論文審査の結果の要旨及び担当者

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論文審査の結果の要旨

近年，RNA修飾およびその生物学的役割への関心が高まり，次世代シークエンサー等の技術力の向上も相まって，epitranscriptomicsの進歩は目覚ましいものがある．特にRNAメチル化酵素methyltransferase-like 3 (METTL3)およびそれにより誘導されるN^6-methyladenosine (m^{6}A)は，腫瘍生物学において重要な知見が報告されている．m^{6}AはmRNAにおいて最も豊富なRNA修飾であるにも関わらず，膨大な個々の遺伝子にとってのm^{6}Aの重要性は定かでなく，またMETTL3の標的およびその下流のパスウェイの制御メカニズムも未だ十分に解明されていない．

本研究は，膵癌細胞において，METTL3が抗癌剤および放射線抵抗性に関与することを明らかにし，加えてMETTL3が膵癌治療の標的となり得る可能性を示した．加えてmicroarrayの網羅的なデータ解析から，METTL3がmitogen-activated protein kinase (MAPK) cascades, ubiquitin-dependent process, RNA splicing, regulation of cellular processといった生物学的に重要な経路に関与することが示唆された．

本研究は，cancer epitranscriptomeのメカニズムを解明するとともに，膵癌の治療抵抗性を克服するために時宜を得た重要な知見を示すものであり，博士（医学）の学位授与に値するものと考えられる．

論　文　内　容　の　要　旨
Synopsis of Thesis

氏　名 Name	竹藤　晃介
論文題名 Title	The epitranscriptome m^6A writer METTL3 promotes chemo- and radioresistance in pancreatic cancer cells　(RNAメチル化酵素METTL3は膵癌細胞の抗癌剤・放射線抵抗性に寄与する)

論文内容の要旨

〔目　的(Purpose)〕

The N6-methyladenosine (m^6A) is known as the most frequently occurring mRNA modification. Recent years have seen significant progress in the field of m6A. Despite that, it has not been clarified how m^6A writer methyltransferase-like 3 (METTL3) regulates its pathways in downstream.

We focused on METTL3 in pancreatic cancer whose prognosis remains not satisfactory and aimed to clarify the functional role of METTL3.

〔方法ならびに成績(Methods/Results)〕

[Methods]

METTL3 knockdown (KD) MIA PaCa-2 cells (human pancreatic cancer) were established using short hairpin RNA. To clarify functional changes in METTL3 KD cells, proliferation assay, sphere formation assay, chemo-sensitivity assay, radio-sensitivity assay, chemoradiosensitivity assay and Annexin V assay were performed.

To screen gene-expression profiling, cDNA microarray analysis was performed, followed by gene ontology (GO) enrichment analysis and Reactome pathway analysis. Protein-protein interaction (PPI) network was constructed. Subsequently, highly interconnected regions in the network were identified as modules, followed by GO analysis of genes in the modules.

[Results]

Although morphologic and proliferative changes were unaffected, METTL3-depleted cells showed higher sensitivity to anticancer reagents such as gemcitabine (GEM), 5-fluorouracil, cisplatin (CDDP) and irradiation. Chemoradiosensitivity assay showed significant synergistic effects of drug (GEM or CDDP) and radiation in control and METTL3 KD cells. Our data suggest that METTL3 is a potent target for enhancing therapeutic efficacy in patients with pancreatic cancer.

Microarray analysis has revealed that METTL3 was associated with mitogen-activated protein kinase cascades, ubiquitin-dependent process and RNA splicing and regulation of cellular process, suggesting functional roles and targets of METTL3.

〔総　括(Conclusion)〕

METTL3 is associated with therapeutic resistance and is a potential therapeutic target of pancreatic cancer. Additionally, our findings suggest several critical pathways, including MAPK cascades, ubiquitin-dependent process, RNA splicing and regulation of cellular process, as possible targets of METTL3.