News Release

Key Points

- The diagnostic process that combined RNA sequencing (RNA-seq) with histopathological examination has shown to improve the diagnostic accuracy of pediatric solid tumors.
- In particular, diagnostic gene mutations were identified in four out of five patients with tumors classified as “undifferentiated sarcoma” without a specific pathological diagnosis, thereby leading to a clear diagnosis.
- We identified a novel *SMARCA4-THOP1* fusion gene and SMARCA4 splice-site mutation in opposite allele, indicating inactivation of the *SAMARCA4* gene, suggesting that it is involved in carcinogenesis.

Summary

A research group led by Professor Yoshiyuki Takahashi, Lecturer Hideki Muramatsu, and Lecturer Yusuke Okuno of the Department of Pediatrics, Nagoya University Graduate School of Medicine; Assistant Professor Yoshie Shimoyama of the Department of Pathology and Laboratory Medicine, Nagoya University Hospital; Daisuke Ichikawa of the Department of Pediatrics, Nagoya Medical Center; and Director Atsuko Nakazawa of the Department of Clinical Research, Saitama Children’s Medical Center performed transcriptome analysis using RNA-seq to assess its clinical utility in the differential diagnosis of pediatric solid tumors.

Pediatric solid tumors are a heterogeneous group of neoplasms with over 100 subtypes. Due to overlapping morphological and immunohistochemical findings and the presence of atypical cases, clinical and histopathological diagnoses remain challenging. We performed RNA-seq in 47 patients with pediatric solid tumors to evaluate the potential utility of including RNA-seq in the diagnostic process. Histopathologists specialized in pediatric cancer re-evaluated pathological specimens to reach a consensus diagnosis; 42 patients were diagnosed with known subtypes of solid tumors, whereas 5 patients were diagnosed with undifferentiated sarcoma that did not match any known pathological features of specific solid tumor subtypes. RNA-seq analysis identified diagnostic genetic variants in four of the five patients with undifferentiated sarcoma. Genetic lesions were detected in 23 patients, including the novel *SMARCA4-THOP1* fusion gene and 22 conventional or recently reported genetic events. These findings suggest that RNA-seq-based genetic analysis may aid in the diagnosis of pediatric solid tumors, and this would be useful for the development of stratified treatment strategies.

Research Background

Pediatric solid tumors are a diverse group of neoplasms, and accurate diagnosis of the tumor subtype is necessary for appropriate patient management. Disease-specific tumor markers are of diagnostic importance; however, most tumors lack specific markers and the diagnosis strongly depends on histopathological evaluation. However, the diagnosis remains challenging for many pediatric solid tumors due to overlapping morphological and/or immunohistochemical features; the presence of unusual clinical, morphological, and immunohistochemical features; and the availability of limited biopsy specimens.

The detection of disease-specific genetic alterations, especially specific fusion genes, can improve the diagnosis of pediatric solid tumors.

In the present study, we performed transcriptome analysis using RNA-seq to assess its clinical utility in the differential diagnosis of suspected pediatric solid tumors.

Research Results

We performed RNA-seq in 47 pediatric patients with solid tumors and detected diagnostic gene mutations in 23 cases, including the novel *SMARCA4-THOP1* fusion gene in 1 case. In this case, in addition to the fusion gene, mutations in the *SMARCA4* gene were also detected in opposite allele, indicating that inactivation of both alleles of the *SMARCA4* gene was involved in carcinogenesis.

In all 47 cases, the histopathological diagnosis was re-evaluated by several pathologists specializing in pediatric solid tumors. Five of the cases were classified as “undifferentiated sarcoma,” because they had no features consistent with the known histological features of the tumor. RNA-seq analysis identified diagnostic genetic variants in four of the five patients with undifferentiated sarcoma. In addition, the RNA-seq results refined the result of histopathological re-evaluation in five additional patients (Fig.1). Overall, RNA-seq changed histopathological diagnosis in nine patients (Fig.1). The detected gene mutations included a *SMARCA4-THOP1* fusion gene, which was not reported previously (Fig.2a). In addition, a splice-site mutation was found on the *SMARCA4* opposite allele in this case (Fig.2b), suggesting that the combination of these mutations causes inactivation of the *SAMARCA4* gene and is involved in carcinogenesis.

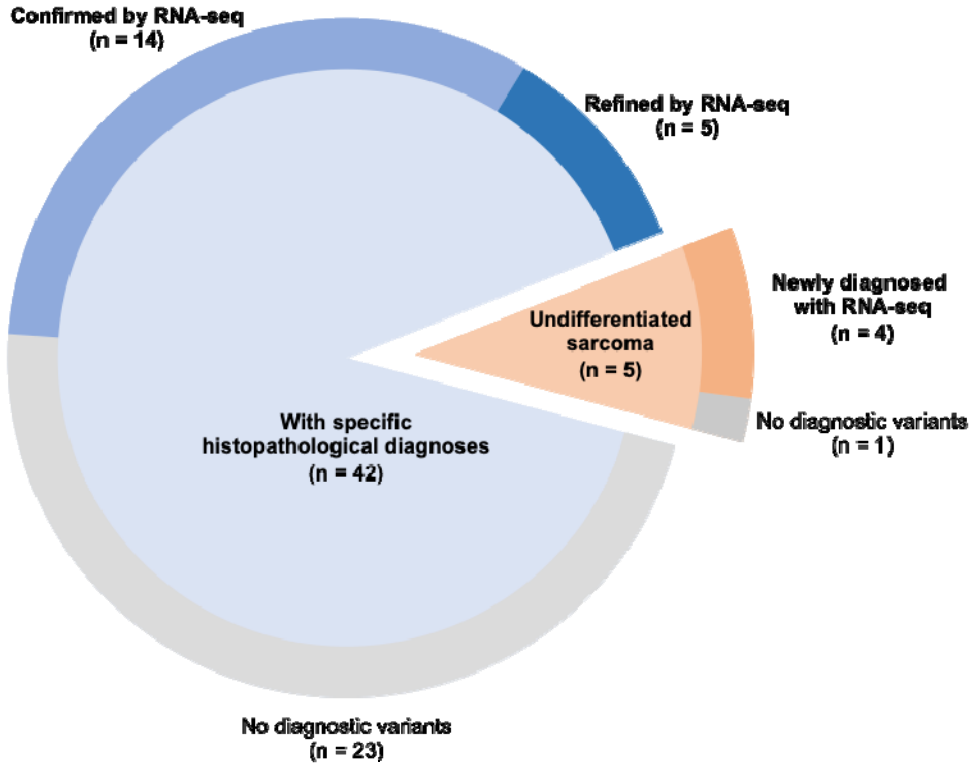

Fig.1

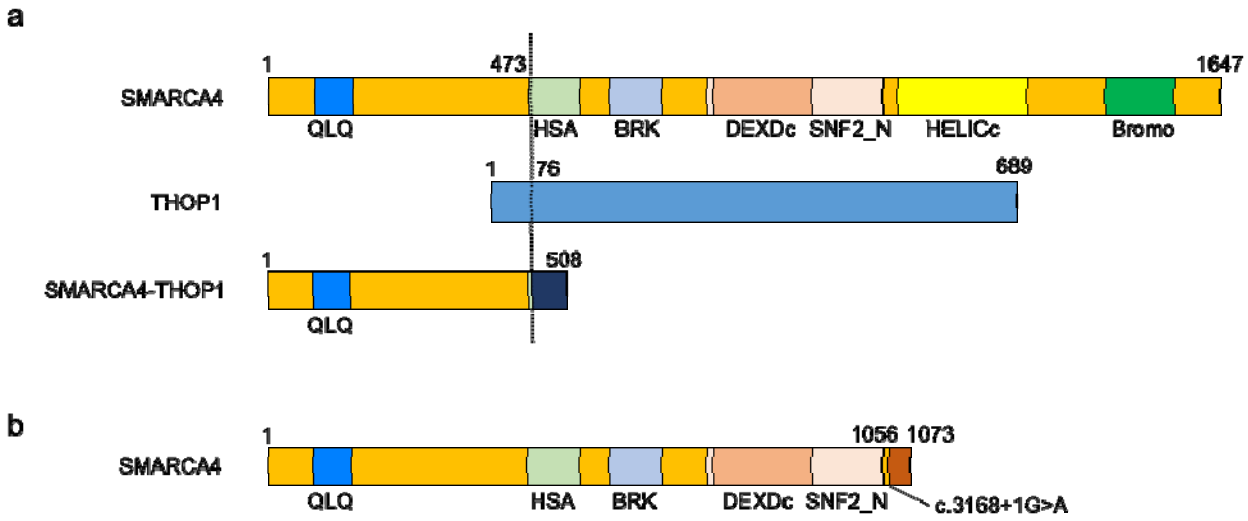

Fig.2

In addition, clustering analysis of some of the cases showed that the cases with rhabdomyosarcoma formed a specific cluster (Fig.3). Gene expression analysis showed that the *MYOG* and *CHRNG* genes were highly expressed in cases with rhabdomyosarcoma (Fig.4). These genes have been reported to be specific to rhabdomyosarcoma, and expression analysis may be useful for diagnosis.

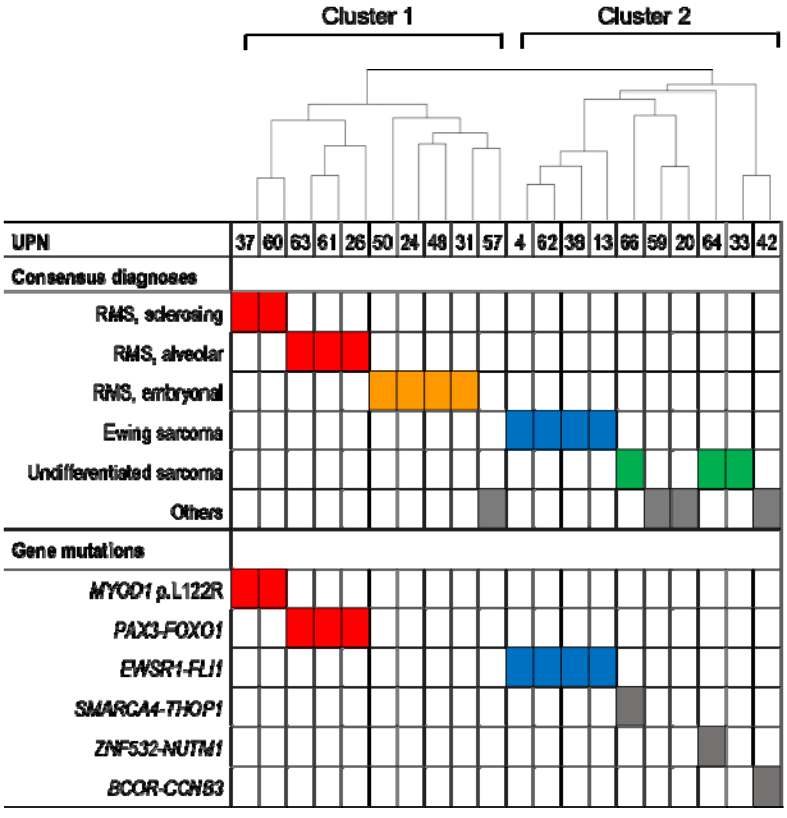

Fig.3

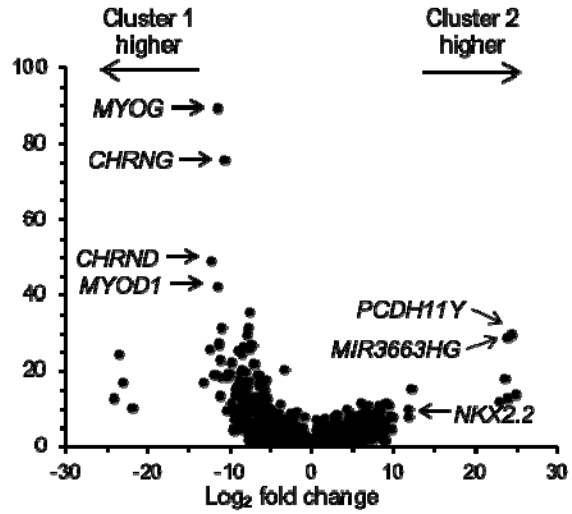

Fig.4

Research Summary and Future Perspective

In the diagnosis of pediatric solid tumors, combining histopathological diagnosis and genetic analysis through RNA-seq will enable more accurate diagnosis. Using the combination of histopathological diagnosis and RNA-seq, we expect to provide appropriate treatments, thereby leading to improved treatment outcomes and reduced treatment complications.

Publication

Journal: npj Genomic Medicine

Title: Integrated diagnosis based on transcriptome analysis in suspected pediatric sarcomas

Daisuke Ichikawa[1], Kyoko Yamashita[2,3], Yusuke Okuno[4], Hideki Muramatsu[1], Norihiro Murakami[1], Kyogo Suzuki[1], Daiei Kojima[1], Shinsuke Kataoka[1], Motoharu Hamada[1], Rieko Taniguchi[1], Eri Nishikawa[1], Nozomu Kawashima[1], Atsushi Narita[1], Nobuhiro Nishio[1,5], Asahito Hama[1], Kenji Kasai[6], Seiji Mizuno[7], Yoshie Shimoyama[8], Masato Nakaguro[8], Hajime Okita[9,10], Seiji Kojima[1], Atsuko Nakazawa[9,11], and Yoshiyuki Takahashi[1*]

[1]Department of Pediatrics, Nagoya University Graduate School of Medicine, Nagoya, Japan.

[2]Department of Pathology and Biological Responses, Nagoya University Graduate School of Medicine, Nagoya, Japan.

[3]Department of Pathology, The Cancer Institute Hospital, Japanese Foundation for Cancer Research, Tokyo, Japan

[4]Medical Genomics Center, Nagoya University Hospital, Nagoya, Japan.

[5]Department of Advanced Medicine, Center for Advanced Medicine and Clinical Research, Nagoya University Hospital, Nagoya, Japan.

[6]Department of Pathology, Aichi Medical University School of Medicine, Nagakute, Japan.

[7]Department of Pediatrics, Central Hospital, Aichi Developmental Disability Center, Kasugai, Japan.

[8]Department of Pathology and Laboratory Medicine, Nagoya University Hospital, Nagoya, Japan.

[9]Department of Pathology, National Center for Child Health and Development, Tokyo, Japan.

[10]Division of Diagnostic Pathology, Keio University School of Medicine, Tokyo, Japan.

[11]Department of Clinical Research, Saitama Children's Medical Center, Saitama, Japan.

DOI：10.1038/s41525-021-00210-y

Japanese ver.

https://www.med.nagoya-u.ac.jp/medical_J/research/pdf/npj_Gen_Med_210615.pdf