

学 位 論 文 要 旨	
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題 目	Studies on the interaction between bacteria and protozoa in the hydroponic environments (水耕栽培環境における大腸菌と原生動物の相互作用に関する研究)
<p>To clarify the threshold cell density for the internalization of <i>Escherichia coli</i> into hydroponic vegetables, <i>E. coli</i> isolated from cow dung was inoculated into hydroponic medium and determined the dynamics of <i>E. coli</i> and protozoa were investigated.</p> <ol style="list-style-type: none">1. To investigate internalization of <i>E. coli</i> into hydroponic leafy vegetables, <i>E. coli</i> was inoculated into hydroponic medium during cultivation. <i>E. coli</i> was revealed to internalize into lettuce, spinach, and Komatsuna plants, while not into small green onion plant. Cell density of <i>E. coli</i> was decreased markedly during cultivation in all plants. To clarify the cause of quick decrease in <i>E. coli</i> population, cell density of <i>E. coli</i> was determined by using plate counting with coliform agar and the μC-FISH method. Cell density of <i>E. coli</i> decreased markedly by as much as 4 to 8 orders of magnitude during 7-day cultivation. Notably, very little difference was observed between counts by the plate method vs. the μC-FISH method. These results suggest that the decrease in <i>E. coli</i> population was not due to a physiological change into the viable but non-culturable state but true death.2. <i>E. coli</i> was inoculated into intact, 0.2 μm-filtrated or 1.0 μm-filtrated hydroponic medium after plant cultivation. As a result, <i>E. coli</i> decreased markedly in the intact hydroponic medium, but totally not in the filtrated hydroponic medium. Furthermore, fluorescent labeled bacteria (FLB) were prepared from <i>E. coli</i> culture and added into hydroponic medium. FLB was observed to be ingested by protozoa (mostly ciliates and flagellates). These findings suggest that protozoa feed on <i>E. coli</i> inoculated in hydroponic medium.3. To elucidate the threshold cell density of <i>E. coli</i> for the internalization into hydroponic vegetables, antiprotozoal agents (Metronidazole and pyrantel pamoate) were selected and used. Protozoan motility was suppressed markedly at 500 to 1,000 mg/L of metronidazole, but no suppression of protozoan predation. Pyrantel pamoate was relatively water-insoluble and thus dissolved in DMSO. However, it was found that lettuce plant died due to toxicity of DMSO at an effective concentration for protozoa. Therefore, attempts were made to inhibit protozoan predation by using a mixture of the two kinds of drugs, however, the mixture was not effective to inhibit protozoan activity.4. To compensate predation by protozoa so as to maintain cell density of <i>E. coli</i> in hydroponic medium, <i>E. coli</i> suspension was added periodically into hydroponic medium. As a result, cell density of <i>E. coli</i> almost unchanged during 5-day incubation in hydroponic medium, resulting in elucidation of the threshold cell density for internalization into lettuce plant with the presence of protozoa. <p>As mentioned above, the interaction between bacteria and protozoa in hydroponic medium is very important. However, there has been no research focusing on protozoa in hydroponic medium. The results of this study are considered to be able to greatly contribute to further progress of the hygiene management of hydroponic vegetables in the future.</p>	