Constitutional mismatch repair deficiency syndrome: clinical description in a French cohort


ABSTRACT

Background Constitutional mismatch repair deficiency (CMMRD) syndrome is a childhood cancer predisposition syndrome involving biallelic germline mutations of MMR genes, poorly recognised by clinicians so far.

Methods Retrospective review of all 31 patients with CMMRD diagnosed in French genetics laboratories in order to describe the characteristics, treatment and outcome of the malignancies and biological diagnostic data.

Results 67 tumours were diagnosed in 31 patients, 25 (37%) Lynch syndrome-associated malignancies, 22 (33%) brain tumours, 17 (25%) haematological malignancies and 3 (5%) sarcomas. The median age of onset of the first tumour was 6.9 years (1.2–33.5). Overall, 22 patients died, 9 (41%) due to the primary tumour. Median survival after the diagnosis of the primary tumour was 27 months (0.26–213.2). Failure rate seemed to be higher than expected especially for T-cell non-Hodgkin’s lymphoma (progression/relapse in 6/12 patients). A familial history of Lynch syndrome was detected in 6/23 families, and consanguinity in 9/23 families. PMS2 mutations (n=18) were more frequent than other mutations (MSH6 (n=6), MLH1 (n=4) and MSH2 (n=3)).

Conclusions In conclusion, this unselected series of patients confirms the extreme severity of this syndrome with a high mortality rate mostly related to multiple childhood cancers, and highlights the need for its early detection in order to adapt treatment and surveillance.

INTRODUCTION

The DNA mismatch repair (MMR) system is a highly conserved biological pathway, maintaining genome-wide instability and cancer development in humans. Heterozygous monoallelic germline loss-of-function mutations in one of the four MMR genes (MLH1, MSH2, MSH6 or PMS2) are responsible for Lynch syndrome (LS), an autosomal-dominant genetic disorder associated with an increased risk of colorectal cancer (CRC), endometrial carcinoma and other malignancies in the fourth and fifth decades of life. LS-associated tumours display somatic loss of the remaining wild-type allele, leading to DNA damage, and particularly to microsatellite instability (MSI).

In contrast to LS, rare individuals carry biallelic homozygous or compound heterozygous deleterious germline mutations in MMR genes leading to the recessive constitutional mismatch repair deficiency (CMMRD) syndrome, now recognised as a distinct childhood cancer predisposition syndrome (OMIM #276300). To date, more than 169 patients from 105 distinct families have been reported to exhibit a broad spectrum of childhood malignancies and a cutaneous phenotype commonly described as an association of café-au-lait macules (CALMs) and other features reminiscent of neurofibromatosis type 1 (NF1). The tumour spectrum of CMMRD can be divided into four main groups: haematological malignancies, brain tumours, LS-associated tumours and other malignancies including embryonic tumours.

Recent molecular findings suggest that CMMRD brain tumours are associated with an exceptionally high rate of somatic mutations, far more than in any other cancer. These ‘ultra-hypermutated’ tumours have been shown to harbour causative mutations in POLE and POLD1 genes, resulting in complete ablation of replication error repair and generation of multiple independent subclones due to dramatic accumulation of somatic mutations.

So far, descriptions of this syndrome were mostly based on case reports and were therefore subject to significant bias, as only the most representative cases were considered. As CMMRD is still poorly recognised by clinicians, we undertook a systematic review of all cases diagnosed in French laboratories in order to describe how CMMRD-associated tumours evolve, define criteria for the early diagnosis of CMMRD and identify particular patterns of response and toxicity due to cytotoxic agents in an unselected series of patients with CMMRD.

PATIENTS AND METHODS

Cancer-bearing patients with CMMRD were retrospectively identified via a survey through French networks of Paediatric Oncology (Société Française des Cancers de l’Enfant) and Oncogenetics (Groupe Génétique et Cancer) and all French genetics laboratories involved in MMR gene testing.
Cases were defined as patients with clinically suspected CMMRD and molecular confirmation of biallelic mutation in one of the four MMR genes (PMS2, MLH1, MSH2 or MSH6) in at least one family member. Borderline cases, with a typical clinical presentation but an MMR allelic variant of unknown significance, were included in the description. Each case had been referred to the genetics clinics and a germline analysis had been performed after obtaining informed consent, consistent with the French law. A questionnaire was sent to the main clinician in charge of each patient in order to collect data concerning1 tumour characteristics and outcome,2 non-malignant features,3 the familial history with a pedigree4 and biological results underpinning the diagnosis of CMMRD. The 3-point scoring system for CMMRD diagnostic criteria established by Wimmer et al5 was applied for each case retrospectively. A historical review was performed on available tumour samples and normal tissue (healthy colonic mucosa or skin biopsy). MSI testing and immunohistochemical (IHC) analyses of PMS2, MLH1, MSH2 and MSH6 were performed on available paraffin-embedded tumour sections. For patients who died before the diagnosis of CMMRD, biological data were either obtained from stored DNA or extrapolated from the diagnosis made in siblings who had developed a cancer and from parental genetic data.

Survival rates (event-free survival (EFS), overall survival (OS)) were estimated, using the Kaplan–Meier method. EFS was defined from the date of diagnosis of the first malignancy to the first event (progression, relapse, second malignancy or death) or the date of the last follow-up, and OS rates were estimated from the date of diagnosis of the first malignancy to death whatever the cause, or the date of the last follow-up. The comparison of the curves was performed using the log rank method.

RESULTS

General characteristics

Between 1999 and 2014, a total of 31 patients (14 males and 17 females) from 23 families were diagnosed with CMMRD in five French laboratories: 11 cases have already been reported7 18–20 and 20 are unpublished (table 1). Median follow-up from the date of diagnosis of the first malignancy was 2.7 years (12 days to 25 years). According to the 3-point scoring system established by Wimmer et al5 all but one patient attained a threshold of 3 at the time of the present analysis (median score 6 (2–11)) and all but two patients if the score had been attributed at the diagnosis of the first tumour (median score 5 (2–11)) (table 1).

Tumour spectrum and outcome

Overall, 67 malignancies were diagnosed in 31 patients: 17 haematological malignancies (n=15 patients), 22 brain or spinal cord tumours (n=21) and 25 LS-associated malignancies (n=15) (table 2). Other malignancies included a dermatofibrosarcoma protuberans and two osteosarcomas.

The median age at diagnosis of haematological malignancies and brain tumours was, respectively, 6.6 (1.2–30.8) and 10.3 (3.3–40) years, whereas LS-associated tumours occurred later (median age at diagnosis: 21.4 years (11.4–36.6)) and were mostly represented by colorectal adenocarcinomas (19/25).

A colonoscopy was performed in 16/31 patients in a context of digestive symptoms at a median age of 20 years (11–33.5). None was normal as all 16 patients had polyps varying in number (median 6 (1–17)) and size (median 17.5 mm (5–33)) and at least one was highly dysplastic. Histology had revealed tubulovillous, villous or tubulovillous adenomas. A synchronous CRC was discovered in 13 patients and an abdominal Burkitt lymphoma in another patient. One 14-year-old patient underwent resection of several polyps, but developed CRC 3 years later. One 11-year-old patient underwent colonoscopy as a surveillance intervention since his 13-year-old sister had developed CRC. He underwent a polyp resection but died 1 year later of glioblastoma.

Treatment of cancer and outcome

All patients were treated according to the procedures available in France at the period of the diagnosis without any change in therapy according to the diagnosis of CMMRD, whenever already known, except for one patient who died before treatment and three patients treated abroad. The general outcome of the population is summarised in figure 1. Among the 22 patients who survived their first malignancy, 19 developed a second malignancy, 10 had a third cancer and seven more than three cancers. Overall, 22 (71%) patients died at a median age of 9.5 years (1.25–33.5). Median survival after the diagnosis of the first malignancy was 27 months (0.3–213.2). The 10-year OS and 10-year EFS rates were, respectively, 39.5% (CI 95% 20.9% to 57.6%) and 3.7% (CI 95% 0.3% to 15.8%) (figure 2A). Death was related to the first malignancy in 45% of the cases. All but one patient died of tumour progression: 12/22 patients died of their brain tumour, six patients of a haematological malignancy and three of a digestive adenocarcinoma. One patient died of toxicity related to graft versus host disease after allogeneic haematopoietic stem cell transplantation (HSCT) for acute myeloblastic leukaemia (AML). Five-year OS after LS-associated malignancy was significantly better than after diagnosis of haematological malignancy (p=0.02) and brain tumour (p<0.007) (figure 2B).

Among the 21 patients treated for brain tumours, 17 experienced tumour progression or relapse. For 4/21 patients, a prolonged complete remission (CR) of the brain tumour was achieved (two glioblastomas, one oligodendroglioma and one medulloblastoma) over a median duration of 6 years (1.4–9.7).

Among the 10 patients with T-cell non-Hodgkin’s lymphoma (T-NHL), five died of lymphoma progression: one before starting treatment, one during induction due to primary chemoresistance and three from a relapse after CR. Three other patients were diagnosed with a second tumour during treatment for T-NHL. Four cases of severe toxicity were reported (neurological disorders due to bleeding of a brain venous malformation and thromboembolism during an asparaginase regimen in one patient and severe infections in three other patients). However, given the toxicity described with the regimen designed for these lymphoma subtypes, these toxic events cannot be classified as unexpected.

Among 15 patients with gastrointestinal tumours, seven patients underwent a total colectomy. Despite this radical treatment, two patients developed both a small bowel and gastric carcinoma after their initial CRC. A new malignancy occurred in all patients but one, either in the digestive tract (11/15) or in another site (two hemopathies, one brain tumour and one urinary tract tumour). Finally, only one patient with coloscopic detection of a colorectal adenocarcinoma is in persistent CR after total colectomy.

Overall, 16 patients underwent radiotherapy. Only one case of grade 3 hematotoxicity was reported in a patient who had received irradiation at a dose of 45 Gy for a spinal cord glioblastoma.

The role of chemotherapy and radiotherapy in the occurrence of second malignancies is difficult to assess in this series. It is noteworthy that two AMLs occurred as second malignancies in children previously treated with chemotherapy and that among

<table>
<thead>
<tr>
<th>Family (Pat.)</th>
<th>Gene</th>
<th>Mutation</th>
<th>Familial history</th>
<th>Score</th>
<th>Malignant tumours/polyps (age at diagnosis)</th>
<th>Cutaneous signs</th>
<th>MSI</th>
<th>IHC</th>
<th>Outcome (age at last follow-up)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 (1)</td>
<td>PMS2</td>
<td>c.[400C&gt;T];[400C&gt;T]; 1579del, p. [Arg134*];[Arg134*];[Arg527Glyfs*68]</td>
<td>Consang. No LS</td>
<td>5</td>
<td>Ganglioglioma (21) CRC/polyps (24)</td>
<td>Hypopigmented macules, congenital naevi, blaschkoil hyperpigmentation</td>
<td>MSI-H</td>
<td>PMS2-</td>
<td>DOD (31)</td>
<td>None</td>
</tr>
<tr>
<td>2 (2)</td>
<td>PMS2</td>
<td>c.[2007–2A&gt;G];[20072A&gt;G], p.?</td>
<td>?</td>
<td></td>
<td>No LS</td>
<td>4</td>
<td>TLL (4)</td>
<td>CALMs, hypopigm. macules, freckling, congenital naevi</td>
<td>NA</td>
<td>PMS2-</td>
</tr>
<tr>
<td>3 (3)</td>
<td>PMS2</td>
<td>c.[2007–2A&gt;G];[20072A&gt;G], p.?</td>
<td>?</td>
<td></td>
<td>No LS</td>
<td>4</td>
<td>TLL (5)</td>
<td>CALMs</td>
<td>MSI-H</td>
<td>PMS2—</td>
</tr>
<tr>
<td>4 (4)</td>
<td>PMS2</td>
<td>c.[137G&gt;T];[354–1G&gt;A], p. [Ser46Ile];[?]</td>
<td>?</td>
<td></td>
<td>Father (bladder)</td>
<td>5</td>
<td>CRC/polyps (19) CRC (20) DLBCL (25)</td>
<td>CALMs</td>
<td>MSI-H</td>
<td>PMS2 + (weak)</td>
</tr>
<tr>
<td>5 (5)</td>
<td>PMS2</td>
<td>c.[989-?_1144+?del];[2249G&gt;A], p. [Glu330_Glu381del];[Gly750Asp]</td>
<td>LS-family</td>
<td>6</td>
<td>CRC (22) CRC (32) Polyps (38)</td>
<td>CALMs</td>
<td>MSI-L</td>
<td>NA</td>
<td>CR (42)</td>
<td>None</td>
</tr>
<tr>
<td>6 (6)</td>
<td>PMS2</td>
<td>c.903G&gt;T, r.804_903del, p.Tyr268*</td>
<td>No LS</td>
<td>3</td>
<td>DFS protuberans (3)Glioblastoma (11) Burkitt lymphoma/polyps (17) CRC (18) Glioblastoma (19)</td>
<td>CALMs, hypopigm. macules, congenital naevi, hamartoma</td>
<td>MSI-H</td>
<td>MLH1/- PMS2-</td>
<td>DOD (20)</td>
<td>None</td>
</tr>
<tr>
<td>7 (7)</td>
<td>PMS2</td>
<td>c.[2113G&gt;A];[7067_903+?del], p. [Glu705Lys];[?]</td>
<td>No LS</td>
<td>4</td>
<td>Glioblastoma (6) TLL (7)</td>
<td>CALMs</td>
<td>MSS</td>
<td>MLH1/- PMS2-</td>
<td>DOD (8)</td>
<td>None</td>
</tr>
<tr>
<td>8 (8)</td>
<td>PMS2</td>
<td>c.[2412_07delinsAAAT];[?_7–23+7del], r.[24_163del];[?]</td>
<td>No LS</td>
<td>4</td>
<td>Glioblastoma (11) TLL (13) CRC (13) Small-bowel carcinoma (20)</td>
<td>CALMs</td>
<td>MSI-H</td>
<td>PMS2-</td>
<td>DOD (21)</td>
<td>Bougeard et al 80</td>
</tr>
<tr>
<td>9 (9)</td>
<td>PMS2</td>
<td>c.[1730dup];[137G&gt;T], p. [Arg578Aialfs*2];[Ser46Ile]</td>
<td>No LS</td>
<td>4</td>
<td>TLL (6) Glioblastoma (9) Polyps (11)</td>
<td>CALMs</td>
<td>MSI-H</td>
<td>PMS2-</td>
<td>DOD (12)</td>
<td>Bougeard et al 80</td>
</tr>
<tr>
<td>9 (11)</td>
<td>PMS2</td>
<td>c.[161T&gt;C];[1831dup], p.[Ile54Thr];[Ile31Anfs*2]</td>
<td>LS-family</td>
<td>2</td>
<td>Osteosarcoma (11)Glioblastoma (13)</td>
<td>NR</td>
<td>NA</td>
<td>PMS2-</td>
<td>DOD (13)</td>
<td>None</td>
</tr>
<tr>
<td>10 (12)</td>
<td>PMS2</td>
<td>c.[161T&gt;C];[1831dup], p.[Ile54Thr];[Ile31Anfs*2]</td>
<td>LS-family</td>
<td>2</td>
<td>Osteosarcoma (11)Glioblastoma (13)</td>
<td>NR</td>
<td>NA</td>
<td>PMS2-</td>
<td>DOD (13)</td>
<td>None</td>
</tr>
<tr>
<td>11 (14)</td>
<td>PMS2</td>
<td>c.[2531C&gt;A];[2531C&gt;A], p.[Pro844His];[Pro844His]</td>
<td>No LS</td>
<td>3</td>
<td>CRC/polyps (20) Glioblastoma (32)</td>
<td>No</td>
<td>MSI-H</td>
<td>PMS2-</td>
<td>DOD (13)</td>
<td>None</td>
</tr>
<tr>
<td>11 (15)</td>
<td>PMS2</td>
<td>c.[137G&gt;T];[137G&gt;T], p.[Ser46Ile];[Ser46Ile]</td>
<td>Consang. LS-family</td>
<td>6</td>
<td>CRC/polyps (22,25) Glioblastoma (34)Endometrial carcinoma (35) Small-bowel carcinoma (36)</td>
<td>CALMs, hypopigm. macules, congenital naevi, hamartoma</td>
<td>MSI-H</td>
<td>NA</td>
<td>CR (39)</td>
<td>None</td>
</tr>
<tr>
<td>12 (16)</td>
<td>PMS2</td>
<td>c.[137G&gt;T];[137G&gt;T], p.[Ser46Ile];[Ser46Ile]</td>
<td>Consang. LS-family</td>
<td>6</td>
<td>CRC/polyps (22,25) Glioblastoma (34)Endometrial carcinoma (35) Small-bowel carcinoma (36)</td>
<td>CALMs, hypopigm. macules, congenital naevi, hamartoma</td>
<td>MSI-H</td>
<td>NA</td>
<td>PMS2-</td>
<td>Tum. progression (37)</td>
</tr>
<tr>
<td>Family (Pat.)</td>
<td>Gene</td>
<td>Mutation</td>
<td>Familial history</td>
<td>Score</td>
<td>Malignant tumours/polyps (age at diagnosis)</td>
<td>Cutaneous signs</td>
<td>MSI</td>
<td>IHC</td>
<td>Outcome (age at last follow-up)</td>
<td>Reference</td>
</tr>
<tr>
<td>--------------</td>
<td>------</td>
<td>----------</td>
<td>------------------</td>
<td>-------</td>
<td>------------------------------------------</td>
<td>----------------</td>
<td>-----</td>
<td>-----</td>
<td>---------------------------------</td>
<td>-----------</td>
</tr>
<tr>
<td>13 (17)</td>
<td>PMS2</td>
<td>c.[22767.<em>*160</em>?<em>del]; [22767.</em>*160_?_del], p.[?];[?]</td>
<td>Aunt (TLL and breast)</td>
<td>5</td>
<td>Spinal glioblastoma (6)</td>
<td>CALMs, congenital naevi, axillary freckling</td>
<td>NA</td>
<td>PMS2-</td>
<td>DOD (7)</td>
<td>None</td>
</tr>
<tr>
<td>14 (18)</td>
<td>PMS2</td>
<td>c.[2007–?_A&gt;G];[20072A&gt;G], p.[?];[?]</td>
<td>Consang. No LS</td>
<td>6</td>
<td>Glioblastoma (6)</td>
<td>CALMs, hypopig. macules</td>
<td>NA</td>
<td>NA</td>
<td>Tum. progression (?)</td>
<td>None</td>
</tr>
<tr>
<td>15 (19)</td>
<td>MLH1</td>
<td>c.[678–?_686del];[678–?_686del], p.[?];[?]</td>
<td>Consang. LS-family</td>
<td>7</td>
<td>Medulloblastoma (5)AML (7)</td>
<td>CALMs</td>
<td>MSS</td>
<td>MLH1</td>
<td>-/PMS2-</td>
<td>DOD (7)</td>
</tr>
<tr>
<td>15 (20)</td>
<td></td>
<td></td>
<td></td>
<td>9</td>
<td>TLL (4) Oligodendroglioma (5)</td>
<td>CALMs</td>
<td>MSS</td>
<td>MLH1</td>
<td>-/PMS2-</td>
<td>DOD (7)</td>
</tr>
<tr>
<td>17 (22)</td>
<td></td>
<td></td>
<td></td>
<td>9</td>
<td>TLL (2)</td>
<td>CALMs</td>
<td>NA</td>
<td>NA</td>
<td>DOD (2)</td>
<td>Wang et al17</td>
</tr>
<tr>
<td>17 (23)</td>
<td>MSH2</td>
<td>c.[1277–?_1386+?]; [1277–?_1386+?], p.[?];[?]</td>
<td>Consang. LS-family</td>
<td>8</td>
<td>TLL (3)</td>
<td>CALMs, hypopig. macules</td>
<td>NA</td>
<td>NA</td>
<td>DOD (4)</td>
<td>None</td>
</tr>
<tr>
<td>18 (24)</td>
<td>MSH2</td>
<td>c.[1–?_1076+?del];[1–?_1076+?del], p.[?];[?]</td>
<td>No LS</td>
<td>5</td>
<td>Glioblastoma (3)</td>
<td>NR</td>
<td>MSS</td>
<td>NA</td>
<td>DOD (3)</td>
<td>Bougeard et al18</td>
</tr>
<tr>
<td>18 (25)</td>
<td></td>
<td></td>
<td></td>
<td>3</td>
<td>TLL (1)</td>
<td>NR</td>
<td>MSS</td>
<td>NA</td>
<td>DOD (1)</td>
<td>Bougeard et al19</td>
</tr>
<tr>
<td>19 (26)</td>
<td>MSH6</td>
<td>c.[2216C&gt;Al];[2216C&gt;A], p.[Thr739Lys];[Thr739Lys]</td>
<td>Consang. Sister (oligodendroglioma)</td>
<td>5</td>
<td>Glioblastoma (6)</td>
<td>CALMs</td>
<td>MSS</td>
<td>MSH6-</td>
<td>DOD (6)</td>
<td>None</td>
</tr>
<tr>
<td>19 (27)</td>
<td></td>
<td></td>
<td></td>
<td>5</td>
<td>Astrocytoma (9)</td>
<td>CALMs</td>
<td>MSS</td>
<td>MSH6+</td>
<td>DOD (9)</td>
<td>None</td>
</tr>
<tr>
<td>20 (28)</td>
<td>MSH6</td>
<td>c.[3991C&gt;T];[3991C&gt;T], p.[Arg1331*];[Arg1331*]</td>
<td>Consang. No LS</td>
<td>6</td>
<td>CRC/polyps (11)</td>
<td>CALMs</td>
<td>NA</td>
<td>NA</td>
<td>DOD (14)</td>
<td>None</td>
</tr>
<tr>
<td>21 (29)</td>
<td>MSH6</td>
<td>c.[3261dup];[2561–?_2563del/p. [Phe1088Leufs*5];[Lys854del</td>
<td>LS-family</td>
<td>11</td>
<td>Polyps (14)CRC (17)CRC (19)</td>
<td>CALMs</td>
<td>MSS</td>
<td>MSH6-</td>
<td>CR (42)</td>
<td>Bougeard et al19</td>
</tr>
<tr>
<td>22 (30)</td>
<td>MSH6</td>
<td>c.[1596_1597dup];[3261del/p. [Glu533Valfs<em>39];[Phe1088Serfs</em>2]</td>
<td>No LS</td>
<td>5</td>
<td>Glioblastoma (7)</td>
<td>CALMs</td>
<td>MSS</td>
<td>MSH6-</td>
<td>DOD (7)</td>
<td>Auclair 2007</td>
</tr>
<tr>
<td>23 (31)</td>
<td>MSH6</td>
<td>c.[1763_1771dup];[1763_1771dup], p. [His588_Pro590dup];[His588_Pro590dup]</td>
<td>Consang. No LS</td>
<td>5</td>
<td>TLL (6)TLL (11) Glioblastoma (14)CRC/polyps (14)</td>
<td>CALMs, hypopig. macules</td>
<td>MSS</td>
<td>MSH2-</td>
<td>DOD (16)</td>
<td>None</td>
</tr>
</tbody>
</table>

AML, acute myeloblastic leukaemia; CALMs, café-au-lait macules; CMMRD, constitutional mismatch repair deficiency; Consang., consanguinity; CR, complete remission; CRC, colorectal cancer; DFS, dermatofibrosarcoma; DLBCL, diffuse large B-cell lymphoma; DOD, dead of disease; DOT, dead of toxicity; Hypopigm., hypopigmented; IHC, immunohistochemistry; LS, Lynch syndrome; MSI, microsatellite instability; MSI-H, MSI-high; MSI-L, MSI-low; MSS, microsatellite stability; NA, not available; NR, not reported; Pat., patient; RMS, rhabdomyosarcoma; Score, score according to Wimmer’s criteria at diagnosis of the first malignancy; TLL, T-cell lymphoblastic lymphoma; tum., tumour.
the nine patients who developed a cancer after radiotherapy, one tumour was in the radiation field.

### Non-malignant features

Information concerning dermatological features was available for 28/31 patients. Cutaneous abnormalities were described in 23/28 (82%) patients and were explicitly absent in five patients. The most frequent lesions were CALMs alone described in 18 patients and hypopigmented macules described in five patients (figure 3). When specified, the number of CALMs over 1 cm in diameter ranged between 2 and more than 10. One case of a giant CALM measuring 15 cm in diameter was reported. CALMs and hypopigmented macules were widespread in 15 patients, lateralised to hemibody in three patients and not specified in two patients. Congenital naevi over 1 cm in diameter were associated with CALMs in four patients. Other NF1 features (Lisch nodules in one case, neurofibromas in the other) were reported in two patients. The NF1 gene had been screened in five patients, none of whom had a germline mutation.

Overall, 5 (16%) cases of brain malformations had been documented: multiple cavernomas in two patients with no history of brain radiotherapy, brain angiomas in two patients and brain hamartomas in one patient.

### Familial histories

The revisited Bethesda criteria for LS were fulfilled in 6/23 families (26%), three families with LS malignancies in two branches and three families in only one branch. Consanguinity was present in 9/23 (39%) families. In 13 families, several siblings had developed a malignancy, including 16 patients analysed in this study and four who had died without any screening for CMMRD. The characteristics of each patient and family are summarised in table 1.

### Molecular characteristics

The results of genetic screening for MMR gene mutations are available in 30/31 patients (at least one individual in all 23 families): 17 patients (12 families) with homozygous biallelic alterations of MMR genes and 13 patients (11 families) with compound heterozygous alterations. These alterations, including six variants of unknown functional significance, involved PMS2 (18 patients), MSH6 (6 patients), MLH1 (4 patients) or MSH2 (3 patients) (table 3).

In one patient, three deleterious PMS2 mutations have been identified. Only one heterozygous mutation of PMS2 was identified in one patient with obvious clinical characteristics of CMMRD. In one case, the first molecular screening had not detected any mutation but further investigations evidenced a large PMS2 rearrangement.

Table 3 summarises clinical and biological CMMRD characteristics for each mutation. Median age at diagnosis of the first malignancy was older in biallelic PMS2 mutation carriers than in patients with mutations in the other MMR genes. As survival was longer in biallelic PMS2 mutation carriers, these patients were likely to develop several malignancies at a significantly higher rate than in the carriers of other mutations (mean 2.6 per patient versus 1.6 per patient, p=0.03 (0.1–1.8), Welsh two sample t test). LS-associated malignancies occurred only in the PMS2 and MSH6 mutation groups, whereas haematological malignancies and brain tumours were equally represented for all types of mutations.
The results of MSI analyses performed on 23 tumours from different patients were heterogeneous, with a high rate of microsatellite stability (MSS) tumours in MLH1, MSH2 and MSH6 mutation carriers, in whom brain tumours were overrepresented, and a majority of MSI tumours in PMS2 mutation carriers, in whom the MSI test had essentially been performed on LS-associated tumours. IHC analysis had been performed on the samples of 22 patients, on CRC, brain

Figure 1  Outcome of the French constitutional mismatch repair deficiency syndrome (CMMRD) cohort (n=31 patients). *Duration of remission.

Figure 2  Overall and event-free survival. (A) Overall survival. (B) Event-free survival. (C) Survival according to tumour type.
tumour or lymphoma and/or on normal tissue. Suppression of protein expression corresponding to a gene mutation in both tumour and normal tissue had been detected in 20 cases, and a weak signal in two other cases (a CRC and a glioblastoma).

**DISCUSSION**

This study conducted in all French patients diagnosed with CMMRD is the first comprehensive nationwide description of the clinical characteristics and outcome of an unselected series of patients with CMMRD. This description strengthens other data previously described about the clinical characteristics of CMMRD patients and brings new comprehensive features about their outcome.

As the tumour spectrum and clinical phenotype of CMMRD overlaps with other cancer predisposition syndromes such as NF1, familial adenomatous polyposis or chromosome breakage disorders, CMMRD is probably underdiagnosed. Thus, the risk of developing second malignancies in CMMRD carriers may be overestimated because CMMRD had rarely been suspected at the diagnosis of the first malignancy, except in the context of malignancy in siblings. In this series, several patients were initially diagnosed as having NF1 because of CALMs, CMMRD had only been suspected after a second tumour.

The spectrum of tumours corroborates data already described in previous reports. Ages at diagnosis are similar to those already published for each tumour type. The high mortality rate among patients with CMMRD is due to the primary tumour in 29% of patients, with a median survival of 27 months from the diagnosis of the first tumour. Consistent with literature, a degree of correlation was evidenced between the genotype and phenotype: patients with a biallelic *PMS2* mutation had an older age at diagnosis and a greater incidence of brain and LS-associated tumours, whereas patients with a biallelic *MLH1*, *MSH2* or *MSH6* mutation had mainly been treated for haematological malignancies and had a higher risk of mortality after their first tumour.

The small sample size precludes any firm conclusions about the efficacy of chemotherapy in each tumour type. However, the high rate of treatment failures (40%) observed in patients with CMMRD-related T-NHL in our series contrasts with the high success rate reported in sporadic T-NHL with 5-year EFS over 80% in most recent protocols. The lower efficacy of

### Table 3  Main clinical and biological characteristics according to molecular diagnosis

<table>
<thead>
<tr>
<th>Gene mutation</th>
<th>PMS2</th>
<th>MLH1</th>
<th>MSH2</th>
<th>MSH6</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nb of patients</td>
<td>18</td>
<td>4</td>
<td>3</td>
<td>6</td>
</tr>
<tr>
<td>Nb of families</td>
<td>14</td>
<td>2</td>
<td>2</td>
<td>5</td>
</tr>
<tr>
<td>Median age at first tumour (years)</td>
<td>15.3 [3.2–33.5]</td>
<td>5.5 [2–6]</td>
<td>2.9 [1.2–3.3]</td>
<td>8.1 [6.1–17.6]</td>
</tr>
<tr>
<td>Mortality (%)</td>
<td>10/18 (56%)</td>
<td>4/4 (100%)</td>
<td>3/3 (100%)</td>
<td>5/6 (83%)</td>
</tr>
<tr>
<td>Median delay (years) between first tumour and death</td>
<td>6.0 [0.9–17.8]</td>
<td>2.1 [0.6–3.8]</td>
<td>0.6 [0.02–1.1]</td>
<td>0.6 [0.4–9.0]</td>
</tr>
<tr>
<td>Tumour type (n=67)</td>
<td>46</td>
<td>7</td>
<td>3</td>
<td>11</td>
</tr>
<tr>
<td>Haematological malignancy</td>
<td>9</td>
<td>4</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>Brain tumour</td>
<td>14</td>
<td>3</td>
<td>1</td>
<td>4</td>
</tr>
<tr>
<td>LS-associated tumour</td>
<td>20</td>
<td>0</td>
<td>0</td>
<td>5</td>
</tr>
<tr>
<td>Other tumours</td>
<td>3</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Result of MSI analysis (n=23)</td>
<td>14</td>
<td>3</td>
<td>1</td>
<td>5</td>
</tr>
<tr>
<td>MSI-H (high)</td>
<td>12</td>
<td>1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>MSI-L (low) or MSS</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>5</td>
</tr>
<tr>
<td>Result of IHC (n=22)</td>
<td>15</td>
<td>2</td>
<td>0</td>
<td>5</td>
</tr>
<tr>
<td>Protein extinction</td>
<td>14</td>
<td>2</td>
<td>0</td>
<td>4</td>
</tr>
<tr>
<td>Protein detected</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>1</td>
</tr>
</tbody>
</table>

IHC, immunohistochemistry; LS, Lynch syndrome; MSI, microsatellite instability; MSI-high; MSI-L, MSI-low; Nb, number.
chemotherapy in this population may probably be explained by the major role of the MMR pathway in the induction of genotoxicity associated with several drugs including thiopurine and alkylating agents,\(^5\) major drugs in current therapeutic protocols for T-NHL. This should prompt us to design specific protocols for those rare cases of CMMRD-associated T-NHL.

The survival of patients with brain tumours is difficult to compare with the outcome of patients with sporadic paediatric malignant gliomas because of the heterogeneity of the histological subtypes, sites and tumour resectability. However, although MMR-deficient cells have been shown to be more tolerant to temozolomide and radiotherapy than MMR-proficient cells,\(^26\) the 5-year OS rate of 21% observed in patients with brain tumours is apparently not lower than that expected in children treated for malignant gliomas.\(^26\)

The toxicity of chemotherapy was clearly not increased in this series of patients. The only toxicity-related death had occurred after allogeneic HSCT, a procedure associated with a significant mortality rate even in the general population. Several patients with T-NHL had developed severe toxicity. However, they were treated according to the protocols known to be associated with high incidence of severe toxic events.\(^24\) We cannot exclude that the three cases of severe infections in this series could be related to the presence of an immune deficiency, which has previously been described in the CMMRD syndrome\(^27\) but was not systematically analysed in this series. The role of chemotherapy or radiotherapy in the occurrence of further malignancies is difficult to assess in this short series of patients treated in different centres at different periods.

One important question is whether patients with CMMRD could be diagnosed before the occurrence of multiple malignancies. The presence of cutaneous abnormalities in a child with a malignancy within the spectrum of CMMRD should probably lead clinicians to investigate CMMRD. Heterogeneous descriptions have been provided for the cutaneous phenotype of CMMRD\(^10\) \(^28\) and this study suggests that common dermatological features at presentation combine widespread or segmental CALMs and hypopigmented macules. Congenital naevi are less common, as well as pilomatricomas, rare non-malignant skin tumours unrelated to NF1.\(^29\) As previously reported,\(^28\) \(^30\) cutaneous abnormalities were absent in rare cases in this series. Brain malformations similar to those reported in the literature\(^31\) are particularly frequent. An analysis of the familial history is not always helpful in suspecting the diagnosis of CMMRD. The absence of consanguinity does not exclude the diagnosis of CMMRD since it was only reported in 30% of cases in this series. Furthermore, a familial history of cancer may be lacking due to the low penetrance of monoallelic PM2 mutations. The 3-point scoring system established by the European Consortium for CMMRD\(^3\) may be helpful in this context. Using this system, most patients already had a score above 3 at the time of the diagnosis of the first malignancy and therefore could have been tested for CMMRD earlier.

Once the diagnosis of CMMRD is suspected, biological confirmation may be challenging. As patients with CMMRD constitutively lack the expression of one of the four MMR genes,\(^16\) the loss of MMR proteins is usually detected by IHC in neoplastic and non-neoplastic tissues such as normal skin. However, in the case of missense mutations, IHC may be normal because of residual protein expression, as it has been found in two cases of this series. Moreover, the procedure for detecting MSI in normal tissues has not been standardised to date. Although MSI analysis follows well-defined protocols for LS screening, its results were unreliable especially for extradigestive tumours as in several previous reports.\(^16\) \(^18\) \(^32\) Thus, MMR genes should be screened if the clinical score threshold has been attained, even in MSS tumours or if there is no loss of MMR protein expression. Even if new methods such as germline microsatellite instability (gMSI) allowing the detection of germline MSI are attempted to facilitate and shorten the time to confirmation,\(^35\) the diagnosis of CMMRD should always be confirmed by genespecific analysis. Reliable methods exist for all four genes including PM2,\(^13\) despite the numerous pseudogenes that interfere with PM2 detection.\(^34\) Eight cases in this series highlight how difficult it is to confirm the diagnosis of CMMRD. Despite a complete CMMRD phenotype, a single heterozygous PM2 mutation has been identified in a patient. Another undetectable PM2 mutation or mechanism of PM2 inactivation is probably involved.\(^19\) Six other patients were carriers of variants of unknown functional significance. In order to facilitate the diagnosis in these questionable cases, a reliable diagnosis method for CMMRD based on the functional properties of MMR-deficient cells has been developed\(^32\) by the European Consortium for CMMRD.

In conclusion, this unselected series of patients confirms the extreme severity of this syndrome with a high mortality rate mostly related to multiple childhood cancers, and highlights the need for its early detection. An adapted therapy may be required in order to overcome chemoresistance especially in patients with haematological malignancies. The use of the 3-point scoring system proposed by the European Consortium for CMMRD should help the clinicians to diagnose the syndrome at the time of diagnosis of the first malignancy and thus allowing adapting therapy and surveillance. However, due to the rarity of this syndrome, an international collaboration is required to constitute a large cohort of patients with CMMRD in order to evaluate guidelines for screening and treatment of malignancies and to explore prevention strategies.
#### Acknowledgements
We are indebted to Lorna Saint-Ange and Vallérie Marechal for editing the manuscript and to Marina Dimaria, Anne-Sophie Defachelles, Dominique Plantaz, Anne Pagnier, Denis Smith, David Malka and Alexander Leis for giving access to clinical and biological data of their patients.

#### Contributors
Manuscript writing: NL and LB. Conception and design: LB and CC. Collection and assembly of data: NL, CC and LB. Pathology review: FA and CC. Dermatological assessments: GS. Molecular analysis: MM, SB, AD, SB-D, JT, TF. Collect clinical data and final approval of manuscript: all authors.

#### Competing interests
None declared.

#### Ethics approval
Gustave Roussy IRB.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### REFERENCES


#### Dermatological assessments: GS. Molecular analysis: MM, SB, AD, SB-D, JT, TF. Collection and assembly of data: NL, CC and LB. Pathology review: FA and CC. Manuscript writing: NL and LB. Conception and design: LB and CC.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.

#### Acknowledgements
None declared.

#### Provenance and peer review
Not commissioned; externally peer reviewed.
Constitutional mismatch repair deficiency syndrome: clinical description in a French cohort


J Med Genet 2015 52: 770-778 originally published online August 28, 2015
doi: 10.1136/jmedgenet-2015-103299