INTRODUCTION

Adult acute myeloid leukemia (AML—OMIM #601626), in general, can be classified into several subgroups according to World Health Organization (WHO 2016 classification of hematologic cancers) (Arber et al., 2016) and the European LeukaemiaNet (ELN) (Döhner et al., 2017). A rare such entity is hereditary AML due to germline-mutated CCAAT/enhancer-binding protein-α (CEBPA—OMIM *116897) gene (Arber et al., 2016). Most of the families reported in the literature demonstrate an autosomal dominant pattern of inheritance, and patients are affected by the malignancy at a younger age than other AML entities (Owen et al., 2008); according to Hackl et al. (2017), AML, in general, has a median age of onset of ~67 years while familial CEBPA-mutated AML affects corresponding family members between 2 and 50 years (Tawana et al., 2017), as is typical for familial cancer syndromes (Rahner & Steinke, 2008).
2.1 Editorial policies and ethical considerations

The father of the studied family agreed with the scientific evaluation and the study was approved by the ethical committee of the Atomic Energy Commission, Damascus, Syria.

2.2 Clinical information

Here a nonconsanguineous Syrian family is reported: among the parents and their six children (four boys and two girls, see Figure 1), overall four are affected by AML.

The father and three of his children were diagnosed with the malignancy, whereas the mother and the remaining of family members were unaffected and are yet clinically healthy. Age of onset of AML was in the father (I-1) 37 years, in son 1 (II-1) 8 years, son 2 (II-2) 2.8 years, and daughter 5 (II-5) 2.5 years.

The father was given standard treatment for AML including 3 + 7 induction chemotherapy (Daunorubicin 60 mg/m² for 3 days and Cytarabine 200 mg/m² for 7 days), and his affected children were treated by ELAM02 induction chemotherapy (Aracytine 200 mg/m² for 5 days and Mithoxantrone 12 mg/m² for 3 days). Bone marrow (BM) examination on day 20 postinduction was consistent with the achievement of complete remission (CR) for the father and his affected children. CR is still present for family members I-1 and II-5. Patient II-2 had one relapse which could be brought to CR again, whereas patient II-1 relapsed 7 years after diagnosis and died (see Table 1).

Interestingly, no CD7 expression was detectable for I-1 and II-2; also, family members showed different FAB subtypes during their causes of AML: The father (I-1) had FAB subtype M2, the eldest boy (II-1), and the youngest affected daughter had FAB subtype M5, whereas the second boy (II-2) had FAB subtype M4.

2.3 Chromosome analysis and fluorescence in situ hybridization (FISH) analysis

Chromosome analysis using GTG-banding was performed on bone marrow (BM) samples prior to chemotherapy and postchemotherapy was performed according to standard protocols (Al-Achkar et al., 2007). Fluorescence in situ hybridization (FISH) using specific probes to detect translocations t(8;21), t(15;17), t(16;16), t(12;21), and deletion del(13q) were applied chromosome preparations of BM samples prior to chemotherapy and postchemotherapy as previously reported (Al-Achkar et al., 2007).
|                | I−1       | II−1      | II−2       | II−5       |
|----------------|-----------|-----------|------------|------------|
| Gender         | M         | M         | M          | F          |
| AML diagnosis  | April 2014| April 2013| April 2010 | December 2014|
| Age at AML diagnosis (years) | 37       | 8         | 2.8        | 2.5        |
| FAB subtypes   | M2        | M5        | M4         | M5         |
| Immunophenotyping | CD45\textsuperscript{dim}, HLADR, MPO, CD33, CD34, CD11c, CD7, CD32, CD38, CD10, CD13 | CD45\textsuperscript{dim}, HLADR, MPO, CD33, CD4, CD11c, CD7, CD32, CD38, D64, CD14 | CD45\textsuperscript{dim}, HLADR, MPO, CD33, CD4, CD11c, CD7, CD32, CD38, CD117, CD15\textsuperscript{dim}, CD13 | CD45\textsuperscript{dim}, HLADR, MPO, CD33, CD4, CD11c, CD7, CD32, CD117, CD14 |
| WBC count      | 36.8 × 10^9/L | 31.3 × 10^9/L | 25.4 × 10^9/L | 57.6 × 10^9/L |
| Plt count × 10^9/L | 31 × 10^9/L | 78 × 10^9/L | 20 × 10^9/L | 67 × 10^9/L |
| HgB, g/dL      | 11        | 10        | 10         | 6.6        |
| LDH, U/L (normal value up to 420) | 277 | 1303 | 1178 | 873 |
| Blasts count in BM | 80% | 55% | 85% | 52% |
| Main clinical features | Purpura, bleeding gums, neck lymphadenopathies (2 × 1 cm) | Bruising’s, several lymphadenopathies (2 × 1 cm) | Purpura, bleeding gums, hepatomegaly (3 cm), splenomegaly (2 cm), several lymphadenopathies (2 × 1 cm) | Bruising’s, pallor, several lymphadenopathies (2 × 2 cm), hepatomegaly (2 cm) |
| Karyotype at AML diagnosis | Normal | Normal | Normal | Normal |
| Germline CEBPA mutation | c.198dupC (Tyr67Leufs*41) | c.198dupC (Tyr67Leufs*41) | c.198dupC (Tyr67Leufs*41) | c.198dupC (Tyr67Leufs*41) |
| Prior chemotherapy treatment protocol | 3 + 7 | ELAM02 | ELAM02 | ELAM02 |
| CR, weeks      | Yes       | Yes       | Yes        | Yes        |
| Time to get CR followed initial chemotherapy treatment | 3 weeks | 3 weeks | 3 weeks | 3 weeks |
| Relapse        | No        | Yes       | Yes        | No         |
| Disease status at last follow-up | CR1 | Relapse March 2020 | CR2 | CR1 |
2.4 | Molecular genetic analysis

Genomic DNA was isolated from peripheral blood (PB) samples after chemotherapy using QIAamp DNA Blood Mini Kit (Qiagen GMBH, Hilden, Germany). Germline mutation analyses were done using next-generation sequencing (NGS) by panel diagnostics in all family members apart from II-6, which was very young at the time of analyses; thus, no blood could be taken. The TrueSight Cancer panel (Illumina; FC-121–0202) was used.

3 | RESULTS

Father (I-1) and his affected children (II-1, II-2, and II-5) had normal results after karyotyping of bone marrow cells (results not shown). Also FISH revealed no evidence of cryptic translocations t(8;21), t(15;17), t(16;16), t(12;21), and/or a deletion del(13q).

However, germline mutation analyses by NGS identified a \( \text{CEBPA} \) gene mutation NM_004364.5:c.198dupC, p.(Tyr67Leufs*41) (described according to HGMD) and according to ACMG a class 5 mutation—see also http://ftp.ebi.ac.uk/pub/databases/lrgex/LRG_456.xml. This mutation was present in all four affected family members, that is, I-1, II-1, II-2, and II-5, and besides also in the yet unaffected third son (II-3).

4 | DISCUSSION

A new familial AML with four affected and a yet unaffected member with the identical germline \( \text{CEBPA} \) mutation is reported. Other unaffected family members did not have this mutation (I-2, II-4) or were not tested (II-6).

The identified c.198dupC mutation has previously been observed in a single case of nonfamilial AML (according to the COSMIC database for somatic samples from hematopoietic and lymphoid tissue, COSV57200711)—see also Dufour et al. (2012). As outlined before, \( \text{CEBPA} \) mutations can occur as acquired somatic and/or germline mutations (Gao et al., 2019). However, only 26 families with germline \( \text{CEBPA} \) mutations have been reported yet (reviewed in Brown et al., 2020). In systematically studied AML cases with normal karyotype, 8–13% of the cases had \( \text{CEBPA} \) gene mutations (Preudhomme et al., 2002; Pabst et al., 2008; Taskesen et al., 2011); among these, 7–11% have \( \text{CEBPA} \) germline mutations (Pabst et al., 2008; Taskesen et al., 2011). As the majority of the AML patients have two \( \text{CEBPA} \) mutations in trans, with both N-terminal and C-terminal frameshift mutations, this might also be suggested here for the reported family, even though it could not be verified. Even though no DNA was
available from the diseased bone marrow of the here presented family members, the fact that an N-terminal frameshift mutation was present here coincidences with other cases from the literature, for which such a germline mutation predisposes to acquire a somatic C-terminal CEBPA mutation on the other allele (Pabst et al., 2008; Tawana et al., 2015).

Also, another observation for the presented family is in concordance with the literature: familial AML with mutated CEBPA gene is inherited in an autosomal dominant way and displays complete or near-complete penetrance for development of AML and disease onset between 2 and 50 years of age (Owen et al., 2008, Renneville et al., 2009, Tawana et al., 2015). As four of six children of the mutated CEBPA gene carrier I-1 have inherited the same CEBPA mutation, it is inherited in an autosomal dominant way; the penetrance is yet four in five gene carriers, suggesting close supervision of blood values of family member II-3 (the family had genetic counseling and II-3 is being monitored with complete blood counts every 6 months), while no special care is necessary for II-4, as he is not a carrier of familial CEBPA mutation. Also, CEBPA mutation screening is indicated of II-6 as soon as possible.

Normally, the clinic-pathologic features of familial CEBPA-mutated AML cases, similar to those of sporadic AMLs with the majority displaying a normal karyotypic, include FAB subtypes M1, M2, and, less frequently, M4 (with Auer rods seen in PB or BM blasts), and aberrant CD7 expression on blasts (Tawana et al., 2015). Thus, it is striking that here in overall four diseased family members, three different FAB subtypes were observed: the expected subtypes M2 and M4 (one time, each) and the yet in this familial AML-type unreported subtype M5—being present in father I-1 and daughter (II-5). Also, the normally in AML with mutated CEBPA gene present CD7 expression was not detectable in family members I-1 and II-1; the underlying mechanisms for both phenomena are yet not understood. Nonetheless and irrespective of the FAB subtype, the clinical outcome was comparatively positive in the reported family. This is also in concordance with literature data; here a favorable prognosis with an overall survival rate of 50–65% compared with 25–40% in normal karyotype AML without germline CEBPA mutation is given (Preudhomme et al., 2002; Fröhling et al., 2004; Bienz et al., 2005; Marcuccio et al., 2008).

In conclusion, we describe here a novel case of a familial AML with four affected members and five of which had a germline mutation c.198dupC CEBPA being never observed in familial AML cases before. This family presents in individual I-1 a new clinical feature such as FAB subtype 5 without CD7 expression but still had a favorable outcome in all but one family member.

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CONFLICT OF INTERESTS
The authors declare that they have no competing interests.

AUTHOR CONTRIBUTIONS
AW, SA, and WA performed banding cytogenetics and provided the clinical data; BA also provided clinical data and chemotherapy plan; AW, FA, and TL performed the molecular cytogenetic analyses; KM did molecular genetic analyses; AA performed immunophenotyping. AW and TL drafted the paper and all authors worked on the final version of the paper. All authors read and approved the final manuscript.

CONSENT FOR PUBLICATION
Written informed consent was obtained from the patient’s brother for publication of this case report and accompanying images. A copy of the written consent is available for review by the Editor-in-Chief of this journal.

DATA AVAILABILITY STATEMENT
All data necessary to understand the results and conclusions are provided in this article.

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