Brief report

A case of Whipple disease diagnosed by polymerase chain reaction analysis (PCR) of formalin-fixed paraffin-embedded tissues (FFPE) from duodenum

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Background : It was very difficult to diagnose Whipple disease caused by the bacterium Tropheryma whipplei reliably. We tried to confirm this bacterium by a genetic analysis of histopahtolgical specimen.

Case report : 76 year-old male with symptoms of diarrhea, abdominal pain, and weight loss was examined with gastrointestinal endoscope, which revealed atrophic gastritis and white granular appearance in the whole distal duodenum, suggesting lymphoid hyperplasia. Microhistopathology revealed marked macrophage infiltrate in duodenum, suggesting Whipple disease. He was diagnosed as Whipple diseases on the basis of the confirmation of Trpheryma whipplei by PCR analysis.

Conclusion : Genetic analysis, especially real-time method, was very useful and convenient tool for the diagnosis of Whipple disease.

Key words : Whipple disease, duodenum, polymerase chain reaction (PCR), PCR, real-time SYBR Green method, formalin-fixed paraffin-embedded tissues (FFPE), immunohistochemistry

Background

Whipple disease was a rare systemic infectious disease caused by the bacterium Tropheryma whipplei, commonly involved duodenum with malabsorption. This life-threatening disease could be treatable with long-term antibiotic therapy, which required the reliable diagnostic method. Genetic confirmation of this bacterium was expected as one of the most reliable diagnostic tools. The FFPE specimens of duodenum was studied genetically by both the usual PCR followed electrophoresis (uPCR with EP) and the real-time SYBR Green method (real-time PCR), the superiority of which was compared.

Case report

76 year-old male with symptoms of diarrhea, abdominal pain, and weight loss was examined with gastrointestinal endoscope. Chronic atrophic gastritis with Helicobaster pylori and Whipple disease were suggested histologically, the latter of which was speculated because of the massive histiocytic infiltrate immunohistochemically and the intracellular digestion-resistant granules on digested Periodic acid-Schiff stain (Fig. 1-3). Final diagnosis of Whipple disease was confirmed by the following genetic studies : (i) uPCR with EP, (ii) real-time PCR. Deoxyribonucleic acid (DNA) extracts were obtained by the previous method (1-3). The condition of PCR in uPCR with EP was same as the report (1, 2). Tropheryma whipplei 16S rDNA gene from 995 to 1260 was amplified with the primers, produced the final product of 266 bps (4). The concrete technique of Real-time PCR was described in our previous report (3). The specific dissociation peak temperature was 89.68℃. The technical simplicity and its required time in Real-time PCR method was more convenient than those in uPCR with EP.

Conclusion

Genetic analysis, especially real-time method, was very useful and convenient tool for the diagnosis of Whipple disease.

Reference

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和　文　抄　録

短報

十二指腸のホルマリン固定パラファフィン包埋材料（FFPE）を使ったポリメラーゼ連鎖反応遺伝子検査（PCR）によりウイップル病と診断された1例

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背景：Tropheryma whipplei 細菌感染に起因するウイップル病を確実に診断することは非常に難しい。我々は、通常病理組織標本の遺伝分析によってこの細菌を確認し、確診できたので報告した。

症例報告：症例は、下痢、腹痛と体重減少を症状とした76歳の男性で、上部消化管内視鏡検査が施行された。萎縮性慢性胃炎と遠位十二指腸のリンパ球増殖を示唆する白色顆粒状像が認められた。病理組織検査上、十二指腸には組織球の顕著な浸潤が明らかにされ、ウイップル病が示唆された。PCR 分析による Trpheryma whipplei の確認により、ウイップル病と確診された。

結論：遺伝学的分析(特にリアルタイム法)は、ウイップル病の診断において非常に役立ち、かつ、便利な検査法であった。

キーワード：ウイップル病、ポリメラーゼ連鎖反応（PCR)、リアルタイム SYBR グリーン方法、ホルマリン固定パラフィン包埋組織（FFPE)、免疫組織化学

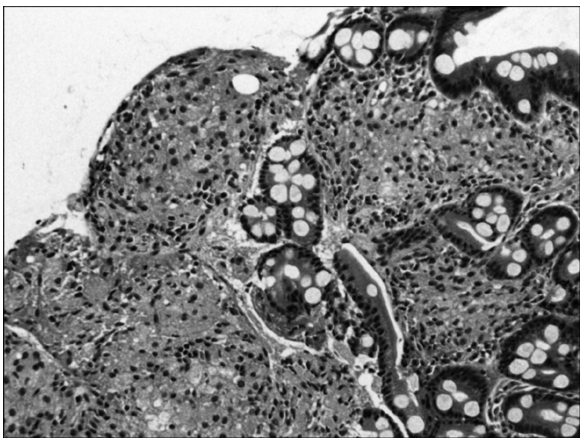

Fig. 1. Marked foamy cell clusters were shown in mucosa by Hematoxyline-eosin stain.

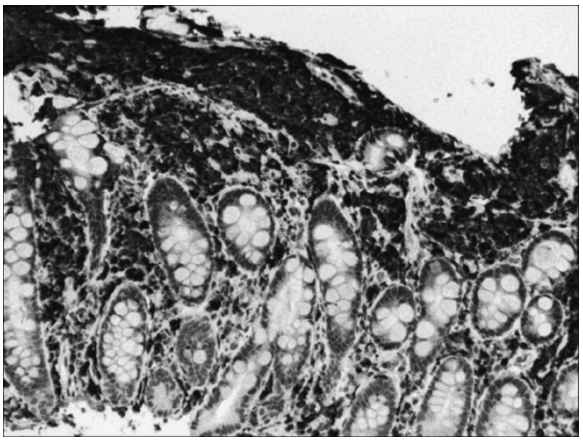

Fig. 2. Marked histiocytic infiltrate was confirmed by immunostain with CD68.

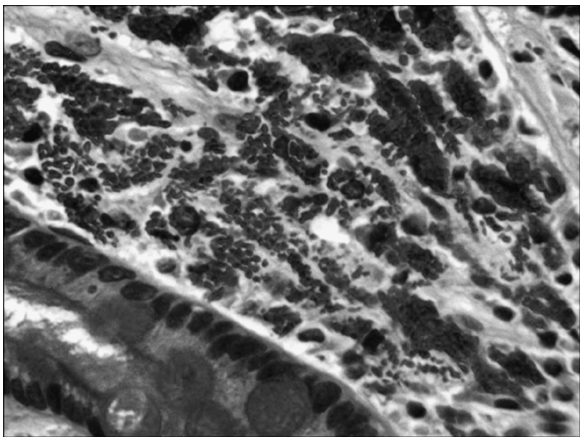

Fig. 3. Positive intracellular granules were found on digested Periodic acid-Schiff stain（PAS）.

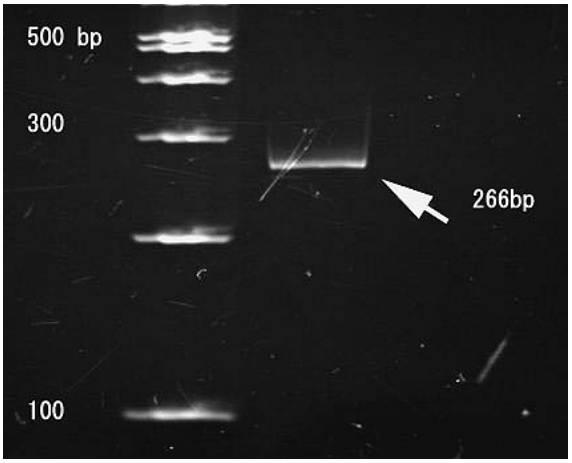

Fig. 4. Single band was found at 266 bp on electrophoresis after polymerase chain reaction.

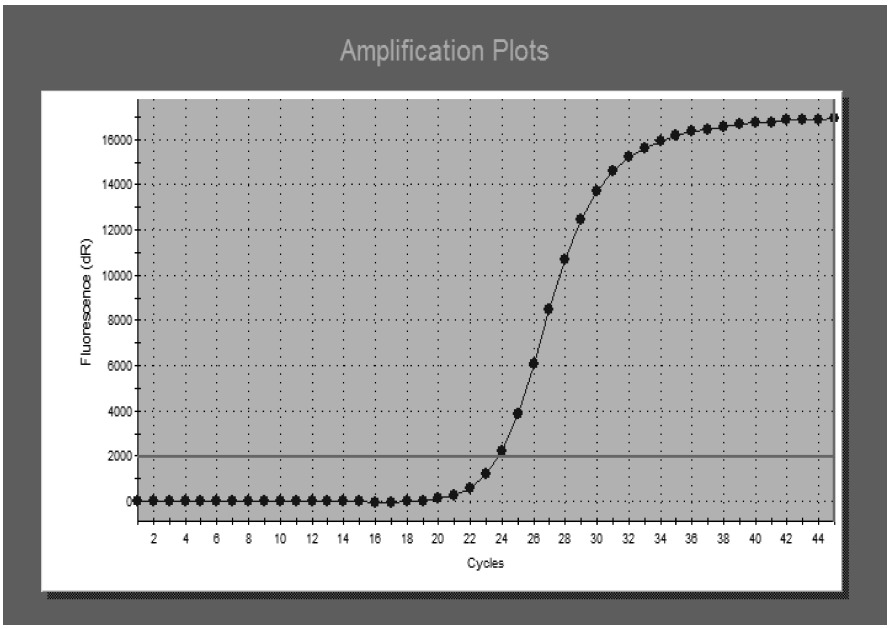

Fig. 5. Amplification elevated at 22 cycles on real-time polymerase chain reaction.

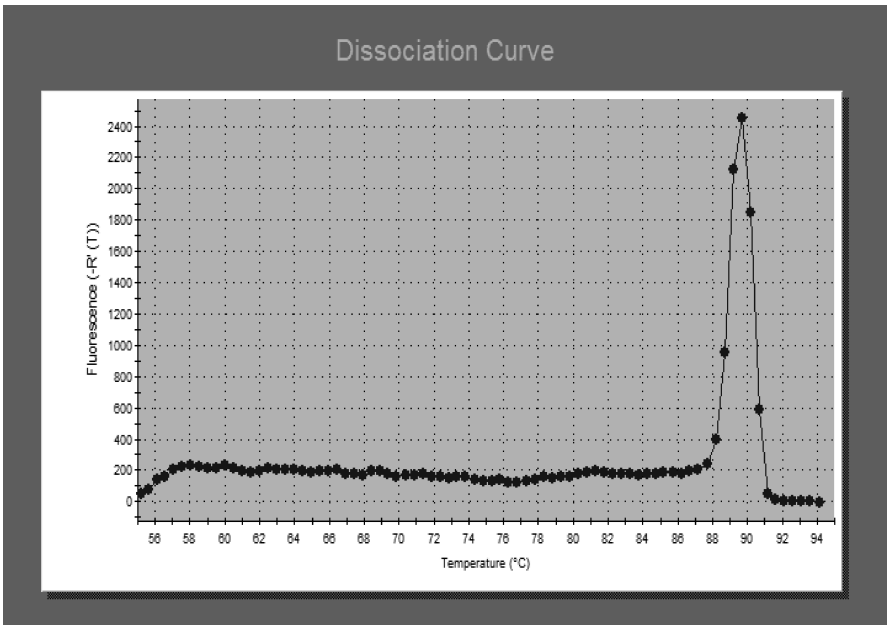

Fig. 6. Dissociation peak temperature was 89.68℃ on real-time polymerase chain reaction.

（2016／12／30受付）