



Brief communication

ATM mutations and protein expression are not associated with familial B-CLL cases

Familial clustering of B-cell chronic lymphocytic leukemia (B-CLL) has been reported [1,2], but to date no major susceptibility gene has been identified thus far. Deletions at chromosome 11q23 are commonly observed [3], and these alterations have been reported to be associated with more aggressive disease among patients who were younger than 55 years of age. Due to the younger age of onset observed in familial CLL cases and increased risk of developing a lymphoproliferative malignancy observed in individuals affected with ataxia-telangiectasia (AT) [4–7], we investigated whether ataxia-telangiectasia mutated (*ATM*) gene plays a role in familial CLL.

We analyzed lymphocytes and DNA from 37 familial CLL cases and 46 unaffected relatives for *ATM* protein expression and mutations in the *ATM* gene region. Table 1 summarizes the family structures between the affected individuals of the 19 kindreds in this study. Of these subjects, 59 had both *ATM* mutation and *ATM* protein analyses performed using previously described techniques [8,9]. Briefly, samples were screened for *ATM* mutations using denaturing high performance liquid chromatography (DHPLC) on a Varian helix system according to manufacturer's recommendation and *ATM* protein expression was quantified in triplicate using a solid-phase plate radioimmunoassay.

Thirty-five of 37 CLL cases had normal *ATM* protein expression levels and no germline mutations were found. Protein expression levels ranged from 0.20 to 1.75 in cases and from 0.50 to 1.48 in controls, and were similar in cases and controls (Fig. 1). Table 2 summarizes the characteristics of the two CLL cases (5.4%) with impaired *ATM* protein expression, defined as having less than half the median expression levels in normal individuals. The two cases are from different CLL families. Patient 1 had large cell lung cancer 10 years prior to her CLL diagnosis, but in regards to CLL, the characteristics including age at onset, stage at diagnosis, and severity of disease were unremarkable.

Both germline and somatic mutations in the *ATM* gene have been reported in sporadic CLL cases [10–13] leading to the suggestion that the familial aggregation observed in CLL may partly be attributable to excess obligate AT heterozygote carriers in these kindreds. However, we found little evidence of a role for *ATM* in familial CLL and were unable to identify any mutations (germline or somatic) in either of the patients with impaired *ATM* protein expression.

A number of reasons may explain the discrepancy between our findings and those reported in the literature. One may be that 11q23 deletions are less common in familial CLL than in sporadic CLL cases where up to 20% of these cases have been reported to have 11q23 deletions [3]. (Preliminary cytogenetic results from our familial CLL patients suggest abnormalities at 13q14 are much more common than deletions at 11q23 and are similar to those reported by Stilgenbauer et al. [14].) Alternatively, pre-screening of sporadic cases for 11q23 deletions (i.e. enriching for those with known loss of heterozygosity (LOH)) may have increased the chance of finding an alteration in this gene [12] in the sporadic cases. It is also possible that some allelic variants may have been missed since the detection rate of most scanning techniques used in identifying mutations is less than 100% [15]. The sensitivity of the screening used in our study has been reported to be around 90% [16].

Familial CLL may also differ biologically from sporadic CLL. We previously reported that the average age of onset was younger in familial cases than in sporadic cases [2]. Although impaired *ATM* protein expression has been reported in 34–40% of sporadic cases [8,13] and particularly in younger cases [8], we did not observe higher frequency of *ATM* impairment in our familial cases. Only 5% of our familial CLL cases were observed to have impaired expression, suggesting that an alternative pathway (i.e. another susceptibility gene) may be involved in this disease.

In summary, we did not find evidence for a role of *ATM* in familial CLL susceptibility. Our results are consistent with the studies conducted by Bevan et al. [17] and Yuille et al. [18]. While our results do not support the hypothesis that the *ATM* gene is a CLL susceptibility gene, it is possible that other genes in the region of 11q23 may play a role in CLL development.

Table 1
Family relationships between affected relatives in 20 CLL families

Number of affected individuals ^a	Relationship	Number of families
2	Siblings only	6
2	Parent–offspring only	3
3	Siblings only	2
3	Parent and two offsprings	2
3	Other ^b	
5	Parent and four offsprings	1

^a Not all affected individuals participated.

^b One of each of the following relationships were observed: three-generational; parent–offspring and cousin; two siblings and an offspring; two siblings and uncle; two siblings and cousin; cousins and nephew.

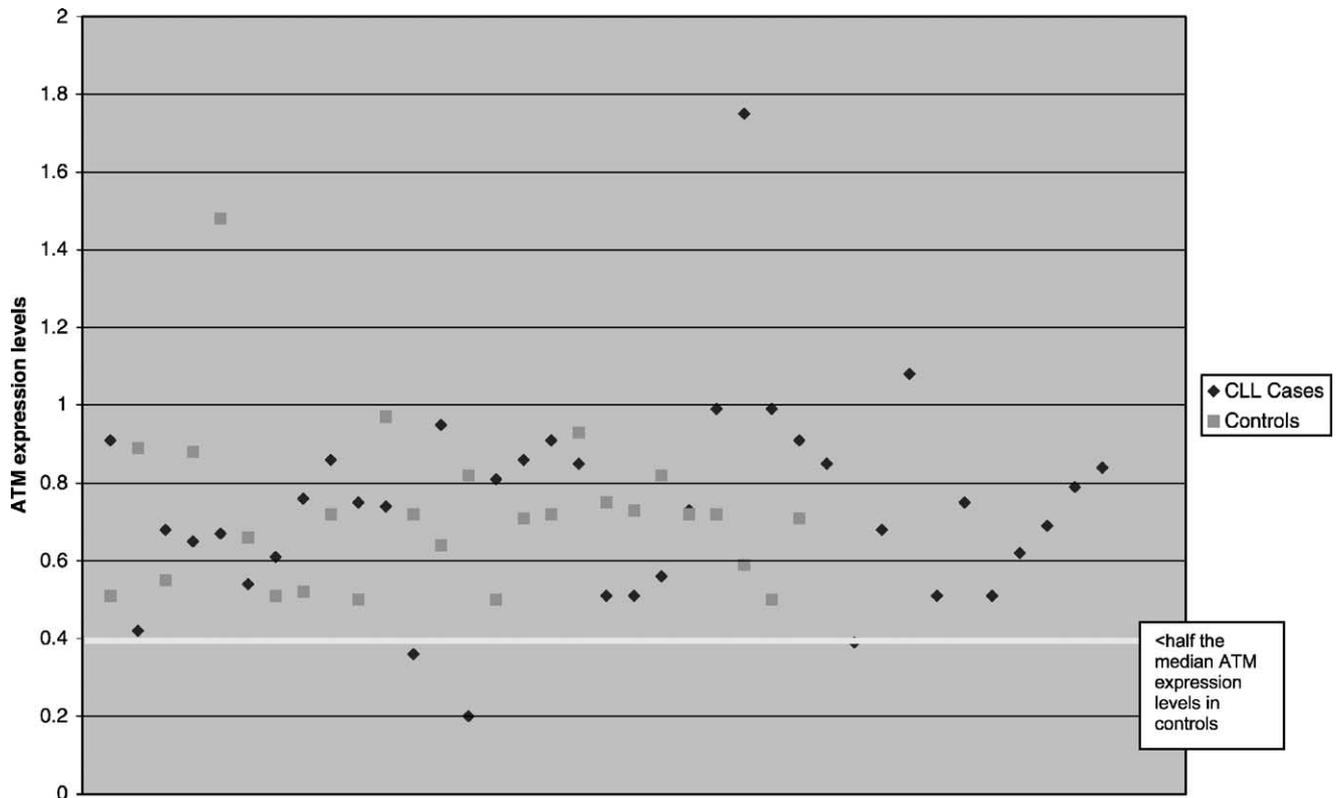


Fig. 1. ATM expression levels by case status.

Table 2
Clinical characteristics of individuals with impaired ATM protein expression

Subject	Gender	Age at diagnosis	Vital status	Current age/ age at death	WBC (% lymphs)	Rai stage	Affected pattern	ATM variants	Second primary tumor
1	F	74	Dead	79	27000 (80)	0	Parent–offspring		Lung cancer, BCC
2	M	50	Dead	53	43000 (NA)	I	Sibling pair	1090 + 26G → T (homozygous)	

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