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Identification of novel somatic fusions of ERG-VEGFA, TMPRSS2-ERG, and VEGFA-TMPRSS2 in prostate cancer treated with anlotinib and androgen deprivation therapy: A case report

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CASE REPORT

CANCER INNOVATION

Identification of novel somatic fusions of *ERG-VEGFA*, *TMPRSS2-ERG*, and *VEGFA-TMPRSS2* in prostate cancer treated with anlotinib and androgen deprivation therapy: A case report

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Abstract

The *TMPRSS2-ERG* fusion gene has frequently been found in prostate cancer and is associated with malignancy. Identifying novel fusions will help to stratify patients and establish patient-tailored therapies. A 78-year-old man presented to our hospital with severe symptoms of urinary urgency and frequency for 2 years, as well as severe bone pain for 1 year. He was diagnosed with metastatic prostate cancer with a Gleason score of 5 + 5. Three gene fusions, *ERG_VEGFA*, *TMPRSS2_ERG*, and *VEGFA_TMPRSS2*, were identified in the patient's prostate cancer tissue. Notably, administration of the tyrosine kinase inhibitor, anlotinib, in combination with a gonadotropin-releasing hormone agonist (GnRHa) and abiraterone, reduced the patient's bone pain and also stabilized his prostate cancer for more than 2 years. This is the first report of somatic fusions among the *VEGFA*, *ERG*, and *TMPRSS2* genes in cancer tissues from a patient with prostate cancer who responded well to antiangiogenic treatment combined with a GnRHa and abiraterone.

KEYWORDS
anlotinib, case report, fusion, prostate cancer, VEGFA

1 | INTRODUCTION

Prostate cancer is the most common cancer and the second most prevalent cause of cancer-related death worldwide in men [1]. Although the localized disease can be treated effectively with surgery and radiation [2], the prognosis of castration-resistant advanced prostate cancer is still disappointing. Androgen deprivation therapy is the cornerstone of first-line treatments for patients with metastatic prostate cancer [3]; however,

Abbreviations: AR, androgen receptor; CRPC, castration-resistant prostate cancer; ERG, erythroblast transformation-specific transcription factor-related gene; GnRHa, gonadotropin-releasing hormone agonist; PARP, poly ADP-ribose polymerase; PSA, prostate-specific antigen; TMPRSS2, transmembrane serine protease 2; VEGFA, vascular endothelial growth factor A.

most metastatic hormone-sensitive prostate cancers become castration-resistant prostate cancer (CRPC) within 18 to 24 months. Despite the efficacies of the second-generation anti-androgen drugs abiraterone and enzalutamide for CRPC [3, 4], significant challenges persist. The development of genetic molecular detection and targeted drug research and development has led to molecularly targeted therapy becoming an indispensable treatment for different types of tumors. Given that angiogenesis plays a major role in the development and spread of prostate cancer, targeting angiogenesis might offer a promising strategy for subtypes of prostate cancer [5]. However, multiple tyrosine kinase inhibitors targeting the vascular endothelial growth factor (VEGF) receptor (VEGFR) and antiangiogenic drugs have demonstrated minimal clinical activity, complicated with severe adverse reactions, in patients with prostate cancer [6]. Further studies are therefore needed to clarify the effects of these drugs in patients with prostate cancer.

A fusion between the genes encoding transmembrane serine protease 2 (*TMPRSS2*) and erythroblast transformation-specific transcription factor-related gene (*ERG*) was reported in prostate cancer in 2005 [7], since then the roles of *TMPRSS2-ERG* fusion in prostate cancer development have attracted wide attention. ERG is a member of the E-26 transformation-specific family with important physiological and pathological roles. ERG binds to DNA at specific sequences to regulate the expression of multiple target genes and is involved in both angiogenesis and vascular homeostasis. *TMPRSS2* is an androgen-regulated gene that is frequently fused to the coding sequence of *ERG*. Under the control of the androgen receptor (AR), high levels of the *TMPRSS2-ERG* fusion transcript can be translated into wild-type or N-terminally truncated ERG with functional domains [8]. Accumulating evidence suggests that approximately 50% of prostate cancers are positive for the *TMPRSS2-ERG* fusion gene [9], and more than 90% of prostate cancers with overexpressed *ERG* result from the *TMPRSS2-ERG* fusion [10]. In addition, prostate cancers with the *TMPRSS2-ERG* fusion gene are more aggressive and lethal [11]. Here we report the case of a patient with prostate cancer with three gene fusions: *ERG* (PMT.. PMT)_*VEGFA* (EX8E.. END), *TMPRSS2* (PMT.. IVS1)_*ERG* (IVS1.. END), and *VEGFA* (PMT.. IVS7)_*TMPRSS2* (IVS1.. END). Notably, administration of the tyrosine kinase inhibitor anlotinib, in combination with the gonadotropin-releasing hormone agonist (GnRHa) goserelin, and abiraterone not only reduced the patient's bone pain but also stabilized his disease for more than 2 years.

2 | CASE PRESENTATION

A 78-year-old man presented to our hospital with severe symptoms of urinary urgency and frequency for 2 years, as well as severe bone pain for 1 year. Laboratory examinations found elevated total prostate-specific antigen (PSA) of 127.01 ng/ml (normal range <4 ng/ml). Positron emission tomography-computed tomography scan revealed a mass in the periphery of the prostate and magnetic resonance imaging showed several masses in his scapulae, humerus, clavicles, and ribs. The bone scan showed multiple lesions. Prostate cancer with multiple metastases was suspected and a 12-needle prostate tissue biopsy confirmed the diagnosis of prostate cancer, Gleason score 5 + 5, in 40%–80% of the biopsies (Figure 1a). Next-generation sequencing using blood and three prostate tissue biopsy samples (Geneplus-Beijing Institute) revealed somatic fusions among the *ERG*, *TMPRSS2*, and *VEGFA* genes in all three biopsy samples, generating *ERG* (PMT.. PMT)_*VEGFA* (EX8E.. END) fusion, *TMPRSS2* (PMT.. IVS1)_*ERG* (IVS1.. END) fusion, and *VEGFA* (PMT.. IVS7)_*TMPRSS2* (IVS1.. END) fusion (Figure 2). We also noted a few additional mutations (Table 1). Considering the effects of the aberrant expression of *VEGFA* and *ERG* on tumor angiogenesis, the patient was prescribed the antiangiogenic drug anlotinib in combination with goserelin and abiraterone. His bone pain reduced dramatically and his PSA levels remained low (<2 ng/ml) (Figure 1b) for

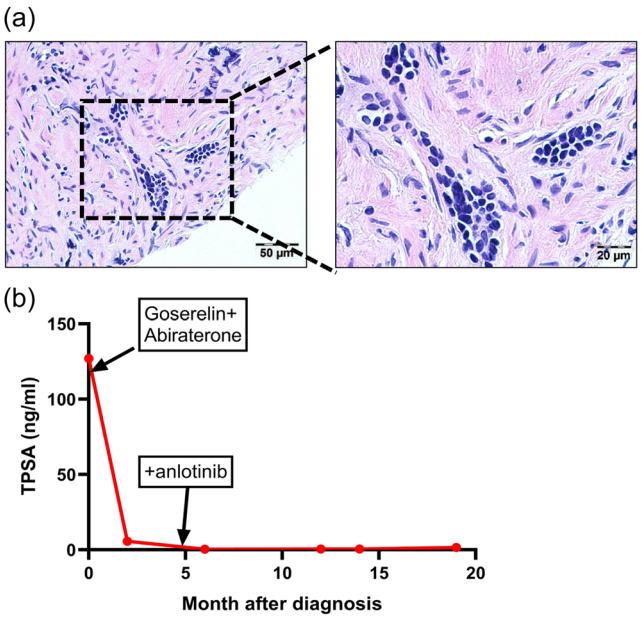

FIGURE 1 (a) Hematoxylin-eosin staining to confirm prostate cancer in the patient samples. (b) Serum levels of total prostate-specific antigen (TPSA) during treatment.

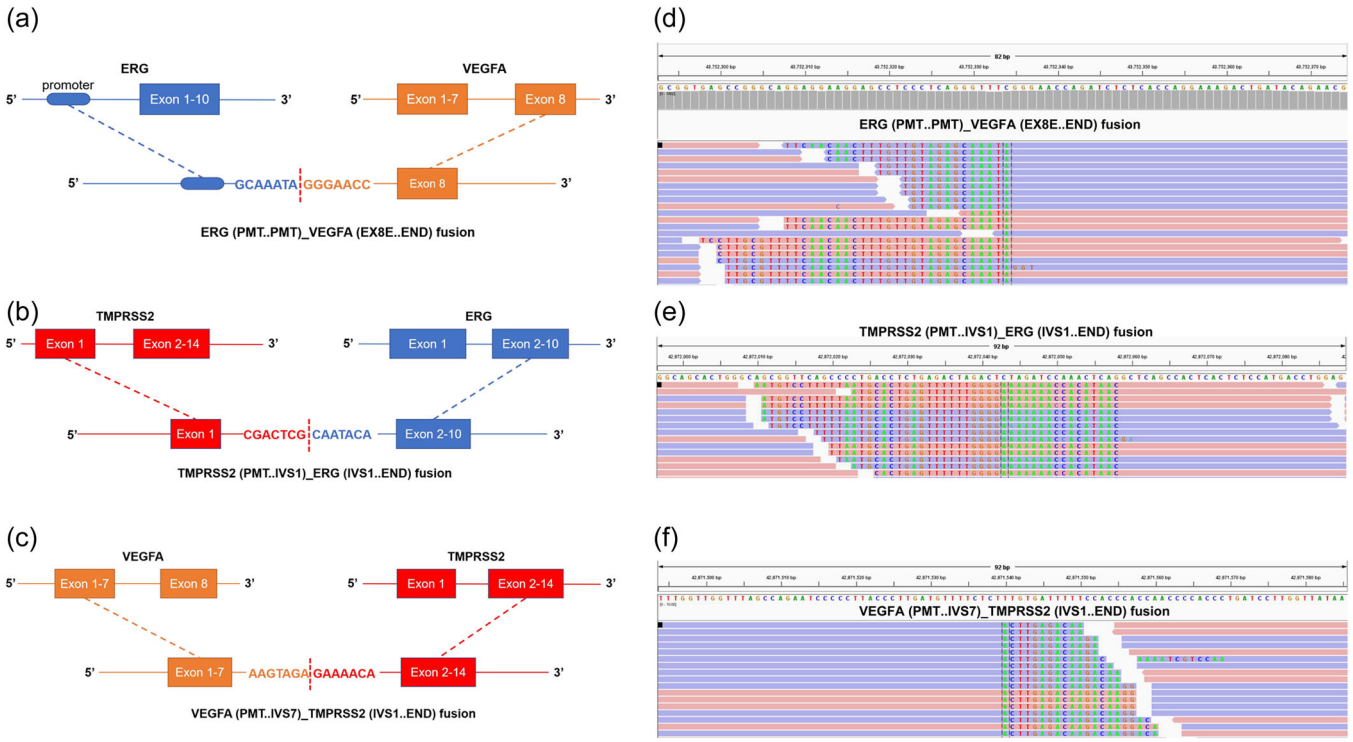

FIGURE 2 Identification of novel somatic fusions among the *ERG*, *TMPRSS2*, and *VEGFA* genes by next-generation sequencing. (a–c) Schematic structure of the genomic DNA sequence with fusion points of the *ERG-VEGFA* (a), *TMPRSS2-ERG* (b), and *VEGFA-TMPRSS2* genes (c). (d–f) DNA sequences flanking the fusion regions using Integrative Genomics Viewer.

TABLE 1 Somatic mutations and fusions in three individual prostate biopsy samples

	Sample 1	Sample 2	Sample 3
ERG-VEGFA	Promoter: EX8E fusion	Promoter: EX8E fusion	Promoter: EX8E fusion
TMPRSS2-ERG	EX1: EX2	EX1: EX2	EX1: EX2
VEGFA-TMPRSS2	EX7: EX2	EX7: EX2	
TBX3	c.547dupA, p.R183Kfs*44	c.547dupA, p.R183Kfs*44	c.547dupA, p.R183Kfs*44
JAK1	c.1625G>A, p.R542H		
CDK12		c.3377_3378delAAinsCC, p. Q1126P	
TPR			c.5093 G>A, p.R1698H

about 2 years. However, the patient finally died in December 2020 as a result of cardiovascular dysfunction.

3 | DISCUSSION AND CONCLUSION

To the best of our knowledge, this is the first report of somatic fusions among the *VEGFA, ERG*, and *TMPRSS2* genes in cancer tissues from a patient with prostate cancer who responded well to antiangiogenic treatment combined with a GnRHa and abiraterone. Although his disease stabilization could not be attributed solely to anlotinib, this rare multiple fusion with a positive response to this specific therapeutic regimen suggests that clinicians should be aware of the existence of these gene fusions in patients with prostate cancer. Further studies of these fusions in prostate cancer will help to stratify patients and to establish patient-tailored therapies.

TMPRSS2-ERG fusions play important roles in multiple stages of prostate cancer development, including premalignant prostatic intraepithelial neoplasia lesions, primary and advanced prostate cancer [8]. Here we present a patient with metastatic prostate cancer with fusions among three genes, *ERG*, *TMPRSS2*, and *VEGFA*,

indicating that these fusions might be associated with more advanced prostate cancer. However, the function of *ERG-VEGFA* needs to be further substantiated. The formation of the *TMPRSS2-ERG* fusion seems to depend on AR signaling. It is possible that activation of AR signaling could induce three-dimensional proximity of the two genomic loci, providing the essential foundation for the fusion following the induction of DNA double-strand breaks and aberrant repair [12]. Further studies are therefore needed to investigate the three-dimensional proximity of the *ERG*, *TMPRSS2*, and *VEGFA* loci under certain conditions.

TMPRSS2 is a target of the AR, suggesting that *TMPRSS2-ERG* fusions in AR-positive prostate cancer would result in ERG overexpression, thus increasing the capacity of the tumor cells to migrate and invade and controlling the expression of genes involved in extracellular matrix remodeling, inflammation, migration, and angiogenesis [13, 14]. Multiple strategies targeting ERG have thus been investigated in patients with *ERG*-fusion prostate cancer [8]. Considering the functional interaction between ERG and the poly ADP-ribose polymerase (PARP) DNA damage-repair protein, Brenner et al. revealed that treatment of ERG-overexpressing cells with the PARP inhibitor olaparib not only decreased ERG-mediated cell invasion and intravasation but also inhibited tumor growth in mouse xenograft models [15]. Several studies also demonstrated that histone deacetylase inhibitors and the small molecules YK-4-279, DB1255, and WP1130 inhibited the proliferation of ERG-overexpressing cells [8]. The current patient with three fusions among the *ERG*, *TMPRSS2*, and *VEGFA* genes benefited from treatment with the antiangiogenic drug anlotinib combined with androgen deprivation therapy. However, further studies are needed to investigate the mechanism underlying the specific therapeutic strategy for patients with *TMPRSS2-ERG* fusions.

VEGF plays important roles in physiological and pathological vasculogenesis and angiogenesis [16]. VEGFA, the prototype member of the VEGF family, is a key regulator of blood vessel growth. The human *VEGFA* gene consists of eight exons separated by seven introns and encodes several isoforms through alternative splicing [17]. The last exon (exon 8) is important for the alternative splicing of *VEGF* pre-mRNA, which is a key element in the balance between proangiogenic and antiangiogenic VEGF isoforms [18]. *VEGFA* exon 8 has been shown to play important roles in the pathogenesis of multiple disease types, including cancer, macular degeneration, nephropathy, pre-eclampsia, and ischemic limb disease [19]. Considering the classical regulation of *VEGFA* exon 8 splicing, the *VEGFA* exon 8 splicing-sensitive fluorescent reporter mouse has been investigated as a novel tool to assess splicing regulation [19]. In this study, we discovered novel fusions among the *VEGFA, TMPRSS2*, and *ERG* genes. Although the *ERG* (PMT. . PMT)_*VEGFA* (EX8E. . END) fusion only encodes exon 8 of *VEGFA*, the effect of this peptide on VEGFA needs to be investigated further.

AUTHOR CONTRIBUTIONS

Qiuli Liu: data curation (lead); formal analysis (lead); investigation (lead); methodology (lead); project administration (lead); writing – original draft (lead); writing – review and editing (supporting). **Shuo Wang**: data curation (supporting); formal analysis (supporting); investigation (supporting); methodology (equal); resources (equal); writing – original draft (supporting). **Ze Wang**: data curation (supporting); formal analysis (supporting); investigation (equal); methodology (equal); resources (supporting); writing – review and editing (supporting). **Peng Tang**: formal analysis (supporting); investigation (equal); methodology (equal); resources (equal); software (equal); visualization (equal). **Dianzheng Zhang**: conceptualization (equal); formal analysis (equal); investigation (equal); supervision (equal); validation (equal); writing – original draft (equal); writing – review and editing (lead). **Weihua Lan**: conceptualization (supporting); data curation (equal); formal analysis (equal); investigation (equal); methodology (supporting); resources (equal). **Jun Jiang**: conceptualization (lead); data curation (lead); funding acquisition (lead); supervision (lead); validation (lead); writing – review and editing (lead).

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

DATA AVAILABILITY STATEMENT

All the data are included in the article. Further inquiries can be directed to the corresponding author.

ETHICS STATEMENT

The study was approved by the institutional ethic review boards of Daping Hospital (2018-28) and was carried out in accordance with The Code of Ethics of the World Medical Association (Declaration of Helsinki) for experiments involving humans. [Correction added on 16 June 2022, after first online publication: In “Ethics Statement” the approval ID “(2018-28)” was missed and included in this version.]

INFORMED CONSENT

Written informed consent was obtained from the patient's next of kin for publication of this case report and any accompanying images.

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REFERENCES

1. Comperat E, Wasinger G, Oszwald A, Kain R, Cancel-Tassin G, Cussenot O. The genetic complexity of prostate cancer. Genes. 2020;11(12):1396. https://doi.org/10.3390/genes11121396
2. Filson CP, Marks LS, Litwin MS. Expectant management for men with early stage prostate cancer. CA Cancer J Clin. 2015;65(4):265–82. https://doi.org/10.3322/caac.21278
3. James ND, de Bono JS, Spears MR, Clarke NW, Mason MD, Dearnaley DP, et al. Abiraterone for prostate cancer not previously treated with hormone therapy. N Engl J Med. 2017;377(4):338–51. https://doi.org/10.1056/NEJMoa1702900
4. Tombal B, Saad F, Penson D, Hussain M, Sternberg CN, Morlock R, et al. Patient-reported outcomes following enzalutamide or placebo in men with non-metastatic, castration-resistant prostate cancer (PROSPER): a multicentre, randomised, double-blind, phase 3 trial. Lancet Oncol. 2019;20(4):556–69. https://doi.org/10.1016/S1470-2045(18)30898-2
5. Melegh Z, Oltean S. Targeting angiogenesis in prostate cancer. Int J Mol Sci. 2019;20(11):2676. https://doi.org/10.3390/ijms20112676
6. Ioannidou E, Moschetta M, Shah S, Parker JS, Ozturk MA, Pappas-Gogos G, et al. Angiogenesis and anti-angiogenic treatment in prostate cancer: mechanisms of action and molecular targets. Int J Mol Sci. 2021;22(18):9926. https://doi.org/10.3390/ijms22189926
7. Park K, Dalton JT, Narayanan R, Barbieri CE, Hancock ML, Bostwick DG, et al. TMPRSS2:ERG gene fusion predicts subsequent detection of prostate cancer in patients with high-grade prostatic intraepithelial neoplasia. J Clin Oncol. 2014;32(3):206–11. https://doi.org/10.1200/JCO.2013.49.8386
8. Lorenzin F, Demichelis F. Past, current, and future strategies to target ERG fusion-positive prostate cancer. Cancers. 2022;14(5):1118. https://doi.org/10.3390/cancers14051118
9. Tomlins SA, Rhodes DR, Perner S, Dhanasekaran SM, Mehra R, Sun XW, et al. Recurrent fusion of TMPRSS2 and ETS transcription factor genes in prostate cancer. Science. 2005;310(5748):644–8. https://doi.org/10.1126/science.1117679
10. Pernar CH, Ebot EM, Pettersson A, Graff RE, Giunchi F, Ahearn TU, et al. A prospective study of the association between physical activity and risk of prostate cancer defined by clinical features and TMPRSS2:ERG. Eur Urol. 2018;76:33–40. https://doi.org/10.1016/j.eururo.2018.09.041
11. Demichelis F, Fall K, Perner S, Andrén O, Schmidt F, Setlur SR, et al. TMPRSS2:ERG gene fusion associated with lethal prostate cancer in a watchful waiting cohort. Oncogene. 2007;26(31):4596–9. https://doi.org/10.1038/sj.onc.1210237
12. Lin C, Yang L, Tanasa B, Hutt K, Ju BG, Ohgi K, et al. Nuclear receptor-induced chromosomal proximity and DNA breaks underlie specific translocations in cancer. Cell. 2009;139(6):1069–83. https://doi.org/10.1016/j.cell.2009.11.030
13. Tomlins SA, Laxman B, Varambally S, Cao X, Yu J, Helgeson BE, et al. Role of the TMPRSS2-ERG gene fusion in prostate cancer. Neoplasia. 2008;10(2):177–88. https://doi.org/10.1593/neo.07822
14. Klezovitch O, Risk M, Coleman I, Lucas JM, Null M, True LD, et al. A causal role for ERG in neoplastic transformation of prostate epithelium. Proc Natl Acad Sci U S A. 2008;105(6):2105–10. https://doi.org/10.1073/pnas.0711711105
15. Brenner JC, Ateeq B, Li Y, Yocum AK, Cao Q, Asangani IA, et al. Mechanistic rationale for inhibition of poly(ADP-ribose) polymerase in ETS gene fusion-positive prostate cancer. Cancer Cell. 2011;19(5):664–78. https://doi.org/10.1016/j.ccr.2011.04.010
16. Ferrara N, Gerber HP, LeCouter J. The biology of VEGF and its receptors. Nature Med. 2003;9(6):669–76. https://doi.org/10.1038/nm0603-669
17. Tischer E, Mitchell R, Hartman T, Silva M, Gospodarowicz D, Fiddes JC, et al. The human gene for vascular endothelial growth factor. Multiple protein forms are encoded through alternative exon splicing. J Biol Chem. 1991;266(18):11947–54.
18. Carter JG, Cherry J, Williams K, Turner S, Bates DO, Churchill AJ. Splicing factor polymorphisms, the control of VEGF isoforms and association with angiogenic eye disease. Curr Eye Res. 2011;36(4):328–35. https://doi.org/10.3109/02713683.2010.548892
19. Stevens M, Star E, Lee M, Innes E, Li L, Bowler E, et al. The VEGF-A exon 8 splicing-sensitive fluorescent reporter mouse is a novel tool to assess the effects of splicing regulatory compounds in vivo. RNA Biol. 2019;16(12):1672–81. https://doi.org/10.1080/15476286.2019.1652522

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