

Injection site reactions correlate to delayed type hypersensitivity tests and suggest that GP2 reverses immune suppression of trastuzumab-treated HER2 positive patients in a phase IIb study evaluating HER2/neu peptide (GLSI-100) vs. GM-CSF after adjuvant trastuzumab in HER2 positive women with breast cancer

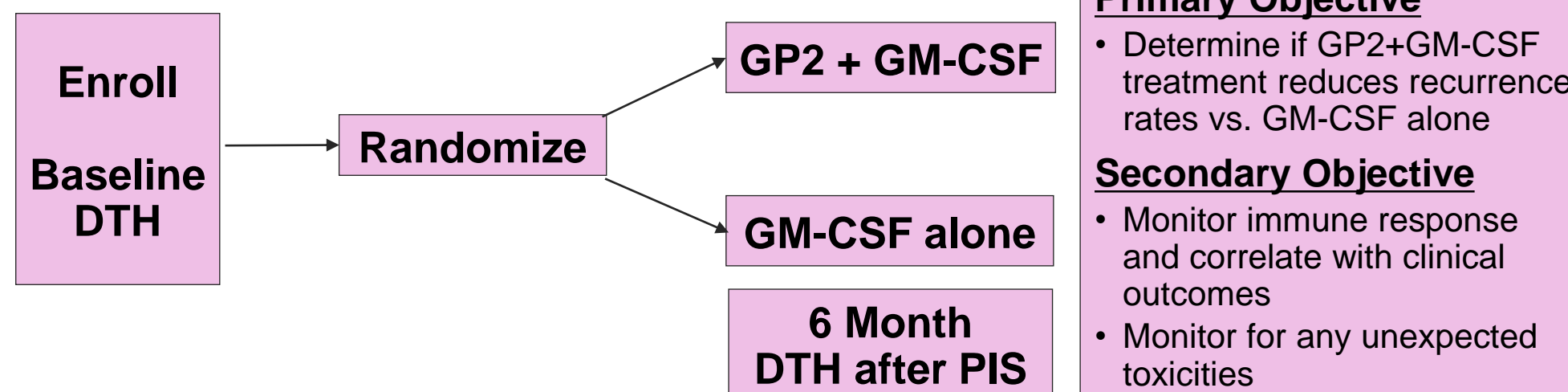
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BACKGROUND

The results of a prospective, randomized, placebo-controlled, single-blinded, multicenter Phase IIb trial investigating GLSI-100 (GP2+GM-CSF) administered in the extended-adjuvant setting to node-positive and high-risk node-negative breast cancer patients with tumors expressing any degree of HER2 (immuno-histochemistry [IHC] 1-3+) (NCT00524277) have been reported. The trial enrolled HLA-A*02 patients randomized to receive GLSI-100 versus GM-CSF alone. It was previously reported that completion of the GLSI-100 Primary Immunization Series (PIS) reduced recurrence rates to 0% over a 5 year follow-up period in HER2 3+ patients, who received a standard course of trastuzumab after surgery. Interim analyses for this trial have been previously reported by Mittendorf et al.

METHODS

Enrolled and consented patients were randomly scheduled to receive a total of 6 GLSI-100 (500 mcg GP2: 125 mcg GM-CSF) or GM-CSF only intradermal injections every 3-4 weeks as part of the PIS for the first 6 months and 4 GLSI-100 or GM-CSF only booster intradermal injections every 6 months thereafter. Injection site reactions (ISR) were measured 48-72 hours after injection. Delayed-type hypersensitivity (DTH) to GP2 was performed at baseline and after 6 months of treatment. For the DTH test, 0.5 mL consisting of 100 mcg of GP2 reconstituted in bacteriostatic saline for injection was placed intradermally. The DTH reaction is measured 48 - 72 hours after injection.



RESULTS

This trial explored HER2 3+ (positive) patients, who generally received a standard course of trastuzumab after surgery, and HER2 1-2+ patients, who did not receive trastuzumab after surgery. The orthogonal mean of the largest diameter of the injection site was measured 48-72 hours after injection. A DTH reaction was used to assess in vivo immune responses in patients prior to exposure to study medication and after 6 months of the first dose. The DTH orthogonal mean was measured 48-72 hours after DTH placement using the sensitive ballpoint-pen method. Previous presentations have reported an increase in DTH reactions after the PIS compared to baseline DTH reactions.

Figure 1: Disease Free Survival and Injection Site Reactions in HER2 3+ (Positive) Population

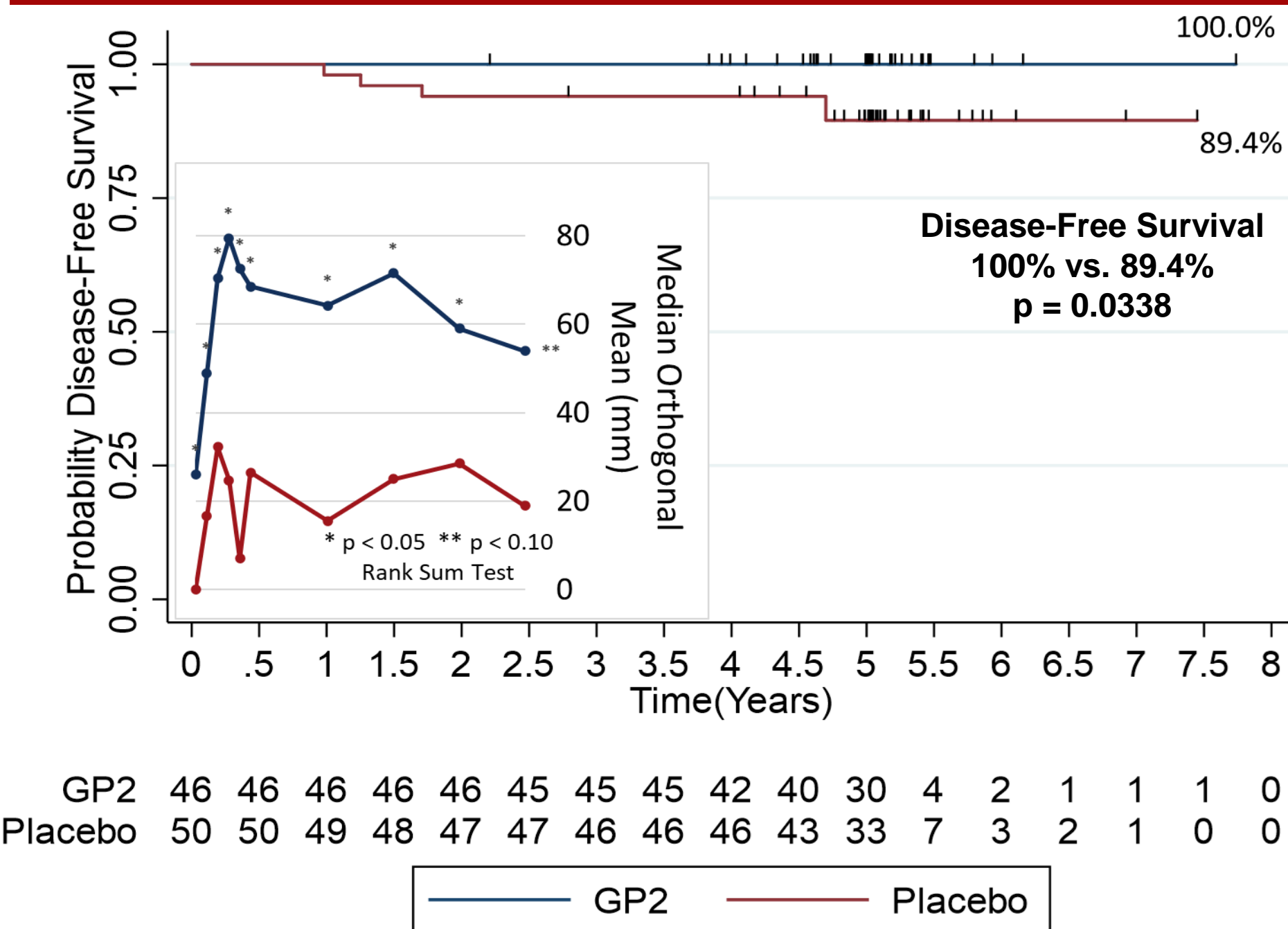


Figure 1: After 5 years of follow-up, the Kaplan-Meier estimated 5-year DFS rate in the 46 HER2 3+ patients treated with GLSI-100, if the patients were treated, followed, and remained disease free over the first six months of treatment, was 100% versus 89.4% (95% CI:76.2, 95.5%) in the 50 placebo patients treated with GM-CSF ($p = 0.0338$). Figure 1 describes the median orthogonal mean of the ISRs over time. Each dose of GLSI-100 generated a significantly larger ISR than the control group. Note that subjects who experienced ISRs larger than 100mm had the subsequent dose of GM-CSF reduced which could have caused the slight reduction in ISRs thereafter.

Figure 2: As shown in Figure 2, the magnitude of ISRs for patients treated with GLSI-100 was similar across HER2 status. However, HER2 3+ control patients had significantly smaller reactions to GM-CSF than HER2 1-2+ control patients did, with an average of 43.1mm difference over all 10 doses. Trastuzumab treated HER2 3+ control patients appeared to be in a suppressed immune state that was reversed by adding GP2 treatment, increasing injection site reactions by an average of 39.2mm over all 10 doses.

Figure 3: In GLSI-100 treated subjects, the DTH response at month 6 was found to be correlated with the ISR at the nearest dose, Dose 6 ($p = 0.41$, $p = 0.0085$). In addition, DTH response at month 6 was found to be correlated with most ISRs.

Figure 2: Median Orthogonal Mean Injection Site Reactions by Treatment and HER2 Status

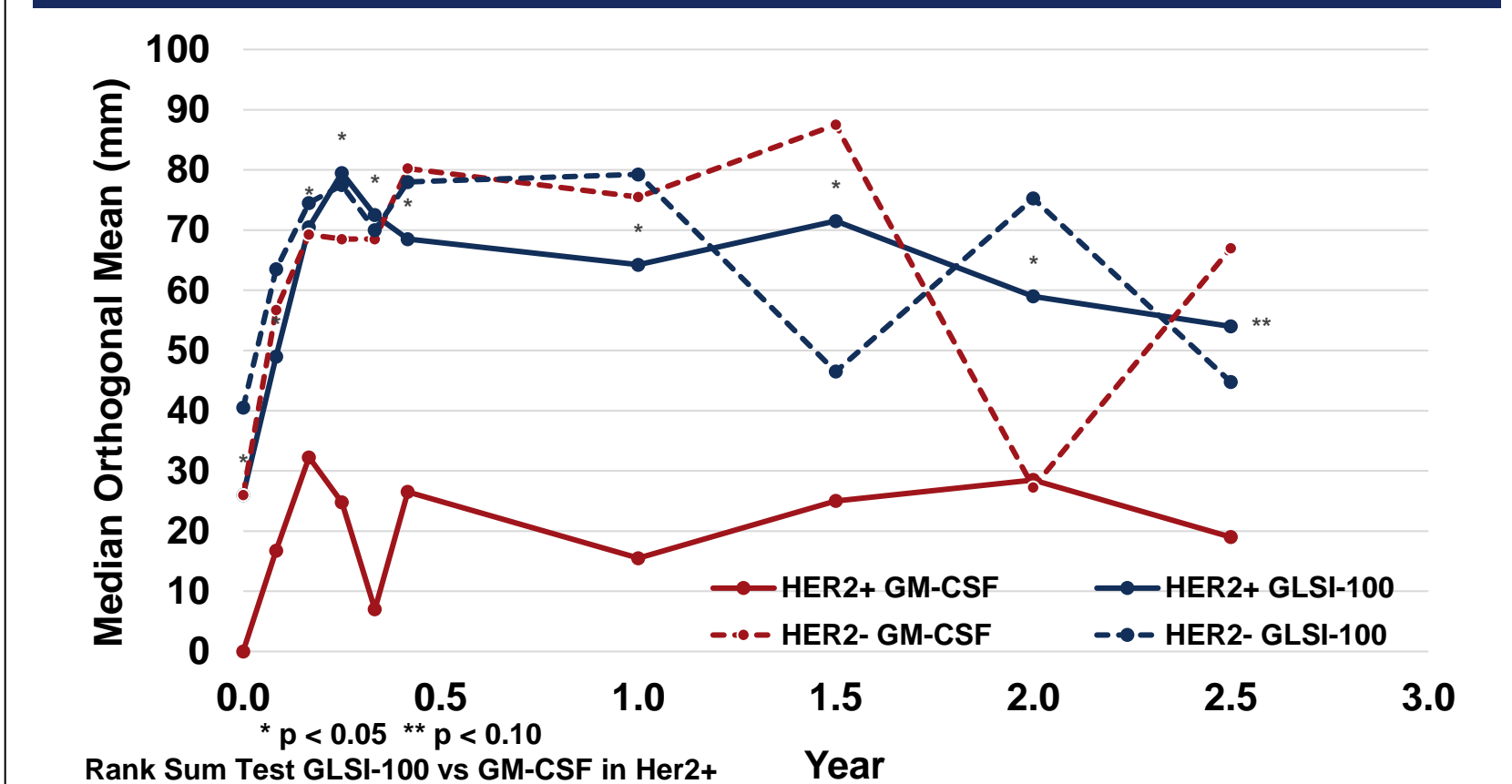
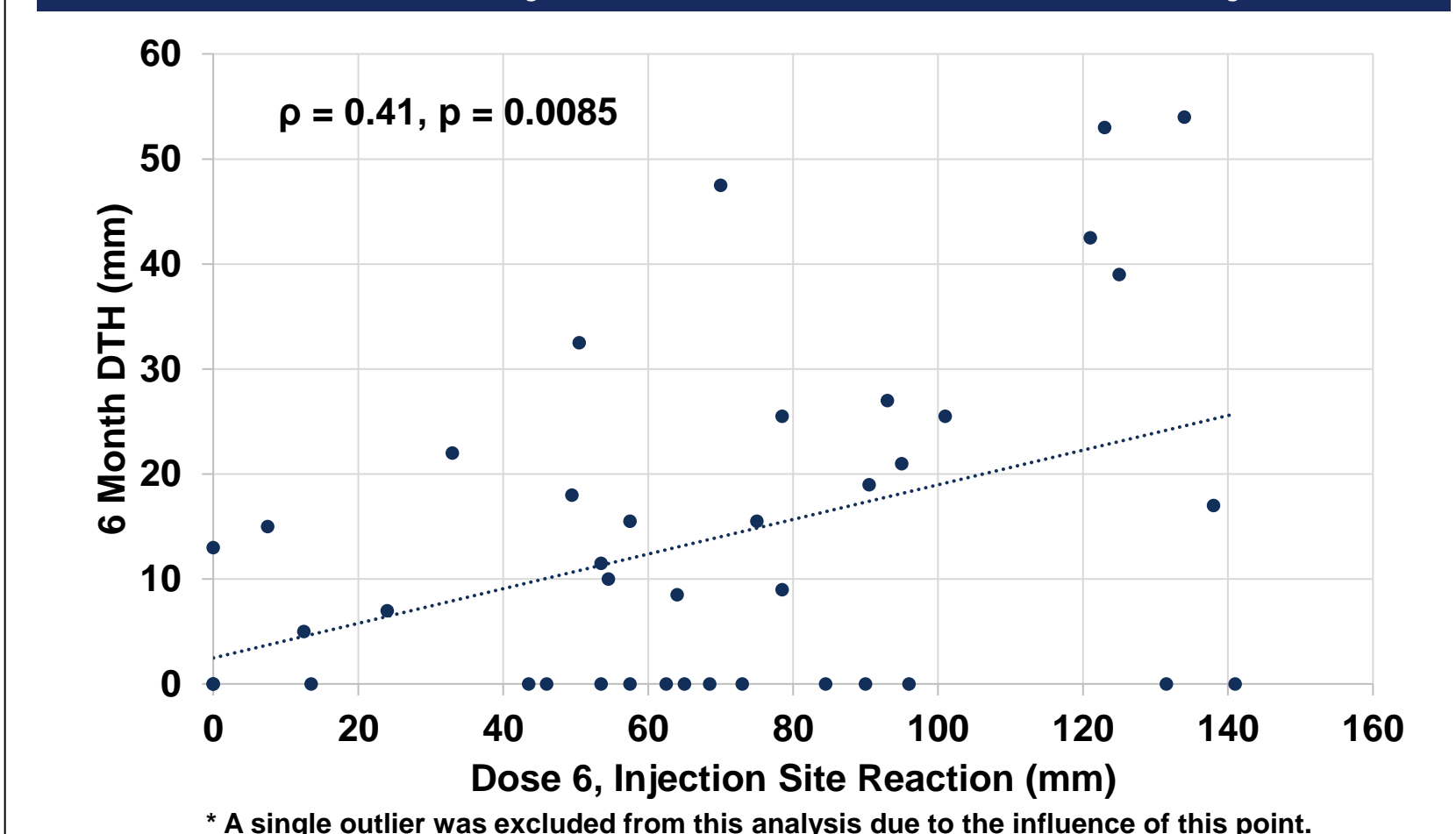


Figure 3: Correlation Between Month 6 DTH and Injection Site Reaction at Dose 6 Injection in GLSI-100 Treated Subjects



DISCUSSION AND CONCLUSIONS

This study demonstrated that GLSI-100 safely elicited a potent immune response, as evidenced by ISRs that correlate to and may serve as a complement to immune response data such as DTH. The lower immune response of the generally trastuzumab treated HER2 positive control population, not evidenced in the low HER2 expressors who did not receive trastuzumab, suggests a reduced immune state potentially related to prior trastuzumab exposure and/or HER2 positivity, which is reversed with the addition of GLSI-100 and warrants further research.

ACKNOWLEDGEMENTS

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