Abstract 573 phase II randomized window of opportunity trial evaluating cytotoxic and immunomodulatory effects of intratumoral INT230-6 in early stage breast cancer: The INVINCIBLE Trial

Arnaout A[1,2], Robertson S[3], Keyhanian K[3], Hopkins M[2], Liao L[2], Talebian V[2], Awan A,[1] Bartlett JMS[4], Pond GR[2,5], Radvanyi L[2], Bender LH[6], Walters IB[6], Lopez Ozuna V[1], Spears M[2,7].

[1]The Ottawa Hospital Research Institute, Ottawa, Ontario, Canada, [2]Ontario Institute of Cancer Research, Toronto, Ontario, Canada. [3]Department of Anatomical Pathology, University of Ottawa, Ottawa, Ontario, Canada, [4]University of Edinburgh, Edinburgh, United Kingdom [5]Faculty of Health Sciences, McMaster University, Hamilton, Ontario, Canada. [6]Intensity Therapeutics Inc., Westport, CT [7] Department of Laboratory Medicine and Pathobiology, University of Toronto, Toronto, Ontario, Canada.

Background:

- Majority of breast cancers outside of the triple negative subtype are considered immune cold and minimally responsive to immunotherapies.
- INT230-6 is a novel product with a unique dual anti-cancer mechanism that achieves both direct cancer cell necrosis and immune cell activation. The drug is comprised of cisplatin (CIS) and vinblastine (VIN) co-formulated with an, cell penetration and unique tissue dispersing enhancing molecule, 8-((2-hydroxybenzoyl)amino) octanoate (SHAO).
- Previous in vitro studies have demonstrated that INT230-6 halts cancer cell replication and induces apoptosis while maturing dendritic cells and recruiting T-cells to the tumor microenvironment.

Objective:

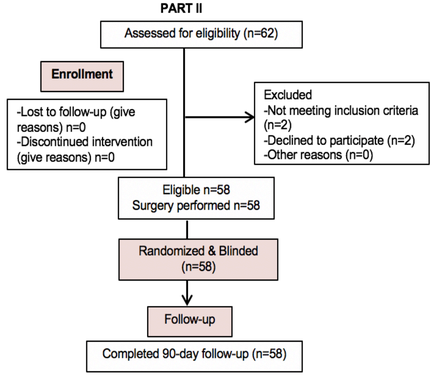
To assess the effect of INT230-6 on the tumour and the tumour microenvironment

Methods:

- We have conducted a randomized, Phase 2 presurgical Window-Of-Opportunity trial for intratumoral (IT) INT230-6 evaluating clinical and BioLogical Effects in patients with early-stage operable Breast Cancer (the INVINCIBLE trial- https://clinicaltrials.gov/ct2/show/NCT04781725).
- 60 women with newly diagnosed operable early-stage intermediate or high-grade T1-T2 invasive breast cancers are randomly allocated (2:1) prior to resection to Intratumoral injections of INT230-6, no treatment or saline sham.
- DNA, RNA were extracted pre- and post-treatment samples
- Mutations and copy number alterations were assessed using the Oncomine Comprehensive Assay plus (Thermofisher).
- Gene expression was measured using Ion AmpliSeq Transcriptome Human Gene Expression Kit (Thermofisher)

- INT230-6 can cause immune priming therapy in historically immune quiescent breast cancers.
- INT230-6 induced TCR signaling, macrophage markers, IL-18 signaling and B cell receptor signaling.

Patient cohort:



Part 2	Total	Injection n= 58
Mean Age (yrs)	58	58.6
Tumor Size Range (cm)	58	1.5-4.0
Mean Tumor Size (cm)	58	2.4
Invasive Ductal	40	68.90%
Invasive Lobular	15	25.86%
Invasive Carcinoma NOS	3	5.17%
ER/PR+ Her2-	40	68.90%
ER/PR+ Her2+	2	3.44%
ER-PR-Her2-	0	0%
1 injection	58	100%
2 injections	0	0%
3 injections	0	0%
Dose Range	58	2-24.5ml
Lumpectomy	48	83%
Mastectomy	10	17%

Figure 1. REMARK diagram and table describing the patient characteristics for part II of the study

Results:

Gene enrichment pathway analysis demonstrated induced signaling in the post-treatment samples.

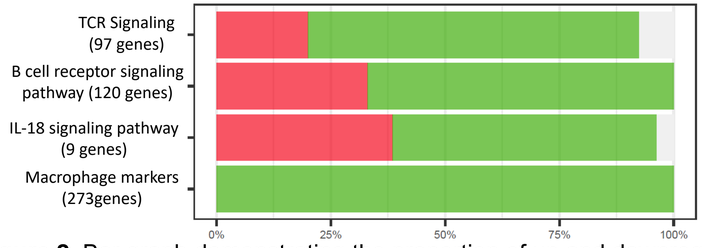


Figure 2. Bar graph demonstrating the proportion of up and downregulated gene in the drug treated patients comparing pre- and post-treatment samples.

Results:

Molecular profiling the cohort

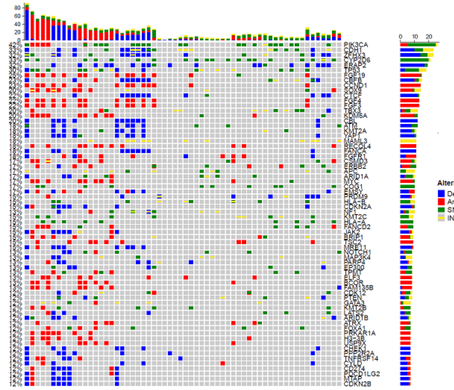


Figure 3. Oncomine Comprehensive Plus Assay was used to profile the pre-treatment samples. Top bar graph: total number of alterations in each patient. Right bar graph: Total number of genomic alterations for each gene.

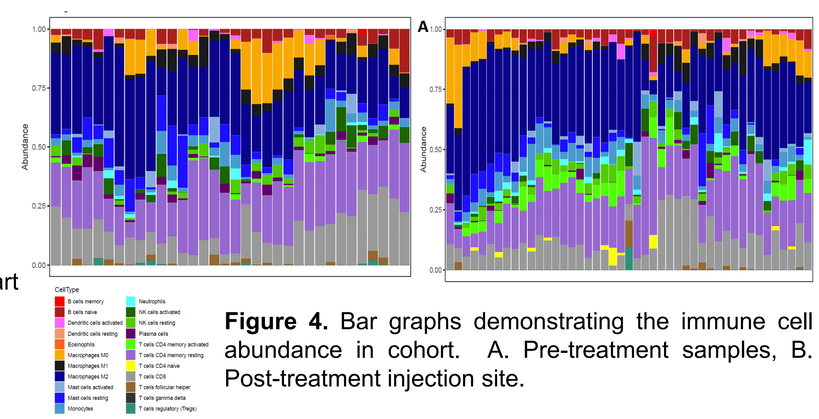


Figure 4. Bar graphs demonstrating the immune cell abundance in cohort. A. Pre-treatment samples, B. Post-treatment injection site.

- Most frequently mutated included PIK3CA (38%), TP53 (23%), CDH1 (22%) and TBX3 (17%).
- Frequent copy number changes in CCND1 (23%), FGFR1 (18%), and CDH1 (17%).
- Post-treatment demonstrated increase expression levels of DC, macrophages and CD4 T cells.
- Pathway analysis demonstrated up-regulation in TCR signaling, macrophage markers, IL-18 signaling and B cell receptor signaling.