（報告様式４）

【16km0405110h0004】
平成 29 年 5 月 31 日

平 成 28 年 度　委 託 研 究 開 発 成 果 報 告 書

I. 基本情報

事　業　名：（日本語）ゲノム医療実現推進プラットフォーム事業
（英　語）Platform Program for Promotion of Genome Medicine

研究開発課題名：（日本語）高齢者発症 AML/MDS における胚細胞変異に基づく個別化医療の確立
（英　語）Personalized medicine for adult-onset AML/MDS based on information of germline variants.

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実 施 期 間：平成 28 年 4 月 1 日　～　平成 29 年 3 月 31 日

分担研究開発課題名：（日本語）バイオバンク・ジャパン検体を用いた胚細胞変異の検索
（英　語）Detection of germline variants using DNA samples of the BioBank Japan

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所属 役職 氏名：（英　語）Michiaki Kubo, Deputy Director, RIKEN Center for Integrative Medical Sciences

分担研究開発課題名：（日本語）小児骨髄悪性腫瘍における胚細胞変異の検索
（英　語）Analysis of germline mutations in pediatric myeloid malignancies

研究開発分担者　（日本語）名古屋大学小児科学講座・助教・村松秀城
所属 役職 氏名：（英　語）Department of Pediatrics, Nagoya University Hospital Assistant Professor, Hideki Muramatsu

分担研究開発課題名：（日本語）コーカサス人種の MDS/AML における胚細胞および体細胞変異の検索
（英　語）Analysis of germline and somatic mutations in Caucasian cases with MDS/AML

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II. 成果の概要（総括研究報告）

和文

京都大学大学院医学研究科腫瘍生物学講座　牧島秀樹講師のグループは、理化学研究所統合生命医科学研究センター　久保充明副センター長のグループと共同して、骨髄異形成症候群(MDS)/急性骨髄性白血病(AML)症例および非悪性腫瘍検体において *DDX41* の胚細胞変異および体細胞変異を検索した。バイオバンクジャパン登録の非悪性腫瘍検体 10,000 人において、欧米とは異なるアジア人独自の胚細胞変異を複数発見し、およそ 1,000 人の MDS/AML 患者のシーケンス結果と比較することにより胚細胞変異の病態への関与を定量的に明らかにした。さらには、およそ 1,000 人の患者検体のシーケンス結果に基づいて特許申請を行った（「ヒトにおける骨髄腫瘍の発症又は発症リスクを検査する指標の取得方法、ヒトにおける *DDX41* 遺伝子の体細胞変異の存在又は将来的な発生を予測する指標の取得方法、並びに、これらの検査又は予測のためのキット」特願 2015-239547）。

続いて、牧島講師は、クリーブランドクリニック　ヤロスヲフ　マチエイェフスキー教授と共同で、欧米人とアジア人において独立して *DDX41* のフレームシフトの胚細胞変異が起こったことを明らかにした。この成果をさらに推し進め、現在、オセアニア地区ではコーカサス人種のアリルが有意にリスクとなっていることを明らかとし、アフリカおよび南アメリカに特異的な独自のリスクアレルが存在することを検出した。さらに、この共同研究グループは、新規胚細胞変異を *GFI1* 遺伝子に検出し、MDS/AML に関して、網羅的なゲノム解析およびマウスモデルを用いた基礎研究により、臨床的・病理学的意義を明らかにし報告した。

さらには、胚細胞変異のみならず体細胞変異もまた計画に沿って研究対象とし、世界最大コホートである、2,500 例の MDS/AML において、網羅的な標的遺伝子解析を京都大学と米国クリーブランドクリニックが共同で実施した。MDS/AML において高頻度に認める体細胞変異に関して病期進行に最も関与するものをバイオマーカーとして抽出した。この成果は Nature Genetics 誌に掲載され(Makishima et al. Nat Genet 2017)、12 月 15 日には AMED と共にプレスリリースを行った(http://www.amed.go.jp/news/release_20161220-01.html)(平成 28 年 12 月 21 日の日本経済新聞に掲載)。

名古屋大学小児科学講座の村松秀城助教のグループは、小児の胚細胞変異の検索を行い、*DDX41* の変異が、先天性の血液免疫疾患においては、有意に濃縮していないことを確認した。さらには、新規胚細胞変異（*RPS15A*、*PIEZO1*、*IL2RG*）を発見し報告した。

京都大学では、新たに発見された *GFI1* 胚細胞変異についてマウスモデルを用いた機能解析研究を行い報告した。さらには、*DDX41* 変異の MDS/AML における病因を明らかにするため動物モデルおよび細胞株を用いた機能解析実験をおこなっている。

英文

Principal investigator, Dr. Makishima, Kyoto University and his group identified novel *DDX41* pathogenic variants in the Asian cases with myelodysplastic syndromes and acute myeloid leukemia (MDS/AML). The group of vice director Kubo, Institute of Physical and Chemical Research sequenced *DDX41* in more than 10,000

healthy donors in Biobank Japan. These Asian variants were different from those reported in US or Europe. Makishima's group compared these variants between healthy donors and more than 1,000 MDS/AML patients to evaluate the effects of the variants on disease predisposition. Clinical implication of these germline mutations of *DDX41* in MDS/AML were patented (#2015-239547).

In international collaboration with professor Maciejewski, Cleveland Clinic, Makishima's group proved that frameshift mutations were originated from independent alleles in Asians and Caucasians. Subsequently, they confirmed that the Caucasian mutant alleles are dominant in Oceania and that original alleles were also in South America and Africa. Their collaboration study also revealed that a novel *GFI1* germline variant was prevalent in MDS/AML cases. They elucidated clinical and pathological impacts of the *GFI1* variant on leukemogenesis by comprehensive genomic analysis and basic functional investigation.

As planned in the grant application, somatic mutations as well as germline mutations were searched in the largest cohort of more than 2,000 cases with MDS/AML. In the collaborative study by Kyoto and Cleveland groups, more frequently mutated genes in secondary AML than MDS were extracted as significant biomarkers for prediction of leukemic evolution. This finding was published in Nature Genetics as an article (Makishima et al. Nat Genet 2017). A press release was made by Kyoto University and Agency for Medical Research and Development (AMED) on Dec. 15, 2016 (http://www.amed.go.jp/news/release_20161220-01.html).

Assistant professor Muramatsu, Nagoya University Department of Pediatrics clarified that *DDX41* germline mutations were not significantly increased in younger MDS/AML. This reproduced the findings in adult MDS/AML cases. Further study of pediatric hematological disorders identified new germline variants in *RPS15A, PIEZO1*, and *IL2RG*.

In Kyoto University, functional significance of *GFI1* and *DDX41* germline mutations were investigated by mouse models. In addition, cell lines with these mutations were utilized for clarification of basic pathogenesis of MDS/AML.

III. 成果の外部への発表

（１）学会誌・雑誌等における論文一覧（国際誌 84 件）

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平成 28 年 12 月 21 日掲載

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