitrite reductase gene *nirS* in denitrification process

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This study explores the use of microbial community analysis to evaluate the processes involved in nitrate attenuation in groundwater. Real-Time PCR (Polymerase chain reaction) is used to quantify nitrite reducing genes (*nirS*). It is suggest that the new method for detecting denitrification activity by comparing the gene dosage that has been detected by Real-Time PCR and the value of the nitrate $\delta^{15}\text{N}$ and $\delta^{18}\text{O}$ is effective. This study focuses on a variation of the nitrite reductase gene (*nirS*) that has been detected by Real-Time PCR through at the denitrification process by the column experiment.

Acrylic column which was used in the experiment is height 70cm, an inner diameter of 7cm. The bottom of the column was packed with crushed Ryukyu limestone, the upper was filled with soil. The analysis items, in addition to the DNA copy number of *nirS*, was selected inorganic nitrogen (NO_3^- , NO_2^- , NH_4^+), Total Organic Carbon (TOC), Inorganic Carbon (IC) and the nitrate $\delta^{15}\text{N}$ and $\delta^{18}\text{O}$.

As a results of the column experiment, oxidative environment had been maintained at the column packed with Ryukyu limestone. On the other hand, the formation of the reducing environment had been confirmed at the column packed with soil.

The variation characteristics of the nitrite reductase gene *nirS* in the denitrification process was understand by column experiment. In addition, a differences as the index of denitrification between *nirS* and the nitrate $\delta^{15}\text{N}$ and $\delta^{18}\text{O}$ was revealed.

Keywords: Denitrification , Nitrite reducing genes (*nirS*), Real Time-PCR