DNA genotek

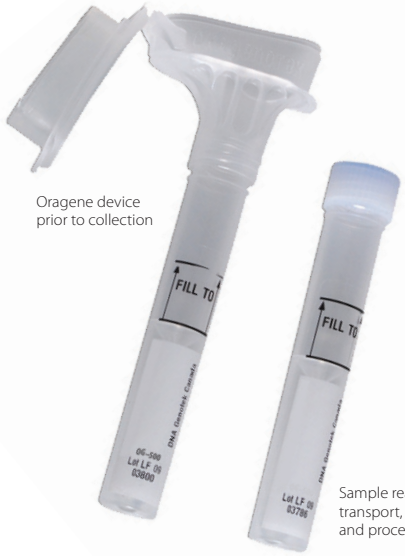
Case study

DNA from saliva offers clues to genetic disorders

Dr. Naomichi Matsumoto
Department of Human Genetics, Yokohama City University Graduate School of Medicine

Study overview

Dr. Naomichi Matsumoto and his team at Yokohama City University's Department of Human Genetics are currently involved in a 3 year project researching genetic diseases, including mendelian disorders. Genetic diseases are caused by a single mutation in the structure of DNA, which results in a single defect with pathologic consequences. More than 4,000 human diseases are caused by single gene defects such as sickle-cell anemia, Tay-Sachs disease and cystic fibrosis. Genetic diseases rarely have effective treatments available. Dr. Matsumoto has been involved in disease-related genome analysis for many years and has seen significant technology advances (exome and Next Generation Sequencing) that will facilitate improved accuracy for gene mutation analysis. It is important to identify the genes responsible for these diseases in individuals and carriers for whom genetic testing and counselling are critical. Dr. Matsumoto plans to analyze 1,000 DNA samples collected from affected individuals and family members over the 3 year period of the study.



Oragene device prior to collection

Sample ready for transport, storage and processing

Main challenges

Affected patients require regular hospital visits and blood draws for comprehensive testing. However, research into genetic disease also requires DNA from family members of the patient. Therefore, the main challenges faced by the research team were collecting samples from family members of affected individuals who did not accompany the patient to the hospital. In addition, the DNA quality had to meet the requirements for PCR sequencing of candidate genes as well as the more stringent requirements of exome sequencing (both the Agilent SureSelect and the Illumina® HiSeq™ 2000 are used for this study). The DNA collection method had to be cost-effective to fall within strict budget guidelines for the project.

Collection methods considered

In addition to whole blood, Dr. Matsumoto looked for alternatives that would facilitate self-collection of DNA samples while meeting the quality standards for his downstream analysis. Oragene®/saliva collection kits were the primary method considered as an alternative to blood sample collection.

DNA Genotek Inc.
2 Beaverbrook Road
Ottawa, ON, Canada K2K 1L1
Subsidiary of OraSure Technologies, Inc.

Toll-free (North America): 1.866.813.6354
Tel.: 613.723.5757 • Fax: 613.723.5057
www.dnagenotek.com
info@dnagenotek.com

Superior samples
Proven performance

Why Oragene/saliva collection kits?

Dr. Matsumoto had used Oragene/saliva collection kits for a previous research project[1]. Based on this experience, he knew the product would meet his requirements for DNA quality and offer a convenient self-collection method for family members. The Oragene kits can also be transported via the standard postal service, facilitating easy and rapid return of the collected sample to the research team for analysis. Oragene offered convenience and increased access to family members.

Results

The Oragene/saliva collection kits are helping Dr. Matsumoto achieve a greater than 95% compliance rate for his study while meeting the requirement for PCR-direct sequencing and even Next Generation Sequencing, when it is required (see Table 1). The data he obtained from the analysis of samples is comparable for results achieved using blood (see Table 2). The data obtained from the samples has already helped him identify culprit mutations in several mendelian diseases including: Coffin-Siris syndrome, spinocerebellar ataxia, HCAHC, early infantile epileptic encephalopathy and more.

Table 1

DNA ID	After purification				
Sample ID	Concentration (ng/µL)	Total amount (ng)	A_{260}/A_{280}	A_{260}/A_{230}	dsDNA concentration (ng/µL)
1	362.9	18145	1.90	2.03	89.2
2	368.1	18405	1.91	1.84	97.4
3	394.7	19735	1.94	1.89	58.8
4	234.2	11710	1.86	2.00	40.7
5	324.2	16210	1.89	1.83	62.3

Measured by Nanodrop
Measured by Qubit (Invitrogen system)

Table 2

Sample ID	Total output	Mean depth	Percent bases above 5	Percent bases above 10	Percent bases above 20	Percent aligned to genome
1	4200958785	125.51	96	94.5	90.6	97.01446718
2	3722946669	111.23	95.7	93.9	89.3	97.39562778
3	4384176351	130.98	95.9	94.5	90.8	95.30240887
4	3690525897	110.26	95.7	94	89.4	96.83854435
5	3720171081	111.14	95.7	94.1	89.6	96.95681714

[1] Yoneda Y., Haginoya K., Arai H., Yamaoka S., Tsurusaki Y., Doi H., Miyake N., (...), Saitsu H. De novo and inherited mutations in COL4A2, encoding the type IV collagen α2 chain cause porencephaly (2012) *American Journal of Human Genetics*, 90 (1), pp. 86-90.

MK-00135 Issue 1/2013-01