

PVD. Histopathological examination of the enucleated eye showed no PVD. Decimal visual acuity improved by 3 lines or more in 6 patients and remained lower than 0.1 in 5 patients.

Conclusion: BRE developed frequently in eyes with no PVD. The absence of PVD may be a risk factor of severe BRE.

P1-07.

Comprehensive genetic analysis of IgG4-related ophthalmic diseases by RNA sequencing

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Purpose: High-throughput RNA sequencing uses massively parallel sequencing that allows unbiased analysis of both genome-wide transcription levels and mutation status of tumors. Immunoglobulin G4-related ophthalmic disease (IgG4-ROD) is a fibroinflammatory disease characterized by enlargement of ocular adnexal tissues, infiltration of IgG4-positive plasmacytes, and elevated serum IgG4 levels. Comprehensive analysis of gene abnormalities in IgG4-ROD may play an important role in discovering new biomarkers. In this study, we analyzed RNA expression levels in biopsy specimens of IgG4-ROD.

Methods: This study included 3 patients who were diagnosed with IgG4-ROD at Tokyo Medical University Hospital. Total RNA was extracted from specimens obtained by biopsy and per-tumor adipose tissues as control, and quantified using NextSeq 500.

Results: By comparing RNA expression levels in the biopsy specimens with those in control tissues and extracting genes with an expression ratio of 16 or more and an expression difference of 16 or more, expression differences were observed in 221 genes. Pathway analysis with these genes revealed a difference in

pathways related to immune systems and extracellular matrix organization. Among them we identified seven genes that were associated with IgG4-ROD.

Conclusion: In biopsy specimens of IgG4-ROD, we identified novel gene abnormalities that are associated with extracellular matrix degradation and B cell receptors. These data may contribute to future development of new biomarkers for diagnosis and molecular-targeted drugs to treat this disease.

P1-08.

Identification of novel microRNAs for distinguishing orbital mucosa-associated lymphoid tissue lymphoma from IgG4-related ophthalmic disease

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Purpose: It is important to distinguish orbital mucosa-associated lymphoid tissue lymphoma (MALT) and benign tumors such as IgG4 related ophthalmic disease (IgG4-ROD) early in the course, since work-up as well as treatment can vary greatly. Although microRNAs (miRNAs) play an important role in the regulation of carcinogenesis and inflammation, the relation between miRNA and orbital lymphoproliferative diseases remains unknown. In this study, we aimed to identify differentially expressed miRNAs and pathways in biopsied specimens and peripheral blood between cases with orbital MALT and IgG4-ROD.

Methods: 38 orbital lymphoproliferative tumors comprising orbital MALT (n=21), IgG4-ROD (n=17) were analyzed by 3D-Gene miRNA microarray.

Results: In serum, IgG4-ROD increased 18 miRNAs, decreased 3 miRNAs compared to MALT. In the tissue, IgG4-ROD increased 23 miRNAs, decreased 3 miRNAs compared to MALT. Pathway analysis with these genes revealed a difference in pathways related to extracellular matrix.

Conclusions : The data indicate that 26 differentially expressed miRNAs in the biopsied 21 of the peripheral blood of orbital lymphoproliferative disorders might be used as biomarkers in the diagnosis of orbital MALT or IgG4-ROD.

P1-09.

化学物質のアレルギー感作性を評価する新規動物実験代替法の開発

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近年、動物福祉の考えから 3R の理念が広まり、感作性試験においても動物を用いない *in vitro* の代替試験法が複数開発され、OECD のテストガイドラインにも採択されている。ところが、これらの代替法は、感作性の有害性発現経路の Key event (KE) 1~3 に相当する初期反応を指標にし、これら複数の方法の組み合わせ (IATA) による評価が必要とされている。我々は、これまでに、生体内の気道上皮を模倣したヒト気道上皮細胞株と末梢血単球由来未成熟樹状細胞 (DC)、繊維芽細胞株を用いた 3 次元 DC 共培養系を開発した。そして、この系を用いて、皮膚と呼吸器の感作性の違いを、DC でのヘルパー CD4⁺ T (Th) 2 分化に重要な OX40 リガンドの発現増強の違いで識別できることを示した (Mizoguchi *et al.* 2017)。本研究では、この系に、さらに、T 細胞を加え、KE4 を指標に、アレルギー感作性・誘発性を評価するための新規 3 次元 DC/T 共培養系を開発することを目的としている。今回は、まず、ヒト末梢血由来の DC と CD4⁺ T 細胞を用いて検討を行った。代表的呼吸器および皮膚感作性化学物質として、Ortho-phtalaldehyde (OPA) と Oxazolone (OXA) を用いて、3 次元 DC 共培養系で刺激 24 時間後、DC の層だけを取り出し、その上にアロジェニックなナイーブ CD4⁺ T 細胞を加え培養後、経時的に RNA を抽出し、リアルタイム RT-PCR により遺伝子発現を解析した。その結果、2 日後には、T 細胞の活性化マーカーである CD69 発現の増強が見られ、OXA 刺激で Th1 分化のマーカーである IFN- γ 発現が上昇したが、OPA 刺激では上がらず Th2 分化マーカーの IL-4 発現が上昇傾

向にあった。5 日後には、逆に、IFN- γ 発現は差が無くなり、OPA 刺激で IL-4 発現が上昇した。7 日後には、どれも、差が殆ど見られなくなった。以上の結果より、この新しい 3 次元 DC/T 共培養系により、KE4 である T 細胞を指標に呼吸器感作性と皮膚感作性の識別が可能である可能性が示された。

P1-10.

Usefulness of newly developed high-speed PCR analysis system called Path-OCTa in the diagnosis of *Clostridioides difficile* infection

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【Introduction】 *Clostridioides difficile* (*C. difficile*) is a pathogen causing antibiotics-associated colitis. Appropriate infection control and management for patients with *C. difficile* is required to prevent nosocomial infection. Immunochromatography with C DIFF QUIK CHEK COMPLETE (Alere Medical) (QUIK CHEK), which detects *C. difficile*-specific glutamate dehydrogenase (GDH) and toxins (CD toxins), is used as the screening test for *C. difficile* infection. However, it has been reported that the sensitivity of QUIK CHEK in detecting CD toxins in stool is relatively low. Therefore, when samples test positive for GDH but negative for CD toxins using QUIK CHEK, stool samples are processed for bacterial culture. After that, if *C. difficile* colonies are obtained, they are tested for CD toxins by the same assay. However, it takes about several days for colonies to form. Therefore, it sometimes leads to delaying diagnosis and inappropriate management of *C. difficile* infection. To solve these problems, we evaluated newly developed high-speed PCR analysis system called Path-OCTa (Metaboscreen) by comparing with real-time PCR analysis as a golden standard for rapid detection of the CD toxin gene.

【Method】 After obtaining approval from the ethics