7014 **Poster Discussion Session (Board #6), Sat, 8:00 AM-12:00 PM and 12:00 PM-1:00 PM**

Use of tumor genomic profiling to reveal mechanisms of resistance to the BTK inhibitor ibrutinib in chronic lymphocytic leukemia (CLL).

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Background: Ibrutinib interacts covalently with cysteine 481 of Bruton tyrosine kinase (BTK), resulting in targeted inhibition of B cell receptor signaling. Early trials of ibrutinib mono- or combination therapy enrolled 246 CLL patients receiving a median of 14 months of ibrutinib. 20 patients (8%) experienced progressive disease (PD), including 8 patients with Richter's transformation. Here we examine changes to the CLL genome in 3 patients that acquired resistance to ibrutinib. **Methods:** Ibrutinib resistance was defined as patients achieving partial response (PR) or better lasting $\geq$ 6 months, then developing PD without Richter's transformation. RNAseq and whole exome sequencing (WES) followed by comparative genome analysis was performed at baseline and after PD and confirmed by Sanger sequencing. RNAseq and WES data were aligned using TopHat and BWA software. Single nucleotide variations (SNVs) were identified using SAMtools mpileup. **Results:** Compared to patients who relapsed from conventional chemotherapy, minimal genomic changes were acquired in ibrutinib resistant patients, reflecting relative genomic stability. SNVs were discovered in 3 patients specific to the relapse sample (Table). 2 out of 3 patients had distinct SNVs that each encode a cysteine-to-serine substitution at position 481of BTK (C481S). Homologous cysteine residues in BMX, ITK, TEC and BLK were wild-type (WT). Ibrutinib inhibited recombinant C481S 25 fold less potently than WT, and could not covalently bind C481S expressed in cells. The third patient had WT BTK, but acquired a potential gain-of-function mutation encoding a R665W substitution in PLCg2, a substrate of BTK, consistent with constitutive PLCg2 activation. **Conclusions:** Although rare, the acquisition of C481S BTK and R665W PLCg2 mutations in the setting of resistance confirms BTK as an important pharmacologic target of ibrutinib, and suggests mechanisms of ibrutinib resistance.

Study	RXn	Duration on ibrutinib	Best response	Mutation
PCYC-04753	Ibrutinib 560 mg daily	575 days	PR	C481S BTK
PCYC-1102	Ibrutinib 420 mg daily	581 days	PR	R665W PLCg2
PCYC-1108	Ibrutinib 420 mg daily + bendamustine/rituximab x 6 cycles	388 days	CR	C481S BTK