TRILACICLIB, A CDK4/6 INHIBITOR, DOSED WITH GEMCITABINE, CARBOPLATIN IN METASTATIC TRIPLE NEGATIVE BREAST CANCER (mTNBC) PATIENTS: PRELIMINARY PHASE 2 RESULTS

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BACKGROUND

- Clinically significant, multi-lineage myelosuppression is a major acute toxicity of cytotoxic chemotherapy leading to hematologic toxicities and subsequent dose reductions and delays
- Trilaciclib, a cyclin-dependent kinase 4 and 6 (CDK4/6) inhibitor, is being developed to reduce the myelosuppressive effects of chemotherapy and preserve immune system function rather than directly target tumor proliferation (Table 1)
- A previously reported randomized, double-blind, Phase 1b/2 trial in small cell lung cancer (SCLC) demonstrated myelopreservation benefits of trilaciclib, including reduced multi-lineage myelosuppression (neutrophils, red blood cells (RBCs), lymphocytes), reduced supportive care requirements, and decreased dose reductions (Dragnev et al. ESMO 2018)
- Metastatic triple negative breast cancer (mTNBC) was chosen to further evaluate myelopreservation and tumor efficacy of trilaciclib because:
  - mTNBC is predominantly a functionally CDK4/6-independent disease, allowing assessment of trilaciclib's effects on the host without any potential direct effects on the tumor
  - Cytotoxic therapy is the backbone of treatment for mTNBC and is often limited by myelotoxicities
- Here, we report randomized, open-label efficacy and safety data of the addition of trilaciclib to standard mTNBC cytotoxic chemotherapy

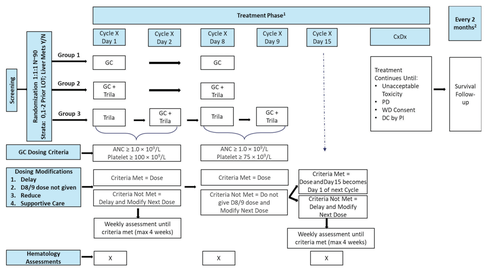
TABLE 1. TRILACICLIB DIFFERS FROM APPROVED CDK4/6 INHIBITORS

	Trilaciclib	Approved CDK4/6 Inhibitors
Target Population	Both CDK4/6-independent and -dependent tumors	CDK4/6-dependent tumors (e.g., HR+ HER2- BC)
Mechanism of Action	Transiently arrests cells in the G1 phase; preserves hematopoietic stem and progenitor cell (HSPC) and immune system function during chemotherapy (myelopreservation); Potentially enhances efficacy of combination treatment	Inhibits tumor proliferation
Route of Administration	IV, intermittent dosing	Oral, chronic dosing
Potential Combination Treatments	Chemotherapy and/or checkpoint inhibitors	Growth-signaling inhibitors (e.g., SERD, EGFRi)
Impact to HSPCs	Transient inhibition of HSPCs reduces chemotherapy-induced myelosuppression across multiple lineages	Chronic inhibition of HSPCs causes myelosuppression

STUDY DESIGN

- Phase 2, open-label, global, multicenter study of the safety, efficacy, and pharmacokinetics of trilaciclib in combination with gemcitabine and carboplatin (GC) chemotherapy in patients with mTNBC
- TNBC is defined as hormone (estrogen and progesterone) receptor negative (local assessment of IHC; < 10% nuclei staining) and HER2-negative per ASCO CAP
- Patients had adequate organ function, ECOG 0 or 1, CNS disease not requiring immediate intervention, and 0-2 prior lines of therapy in the locally recurrent/metastatic setting: systemic therapy in the neoadjuvant/adjuvant setting was considered a line of therapy when disease recurred ≤ 12 months after treatment
- Prophylactic growth factors were not allowed in cycle 1; otherwise, supportive care was allowed as needed
- Endpoints were pre-specified to assess the effect of trilaciclib on:
  - Multi-lineage myelosuppression endpoints: the occurrence of severe (Grade 4) neutropenia, RBC transfusions, granulocyte-colony stimulating factor (GCSF) administrations, platelet transfusions, and the duration of severe neutropenia
  - Adverse events (AEs) and additional safety endpoints
  - Pre-specified exploratory composite endpoint: Major Adverse Hematologic Events (MAHE)
  - Antitumor efficacy: evaluated based on RECIST, Version 1.1 for best overall response (BOR), objective response rate (ORR), and progression-free survival (PFS); overall survival (OS)
- Data presented here are from the following data cuts: 30Jul2018 for multi-lineage myelosuppression endpoints and 01Nov2018 for tumor efficacy endpoints

FIGURE 1. STUDY SCHEMATIC



[1] 21 day cycles: Trilaciclib administered IV prior to GC infusions. Trilaciclib 240 mg/m²; Gemcitabine 1000 mg/m²; Carboplatin AUC 2

[2] Survival Follow-Up continues until at least 50% of events have occurred; 80% target for mature OS

Peripheral blood samples were collected predose, on Day 1 of odd cycles, at the PTV, and at the first Survival FU for flow cytometric analysis

Tumor assessments completed every 9 weeks through Week 39 and every 12 weeks thereafter until PD, WD consent, starting off-study anticancer therapy, or study completion

LOT=Lines of Therapy, Trila=Trilaciclib, PD=Progressive Disease, WD=Withdrew, DC=discontinued, PI=Principal Investigator, ANC=absolute neutrophil count, IV=Intravenous, AUC=area under curve, OS=Overall Survival, PTV=Post Treatment Visit, FU=Follow Up

RESULTS

TABLE 2. DEMOGRAPHICS AND BASELINE DISEASE CHARACTERISTICS

	Group 1 (GC)	Group 2 (GC + trila x 1)	Group 3 (GC + trila x 2)	Total
Patients randomized, n	34	33	35	102
Number of prior lines of systemic therapy in recurrent/metastatic setting per protocol, n (%)				
0	21 (61.8)	22 (66.7)	21 (60.0)	64 (62.7)
1 or 2	13 (38.2)	11 (33.3)	14 (40.0)	38 (37.3)
Liver involvement, n (%)				
Yes	8 (23.5)	8 (24.2)	10 (28.6)	26 (25.5)
No	26 (76.5)	25 (75.8)	25 (71.4)	76 (74.5)
Brain scan results, n (%)				
Brain metastases	3 (8.8)	2 (6.1)	1 (2.9)	6 (5.9)
No brain metastases	14 (41.2)	13 (39.4)	16 (45.7)	43 (42.2)
Not completed	17 (50.0)	18 (54.5)	18 (51.4)	53 (52.0)
Tumor subtyping, n (%)	28 (82.4)	30 (90.9)	32 (91.4)	90 (88.2)
Basal-like	14 (50.0)	17 (56.7)	16 (50.0)	47 (52.2)

Four patients in Group 1 were randomized but withdrew consent and were never treated. All patients randomized were female with the exception of 1 male randomized to Group 2. Basal-like was defined using the PAM50 analysis and frequency determined using patients with available RNA expression data.

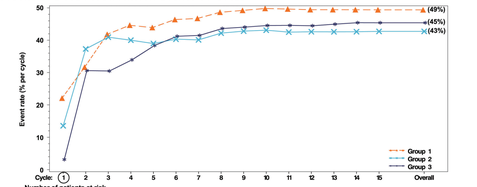
- Overall demographic data are similar across the groups: mean age (55-58 years), race distribution (66.7-82.4% white), country (15.2-22.9% ex-US), ECOG (40.0-55.9% ECOG=1), prior systemic anticancer treatment (75.8-85.7% any treatment), breast cancer gene (BRCA) classification (61.8-68.6% untested)
- 4 (Group 1),10 (Group 2), and 9 (Group 3) patients are still on treatment
- Treatment discontinuation due to adverse events is similar across groups: 7 (Group 1), 6 (Group 2), and 6 (Group 3)
- 29% of patients had died at the time of the tumor efficacy data cut (11 (Group 1), 9 (Group 2), and 10 (Group 3)); all but 3 deaths were due to disease progression (one ventricular failure, one influenza A (Group 1) and one unknown (Group 2))

TABLE 3. SUMMARY OF DOSE EXPOSURE AND DOSE MODIFICATIONS

	Group 1 (GC)	Group 2 (GC + trila x 1)	Group 3 (GC + trila x 2)
Safety Analysis Set, n	30	33	35
Duration of exposure			
Weeks, median (min, max)	14.4 (3, 36)	20.0 (6, 49)	19.0 (3, 47)
Cycles, median (min, max)	4 (1, 11)	7 (2, 15)	6 (1, 14)
Cumulative Dose			
Gemcitabine dose, median mg/m²	7306.2	9643.7	10959.6
min, max	1000.0, 20000.0	3000.0, 29400.0	2000.0, 25204.5
Carboplatin dose, median AUC	15	20	20
min, max	2.0, 40.0	6.0, 60.0	4.0, 51.0
Cycles where D8/9 dose was not given (%)	16.7	15.9	8.9
Days to first D8/9 dose not given, mean (min, max)	48.7 (8, 190)	33.0 (8, 80)	84.3 (9, 184)

- Relative dose intensity of GC is similar across Groups (80.4% - 84.3%) (data not shown)
- The addition of trilaciclib to GC increases the duration of exposure and cumulative dose of GC compared to patients treated with GC alone

FIGURE 2. DOSE MODIFICATION EVENTS BY CYCLE



Dose modifications are defined as (1) dose reductions, (2) cycle delays, and (3) D8/9 dose not given.

- The addition of trilaciclib to GC decreases the rate of dose modification events over time with the greatest decrease in cycle 1 compared to GC alone

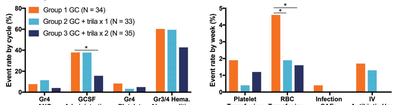
TABLE 4. PROSPECTIVELY DEFINED MYELOSUPPRESSION ENDPOINTS

	Group 1 (GC)	Group 2 (GC + trila x 1)	Group 3 (GC + trila x 2)
Intent to treat, n	34	33	35
Total weeks of treatment, n	522	702	752
Patients with severe neutropenia, n (%)	9 (26.5)	12 (36.4)	8 (22.9)
Patients receiving RBC transfusions on/after 5 weeks, n (%)	12 (35.3)	11 (33.3)	8 (22.9)
Patients receiving GCSF, n (%)	16 (47.1)	21 (63.6)	14 (40.0)
Patients receiving platelet transfusions, n (%)	4 (11.8)	3 (9.1)	6 (17.1)
Days of severe neutropenia in Cycle 1, mean (SD)	1 (2.4)	2 (3.5)	1 (2.6)

SD = standard deviation

- While the number of patients experiencing myelosuppression events is not significantly different across the Groups, this does not take into account the increase in drug exposure observed in patients receiving GC + trilaciclib compared to patients receiving GC alone
- Although the number of patients that received RBC transfusions is similar across Groups, the mean units/patient is lower in patients that received GC + trilaciclib (2 units per Group) compared to GC alone (3 units)
- Although the number of patients that received platelet transfusions is similar across Groups, the mean units/patient is lower in patients that received GC + trilaciclib (4 units in Group 2, 3 units in Group 3) compared to GC alone (8 units)

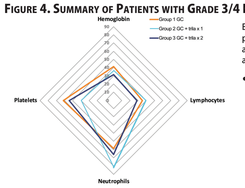
FIGURE 3. FREQUENCY OF MYELOSUPPRESSION EVENTS



Event unit varies due to the timing in which events may occur for the patient (cycle vs week). Events occurring by cycle are limited by the lab sampling schedule within the cycle (total # cycles with event/total # cycles administered). Events occurring by week are events that can occur at any time during study participation (total # events/total duration of treatment in weeks), with the exception of RBC transfusions, which were included only if occurring on/after 5 weeks. A negative binomial regression model was used to evaluate the statistical differences by Group. *p≤0.05.

- The addition of trilaciclib to GC decreases the frequency of occurrence of myelosuppression events and their consequences compared to GC alone

FIGURE 4. SUMMARY OF PATIENTS WITH GRADE 3/4 HEMATOLOGIC LABORATORY ABNORMALITIES



Each axis of the chart represents the proportion (%) of patients with a grade 3/4 hematologic laboratory abnormality. These abnormalities were compared across treatment groups using a multivariate test.

- Not only do patients that receive GC + trilaciclib have higher exposure to GC, they also experience fewer grade 3/4 lab abnormalities, with the exception of neutropenia, compared to patients receiving GC alone

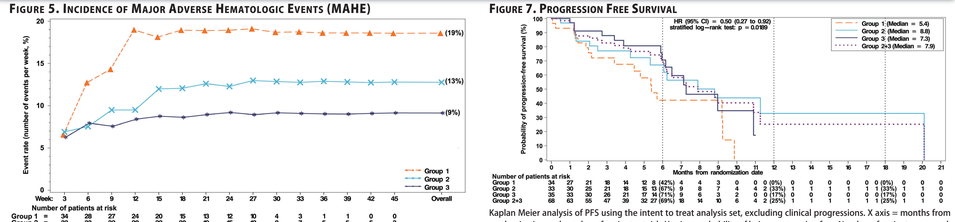
TABLE 5. TREATMENT EMERGENT ADVERSE EVENTS (TEAES) (≥20% OF PATIENTS)

	Group 1 (GC) N=30		Group 2 (GC + trila x 1) N=33		Group 3 (GC + trila x 2) N=35	
Preferred Term	All Grades	Grade ≥ 3	All Grades	Grade ≥ 3	All Grades	Grade ≥ 3
Any TEAEs, n (%)	30 (100.0)	25 (83.3)	33 (100.0)	29 (87.9)	34 (97.1)	30 (85.7)
Neutropenia	19 (63.3)	18 (60.0)	27 (81.8)	26 (78.8)	22 (62.9)	19 (54.3)
Thrombocytopenia	17 (56.7)	13 (43.3)	15 (45.5)	8 (24.2)	21 (60.0)	14 (40.0)
Anemia	20 (66.7)	12 (40.0)	15 (45.5)	6 (18.2)	15 (42.9)	11 (31.4)
Fatigue	11 (36.7)	1 (3.3)	14 (42.4)	1 (3.0)	14 (40.0)	2 (5.7)
Nausea	6 (20.0)	0	14 (42.4)	0	16 (45.7)	1 (2.9)
Vomiting	8 (26.7)	0	7 (21.2)	1 (3.0)	11 (31.4)	0
Headache	5 (16.7)	0	7 (21.2)	0	13 (37.1)	0
Constipation	5 (16.7)	0	7 (21.2)	0	9 (25.7)	0

Note: Across all groups, TEAE Grade ≥3 data include only one Grade 5 TEAE (Group 1, right ventricular failure, unrelated)

- The most frequent TEAEs are common events attributable to cytotoxic chemotherapy
- No trilaciclib related serious adverse events (SAEs) reported to date

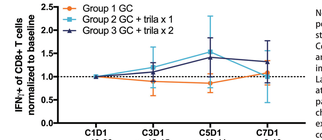
FIGURE 5. INCIDENCE OF MAJOR ADVERSE HEMATOLOGIC EVENTS (MAHE)



MAHE is a pre-specified exploratory composite measure of trilaciclib effects, which include the following individual components: all-cause hospitalizations, all-cause dose reductions, febrile neutropenia, prolonged severe neutropenia (G4 ≥ 5 days), RBC transfusions on/after 5 weeks, and platelet transfusions. The number of events for each component is derived as the number of episodes with a unique start date during the treatment period, with the exception of prolonged severe neutropenia and all-cause dose reduction, which are reported as number of cycles with any episodes. The graph depicts mean cumulative instances over time. A negative binomial regression model was used to evaluate the statistical differences between Group 1 and Group 3.

- The addition of trilaciclib to GC significantly decreases the rate of occurrence of MAHE (p=0.0181) compared to GC alone, with significant decreases in all-cause hospitalizations (p=0.0099), prolonged severe neutropenia (G4 ≥ 5 days) (p=0.0406), and RBC transfusions on/after 5 weeks (p=0.0197)

FIGURE 6. FLOW CYTOMETRY



Normalized mean frequency of IFNγ+ population of CD8+ T cells after *ex vivo* stimulation (IFNγ+IL-17A-(CD3+CD8+)). Cell populations in whole blood were analyzed by flow cytometry at the indicated time points at Covance Central Laboratories. Analyses were completed at Fios Genomics and included only patients who received 3+ cycles of chemotherapy with statistical outliers excluded. Error bars represent 95% confidence interval (CI).

- The ratio of total CD8+ T cells to regulatory T cells in peripheral blood is similar across Groups (data not shown)
- After *ex vivo* stimulation, there is a higher frequency of CD8+ T cells producing IFNγ in patients that received GC + trilaciclib compared to GC alone, suggesting a more functional lymphocyte population

TABLE 6. TUMOR RESPONSE AND PROGRESSION FREE SURVIVAL

	Group 1 (GC)	Group 2 (GC + trila x 1)	Group 3 (GC + trila x 2)
Response Evaluable Set, n	24	30*	30
Best Overall Response (BOR), n (%)			
Complete Response (CR)	0	0	0
Partial Response (PR)	7 (29.2)	13 (43.3)	11 (36.7)
Stable Disease (SD)	11 (45.8)	10 (33.3)	16 (53.3)
Progressive Disease (PD)	6 (25.0)	6 (20.0)	3 (10.0)
Objective Response Rate (ORR)			
CR+PR, n (%)	7 (29.2)	13 (43.3)	11 (36.7)
95% CI	12.6%, 51.1%	25.5%, 62.6%	19.9%, 56.1%
Intent to Treat, n	34	33	35
Median PFS (months, 95% CI)	5.4 (3.4, 9.2)	8.8 (5.3, 20.1)	7.3 (6.0, 10.9)
HR (vs Group 1, 95% CI)	N/A	0.52 (0.25, 1.09)	0.49 (0.24, 1.03)
Two-sided p-value	N/A	0.0669	0.0546

*Includes one patient that died due to disease progression before their first post-baseline tumor assessment.

PFS = progression free survival; HR = hazard ratio

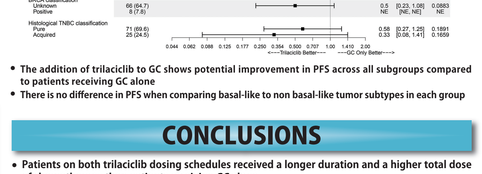
- Patients receiving GC + trilaciclib have a higher response rate and longer PFS compared to GC alone

FIGURE 7. PROGRESSION FREE SURVIVAL

Kaplan Meier analysis of PFS using the intent to treat analysis set, excluding clinical progressions. X axis = months from randomization and number of patients at risk; Y axis = probability of being progression free; Number of patients censored, n (%): Group 1 = 17 (50.0%), Group 2 = 18 (54.5%), Group 3 = 21 (60.0%); HR and p-values shown for Groups 2+3 vs Group 1

- Median PFS is longer when trilaciclib is added to GC compared to GC alone (5.4 months (Group 1), 7.9 months (Groups 2+3)) with a HR (95% CI) = 0.50 (0.27, 0.92), p=0.0189
- Analysis of 6-month PFS demonstrates the probability of patients remaining progression free at 6 months is higher for patients receiving GC + trilaciclib compared to patients receiving GC alone (42% (Group 1), 69% (Groups 2+3), p=0.0169), suggesting that adding trilaciclib to GC extends PFS

FIGURE 8. FOREST PLOT FOR PFS



- The addition of trilaciclib to GC shows potential improvement in PFS across all subgroups compared to patients receiving GC alone
- There is no difference in PFS when comparing basal-like to non basal-like tumor subtypes in each group

CONCLUSIONS

- Patients on both trilaciclib dosing schedules received a longer duration and a higher total dose of chemotherapy than patients receiving GC alone
- Adjusting for the duration of chemotherapy, trilaciclib demonstrated multi-lineage myelopreservation benefits (neutrophils, RBCs, and platelets)
- Patients receiving GC + trilaciclib had higher tumor response rates and longer PFS than patients receiving GC alone; OS is immature
- Patients receiving GC + trilaciclib had a lower rate of occurrence of MAHE, an exploratory composite measure, than patients receiving GC alone
- Trilaciclib was well tolerated and the overall adverse event profile was consistent with that of GC; no trilaciclib related serious adverse events were reported
- Trilaciclib is being evaluated concurrently in three other randomized Phase 2 studies: 1st line SCLC (+ etoposide/carboplatin NCT02499770), 1st line SCLC (+atezolizumab/etoposide/carboplatin; NCT03041311), and 2nd/3rd line SCLC (+topotecan; NCT02514447)

ACKNOWLEDGEMENTS

We thank and acknowledge all of the patients, their families and site personnel for participating in the study.