4610 S. 44th Place; Phoenix, AZ 85040; Ph: 800.901.5177, Fax: 866.479.4925

Sugarbaker David, MD
The Clearity Foundation
75 Francis Street
Boston, MA 02115

Reference : Walker, Samantha (TN12-106698)

Dear Dr. David,

Please find attached the results for your patient, **Samantha Walker**. The Caris Target Now™ final report provides detailed evidence-based molecular profiling information. This information includes expression levels of specific biomarkers, for the tumor specimen submitted for analysis, and identifies therapeutic options that may be associated with possible clinical benefit, as well as lack of clinical benefit, based upon scientific and medical literature.

Caris Target Now™ combines comprehensive molecular pathology and tumor profiling with an extensive evidence-based review of the latest clinical literature on biomarkers and their correlation with drug response. This service provides patient-specific information that can be used to personalize cancer therapy for your patients.

Additionally, we recognize that certain therapies associated with clinical benefit may not be covered by insurance or may require addition information for coverage. Given that the therapies identified in the literature and provided in the Caris Target Now™ report are evidence-based, we offer assistance in obtaining insurance coverage for patients who have been denied therapy or for patients who require advanced approval for non-standard therapy.

If you would like more information about our reimbursement services, or would like to discuss the results for this case, please contact Client Services at 800-901-5177.

Sincerely,

Michael Castro, MD
Medical Oncologist,
Senior Medical Director, Caris Target Now™
Caris Life Sciences

Sandeep Reddy, MD
Medical Oncologist,
Senior Medical Director, Caris Target Now™
Caris Life Sciences

cc: of Pathology, Department, Brigham and Women's Hospital, Dept. of Pathology

Patient Information	Specimen Information	Ordered By
Walker, Samantha **Case Number:** TN12-106698 **Date Of Birth:** [redacted] **Sex:** Female	**Primary Tumor Site:** Ovary **Specimen Site:** Pleura **Specimen Collected:** 05/10/2012 **Specimen Received:** 06/05/2012 **Date Reported:** 06/07/2012	**Sugarbaker David, MD** **The Clearity Foundation** 75 Francis Street Boston, MA 02115 (617) 732-7510

Caris Target Now Final Report

TN01-2012-04-23.0

Clinical History

Per the submitted surgical pathology report, the patient is a 42 year-old female with a history of metastatic mucinous adenocarcinoma.

Pathologic Diagnosis

Visceral pleura: Metastatic mucinous adenocarcinoma.

Agents Associated WITH CLINICAL BENEFIT

ON NCCN COMPENDIUM™

- **paclitaxel, docetaxel**
- **cisplatin, carboplatin**
- **nab-paclitaxel**
- **liposomal-doxorubicin**

OFF NCCN COMPENDIUM™

- **doxorubicin**

Agents Associated With LACK OF CLINICAL BENEFIT

- **topotecan, irinotecan**
- **trastuzumab**
- **gemcitabine**

Caris Target Now is an evidence-based molecular profiling service that associates biomarker status to agents with potential clinical benefit or potential lack of clinical benefit. Agents associated with clinical benefit are presented based on NCCN Compendium™ inclusion, relevance of tumor lineage, level of published evidence and strength of biomarker expression. The agents are not ranked in order of potential or predicted efficacy. The information in this report must be considered in conjunction with all other relevant information in respect of a given patient before determining the appropriate course of treatment. The agents identified may not be suitable for use with a particular patient and the report does not guarantee that any particular agent will be effective with the treatment of any particular condition. The selection of any, all or none of the matched agents resides with the discretion of the treating physician. Caris Life Sciences does not represent that any patient will be reimbursed or paid for by any healthcare provider or insurer.

Caris Life Sciences has exercised all reasonable skill and care in the preparation of this report and believes that its findings will assist in the selection of appropriate treatments. Caris Life Sciences expressly excludes, all other representations, warranties, conditions and terms (whether express or implied), and all liability to any physician or patient, to the fullest extent permitted by law, except that the liability of Caris Life Sciences in respect of any death or personal injury caused by our negligence shall not be restricted.

The selection of agents for retreatment from prior chemotherapy regimens lies with the treating physician and must be done with careful consideration.

** FINAL REPORT **

An expert oncology consultation can be arranged if a request is made through our Client Services Department at 1-800-901-5177.

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Specimens Received

The specimens consist of:

55 (A-A54) Tissue Biopsy Slide unstained - Client ID(S12-21973-H1) from Brigham and Women's Hospital, Boston, MA, with the corresponding surgical pathology report labeled "BS-12-A21973".

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TARGET NOW® SUMMARY - AGENTS ASSOCIATED WITH CLINICAL BENEFIT

Agents Associated With CLINICAL BENEFIT	Biomarker	Result	Method	Summary Statement
paclitaxel, docetaxel	PGP	Above Threshold	IHC	Although PGP is above threshold, taxane-based therapy may be of potential benefit due to high expression of TLE3.
	TLE3	Above Threshold	IHC	
cisplatin, carboplatin	ERCC1	Negative	IHC	Low expression of ERCC1 has been associated with benefit from platinum analogs.
nab-paclitaxel	SPARC Polyclonal	Above Threshold	IHC	High expression of SPARC has been associated with benefit from nab-Paclitaxel.
	SPARC Monoclonal	Negative	IHC	
doxorubicin, liposomal-doxorubicin	TOP2A	Above Threshold	IHC	High expression of TOPO2A has been associated with benefit from anthracycline-based therapy.

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TARGET NOW® SUMMARY - Agents Associated with LACK OF CLINICAL BENEFIT

Agents Associated With LACK OF CLINICAL BENEFIT	Biomarker	Result	Method	Summary Statement
topotecan, irinotecan	TOPO1	Negative	IHC	Low expression of TOPO1 has been associated with lack of benefit from Topoisomerase I inhibitors.
trastuzumab	Her2/Neu	Negative	IHC	HER2 targeted antibodies are potentially of minimal benefit due to low expression of HER2.
gemcitabine	RRM1	Above Threshold	IHC	High expression of RRM1 has been associated with lack of benefit from gemcitabine.

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IHC Biomarker Detail

Biomarker	Agent Association **	Patient Tumor		Threshold *
		Staining Intensity	Percent Staining	Biomarker Intensity/Percentage
MRP1		2	90	=0+ or =1+ and <10% or ≥1+ and ≥10%
BCRP		2	80	=0+ or =1+ and <10% or ≥1+ and ≥10%
SPARC Polyclonal	✔	2	60	<30% or <2+ or ≥2+ and ≥30%
IGF1R		2	50	=0+ and =100% or ≥2+ and ≥30%
RRM1	✔	2	50	=0+ or ≥2+ and ≥50%
TLE3	✔	2	45	=0+ or ≥2+ and ≥30%
TUBB3		2	20	=0+ and =100% or ≥2+ and ≥30%
TOP2A	✔	2	10	=0+ or <10% or ≥1+ and ≥10%
SPARC Monoclonal	✔	2	5	<30% or <2+ or ≥2+ and ≥30%
TS		2	5	=0+ or =1+ and ≤25% or ≥2+ and ≥30%
PGP	✔	1	50	=0+ or =1+ and <10% or ≥1+ and ≥10%
MGMT		1	30	=0+ or =1+ and <10% or ≥1+ and ≥50%
TOPO1	✔	1	25	=0+ or <30% or <2+ or ≥2+ and ≥30%
PTEN		1	5	=0+ or =1+ and ≤10% or ≥2+ and ≥10%
ERCC1	✔	1	2	<2+ or =2+ and <50% or ≤3+ and <10% or ≥2+ and ≥50% or ≥3+ and ≥10%
Androgen Receptor		0	100	=0+ and =100% or ≥1+ and ≥10%
ER		0	100	<75% or <2+ or ≥2+ and ≥75%
Her2/Neu	✔	0	100	≤1+ or =2+ and <10% or ≥3+ and >30%
PR		0	100	<10% or <1+ or ≥1+ and ≥10%
cMET		0	100	=0+ and =100% or ≥2+ and ≥30%

* Caris Life Sciences has defined threshold levels of reactivity of IHC to establish cutoff points based on published evidence. Clones used: MRP1(33A6), BCRP(6D171), SPARC Polyclonal(polyclonal), RRM1(polyclonal), TOP2A(3F6), SPARC Monoclonal(12251), TS(TS106), PGP(C494), MGMT(MT23.3), TOPO1(1D6), PTEN(6H2.1), ERCC1(8F1), Androgen Receptor(AR27), ER(SP1), Her2/Neu(4B5), PR(1E2).
** All Agent Associations are reflected in the Target Now Summary.

P. Gupta MD.

Electronic Signature
Pushpa Gupta, MD
06/07/2012

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Appendix

BIOMARKER DESCRIPTION

Target	Biomarker Description
ERCC1	Nucleotide excision repair (NER) is a DNA repair mechanism necessary for the repair of DNA damage from a vast variety of sources including chemicals and ultraviolet (UV) light from the sun. NER is a particularly important mechanism by which cells prevent unwanted and potentially cancer-causing mutations. ERCC1 (excision repair cross-complementation group 1) is an important enzyme in the NER pathway. Some anticancer drugs kill cancer cells by causing DNA damage and hence need to overcome the effects of the DNA repair pathways to be effective. For example, platinum based drugs induce DNA cross-links that interfere with DNA replication and transcription. Tumors with low ERCC1 expression are more likely to benefit from platinum based DNA damaging agents while tumors that overexpress ERCC1 are more likely to be resistant to such drugs
Her2/Neu	ErbB2/Her2 encodes a member of the epidermal growth factor (EGF) receptor family of receptor tyrosine kinases. Her2 has no ligand binding domain of its own and therefore cannot bind growth factors. It does however bind tightly to other ligand-bound EGF receptor family members to form a heterodimer and enhances kinase-mediated activation of downstream signaling pathways leading to cell proliferation. Her2 is overexpressed in 15-30% of newly diagnosed breast cancers. Clinically, Her2 is a target for the monoclonal antibody Trastuzumab/Herceptin (which binds and blocks the receptor extracellularly) and the kinase inhibitor Lapatinib/Tykerb (which binds and blocks the receptor intracellularly).
PGP	P-glycoprotein (MDR1, ABCB1) is an ATP-dependent, transmembrane drug efflux pump with broad substrate specificity, which pumps antitumor drugs out of cells. Its expression is often induced by chemotherapy drugs and is thought to be a major mechanism of chemotherapy resistance. Overexpression of p-gp can be a negative predictive factor for various drugs such as anthracylines (doxorubicin, epirubicin), paclitaxel, vinblastine etc. P-gp remains the most important and dominant representative of Multi Drug Resistance phenotype and is correlated with disease state and resistant phenotype.
RRM1	Ribonucleotide reductase subunit M1 (RRM1) is a component of the ribonucleotide reductase holoenzyme consisting of M1 and M2 subunits. The ribonucleotide reductase is a rate-limiting enzyme involved in the production of nucleotides required for DNA synthesis. Gemcitabine is a deoxycitidine analogue which inhibits ribonucleotide reductase activity. Based on the literature, RRM1 levels are a predictor of patient response when treated with gemcitabine.
SPARC Monoclonal	SPARC Monoclonal (secreted protein acidic and rich in cysteine) is a calcium-binding matricellular glycoprotein secreted by many types of cells. It has a normal role in wound repair, cell migration, and cell-matrix interactions. Its over-expression is thought to have a role in tumor invasion and angiogenesis. A few studies indicate that SPARC over-expression improves the response to the anti cancer drug, nab-paclitaxel. The improved response is thought to be related to SPARC's role in accumulating albumin and albumin targeted agents within tumor tissue.
SPARC Polyclonal	SPARC Polyclonal (secreted protein acidic and rich in cysteine) is a calcium-binding matricellular glycoprotein secreted by many types of cells. It has a normal role in wound repair, cell migration, and cell-matrix interactions. Its over-expression is thought to have a role in tumor invasion and angiogenesis. A few studies indicate that SPARC over-expression improves the response to the anti cancer drug, nab-paclitaxel. The improved response is thought to be related to SPARC's role in accumulating albumin and albumin targeted agents within tumor tissue.
TLE3	TLE3 is a member of the transducin-like enhancer of split (TLE) family of proteins that have been implicated in the tumorigenesis and classification of sarcomas. TLE3 is a nuclear expressed protein originally identified in Drosophila as required for epithelial cell fate determination through interacting with members of the notch and wnt pathway. It acts downstream of APC and β-catenin to repress transcription of a number of oncogenes, which influence growth and microtubule stability. Recent studies indicate that TLE3 expression is associated with response to taxane therapy in triple negative breast cancer patients.
TOP2A	TOPOIIA is an enzyme that alters the super coiling of double stranded DNA and allows chromosomal segregation into daughter cells. Due to its essential role in DNA synthesis and repair, and frequent over expression in tumors, TOPOIIA is an ideal target for antineoplastic agents. High expression and/or co-amplification of TOPOIIA and HER2 have been associated with benefit from anthracycline based therapy.
TOPO1	Topoisomerase I is an enzyme that alters the supercoiling of double-stranded DNA. TopoI acts by transiently cutting one strand of the DNA to relax the coil and extend the DNA molecule. The regulation of DNA supercoiling is essential to DNA transcription and replication, when the DNA helix must unwind to permit the proper function of the enzymatic machinery involved in these processes. Higher expression of TopoI has been associated with response to first line chemotherapy containing Irinotecan, a TopoI inhibitor.

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LITERATURE LEVEL of EVIDENCE

Agents Associated With CLINICAL BENEFIT	Reference	Level of Evidence
paclitaxel, docetaxel	Penson, R. T., E. Oliva, et al. (2004). "Expression of multidrug resistance-1 protein inversely correlates with paclitaxel response and survival in ovarian cancer patients: a study in serial samples." Gynecol Oncol 93(1): 98-106.	**II-3 / Fair**
paclitaxel, docetaxel	Ross, DT. et al. TLE3 expression is predictive of response to chemotherapy in NSCLC, AACR 2010.	**III / Good**
paclitaxel, docetaxel	Fauci, JM, Obrien, P, TLE3 predicts sensitivity to taxane therapy in ovarian carcinoma, SGO Annual meeting poster presentation, March 2010.	**III / Good**
paclitaxel, docetaxel	Kulkarni, S., D. Hicks, et al. (2009)."TLE3 as a candidate biomarker of response to taxane therapy." Breast Cancer Research 11(2):R17	**II-2 / Good**
cisplatin, carboplatin	Steffensen, K.D., A. Jakobsen, et al. (2009). "The Relationship of Platinum Resistance and ERCC1 Protein Expression in Epithelial Ovarian Cancer." Int J Gynecol Cancer 19: 820-825.	**II-3 / Good**
cisplatin, carboplatin	Lin, K., X. Xie, et al. (2008). "Protein expression levels of excision repair cross-complementation group 1 and xeroderma pigmentosum D correlate with response to platinum-based chemotherapy in the patients with advanced epithelial ovarian cancer." Int J Gynecol Cancer 18: 1007-1012.	**II-3 / Fair**
cisplatin, carboplatin	Scheil-Bertram, S., A. Fisseler-Eckhoff, et al. (2010). "Excision repair cross-complementation group 1 protein overexpression as a predictor of poor survival for high-grade serous ovarian adenocarcinoma." Gynecologic Oncology 119(2): 325-331.	**II-2 / Good**
nab-paclitaxel	Desai, N., Soon-Shiong, P., et al. (2009). "SPARC Expression Correlates with Tumor Response to Albumin-Bound Paclitaxel in Head and Neck Cancer Patients." Translational Oncology 2(2): 59-64.	**II-3 / Good**
nab-paclitaxel	Raefsky, E., et al. Phase II study of neoadjuvant bevacizumab and trastuzumab administered with albumin-bound paclitaxel (nab paclitaxel) and carboplatin in HER2+ locally advanced breast cancer. J Clin Oncol (May 20 suppl; abstr 627), 2008.	**III / Good**
nab-paclitaxel	Yardley, D.A., et al. Phase II study of neoadjuvant gemcitabine, epirubicin, and albumin-bound nab paclitaxel (GEA) in locally advanced breast cancer with SPARC tumor assessments. J Clin Oncol (May 20 suppl; abstr 603), 2008. 26.	**III / Good**
doxorubicin, liposomal-doxorubicin	Mukherjee, A., S. Chan, et al. (2010). "TOPO2A protein expression predicts response to anthracycline combination neo-adjuvant chemotherapy in locally advanced primary breast cancer." Br J Cancer Advance online publication, 9 November 2010.	**II-3 / Good**
doxorubicin, liposomal-doxorubicin	Durbecq, V., M. Paesmans, et al. (2004). "Topoisomerase-II alpha expression as a predictive marker in a population of advanced breast cancer patients randomly treated either with single-agent doxorubicin or single-agent docetaxel." Mol Cancer Ther 3(10): 1207-14.	**II-2 / Good**

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LITERATURE LEVEL of EVIDENCE

Agents Associated With LACK OF CLINICAL BENEFIT	Reference	Level of Evidence
topotecan, irinotecan	Kigawa, J., N. Terakawa, et al. (1999). "Topoisomerase-I activity and response to second-line chemotherapy consisting of camptothecin-11 and cisplatin in patients with ovarian cancer." Int J Cancer 84(5): 521-4.	**II-3 / Fair**
topotecan, irinotecan	Litzow, M.R., S.H. Kaufmann, et al. (2010). "Phase I trial of autologous hematopoietic SCT with escalating doses of topotecan combinated with CY and carboplatin in patients with relapsed or persistent ovarian or primary peritoneal carcinoma." Bone Marrow Transplantation 45: 490-497.	**II-3 / Good**
topotecan, irinotecan	Naniwa, J., N. Terakawa, et al. (2007). "Genetic diagnosis for chemosensitivity with drug-resistance genes in epithelial ovarian cancer." Int J Gynecol Cancer 17(1): 76-82.	**II-3 / Fair**
trastuzumab	Bookman, M. A., I.R. Horowitz, et al. (2003). "Evaluation of monoclonal humanized anti-HER2 antibody, Trastuzumab, in patients with recurrent or refractory ovarian or primary peritoneal carcinoma with over expression of HER2: A phase II trial of the gynecologic oncology group." J Clin Oncol 21:283-90.	**II-3 / Good**
trastuzumab	McAlpine, J.N., D.M. Miller, et al. (2009). "HER2 over expression and amplification is present in a subset of ovarian mucinous carcinomas and can be targeted with trastuzumab therapy." BMC Cancer 9:433.	**II-3 / Good**
gemcitabine	Bepler, G., I. Kusmartseva, et al. (2006). "RRM1 modulated in vitro and in vivo efficacy of gemcitabine and platinum in non-small-cell lung cancer." J Clin Oncol 24(29): 4731-7.	**II-3 / Good**
gemcitabine	Nakahira, S., S. Nakamori, et al. (2007). "Involvement of ribonucleotide reductase M1 subunit overexpression in gemcitabine resistance of human pancreatic cancer." Int J Cancer 120(6): 1355-63.	**II-3 / Good**
gemcitabine	Rosell, R., E. Felip, et al. (2004). "Gene expression as a predictive marker of outcome in stage IIB-IIIA-IIIB non-small cell lung cancer after induction gemcitabine-based chemotherapy followed by resectional surgery." Clin Cancer Res 10(12 Pt 2): 4215s-4219s.	**II-3 / Good**

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LITERATURE LEVEL OF EVIDENCE ASSESSMENT FRAMEWORK*

Study Design

Hierarchy of Design	Criteria
I	Evidence obtained from at least one properly designed **randomized controlled trial**.
II-1	Evidence obtained from well-designed controlled trials **without randomization**.
II-2	Evidence obtained from well-designed **cohort** or **case-control** analytic studies, preferably from more than one center or research group.
II-3	Evidence obtained from multiple time series with or without the intervention. Dramatic results in uncontrolled trials might also be regarded as this type of evidence.
III	Opinions of respected authorities, based on clinical experience, descriptive studies, or reports of expert committees.

Study Validity

Grade	Criteria
Good	The study is judged to be valid and relevant as regards results, statistical analysis, and conclusions and shows no significant flaws.
Fair	The study is judged to be valid and relevant as regards results, statistical analysis, and conclusions, but contains at least one significant but not fatal flaw.
Poor	The study is judged to have a fatal flaw such that the conclusions are not valid for the purposes of this test.

* Adapted from Harris, T., D. Atkins, et al. (2001). "Current Methods of the U.S. Preventive Services Task Force." Am J Prev Med 20(3S)[9]

Disclaimer

This test was developed by Caris Life Sciences and their performance characteristic was determined by Caris Life Sciences. It has not been cleared or approved by the U. S. Food and Drug Administration (FDA). These tests are permitted for clinical purposes and should not be regarded as purely investigational or for research only. Caris Life Sciences is certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA) as qualified to perform high-complexity clinical testing.

By requesting and/or utilizing this test and the report, you agree that the associated analysis, interpretation, and intellectual property generated by the utilization or included in the report is copyright protected, proprietary, and owned by Caris Life Sciences. Caris Life Sciences grants to the physician a limited right to use the information to care for the associated patient, but for no other purpose, including but not limited to validating or creating a similar test, program, or report, which is strictly prohibited unless by the express written permission of Caris Life Sciences and with appropriate patient consents. By requesting and/or utilizing this report, you agree that in the event of a breach of these provisions, Caris Life Sciences shall be entitled to receive as liquidated damages an immediate payment of $100,000 for each violation and 1% of the total amount for each day that such a breach continues while acknowledging and agreeing that actual damages for such a breach would be difficult to calculate, and that such an amount constitutes a reasonable estimate necessary to compensate Caris Life Sciences for the damage suffered by it as a result of the breach of these provisions that are intended to limit the use of the test for the care of the associated patient.

Decisions on care and treatment should be based on the independent medical judgment of the treating physician taking into consideration all available information concerning the patient's condition, including other laboratory tests, in accordance with the standard of care in a given community. Decisions regarding care and treatment should not be based on a single test such as this test. The finding of a biomarker expression does not necessarily indicate pharmacologic effectiveness or lack thereof. If a patient's tumor has previously progressed on an agent identified as associated with clinical benefit on this report, the patient should not be re-treated with this agent.

P. Gupta M.D.

Electronic Signature
Pushpa Gupta, MD
06/07/2012

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