HHS Public Access

Author manuscript

JAMA Oncol. Author manuscript; available in PMC 2016 July 01.

Published in final edited form as:

JAMA Oncol. 2015 July ; 1(4): 486–494. doi:10.1001/jamaoncol.2015.1432.

Association of Somatic Mutations of *ADAMTS* Genes With Chemotherapy Sensitivity and Survival in High-Grade Serous Ovarian Carcinoma

Yuexin Liu, PhD, **Maya Yasukawa, MD**, **Kexin Chen, MD, PhD**, **Limei Hu, MD**, **Russell R. Broaddus, MD, PhD**, **Li Ding, PhD**, **Elaine R. Mardis, PhD**, **Paul Spellman, PhD**, **Douglas A. Levine, MD**, **Gordon B. Mills, MD, PhD**, **Ilya Shmulevich, PhD**, **Anil K. Sood, MD, PhD**, and **Wei Zhang, PhD**

Department of Pathology, University of Texas MD Anderson Cancer Center, Houston (Liu, Yasukawa, Hu, Broaddus, Zhang); Institute for Systems Biology/MD Anderson Cancer Center Genome Data Analysis Center, The Cancer Genome Atlas, Bethesda, Maryland (Liu, Shmulevich, Zhang); Department of Obstetrics and Gynecology, Showa University School of Medicine, Shinagawa-ku, Tokyo, Japan (Yasukawa); Department of Epidemiology and Biostatistics, National Clinical Research Center for Cancer, Key Laboratory of Cancer Prevention and Therapy of Tianjin, Tianjin Medical University Cancer Hospital and Institute, Tianjin, PR China (Chen); Genome Institute, Washington University, St Louis, Missouri (Ding, Mardis); Department of Molecular and Medical Genetics, Oregon Health and Science University, Portland (Spellman); Gynecology Service, Department of Surgery, Memorial Sloan-Kettering Cancer Center, New York, New York (Levine); Department of Systems Biology, University of Texas MD Anderson Cancer Center, Houston (Mills); Institute for Systems Biology, Seattle, Washington (Shmulevich); Department of Gynecologic Oncology and Reproductive Medicine, University of Texas MD Anderson Cancer Center, Houston (Sood); Department of Cancer Biology, University of Texas MD Anderson Cancer Center, Houston (Sood)

Abstract

Corresponding Author: Wei Zhang, PhD, Department of Pathology, Unit 85, University of Texas MD Anderson Cancer Center, 1515 Holcombe Blvd, Houston, TX 77030 (wzhang@mdanderson.org).

Supplemental content at jamaoncology.com

Conflict of Interest Disclosures: None reported.

Additional Contributions: We thank Ann Sutton, BA, Department of Scientific Publications, University of Texas MD Anderson Cancer Center, for editing the manuscript. She received no specific compensation for editing this article.

Author Contributions: Dr Zhang had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.
Study concept and design: Liu, Yasukawa, Mills, Sood, Zhang.
Acquisition, analysis, or interpretation of data: Liu, Yasukawa, Chen, Hu, Broaddus, Ding, Mardis, Spellman, Levine, Shmulevich, Sood, Zhang.
Drafting of the manuscript: Liu, Yasukawa, Zhang.
Critical revision of the manuscript for important intellectual content: Liu, Yasukawa, Chen, Hu, Broaddus, Ding, Mardis, Spellman, Levine, Mills, Shmulevich, Sood, Zhang.
Statistical analysis: Liu, Yasukawa, Shmulevich
Obtained funding: Zhang.
Administrative, technical, or material support: Chen, Hu, Broaddus, Ding, Spellman, Levine, Mills, Zhang.
Study supervision: Mardis, Mills, Sood, Zhang.

IMPORTANCE—Chemotherapy response in the majority of patients with ovarian cancer remains unpredictable.

OBJECTIVE—To identify novel molecular markers for predicting chemotherapy response in patients with ovarian cancer.

DESIGN, SETTING, AND PARTICIPANTS—Observational study of genomics and clinical data of high-grade serous ovarian cancer cases with genomic and clinical data made public between 2009 and 2014 via the Cancer Genome Atlas project.

MAIN OUTCOMES AND MEASURES—Chemotherapy response (primary outcome) and overall survival (OS), progression-free survival (PFS), and platinum-free duration (secondary outcome).

RESULTS—In 512 patients with ovarian cancer with available whole-exome sequencing data, mutations from 8 members of the *ADAMTS* family (*ADAMTS* mutations) with an overall mutation rate of approximately 10.4% were associated with a significantly higher chemotherapy sensitivity (100% for *ADAMTS*-mutated vs 64% for *ADAMTS* wild-type cases; $P < .001$) and longer platinum-free duration (median platinum-free duration, 21.7 months for *ADAMTS*-mutated vs 10.1 months for *ADAMTS* wild-type cases; $P = .001$). Moreover, *ADAMTS* mutations were associated with significantly better OS (hazard ratio [HR], 0.54 [95% CI, 0.42–0.89]; $P = .01$ and median OS, 58.0 months for *ADAMTS*-mutated vs 41.3 months for *ADAMTS* wild-type cases) and PFS (HR, 0.42 [95% CI, 0.38–0.70]; $P < .001$ and median PFS, 31.8 for *ADAMTS*-mutated vs 15.3 months for *ADAMTS* wild-type cases). After adjustment by *BRCA1* or *BRCA2* mutation, surgical stage, residual tumor, and patient age, *ADAMTS* mutations were significantly associated with better OS (HR, 0.53 [95% CI, 0.32–0.87]; $P = .01$), PFS (HR, 0.40 [95% CI, 0.25–0.62]; $P < .001$), and platinum-free survival (HR, 0.45 [95% CI, 0.28–0.73]; $P = .001$). *ADAMTS*-mutated cases exhibited a distinct mutation spectrum and were significantly associated with tumors with a higher genome-wide mutation rate than *ADAMTS* wild-type cases across the whole exome (median mutation number per sample, 121 for *ADAMTS*-mutated vs 69 for *ADAMTS* wild-type cases; $P < .001$).

CONCLUSIONS AND RELEVANCE—*ADAMTS* mutations may contribute to outcomes in ovarian cancer cases without *BRCA1* or *BRCA2* mutations and may have important clinical implications.

Ovarian cancer remains the leading cause of mortality from gynecologic cancer.[1,2] Despite aggressive surgery and chemotherapy, most patients eventually experience relapse with generally incurable disease mainly due to emergence of chemotherapy resistance.[3,4] Early identification and differentiation of patients with chemotherapy-resistant disease could allow enrollment in clinical trials with alternative therapeutics rather than ineffective chemotherapy.

Patients with ovarian cancer with germline or somatic *BRCA1* or *BRCA2* mutations are recognized to have better response to platinum-based treatment and substantially longer survival than noncarriers.[5] Recent analyses showed that *BRCA2* mutation demonstrated a stronger association with improved survival and chemotherapy response among women with ovarian cancer than *BRCA1* mutation across multiple data sets.[6,7]

BRCA1 or *BRCA2* mutations including both germline and somatic mutations have been found in 20.3% of the Cancer Genome Atlas (TCGA) patients with ovarian cancer,[8] which is similar to the mutation rates reported in previous studies.[9,10] However, the clinical chemosensitive rates to platinum-based therapy regimens are approximately 70%,[11] suggesting that events other than *BRCA1* or *BRCA2* mutations exist that predict chemotherapy response. In this study, we examined TCGA genomic and clinical data to determine the association between novel gene mutations in ovarian cancer and patient overall survival (OS), progression-free survival (PFS), and chemotherapy response.

Methods

Patients and Study Design

We obtained the whole-exome sequencing data for 512 patients with high-grade serous ovarian cancer from TCGA.[8] The specimens were obtained prior to systemic therapy and all patients received platinum-based chemotherapy. The entire TCGA cohort was divided into a discovery set of 210 cases (hereafter referred to as the discovery cohort) and a validation set of 302 cases (hereafter referred to as the validation cohort). The separation of discovery and validation cohorts is described in detail in the eMethods in the Supplement. Details about patient characteristics and study design are described in the eMethods, eFigure 1, and eTables 1, 2, and 3 in the Supplement. Access to TCGA database was approved by the National Cancer Institute (https://tcga-data.nci.nih.gov/tcga). The study was approved by the institutional review board at the University of Texas MD Anderson Cancer Center. The need for consent was waived because of the retrospective nature of the study.

Whole-Exome Sequencing Data Analysis

We analyzed the whole-exome sequencing data for the 210 TCGA cases in the discovery cohort that had explicitly defined response status to chemotherapy (sensitive or resistant). To quantify the association of gene mutation with response status, we calculated for each individual gene the number of mutations in the sensitive (N_s) or resistant (N_r) samples, respectively. We further selected the genes associated with chemosensitivity by applying both of the following criteria: (1) $N_r = 0$; (2) $N_s \geq 2$.

We calculated the mutation frequency in terms of the total number of mutations including single-nucleotide substitution or insertion-deletion (indel) per sample. Fractions of mutations (indels were excluded) in the 6 possible mutation classes (ie, C>T, C>A, C>G, A>G, A>C, and A>T) were calculated for each sample. Details of whole-exome sequencing and chemotherapy response data analyses are provided in the eMethods in the Supplement.

Statistical Analysis

Survival differences were assessed using the log-rank test or Wald test (details are described in the eMethods in the Supplement). Other standard statistical tests were used to analyze the clinical and genomic data, including the Mann-Whitney, Fisher exact, and χ^2 tests. All statistical tests were 2 sided, and $P < .05$ was considered statistically significant. Statistical analyses were performed using scientific software such as Matlab (MathWorks), SPSS version 18 (SPSS Inc), and GraphPad Prism, version 6 (Graphpad Software Inc).

Results

ADAMTS Mutations in TCGA Ovarian Cancer Patients

Whole-exome capture and sequencing of TCGA ovarian cancer samples targeted approximately 180 000 exons from 18 500 genes.[8] Of the 210 patients with explicit chemotherapy response status in TCGA discovery cohort (eMethods and eFigure 1 in the Supplement), 141 were designated as sensitive and 69 as resistant. Mutation analysis showed that 2118 genes including *BRCA2* were mutated in at least 2 chemosensitive samples ($N_s \geq 2$), but not in any of the chemoresistant cases ($N_r = 0$) (eFigure 2 and eTable 4 in the Supplement). The majority of these genes had small numbers of mutations, which is consistent with the somatic mutation frequency of any gene other than *TP53* being relatively low in high-grade serous ovarian cancer.[8] *ADAMTS16*, a member of the *ADAMTS* (a disintegrin and metalloproteinase with thrombospondin motifs) superfamily,[12] is one of the most frequently mutated genes (eTable 5 in the Supplement). Because members from the gene family share common protein structural domains and demonstrate functional redundancy,[13,14] we next examined whether any other member(s) of the *ADAMTS* family was associated with chemosensitivity. Interestingly, we found that 6 *ADAMTS* family members in addition to *ADAMTS16* demonstrated a mutation bias in platinum-sensitive patients (eFigure 2 in the Supplement). Furthermore, gene set enrichment analysis of the responder-related genes showed that members of this gene family were significantly enriched in the list ($P = .02$, χ^2 test). The mutated members consisted of *ADAMTS16* (~4.3%), *ADAMTSL1* (~2.9%), and *ADAMTS1*, *ADAMTS15*, *ADAMTS6*, *ADAMTS9*, and *ADAMTS18* (~1.0% each). To obtain a more comprehensive view of gene mutations from this family, we included *ADAMTS13* in the downstream analysis although it was mutated in only 1 chemosensitive sample (Figure 1). We use the term "*ADAMTS* mutations" to refer to the mutations of these 8 members, unless specified otherwise.

Together, *ADAMTS* mutations were found in a total of 23 ovarian cancer samples (Figure 1); most of these were missense (eFigure 3 in the Supplement). Forty-two samples harbored *BRCA1* or *BRCA2* mutations, 66.7% of which were germline mutations (Figure 1). The *BRCA* and *ADAMTS* mutations were not correlated with each other ($P = .26$, Fisher exact test). Except for a significant correlation with chemotherapy response status, *ADAMTS* mutations were not correlated with age or clinical characteristics such as stage, grade, and residual tumor (Figure 1 and eTable 6 in the Supplement).

Association of *ADAMTS* Mutations With Patient Survival

We next determined the relationship between the prevalence of *ADAMTS* mutations and patient outcome (Figure 2). Kaplan-Meier survival analysis revealed that patients with *ADAMTS* mutations had a 5-year survival rate of approximately 59% and exhibited significantly longer OS than those without (median OS, not reached vs 44.4 months; log-rank $P = .007$; hazard ratio [HR], 0.37 [95% CI, 0.29–0.82]) (Figure 2A). Similarly, the median PFS of the *ADAMTS*-mutated cases was almost twice as long as that of the *ADAMTS* wild-type cases (26.8 vs 14.0 months; log-rank $P = .002$; HR, 0.47 [95% CI, 0.38–0.80]) (Figure 2B).

To test whether this result was independent of known predictive variables such as *BRCA1* or *BRCA2* mutation status, residual tumor size (eFigures 4 and 5 in the Supplement), stage, orage,[15,16] we applied multivariate analysis using a Cox proportional hazards model with *ADAMTS* mutation status and known predictors as covariates. After adjustment by *BRCA1* or *BRCA2* mutation, stage, residual tumor, and patient age, *ADAMTS* mutation was significantly associated with longer OS (HR, 0.32[95% CI, 0.14–0.69]; $P = .004$) and PFS (HR, 0.42 [95% CI, 0.25–0.71]; $P = .001$) (Table). Furthermore, we observed no correlation between *ADAMTS* mutation and these covariates (eTable 6 in the Supplement). These data suggested that *ADAMTS* mutation is an independent predictor of survival in patients with ovarian cancer.

Association of *ADAMTS* Mutations With Chemotherapy Response

All *ADAMTS*-mutated cases in the discovery cohort were designated as chemosensitive. We next determined the association between *ADAMTS* mutations and platinum-free duration after treatment, a parameter characterizing platinum-based chemotherapy response. As shown in Figure 2C, 41% of patients with *ADAMTS* mutations had a 2-year platinum-free duration. Patients with *ADAMTS* mutations exhibited a significantly longer platinum-free duration than those with wild-type *ADAMTS* (median platinum-free duration, 21.7 vs 8.7 months; log-rank $P = .004$; HR, 0.49 [95% CI, 0.38–0.83]). Multivariate Cox proportional hazards model analysis showed that *ADAMTS* mutation had a significant association with platinum-free duration (HR, 0.43 [95% CI, 0.26–0.73]; $P = .002$), independent of other known predictors such as *BRCA1* or *BRCA2* mutations, stage, and residual tumor volume (Table).

Association of *ADAMTS* Mutations With Mutation Spectra

Using whole-exome sequencing data, we further examined the association between *ADAMTS* mutations with mutation spectra in the ovarian cancer exome. Using the method as previously described,[7] we found that the *ADAMTS*-mutated cases were significantly enriched in the hypermutated samples ($P < .01$) (eFigure 6 in the Supplement). The median number of mutations was 183 for *ADAMTS*-mutated vs 69 for *ADAMTS* wild-type cases ($P < .001$, Mann-Whitney test) (Figure 3A). In contrast, *BRCA2* mutation ($P = .02$) but not *BRCA1* mutation ($P = .73$) was significantly associated with mutation rate in this cohort (eFigure 7 in the Supplement). Moreover, *ADAMTS*-mutated cases had a significantly lower percentage of C>T transition ($P = .003$) but a significantly higher percentage of A>T transversion ($P = .03$) than *ADAMTS* wild-type cases (Figure 3B and eFigure 8 in the Supplement). The proportion of C>T transitions was negatively correlated with the mutation rate, whereas the proportion of A>T transversions showed a significant correlation but in the opposite direction (eFigure 9 in the Supplement). Because high mutation rate was previously reported to be associated with better prognosis in endometrial[17] and colorectal[18] cancer, we hypothesized that the association of *ADAMTS* mutations with better survival in ovarian cancer could be a consequence of genetic instability. Supporting this notion, we found that patients with higher mutation rates exhibited significantly longer OS ($P = .002$) and PFS ($P = .05$) but no significant difference in platinum-free interval ($P = .09$) as compared with those with lower mutation rates (Figure 3C).

Multifaceted Validation of *ADAMTS* Mutations

To determine whether the association of *ADAMTS* mutations with patient outcome is due to chance, we attempted to examine whether any gene combinations from the original 2118 responder-related genes were also significantly associated with patient outcome. We randomly selected 8 genes (to match the 8 *ADAMTS* genes) from the list and then performed survival analysis between patients stratified by mutation status in the 8 selected genes. This process was repeated 10^5 times (eMethods and eFigure 10 in the Supplement). We found that mutations in 58.8%, 69.3%, or 64.0% of gene combinations were not significantly associated with OS, PFS, or platinum-free survival, respectively ($P > .05$). Only 3.4% of gene combinations were simultaneously correlated with OS, PFS, and platinum-free survival ($P < .01$ for all 3 survival categories), similar to those of *ADAMTS* mutations (eFigure 11 in the Supplement). The *P* values of these random selections generated a null distribution for association of the 8-gene combination with outcome, from which we can calculate the nominal *P* value of association of *ADAMTS* mutations with outcome relative to this null distribution. This analysis showed that the association of *ADAMTS* mutations with outcome was statistically significant for PFS (nominal $P = .02$) and platinum-free survival (nominal $P = .05$) but not for OS (nominal $P = .09$), as compared with the background statistical significance levels (eMethods and eFigure 11 in the Supplement).

Next we validated the predictive value of *ADAMTS* mutations in a separate TCGA validation cohort that comprised 302 ovarian cancer samples (eMethods, eFigure 12, and eTable 7 in the Supplement), as evaluated by the associations of *ADAMTS* mutations with chemosensitivity, survival, and mutation spectra. In this validation cohort, 30 cases had *ADAMTS* mutations that were not correlated with *BRCA1* or *BRCA2* mutations ($P = .24$, Fisher exact test) (eFigures 13 and 14 in the Supplement) (only somatic mutation data were available for the second batch). *ADAMTS* mutations were significantly associated with hypermutated samples ($P < .001$, Mann-Whitney test) and had a significantly lower percentage of C>T transition ($P < .001$) but higher percentage of A>T transversion ($P = .003$) (Figure 4A and eFigures 15–17 in the Supplement). Among those with known chemotherapy response status, all *ADAMTS*-mutated cases were sensitive and none were resistant (Figure 4A). These results were consistent with the findings from the discovery cohort. Additionally, patients with *ADAMTS* mutation had significantly longer PFS than those without (HR, 0.36 [95% CI, 0.27–0.81]; log-rank $P = .008$) (Figure 4B). *ADAMTS* mutations exhibited no significant difference in OS and platinum-free interval; this could have resulted from the short OS follow-up duration and smaller size of analyzed samples with platinum-free survival data (eFigure 18 and eTable 8 in the Supplement). Likely for the same reason, the known out come predictor, *BRCA1* or*BRCA2* mutation status, unexpectedly was not significantly associated with OS and platinum-free survival (eFigure 19 in the Supplement).

We pooled the 2 TCGA cohorts and analyzed *ADAMTS* mutations in the combined cohort of 512 patients with ovarian cancer. A total of 53 cases had *ADAMTS* mutations, which corresponded to an overall mutation rate of approximately 10.4%; all were chemotherapy sensitive (eFigure 20 and eTable 9 in the Supplement). Consistently, *ADAMTS* mutations were significantly associated with hypermutated samples ($P < .001$, Mann-Whitney test),

low C>T transition ($P < .001$), and high A>T transversion ($P < .001$) but were not significantly correlated with *BRCA1* or *BRCA2* mutations ($P = .07$, Fisher exact test) (eFigures 20 and 21 in the Supplement). With an increased clinical follow-up and more samples included in the platinum-free survival analysis (eTable 8 in the Supplement), *BRCA1* or *BRCA2* mutations, as anticipated, exhibited a significant correlation with clinical outcome in this combined cohort (eFigure 22 in the Supplement). For the same reason, patients with *ADAMTS* mutations exhibited significantly longer OS (HR, 0.54 [95% CI, 0.42–0.89]; log-rank $P = .01$) and platinum-free interval (HR, 0.48 [95% CI, 0.39–0.80]; log-rank $P = .001$) than did those without (Figure 4C), similar to PFS (HR, 0.42[95% CI, 0.38–0.70]) (eFigure 23 in the Supplement). In an adjusted model, *ADAMTS* mutation was significantly associated with longer OS (HR, 0.53 [95%CI, 0.32–0.87]; $P = .01$), PFS (HR, 0.40 [95% CI, 0.25–0.62]; $P < .001$), and platinum-free survival (HR, 0.45 [95% CI, 0.28–0.73]; $P = .001$) independent of *BRCA1* or *BRCA2* mutation, stage, residual tumor, and age (eTable 10 in the Supplement). Moreover, *ADAMTS* nonsilent mutations consistently exhibited significant association with longer OS (HR, 0.57 [95% CI, 0.41–0.98]; $P = .04$), PFS (HR, 0.49 [95% CI, 0.40–0.82]; $P = .002$), and platinum-free survival (HR, 0.52 [95% CI, 0.40–0.89]; $P = .01$) even when silent mutations were excluded (eFigure 24 in the Supplement).

Discussion

Drug resistance is a major cause of treatment failure in ovarian cancer and primarily contributes to the disease's high mortality rate. The early identification of patients who are (or are not) benefiting from platinum-based therapy is central to advancing ovarian cancer management and represents an important step toward the goal of personalized treatment. In this study, we found that patients with *ADAMTS* mutations were significantly correlated with an improved chemotherapy sensitivity and exhibited a significantly longer platinum-free duration than those with *ADAMTS* wild-type tumors. Moreover, *ADAMTS* mutation status was an independent predictor of OS and PFS in patients with ovarian cancer regardless of *BRCA1* or *BRCA2* mutations, stage, residual tumor, and age. Thus, taken together, patients with either *ADAMTS* or *BRCA1* or *BRCA2* mutations are more likely to benefit from platinum-based therapy. Nevertheless, additional predictors of sensitivity remain to be detected because many patients without *ADAMTS* or *BRCA1* or *BRCA2* mutations are chemosensitive.

Alterations in the *ADAMTS* genes have been detected in cancers[8,13] and other diseases.[12] A variant at *ADAMTS6* or*ADAMTS16* was reported to be associated with susceptibility toosteosarcoma[19] or with premature ovarian failure.[20] *ADAMTS13* mutation was recognized to cause thrombotic thrombocytopenic purpura.[21] For the first time, to our knowledge, we identified an association between *ADAMTS* mutations and clinical outcome in patients with ovarian cancer. The *ADAMTS* genes consist of a protease domain and an ancillary domain, each of which provides substrate-binding or cleavage-site specificity. Similar to *BRCA1* or*BRCA2*, which have no "hot spot" (recurrent) somatic mutations,[7,8] there is no common domain that is mutated across the *ADAMTS* genes. However, unlike *BRCA1* or *BRCA2*, *ADAMTS* genes are rarely reported to be mutated in the germ line.

Functionally, the ADAMTS proteases are a distinct group of enzymes with broad catalytic activity against a range of substrates[22] and have been demonstrated to have important roles in angiogenesis, cell migration, coagulation, andinflammation.[23] In particular, *ADAMTS1*,[24] *ADAMTS9*,[25,26] *ADAMTS15*,[27] and *ADAMTS18*[28,29] have been reported to function as tumor suppressor genes and to inhibit angiogenesis[26,30,31] in several cancers. *ADAMTSL1* was previously shown to be involved in ovary development.[32] *ADAMTS15* mutations restrained tumor growth and invasion in colorectal cancer. *Adamts16*-mutant rats exhibited a longer survival rate than did control rats by alteration in the vasculature.[33] Collectively, the *ADAMTS* genes, to a large extent, have been demonstrated to play a critical role in the development of vasculature, which is known to be heavily implicated in ovarian cancer prognosis.[34,35] This may explain our observation of a better survival among *ADAMTS*-mutated patients.

ADAMTS mutations were significantly associated with tumors with a high mutation rate, which was similar to what has been observed for *BRCA2* mutations. [7] A recent study showed that some ovarian cancer cases had a mutation signature similar to that found in *BRCA1*- or *BRCA2*-mutated cases but did not harbor *BRCA1* or *BRCA2* mutations, indicating that abnormalities of genes other than *BRCA1* or *BRCA2* may contribute to this mutation pattern.[36] Previous studies suggested that genetic in stability could result in the sensitization to DNA-damaging agents.[37] Platinum based treatment induces cross-linking and single-strand or double-strandbreaks.[38] Cells that have more mutations in the genome may have compromised DNA repair and altered DNA replication capacities, contributing to an increased sensitivity to apoptosis triggered by platinum-induced DNA damage. In support of this notion, we recently showed that *BRCA2* mutations but not *BRCA1* mutations were significantly associated with high mutation rate and significantly correlated with an improved chemosensitivity in patients with ovarian cancer, as compared with *BRCA1* or *BRCA2* wild-type cases.[7] Prominently, we further found that the association of mutation rate with *ADAMTS* mutations is also statistically significant. High mutation rate is associated with better prognosis in ovarian cancer, similar to findings in endometrial[17] andcolorectal[18] cancer. These data together with a small overlap between tumors with *ADAMTS* and *BRCA1* or *BRCA2* mutations suggest that *ADAMTS* mutations may play a similar role in response to DNA-damaging agents, leading to better survival and improved chemosensitivity in the patients with ovarian cancer whose tumors did not harbor *BRCA1* or *BRCA2* mutations. However, the molecular mechanism underlying the association of *ADAMTS* mutations with hypermutated samples remains unclear and requires in-depth studies.

Conclusions

Using whole-exome sequencing, we have, for the first time to our knowledge, reported a novel association of *ADAMTS* mutations with longer survival and improved chemotherapy sensitivity in patients with ovarian cancer. The finding has important implications for clinical prediction and trial design and may be a useful addition to *BRCA* mutation assessment for patients with ovarian cancer.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

Funding/Support: This study was partially supported by a Sprint for Life Research Award from the Blanton-Davis Ovarian Cancer Research Program (Dr Liu), grants from the Program for Changjiang Scholars and Innovative Research Team in University in China and National Key Scientific and Technological Project (2011ZX0 9307–001–04 to Dr Chen), the Ovarian Cancer SPORE (P50 CA083639 to Dr Sood), U54 CA151668 (Dr Sood), funding for the Genome Data Analysis Centers from the National Institutes of Health (U24 CA143835 to Drs Shmulevich and Zhang), and funding for the Cancer Systems Informatics Center from the National Foundation for Cancer Research (Dr Zhang).

Role of the Funder/Sponsor: The funders had no role in the design and conduct of the study; collection, management, analysis, and interpretation of the data; preparation, review, or approval of the manuscript; and decision to submit the manuscript for publication.

References

1. Siegel R, Naishadham D, Jemal A. Cancer statistics, 2013. CA Cancer J Clin. 2013; 63(1):11–30. [PubMed: 23335087]
2. Spentzos D, Levine DA, Kolia S, et al. Unique gene expression profile based on pathologic response in epithelial ovarian cancer. J Clin Oncol. 2005; 23(31):7911–7918. [PubMed: 16204010]
3. Cannistra SA. Cancer of the ovary. N Engl J Med. 2004; 351(24):2519–2529. [PubMed: 15590954]
4. Selvanayagam ZE, Cheung TH, Wei N, et al. Prediction of chemotherapeutic response in ovarian cancer with DNA microarray expression profiling. Cancer Genet Cytogenet. 2004; 154(1):63–66. [PubMed: 15381375]
5. Chetrit A, Hirsh-Yechezkel G, Ben-David Y, Lubin F, Friedman E, Sadetzki S. Effect of BRCA1/2 mutations on long-term survival of patients with invasive ovarian cancer: the national Israeli study of ovarian cancer. J Clin Oncol. 2008; 26(1):20–25. [PubMed: 18165636]
6. Bolton KL, Chenevix-Trench G, Goh C, et al. EMBRACE; kConFab Investigators; Cancer Genome Atlas Research Network. Association between BRCA1 and BRCA2 mutations and survival in women with invasive epithelial ovarian cancer. JAMA. 2012; 307(4):382–390. [PubMed: 22274685]
7. Yang D, Khan S, Sun Y, et al. Association of BRCA1 and BRCA2 mutations with survival, chemotherapy sensitivity, and gene mutator phenotype in patients with ovarian cancer. JAMA. 2011; 306(14):1557–1565. [PubMed: 21990299]
8. Cancer Genome Atlas Research Network. Integrated genomic analyses of ovarian carcinoma. Nature. 2011; 474(7353):609–615. [PubMed: 21720365]
9. Hennessy BT, Timms KM, Carey MS, et al. Somatic mutations in BRCA1 and BRCA2 could expand the number of patients that benefit from poly (ADP ribose) polymerase inhibitors in ovarian cancer. J Clin Oncol. 2010; 28(22):3570–3576. [PubMed: 20606085]
10. Pal T, Permuth-Wey J, Betts JA, et al. BRCA1 and BRCA2 mutations account for a large proportion of ovarian carcinoma cases. Cancer. 2005; 104(12):2807–2816. [PubMed: 16284991]
11. Rustin GJ, Nelstrop AE, McClean P, et al. Defining response of ovarian carcinoma to initial chemotherapy according to serum CA 125. J Clin Oncol. 1996; 14(5):1545–1551. [PubMed: 8622070]
12. Apte SS. A disintegrin-like and metalloprotease (reprolysin-type) with thrombospondin type 1 motif (ADAMTS) superfamily: functions and mechanisms. J Biol Chem. 2009; 284(46):31493–31497. [PubMed: 19734141]
13. Kan Z, Jaiswal BS, Stinson J, et al. Diverse somatic mutation patterns and pathway alterations in human cancers. Nature. 2010; 466(7308):869–873. [PubMed: 20668451]
14. Seshagiri S, Stawiski EW, Durinck S, et al. Recurrent R-spondin fusions in colon cancer. Nature. 2012; 488(7413):660–664. [PubMed: 22895193]

15. Landrum LM, Java J, Mathews CA, et al. Prognostic factors for stage III epithelial ovarian cancer treated with intraperitoneal chemotherapy: a Gynecologic Oncology Group study. Gynecol Oncol. 2013; 130(1):12–18. [PubMed: 23578540]
16. Chan JK, Tian C, Monk BJ, et al. Gynecologic Oncology Group. Prognostic factors for high-risk early-stage epithelial ovarian cancer: a Gynecologic Oncology Group study. Cancer. 2008; 112(10):2202–2210. [PubMed: 18348296]
17. Kandoth C, Schultz N, Cherniack AD, et al. Cancer Genome Atlas Research Network. Integrated genomic characterization of endometrial carcinoma. Nature. 2013; 497(7447):67–73. [PubMed: 23636398]
18. Cancer Genome Atlas Network. Comprehensive molecular characterization of human colon and rectal cancer. Nature. 2012; 487(7407):330–337. [PubMed: 22810696]
19. Savage SA, Mirabello L, Wang Z, et al. Genome-wide association study identifies two susceptibility loci for osteosarcoma. Nat Genet. 2013; 45(7):799–803. [PubMed: 23727862]
20. Pyun JA, Kim S, Cha DH, Kwack K. Epistasis between polymorphisms in TSHB and ADAMTS16 is associated with premature ovarian failure. Menopause. 2014; 21(8):890–895. [PubMed: 24366283]
21. Levy GG, Nichols WC, Lian EC, et al. Mutations in a member of the ADAMTS gene family cause thrombotic thrombocytopenic purpura. Nature. 2001; 413(6855):488–494. [PubMed: 11586351]
22. Gao W, Zhu J, Westfield LA, Tuley EA, Anderson PJ, Sadler JE. Rearranging exosites in noncatalytic domains can redirect the substrate specificity of ADAMTS proteases. J Biol Chem. 2012; 287(32):26944–26952. [PubMed: 22707719]
23. Stanton H, Melrose J, Little CB, Fosang AJ. Proteoglycan degradation by the ADAMTS family of proteinases. Biochim Biophys Acta. 2011; 1812(12):1616–1629. [PubMed: 21914474]
24. Martino-Echarri E, Fernández-Rodríguez R, Rodríguez-Baena FJ, et al. Contribution of ADAMTS1 as a tumor suppressor gene in human breast carcinoma: linking its tumor inhibitory properties to its proteolytic activity on nidogen-1 and nidogen-2. Int J Cancer. 2013; 133(10): 2315–2324. [PubMed: 23681936]
25. Du W, Wang S, Zhou Q, et al. ADAMTS9 is a functional tumor suppressor through inhibiting AKT/mTOR pathway and associated with poor survival in gastric cancer. Oncogene. 2013; 32(28):3319–3328. [PubMed: 22907434]
26. Lo PH, Lung HL, Cheung AK, et al. Extracellular protease ADAMTS9 suppresses esophageal and nasopharyngeal carcinoma tumor formation by inhibiting angiogenesis. Cancer Res. 2010; 70(13): 5567–5576. [PubMed: 20551050]
27. Porter S, Span PN, Sweep FC, et al. ADAMTS8 and ADAMTS15 expression predicts survival in human breast carcinoma. Int J Cancer. 2006; 118(5):1241–1247. [PubMed: 16152618]
28. Jin H, Wang X, Ying J, et al. Epigenetic identification of ADAMTS18 as a novel 16q23.1 tumor suppressor frequently silenced in esophageal, nasopharyngeal and multiple other carcinomas. Oncogene. 2007; 26(53):7490–7498. [PubMed: 17546048]
29. Nordgard SH, Johansen FE, Alnaes GI, et al. Genome-wide analysis identifies 16q deletion associated with survival, molecular subtypes, mRNA expression, and germline haplotypes in breast cancer patients. Genes Chromosomes Cancer. 2008; 47(8):680–696. [PubMed: 18398821]
30. Inagaki J, Takahashi K, Ogawa H, et al. ADAMTS1 inhibits lymphangiogenesis by attenuating phosphorylation of the lymphatic endothelial cell-specific VEGF receptor. Exp Cell Res. 2014; 323(2):263–275. [PubMed: 24631293]
31. Kelwick R, Wagstaff L, Decock J, et al. Metalloproteinase-dependent and -independent processes contribute to inhibition of breast cancer cell migration, angiogenesis and liver metastasis by a disintegrin and metalloproteinase with thrombospondin motifs-15. Int J Cancer. 2015; 136(4):E14–E26. [PubMed: 25099234]
32. Carré GA, Couty I, Hennequet-Antier C, Govoroun MS. Gene expression profiling reveals new potential players of gonad differentiation in the chicken embryo. PLoS One. 2011; 6(9):e23959. [PubMed: 21931629]
33. Gopalakrishnan K, Kumarasamy S, Abdul-Majeed S, et al. Targeted disruption of Adamts16 gene in a rat genetic model of hypertension. Proc Natl Acad Sci U S A. 2012; 109(50):20555–20559. [PubMed: 23185005]

34. Brown HM, Russell DL. Blood and lymphatic vasculature in the ovary: development, function and disease. Hum Reprod Update. 2014; 20(1):29–39. [PubMed: 24097804]

35. Stone RL, Nick AM, McNeish IA, et al. Paraneoplastic thrombocytosis in ovarian cancer [published correction appears in *N Engl J Med*. 2012;367(18):1768]. N Engl J Med. 2012; 366(7): 610–618. [PubMed: 22335738]

36. Alexandrov LB, Nik-Zainal S, Wedge DC, et al. Australian Pancreatic Cancer Genome Initiative; ICGC Breast Cancer Consortium; ICGC MMML-Seq Consortium; ICGC PedBrain. Signatures of mutational processes in human cancer [published correction appears in *Nature*. 2013;502(7470): 258]. Nature. 2013; 500(7463):415–421. [PubMed: 23945592]

37. Meuth M. The molecular basis of mutations induced by deoxyribonucleoside triphosphate pool imbalances in mammalian cells. Exp Cell Res. 1989; 181(2):305–316. [PubMed: 2647496]

38. Bhattacharyya A, Ear US, Koller BH, Weichselbaum RR, Bishop DK. The breast cancer susceptibility gene BRCA1 is required for subnuclear assembly of Rad51 and survival following treatment with the DNA cross-linking agent cisplatin. J Biol Chem. 2000; 275(31):23899–23903. [PubMed: 10843985]

At a Glance

- Chemotherapy response in the majority of high-grade serous ovarian cancer patients remains unpredictable.
- *ADAMTS* mutations are significantly associated with improved chemotherapy sensitivity ($P < .001$) and a longer platinum-free duration ($P = .001$).
- *ADAMTS* mutations are significantly associated with longer overall survival ($P = .01$) and progression-free survival ($P < .001$), independent of *BRCA1* or *BRCA2* mutations, tumor stage, residual tumor size, and age.
- There is no statistically significant correlation between *ADAMTS* and *BRCA1* or *BRCA2* mutations.
- *ADAMTS* mutations are significantly associated with patients with ovarian cancer with a higher mutation rate ($P < .001$).

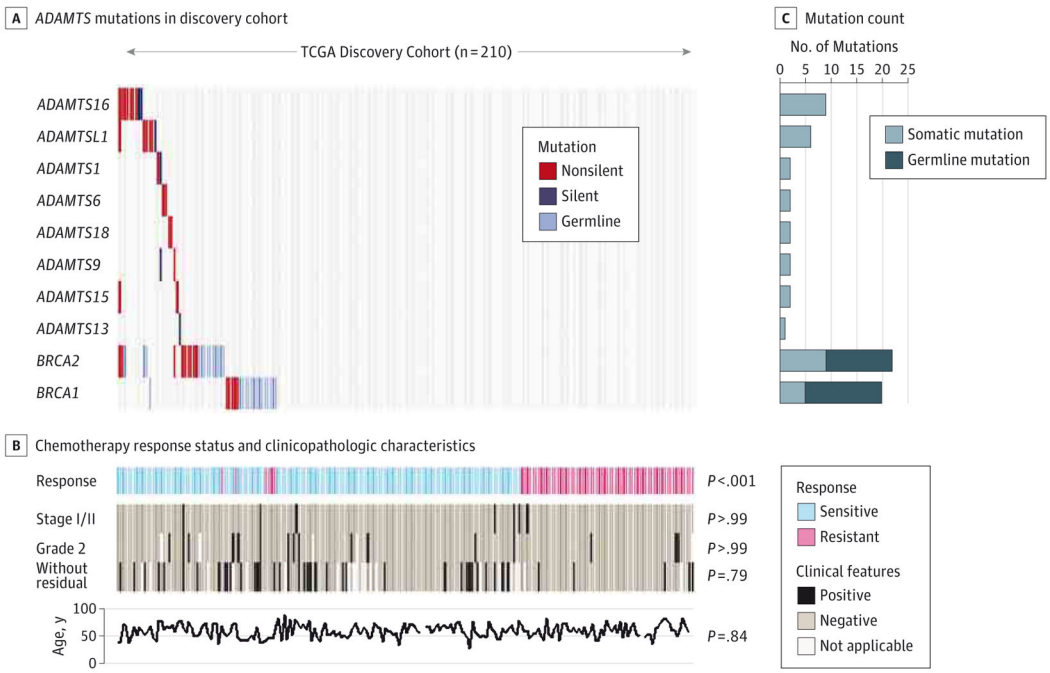

Figure 1. *ADAMTS* Mutations in the Cancer Genome Atlas (TCGA) Discovery Cohort

ADAMTS and *BRCA1* or *BRCA2* mutations that were detected in the 210 TCGA patients with ovarian cancer in the discovery cohort. A, For each gene (row) indicated, tumors (columns) with mutations are labeled with red (nonsilent mutations), dark blue (silent mutations), or light blue (germline mutations) bars. The locations of the residuals altered by *ADAMTS* mutations are detailed in eFigure 3 in the Supplement. B, Chemotherapy response status and clinicopathologic characteristics for each individual patient. “Without residual” denotes a tumor with no macroscopic disease. The *P* values show the comparison between the *ADAMTS*-mutated cases vs *ADAMTS* wild-type cases. C, Mutation count for each individual gene shown in panel A.

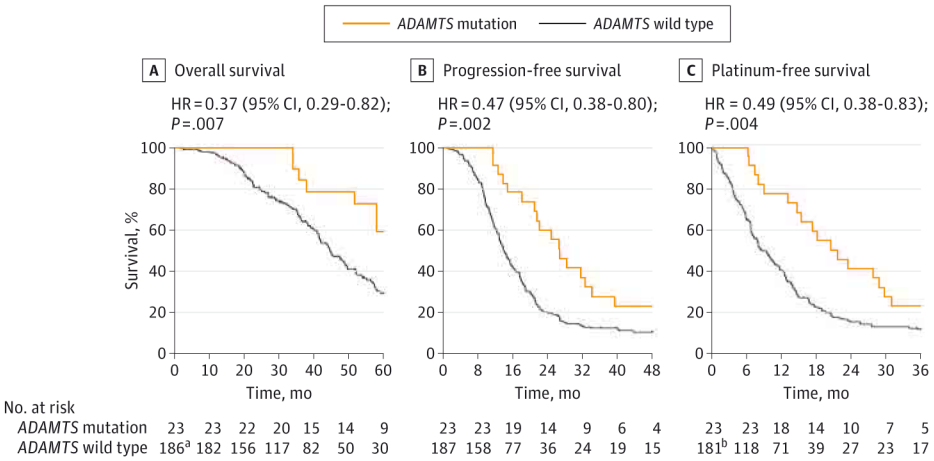

Figure 2. Association of *ADAMTS* Mutations With Clinical Outcome and Chemotherapy Response

Estimates of clinical outcome and chemotherapy response were performed among patients that were stratified on the basis of *ADAMTS* mutations. Subgroups were compared with the use of the log-rank test. Kaplan-Meier analyses of overall survival (A), progression-free survival (B), and platinum-free survival (C) of individuals with ovarian cancer in the Cancer Genome Atlas (TCGA) discovery cohort are shown. A and B, The percentage probability is plotted vs time since diagnosis in months. C, The percentage probability is plotted vs time since the end of adjuvant therapy. The number of patients at risk is shown below each curve at various time points.

[a]One case is not included in this analysis because of missing overall survival data in TCGA database.

[b]Six cases are excluded from this analysis because the patients underwent platinum treatment after progression or recurrence.

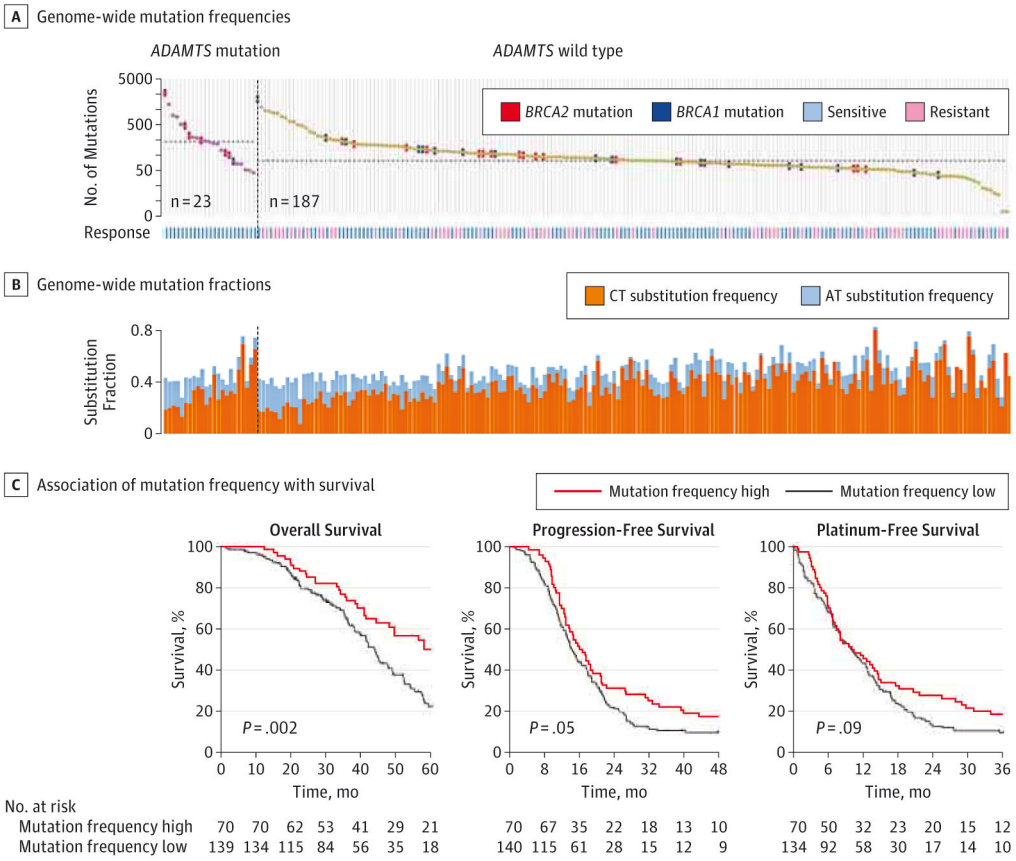

Figure 3. Association of *ADAMTS* Mutations With Mutation Spectra

A, Genome-wide mutation frequencies in terms of the number of mutations (vertical axis) detected for each tumor (horizontal axis) in order of descending number of mutations in each patient group stratified according to *ADAMTS* mutations. The median number of mutations in the *ADAMTS*-mutated (183) and wild-type groups (69) are indicated by the horizontal dashed lines. Samples with *BRCA1* or *BRCA2* mutations are also indicated. Patients' response status to chemotherapy is also shown. B, Fractions (vertical axis) of C>T transition and A>T transversion for each tumor (horizontal axis) in the same order as in A. C, Kaplan-Meier analyses of overall survival, progression-free survival, and platinum-free survival in patients stratified by mutation frequency. The ovarian cancer tumors were dichotomously categorized on the basis of patient mutation rate into 2 groups, high (highest one-third, n = 70) and low (rest of cohort, n = 140) mutation frequency. Subgroups were compared with the use of the log-rank test.

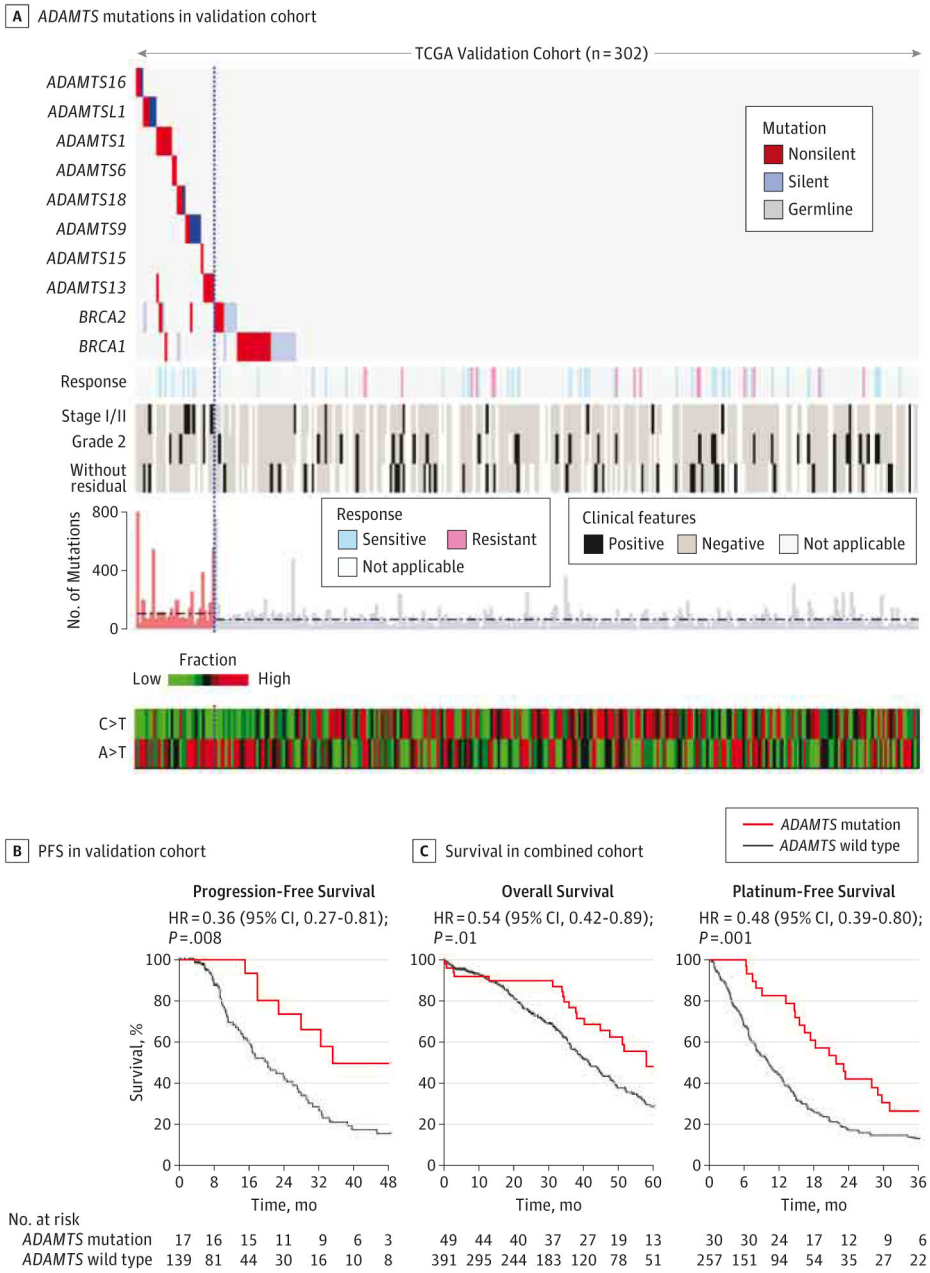

Figure 4. Validation of *ADAMTS* Mutations

A, Association of *ADAMTS* mutations with chemotherapy response status, clinicopathologic characteristics, and mutation spectra in the Cancer Genome Atlas (TCGA) validation cohort (n = 302). "Without residual" denotes a tumor with no macroscopic disease. The median number of mutations in the *ADAMTS*-mutated (111) and wild-type groups (69) are indicated by the horizontal dashed lines. The vertical dashed line highlights the separation of the *ADAMTS*-mutated samples from the ADAMTS wild-type cases. B, Kaplan-Meier analysis of progression-free survival in patients stratified by *ADAMTS* mutations in the validation

cohort. C, Kaplan-Meier analyses of overall survival and platinum-free survival in patients stratified by *ADAMTS* mutations in TCGA combined cohort.

Table

Univariate and Multivariate Models for Overall Survival, Progression-Free Survival, and Platinum-Free Survival in Women With Ovarian Cancer in the Cancer Genome Atlas (TCGA) Discovery Cohort[a]

	Univariate Analysis		Multivariate Analysis[b]	
Characteristic	**HR (95% CI)**	***P* Value[c]**	**HR (95% CI)**	***P* Value[c]**
Overall Survival				
ADAMTS status				
Wild type	1 [Reference]	.01	1 [Reference]	.004
Mutation	0.36 (0.17–0.79)		0.32 (0.14–0.69)	
BRCA1 or *BRCA2* status				
Wild type	1 [Reference]	.001	1 [Reference]	.002
Mutation	0.36 (0.20–0.64)		0.40 (0.22–0.72)	
Tumor stage				
II	1 [Reference]	.24	1 [Reference]	.24
III or IV	1.27 (0.40–4.00)		2.32 (0.56–9.50)	
Residual tumor size, mm				
0[d]	1 [Reference]		1 [Reference]	
1–20	2.07 (1.17–3.66)	.01	1.91 (1.08–3.39)	.03
>20	2.17 (1.08–4.37)	.03	1.59 (0.79–3.24)	.20
Age at diagnosis, y	1.01 (0.99–1.03)	.20	1.02 (1.00–1.03)	.09
Progression-Free Survival				
ADAMTS status				
Wild type	1 [Reference]	.003	1 [Reference]	.001
Mutation	0.46 (0.28–0.77)		0.42 (0.25–0.71)	
BRCA1 or *BRCA2* status				
Wild type	1 [Reference]	.006	1 [Reference]	.009
Mutation	0.59 (0.40–0.86)		0.58 (0.39–0.88)	
Tumor stage				
II	1 [Reference]	.26	1 [Reference]	.18

	Univariate Analysis		Multivariate Analysis[b]	
Characteristic	**HR (95% CI)**	***P* Value[c]**	**HR (95% CI)**	***P* Value[c]**
III or IV	1.67 (0.69–4.08)		2.00 (0.73–5.47)	
Residual tumor size, mm				
0[d]	1 [Reference]		1 [Reference]	
1–20	1.89 (1.26–2.83)	.002	1.90 (1.26–2.86)	.002
>20	1.82 (1.11–2.98)	.02	1.56 (0.94–2.60)	.08
Age at diagnosis, y	1.00 (0.98–1.01)	.56	1.00 (0.98–1.01)	.47
Platinum-Free Survival				
ADAMTS status				
Wild type	1 [Reference]	.005	1 [Reference]	.002
Mutation	0.48 (0.29–0.80)		0.43 (0.26–0.73)	
BRCA1 or *BRCA2* status				
Wild type	1 [Reference]	.01	1 [Reference]	.01
Mutation	0.60 (0.41–0.89)		0.59 (0.39–0.89)	
Tumor stage				
II	1 [Reference]	.18	1 [Reference]	.21
III or IV	1.83 (0.75–4.47)		1.91 (0.70–5.23)	
Residual tumor size, mm				
0[d]	1 [Reference]		1 [Reference]	
1–20	1.86 (1.24–2.79)	.003	1.88 (1.25–2.83)	.03
>20	1.84 (1.11–3.03)	.02	1.60 (0.96–2.69)	.07
Age at diagnosis, y	1.00 (0.98–1.01)	.63	1.00 (0.98–1.01)	.50

Abbreviation: HR, hazard ratio.

[a] Included are data from the 210 TCGA patients with ovarian cancer who had an explicitly defined chemotherapy response status. Patient characteristics are detailed in eTable 1 in the Supplement. *BRCA* mutations include somatic and germline mutations of *BRCA1* and *BRCA2*. Both *ADAMTS* and *BRCA1* and *BRCA2* mutations are depicted in Figure 1.

[b] Based on a multivariate Cox proportional hazards model, including all variables in the table.

[c] Wald test.

[d] Patients with no macroscopic disease are categorized as 0 mm.