

Supplemental Data

Table S1, related to Table 2. Characteristics of Patients with Aspergillosis

#	Patient # at study entry	Age (Year)	Sex	Prior # Rx	Ibrutinib dose (mgs)	Pre-Rx steroids	Onset		Site	Attribution	Aspergillus Antigen (n ≤ 0.5)			Outcome and aspergillosis attribution
							Ibrutinib alone	TEDDi-R			BAL	Serum	CSF	
1 ¹	9	76	F	1	700	Yes	Day 13		Lung Brain	Proven	>3.75	>1.79	>9.5	Death aspergillosis definitely related
2 ²	13	65	M	4	840	Yes	Day 13		Lung Brain	Proven	>3.75	N/A	>3.24	Death aspergillosis definitely related
3 ³	4	87	F	0	560	No		Cycle 4, Day 18	Lung Brain	Proven	>1.738	>0.964	N/A	Death PCNSL definitely related
4 ⁴	11	60	M	0	840	Yes		Cycle 5, Day 16	Lung	Possible	<0.5	N/A	N/A	Resolution probably related
5 ⁵	14	53	M	1	840	No		Cycle 3, Day 1	Lung	Possible	<0.5	<0.5	N/A	Resolution probably related
6 ⁶	15	64	M	2	840	Yes		Cycle 2, Day 1	Lung	Possible	<0.5	<0.5 ²	N/A	Resolution probably related
7 ⁷	18	49	M	2	840	Yes		Cycle 1, Day 13	Lung Brain	Probable	<0.5	N/A	0.64	Resolved definitely related

Abbreviations: Rx-regimens; BAL-broncho-alveolar lavage; CSF-cerebral spinal fluid.

¹ Patient #1 was a 76-year-old female with primary-refractory PCNSL. Prior to enrollment, she received dexamethasone 16 mgs/day for 4 weeks, which was continued for symptom control. Pre-treatment staging showed PCNSL in the right hemisphere and left temporal lobe, and normal body computerized tomography (CT) and FDG-PET-CT scans. On day 10 of ibrutinib, she became unresponsive. A brain MRI revealed a major reduction in the lymphomatous mass but new diffusion restricted lesions consistent with intracranial abscesses throughout both hemispheres (Figure 5B of manuscript). A chest CT scan revealed new right peri-hilar and left basilar infiltrates, and a broncho-alveolar lavage (BAL) demonstrated *A. fumigatus* and *P. jirovecii*. Aspergillus antigen was positive in the BAL, CSF and serum. Despite broad antifungal and antibiotic coverage, the patient died 14 days later from aspergillosis. Autopsy showed *A. fumigatus* in the lungs and brain.

² Patient #2 is a 65-year-old male with relapsed/refractory PCNSL after 4 prior therapies. Prior to enrollment, he received dexamethasone 40 mgs/day for 2 weeks, which was tapered to 16 mgs/day by study enrollment. Pre-treatment staging showed PCNSL in the periventricular area,

and normal body CT and FDG-PET-CT scans. On day 15 of ibrutinib, he developed worsening neurological signs and a brain MRI demonstrated a major reduction in the lymphomatous lesions but new lesions throughout the subarachnoid space with diffusion restriction. A chest CT scan showed a new 5 cm cavitating right pulmonary lesion, proven to be *A. fumigatus* by BAL (Figure 5C of manuscript). Aspergillus antigen was positive in the BAL and CSF. Despite broad antifungal coverage, the patient died 11 days later from complications of aspergillosis and an autopsy revealed evidence of *A. fumigatus* in the lung and brain.

³ Patient #3 is an 87-year-old female with treatment naïve PCNSL. Pre-treatment staging showed PCNSL in the corpus collosum and left periaxial white matter, and normal body CT and FDG-PET-CT scans. At the end of cycle 4 of DA-TEDDi-R, she developed pulmonary symptoms and a chest CT scan showed bilateral pulmonary nodules up to 4 cm in diameter and a new cavitary lesion in the brain. During therapy, she only had two days of neutropenia (< 500 cells/ μ l), which occurred on the first cycle. A BAL confirmed aspergillus, and aspergillus antigen was found in the BAL and serum. CSF β -D-glucan was negative. The patient died 18 days later from progressive disease and an autopsy revealed pulmonary *A. fumigatus*.

⁴ Patient #4 is a 60-year-old male with treatment naïve PCNSL. He received dexamethasone 8 mgs/day for 3-weeks before enrollment, which was tapered to replacement hydrocortisone. Pre-treatment staging showed PCNSL in the corpus collosum, frontal and left temporal lobes, and normal body CT and FDG-PET-CT scans. At the end of cycle 5 of DA-TEDDi-R, he developed pulmonary symptoms and a chest CT scan showed two new left pulmonary nodules up to 1.5 cm in diameter. He only had significant neutropenia (< 500 cells/ μ l) on cycle 5, which lasted 8 days. Although the BAL and serum were negative for infectious etiologies, anti-fungal therapy with liposomal amphotericin and voriconazole were instituted due to a high suspicion of aspergillosis. A repeat chest CT three weeks later showed significant improvement with resolution after 5 weeks of treatment.

⁵ Patient #5 is a 53-year-old male with primary-refractory PCNSL. Pre-treatment staging showed PCNSL in the left parieto-occipital and posterior medullary areas, and normal body CT and FDG-PET-CT scans. Due to concern regarding *A. fumigatus*, we began surveillance chest CT

scans after each treatment cycle. A surveillance chest CT scan at cycle 3 of DA-TEDDi-R showed a new 1.2 cm right pulmonary nodule. During the first 2 treatment cycles, he had one day of neutropenia (< 500 cells/ μ l). Although the BAL and serum were negative for infectious etiologies, anti-fungal therapy with caspofungin and voriconazole was instituted due to a high suspicion of aspergillosis. A repeat chest CT three weeks later showed significant improvement with resolution after 3 weeks of treatment.

⁶ Patient #6 is a 64-year-old male with primary-refractory PCNSL. He began dexamethasone 16 mg/day 4-weeks before enrollment, which was reduced to 4 mg/day and tapered off over the next several weeks. Pre-treatment staging showed PCNSL in the choroid plexus of the lateral and 4th ventricles and normal body CT and FDG-PET-CT scans. A surveillance chest CT scan at cycle 2 of DA-TEDDi-R showed new bilateral pulmonary nodules, including a 1.4 cm right lower cavitory lesion, and diffuse pulmonary infiltrates. During the first treatment cycle, he had 4 days of neutropenia (< 500 cells/ μ l). A BAL was positive for *P. jirovecii* and the serum beta-D-glucan was elevated at >500 pg/ml (normal < 60 pg/ml). Three weeks after treatment for *P. jirovecii* with atovaquone, and aspergillosis with caspofungin, liposomal amphotericin and voriconazole, the diffuse pulmonary infiltrates resolved and the cavitory nodule reduced in size. A repeat chest CT showed almost complete resolution of the pulmonary nodules after 6 weeks of treatment.

⁷ Patient #7 is a 49-year-old male with refractory PCNSL after 3 prior treatments. He began dexamethasone 6 mgs/day 2-weeks before enrollment, which was tapered to replacement prednisone. Pre-treatment staging demonstrated PCNSL in the right frontal, left parietal, and bilateral temporal lobes and normal body CT and FDG-PET-CT scans. On day 13 of cycle 1 of DA-TEDDi-R, he was admitted with febrile neutropenia and worsening mental status. He had 5 documented days of neutropenia (< 500 cells/ μ l). A chest CT revealed bilateral lung nodules, up to 2.2 cm in diameter, and an MRI showed a new diffusion restricted lesion in the left caudate head. A BAL culture showed *Aspergillus fumigatus* and CSF was positive for aspergillus galactomannan antigen. He began antifungal treatment and the lesions resolved.

Table S2, related to Table 3. Ibrutinib C_{max} (nM) Before and During DA-TEDDi-R

Pt Number	Ibrutinib Dose Level	Cycle 1, Day -14 2 hours	Cycle 1, Day -1 2 hours	Cycle 2, Day 5 2 hours	Cycle 4, Day 5 2 hours
Plasma					
1	560 mg	247	-	-	-
2	560 mg	145	-	-	-
3	560 mg	77	118	-	-
4	560 mg	44	-	-	-
5	560 mg	163	368	-	35
6	560 mg	88	198	-	221
7	700 mg	522	327	34	-
8	700 mg	411	12	5	25
9	700 mg	164	-	-	-
10	700 mg	577	316	208	180
11	840 mg	114	40	167	-
12	840 mg	247	465	98	53
13	840 mg	427	-	-	-
14	840 mg	270	933	1058	151
15	840 mg	122	243	137	225
16	840 mg	151	415	-	-
17	840 mg	10	58	318	-
18	840 mg	97	-	-	-
Median		157	280	208	151
Range		10-522	12-933	5-1058	25-225
CSF					
1	560 mg	2.0	-	-	-
2	560 mg	0.7	-	-	-
3	560 mg	2.4	-	-	-
4	560 mg	1.2	-	-	-
5	560 mg	2.0	5.1	-	0.4
6	560 mg	0.7	1.3	-	1.6
7	700 mg	11.1	4.2	0.5	-
8	700 mg	1.6	0.4	-	0.3
9	700 mg	0.5	-	-	-
10	700 mg	2.4	2.4	2.3	3.5
11	840 mg	1.7	0.3	2.1	-
12	840 mg	1.4	3.8	0.9	-
13	840 mg	1.0	-	-	-
14	840 mg	2.7	5.3	5.6	1.2
15	840 mg	0.5	0.7	1.1	2.9
16	840 mg	0.7	1.4	-	-
17	840 mg	NA	0.5	2.7	-
18	840 mg	0.5	-	-	-
Median		1.4	1.4	2.1	1.4
Range		0.5-11.1	0.3-5.3	0.5-5.6	0.3-3.5

Table S3, related to Table 3. Metabolite PCI-45227 C_{max} (nM) Before and During DA-TEDDi-R

Pt Number	Ibrutinib Dose Level	Cycle 1, Day -14 2 hours	Cycle 1, Day -1 2 hours	Cycle 2, Day 5 2 hours	Cycle 4, Day 5 2 hours
Plasma					
1	560 mg	202	-	-	-
2	560 mg	313	-	-	-
3	560 mg	111	-	-	-
4	560 mg	133	-	-	-
5	560 mg	172	414	-	65
6	560 mg	188	94	-	290
7	700 mg	466	491	46	-
8	700 mg	286	31	14	78
9	700 mg	265	-	-	-
10	700 mg	456	252	361	357
11	840 mg	296	152	433	-
12	840 mg	214	349	199	83
13	840 mg	536	-	-	-
14	840 mg	288	473	342	148
15	840 mg	235	328	117	286
16	840 mg	508	594	-	-
17	840 mg	55	159	357	-
18	840 mg	149	-	-	-
Median		250	328	271	148
Range		55-536	31-594	14-433	65-357
CSF					
1	560 mg	4.3	-	-	-
2	560 mg	5.0	-	-	-
3	560 mg	2.1	-	-	-
4	560 mg	2.4	-	2.5	-
5	560 mg	2.9	9.7	1.5	0.2
6	560 mg	3.0	1.9	-	5.4
7	700 mg	13.6	9.9	2.1	-
8	700 mg	2.7	1.9	1.9	1.8
9	700 mg	1.0	0.0	-	-
10	700 mg	4.0	5.4	10.7	10.6
11	840 mg	9.4	2.6	6.9	-
12	840 mg	2.4	8.8	3.1	-
13	840 mg	2.4	-	-	-
14	840 mg	5.5	6.3	7.0	3.5
15	840 mg	1.9	5.8	2.7	5.7
16	840 mg	3.0	4.3	-	-
17	840 mg	1.2	3.4	5.8	-
18	840 mg	1.5	-	-	-
Median		2.8	5.4	2.9	4.5
Range		1-13.6	1.9-9.9	1.5-10.7	0.2-10.6