Monostotic fibrous dysplasia is a single disorder caused by somatic mosaic activating mutations in GNAS

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Take Home Message
Monostotic fibrous dysplasia is a single disorder caused by somatic mosaic activating mutations in GNAS

Introduction

- Monostatic fibrous dysplasia (MFD) is thought to be caused by somatic mosaic activating mutations in GNAS
- In previous GNAS mutation analyses of MFD patients, direct sequencing using paraffin embedded bone sample detected activating GNAS mutations only in 21 of 40 cases (52.5%)1
- We reported that next generation sequencing (NGS) detected somatic activating GNAS mutations sensitively from peripheral blood leucocytes (PBL) samples in McCune-Albright syndrome2

Objective

To determine if we could detect somatic activating GNAS mutations in MFD patients using direct sequencing of bone samples and NGS of peripheral blood samples

Methods

Participants

< Inclusion criteria>
- Diagnosed as having MFD by pathological study
- Underwent operation at our institution between April 2012 and July 2015

< Exclusion criteria>
We excluded patients with any of the following
- Café-au-lait skin spots
- Endocrine disorder
- More than one lesion of FD on X-ray examination

Detection of somatic activating GNAS mutations

< Direct sequencing of bone samples>
- Material: frozen bone or formalin fixed paraffin embedded sample (FFPE) decalcified by formic acid
- GNAS analyses: Direct sequencing

< NGS of blood samples>
- Material: PBL
- GNAS analyses: NGS and combinatory method of peptide nucleic acids (PNA) probe with NGS

Results

Table 1. Characteristics of 8 patients with MFD

<table>
<thead>
<tr>
<th>Age (years)</th>
<th>Sex</th>
<th>Material</th>
<th>GNAS</th>
<th>GAPDH</th>
<th>Detection of GNAS mutations</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>DS-Bone</td>
</tr>
<tr>
<td>1</td>
<td>10</td>
<td>M</td>
<td>FFPE</td>
<td>-</td>
<td>ND</td>
</tr>
<tr>
<td>2</td>
<td>14</td>
<td>F</td>
<td>FFPE</td>
<td>-</td>
<td>ND</td>
</tr>
<tr>
<td>3</td>
<td>19</td>
<td>M</td>
<td>Frozen sample</td>
<td>+</td>
<td>ND</td>
</tr>
<tr>
<td>4</td>
<td>23</td>
<td>F</td>
<td>Frozen sample</td>
<td>+</td>
<td>ND</td>
</tr>
<tr>
<td>5</td>
<td>34</td>
<td>M</td>
<td>FFPE</td>
<td>-</td>
<td>ND</td>
</tr>
<tr>
<td>6</td>
<td>41</td>
<td>M</td>
<td>Frozen sample</td>
<td>+</td>
<td>ND</td>
</tr>
<tr>
<td>7</td>
<td>42</td>
<td>F</td>
<td>Frozen sample</td>
<td>+</td>
<td>ND</td>
</tr>
<tr>
<td>8</td>
<td>67</td>
<td>F</td>
<td>Frozen sample*</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

DS: direct sequencing, ND: not done

Discussion

- Somatic activating GNAS mutations were detected in all cases by direct sequencing of bone samples and/or by NGS of PBL samples
  - This result indicates that MFD is a single disorder caused by somatic mosaic activating mutations in GNAS
- In cases 1, 2, 5, and 8, neither GNAS nor GAPDH were amplified by PCR using bone samples
  - In cases 1, 2 and 5, formic acid used for decalcification might cause DNA degradation
  - In case 8, severely calcified bone due to repeated surgery might not contain enough DNA to be amplified
- There was a discrepancy in detection probabilities of somatic activating GNAS mutations between previous study and present study
  - Materials (e.g., formic acid, hydrochloric acid) used for decalcification of paraffin embedded bone sample in previous study might cause this discrepancy

< Reference>

Nothing to disclose