TITLE: A patient-driven clinicogenomic partnership through the Metastatic Prostate Cancer Project

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ABSTRACT

Molecular profiling studies have enabled numerous discoveries for metastatic prostate cancer (MPC), but they have mostly occurred in academic medical institutions focused on select patient populations. We developed the Metastatic Prostate Cancer Project (MPCproject, mpcproject.org), a patient-partnered initiative to empower MPC patients living anywhere in the U.S. and Canada to participate in molecular research and contribute directly to translational discovery. Here we present clinicogenomic results from our partnership with the first 706 MPCproject participants. We found that a patient-centered and remote research strategy enhanced engagement with patients in rural and medically underserved areas. Furthermore, patient-reported data achieved 90% consistency with abstracted health records for therapies and provided a mechanism for patient-partners to share information about their cancer experience not documented in medical records. Among the molecular profiling data from 333 patient-partners (n = 573 samples), whole exome sequencing of 63 tumor samples obtained from hospitals across the U.S. and Canada and 19 plasma cell-free DNA (cfDNA) samples from blood donated remotely recapitulated known findings in MPC and enabled longitudinal study of prostate cancer evolution. Inexpensive ultra-low coverage whole genome sequencing of 318 cfDNA samples from donated blood revealed clinically relevant genomic changes like *AR* amplification, even in the context of low tumor burden. Collectively, this study illustrates the power of a longitudinal partnership with patients to generate a more representative clinical and molecular understanding of MPC.

Note: To assist our patient-partners and the wider MPC community interpret the results of this study, we have attached a supplemental glossary of terms.

INTRODUCTION

22 Prostate cancer is the second most diagnosed cancer in men, with nearly 200,000 men
23 diagnosed in 2020 alone in the U.S.[1] Survival rates for localized disease are high, but the five-
24 year survival rate for the over 300,000 men currently living with metastatic prostate cancer
25 (MPC) is only 31%, representing the third leading cause of death for men[1,2]. Because prostate
26 cancer is largely driven by alterations to DNA, genomic sequencing studies have enabled
27 discoveries of its molecular drivers and new therapeutic targets in both primary and metastatic
28 clinical settings[3–6]. However, obtaining large cohorts of tumor biopsies from MPC patients for
29 molecular study has been challenging. MPC most commonly spreads to bone, and sampling
30 osseous lesions necessitates painful and technically challenging procedures that are not widely
31 accessible or feasible in clinical care. Because prostate cancer can shed cell-free DNA (cfDNA)
32 into the bloodstream, blood biopsies that sample this circulating tumor DNA have proven to be a
33 useful alternative for the study of MPC[7,8].

34 Historically, quaternary care academic medical institutions have had the necessary
35 infrastructure and expertise to lead clinically integrated MPC sequencing studies through clinical
36 trials. However, the resulting clinical and genomic data is often siloed within these institutions,
37 leading many to push for mandatory data sharing[9,10]. These efforts, while critical to
38 democratizing genomic research, do not directly improve access to molecular research programs
39 and do not address underlying ethnic, socioeconomic, and geographic patient disparities in such
40 studies, which threaten to bias findings and eventually care towards select patient populations[11–
41 14]. Commercial sequencing options for prostate cancer are emerging, but such approaches are
42 often proprietary, only available to patients with appropriate insurance, and regularly
43 inaccessible for wider research use[15–17]. Indeed, despite growing interest in clinical and research-

44 based genomic sequencing within the MPC patient community, there are only limited
45 mechanisms for these patients to participate in molecular profiling studies and partner with the
46 research community to accelerate discoveries[18–20].

47 We hypothesized that a patient-partnered framework that empowers MPC patients to
48 share their biological samples, clinical histories, and lived experiences directly with researchers
49 regardless of geographic location or hospital affiliation would lead to new clinicogenomic
50 discoveries and begin to address demographic inequities and data access barriers in molecular
51 studies for this disease. Thus, we established the Metastatic Prostate Cancer Project
52 (MPCproject, mpcproject.org), a research model that leverages patient advocacy and social
53 media to enable MPC patients to participate in genomic research remotely at no personal cost.

RESULTS

54 *Development of a patient-partnered metastatic prostate cancer research model*

55 Working with patients, loved ones, and advocates, we established an MPCproject
56 enrollment process for men living with MPC in the U.S. and Canada (Fig. 1a). The MPCproject
57 outreach model is community-centered and utilizes advocacy partnerships, social media
58 campaigns, and educational initiatives to engage patients (Supplementary Fig. 1). Should they
59 choose to register, patient-partners complete an online survey describing their experience with
60 MPC, followed by signing electronic consent and medical release forms, which allow the
61 MPCproject team to contact their hospitals to request medical records for abstraction and
62 optionally archival tumor tissue for research-grade genomic sequencing (Supplementary Fig. 2).
63 Additionally, enrolled patients can use a mailed kit to donate saliva and/or blood at routine blood
64 draws at no cost, and these samples are sequenced to assess germline DNA and cfDNA,
65 respectively (Supplementary Fig. 3, 4).

66 Our partnership with patients is reciprocal and continuous. Patient-partners and advocates
67 are involved in every step of the project's design and execution—we respond directly to their
68 feedback and keep them informed of our progress and findings (Supplementary Fig. 5). We work
69 with men who choose to continue donating blood to help the research community understand the
70 evolution of metastatic disease, and we regularly release prepublication, deidentified genomic
71 and clinical data in public repositories for research use.

72 ***Partnering with a demographically distinct patient population***

73 To date, the MPCproject has partnered with over 1,000 patients in the U.S. and Canada
74 and has orchestrated three public data releases (Fig. 1b). The analyses presented here are based
75 on the 706 men from the U.S. and Canada who had enrolled (completed consent forms) as of
76 June 1, 2020 (Supplementary Fig. 6).

77 Using patient-reported survey data, we assessed the geographical diversity of our patient-
78 partners. Hailing from 49 U.S. states and 6 Canadian provinces, patient-partners reported
79 receiving care for their prostate cancer at over 1,000 distinct medical institutions, 91% of which
80 were reported by two or fewer patients (Fig. 1c). We found that 55% of patient-partners have
81 never received care at an NCI-designated cancer center, where genomic research is traditionally
82 conducted (Supplementary Table 1). These patient-partners were three times less likely to report
83 participating in a clinical trial, indicating the understudied nature of our cohort and barriers MPC
84 patients face in access to clinical trials (7% vs. 20%, $P = 1 \times 10^{-6}$, Fisher's exact test).

85 Patients in rural and medically underserved areas face unique obstacles and disparities in
86 clinical cancer care[21,22]. To better understand the challenges faced by our patient-partners, we
87 identified the census tracts of patient-reported U.S. home addresses and examined their
88 geographic characteristics (n = 628/706 participants provided U.S. addresses, Methods). We

89 found that 13% of patient-partners live in rural areas defined by the USDA, a proportion
90 consistent with MPC patients in the U.S. generally (11%)[23]. We then examined primary care
91 health physician shortage areas (HPSAs) and medically underserved areas (MUAs) defined by
92 the Health Resources and Services Administration (Methods). We found that 38% of patient-
93 partners live in HPSAs (29%) or MUAs (23%) (Fig. 1d)[24]. These proportions could not be
94 compared with MPC patients in the U.S. due to a lack of published data, but they are
95 significantly enriched compared to the general U.S. population (25% HPSA, 5% MUA, $P = 0.03$
96 and 1×10^{-82} respectively, Fisher's exact test)[25,26]. While living in a rural area was associated
97 with being in a MUA or HPSA, 23% of MPCproject patient-partners live in urban primary care
98 MUAs or HPSAs ($P = 5.7 \times 10^{-13}$, Fisher's exact test).
99 We found that home addresses in rural areas were a median of 160 km farther from
100 institutions where those patients reported receiving treatment, compared to home addresses in
101 urban areas ($P < 10^{-11}$, Mann-Whitney U test) (Methods, Fig. 1e). Although we cannot determine
102 if home addresses changed during treatment, this suggests that patient-partners in rural areas
103 travel significantly farther for cancer care. We did not observe significant differences in baseline
104 clinical factors, therapies received, or likelihood to participate in a clinical trial across patients in
105 rural areas, MUAs, or HPSAs.
106 The combination of the MPCproject's online enrollment and patient-centered outreach
107 through advocacy partnerships enabled the creation of a geographically distinct prostate cancer
108 research program. Despite the project's geographical diversity, however, fewer than 10% of
109 patient-partners self-identify as non-white. While similar to existing studies, this representation
110 remains below the proportion of minority prostate cancer patients generally (20%), a racial

111 imbalance that has spurred new MPCproject initiatives to connect with patients of color
112 (Supplementary Table 2, Discussion)[23].

113 ***Patient-reported data augment medical records to amplify patient stories***

114 Through the patient-reported data, we sought to understand the experiences of those
115 living with MPC. 45% of patient-partners report being diagnosed with *de novo* metastatic
116 disease, with bone (48%) and lymph node (39%) lesions as the most common metastatic sites
117 (Fig. 2a, b). 48% of patient-partners reported a family history of prostate or breast cancer, while
118 24% reported having at least one other cancer diagnosis in their lifetime, 30% of which was a
119 non-skin form of cancer (Fig. 2c, d). The average age at diagnosis was significantly younger than
120 the national average (61 vs. 65 years old, $P < 10^{-39}$, t-test), and 24% of participants were
121 diagnosed with early-onset prostate cancer (≤ 55 years at diagnosis, Supplementary Table 2)[27].

122 We used the MPCproject's comprehensive abstracted medical records taken from
123 medical documentation together with patient-reported data to evaluate the treatments received in
124 this real-world cohort (Methods, Fig. 2e). Patient-partners reported taking an average of 2.8
125 therapies (range 1-13) to treat their prostate cancer. 119 (17%) patient-partners had abstracted
126 medical records at the time of writing, and there was 90% concordance between therapies noted
127 in formal medical records and therapies reported by patients. The overlap was lowest for
128 treatments typically given earlier in the therapeutic timeline (first line androgen deprivation
129 therapy, 83%), supportive care therapies (64%), or treatments abandoned quickly due to side-
130 effects (Fig. 2e). This finding illustrates the value of patient-reported data obtained via surveys
131 for MPC, particularly in the absence of a complete medical record.

132 We also used the patient-reported data to assess how living with prostate cancer has
133 changed the daily lives of our patient-partners. For example, in the survey, we asked participants

134 to list additional medications, alternative medications, or lifestyle changes since their diagnosis
135 of prostate cancer. 56% of patient-partners reported a lifestyle change because of living with
136 their cancer, with the most common being a change in diet or exercise (Fig. 2f). Common
137 nutritional supplements reported include Vitamin D and antioxidant-based supplements, while
138 common non-cancer medications included metformin and statins. Collectively, these results
139 demonstrate the impact of metastatic prostate cancer on patient lifestyles and that patients often
140 pursue supplemental therapies that are not regularly documented in the medical record.

141 ***Whole exome sequencing of a real-world MPC patient cohort***

142 To date, we have completed molecular profiling of 573 samples from 333 patient-
143 partners, including: ultra-low pass whole genome sequencing (ULP-WGS, average depth of
144 0.1x) of cfDNA from 319 donated blood samples; whole exome sequencing (WES) of cfDNA
145 from 47 of those blood samples; WES of 106 tumor samples; and WES of 148 germline samples
146 from donated saliva or blood buffy coat. cfDNA samples underwent WES if ULP-WGS detected
147 a tumor fraction above 0.03 (Methods). In total, 82 exome-sequenced samples (63 tumor and 19
148 cfDNA) from 79 patient-partners enrolled before June 1, 2020 were included in downstream
149 genomic analyses after assessment of sufficient tumor purity ($\geq$10%) and coverage (Methods).

150 Exome sequencing from the tumor and cfDNA samples recapitulated known genomic
151 patterns in metastatic prostate cancer (Fig. 3a). *TP53* and *SPOP* were recurrently altered,
152 consistent with previous studies of both metastatic and primary prostate cancer ($q < 0.1$ via
153 MutSig2CV)[3,4,6]. In primary tumor samples from this cohort, the mutation frequency of *TP53*
154 (30%) was more consistent with metastatic cohorts than those of primary prostate cancer[3,6]. 17
155 (27%) primary tumor samples were from men diagnosed with *de novo* metastatic disease, and
156 samples from these patient-partners were more likely to carry *TP53* mutations ($P = 0.04$, Fisher's

157 exact test). We also observed known patterns of copy number alteration in prostate cancer (Fig.
158 3a). Analysis of gene copy number alterations using GISTIC2.0 revealed recurrent
159 amplifications of *AR* and *FOXA1*, as well as recurrent deletions of *PTEN* ($q < 0.1$)[28]. Whole-
160 genome doubling was present in 5/63 tumor samples and 3/19 cfDNA samples, including in two
161 tumor samples from patient-partners initially diagnosed with localized prostate cancer. In both
162 cases, the patients were diagnosed with metastatic disease within a few months of their initial
163 diagnosis.

164 To understand the mutational processes in this cohort's exome-sequenced samples, we
165 used a mutation-based method (deconstructSigs) to determine the contribution of COSMIC v2.0
166 signatures to each sample[29,30] (Fig. 3b, Methods). We detected the presence of aging-associated
167 clock-like signature 1 in all samples and the presence of signature 3 (associated with homologous
168 recombination deficiency, HRD) and signature 6 (associated with mismatch repair deficiency,
169 MMR) in a subset of samples. These results are consistent with previous studies implicating
170 these signatures in prostate cancer, although they likely overestimate the prevalence of signature
171 6 in tumor samples due to formalin-induced deamination artifacts[31,32]. We found that the
172 presence of signature 3 was enriched in metastasis-associated samples (cfDNA and primary
173 tumors obtained in the metastatic setting) relative to tumor tissue from patients with strictly
174 localized tumors at time of resection ($P < 0.02$, Fisher's exact test). While some samples with
175 signature 3 had alterations in *BRCA1*, *BRCA2*, or another DNA repair gene, this association was
176 not statistically significant, potentially highlighting the presence of HRD-positive tumors without
177 a causative molecular alteration as previously reported in studies of prostate and breast
178 cancer[5,33–36].

179 In 10% of samples (8/82), we observed contributions from COSMIC signatures 2 and 13,
180 which are driven by APOBEC cytidine deaminases and known to operate at a baseline level in
181 prostate cancer[31,37]. APOBEC-driven mutagenesis has been implicated in kataegis—rare,
182 localized hypermutation in specific nucleotide contexts that is associated with genomic
183 instability and increased Gleason score in prostate cancer[38,39]. In a cfDNA sample donated by
184 one patient-partner (patient-partner 0203), we detected eight distinct mutations within a 2 kB
185 window in *KMT2C*, a known driver of prostate cancer (Fig. 3c)[3]. Six of these mutations were in a
186 T(C>T)A nucleotide context, and this sample had a detectable contribution from COSMIC
187 signature 13. We found that two pairs of the mutations, p.S1947F/p.S1954F and
188 p.Q2325*/p.S2337Y, were each present on individual sequencing reads, confirming that these
189 mutations existed within the same cell and strongly implicating *KMT2C* disruption through
190 kataegis (Supplementary Fig. 7). These findings illustrate the ability to detect both frequent and
191 rare clinically relevant molecular events in MPC across diverse contexts using a patient-
192 partnered model.
193 Given the strong heritability of prostate cancer, we also sought to assess inherited
194 germline alterations and their overlap with self-reported family history of cancer[40]. We found
195 that among the 132 patient-partners (19%) with WES of donated saliva or blood buffy coat, 15
196 had pathogenic germline alterations in select genes implicated in prostate cancer heritability (Fig.
197 3d, Supplementary Table 3)[41]. 14% of men that reported a family history of prostate or breast
198 cancer had at least one pathogenic germline alteration, compared to 7% of men that reported no
199 family history, although this difference was not statistically significant ($P = 0.38$, Fisher's exact
200 test). The most mutated gene was *CHEK2* (8 patient-partners), followed by *BRCA2* (4 patient-
201 partners). In three cases, we detected an accompanying somatic loss of a germline-mutated gene

202 (Fig. 3d). These results emphasize the need to further characterize the drivers of germline
203 susceptibility in men with MPC and to expand clinical germline testing beyond *BRCA2* in
204 diverse clinical settings.

205 ***Longitudinal blood biopsies enable study of tumor evolution in a patient-partnered model***

206 Ten patient-partners had WES from both tumor tissue and cfDNA, and three patient-
207 partners had both samples pass quality control metrics. Using the molecular data and abstracted
208 medical records, we sought to explore the evolutionary relationships between these longitudinal
209 samples in the context of patient clinical trajectories. Like most men with MPC, one participant,
210 patient-partner 0495, received a diverse range of treatments between biopsy timepoints (Fig. 4a).
211 After responding to first line anti-androgen therapy (leuprolide + bicalutamide), they took
212 second-generation anti-androgen inhibitors (abiraterone, enzalutamide), as well as experimental
213 radiotherapy and immunotherapy. To explore the relationship between samples, we utilized
214 PhylogicNDT, an algorithm that clusters mutations based on their prevalence in the tumor
215 (cancer cell fraction) into evolutionarily related subclones (Methods)[42]. In the cfDNA sample of
216 patient-partner 0495 but not the primary tumor, we observed two distinct frameshift mutations in
217 *ASXL2*, a gene implicated in castration-resistant metastatic prostate cancer, as well as an
218 amplification of *AR*, a known resistance mechanism to abiraterone and enzalutamide[43,44]. Patient-
219 partner 0093's tumor had clonal mutations in *TP53* and *KMT2D* but harbored an *NF2* mutation
220 solely in the cfDNA sample. Patient-partner 0213's tumor had a *TP53* mutation and APOBEC-
221 associated COSMIC signature 13 detected exclusively in the cfDNA sample.

222 Two of these patient-partners, 0495 and 0093, were initially diagnosed with primary
223 prostate cancer (Gleason score 4 + 3 and 5 + 4, respectively), while patient-partner 0213 was
224 diagnosed with *de novo* metastatic disease. The primary tumor tissues of these participants were

225 obtained at the time of diagnosis and separated from their donated blood samples by a range of
226 years, ranging from 2 to 10 years. Despite these varied disease presentations, clinical trajectories,
227 and biopsy timelines, we observed similar patterns of a "clonal switch" between the primary
228 tumor and cfDNA, wherein different subclones were dominant each sample (Fig. 4b,
229 Supplementary Fig. 8). We did not, however, observe primary tumor-specific copy number
230 alterations, bolstering previous claims that subclonal diversification in MPC via mutations may
231 happen after acquisition of ancestral copy number alterations (Supplementary Fig. 9)[45].
232 Furthermore, we observed primary tumor-specific mutations across all seven other patient-
233 partners with both tumor and cfDNA samples, although their exact clonal structure could not be
234 resolved due to low purity (Supplementary Fig. 10). While we cannot account for the sampling
235 bias of tumor biopsies, these results suggest that such clonal switches may be common in the
236 development of metastatic disease.

237 In two of the three patient-partners with tumor and cfDNA samples that passed quality
238 control, we detected the emergence of an amplification in the androgen receptor (*AR*) between
239 the initial diagnosis and metastatic blood sample that was accurately captured using ULP-WGS
240 of cfDNA (example patient-partner shown in Fig. 4c). This led us to examine *AR* copy number
241 using ULP-WGS of cfDNA samples across the entire cohort, including those that did not have
242 exome sequencing (n = 300 patient-partners, 318 samples, Fig. 4d). We found that patient-
243 partners who reported taking enzalutamide or abiraterone had significantly higher *AR* log copy-
244 ratios across a range of tumor fractions ($P < 0.001$, linear regression). Men who had taken
245 enzalutamide or abiraterone also had significantly higher tumor fractions, likely reflecting a
246 more advanced disease state and subsequent higher tumor burden in blood ($P < 0.001$, Mann-
247 Whitney U test)[46]. We observed that *AR* amplifications are often detectable in ULP-WGS of

248 cfDNA even when the tumor fraction is below 0.03 (Fig. 4e, f). For one patient-partner, the
249 tumor fraction within their donated blood was inferred as undetectable, but we nevertheless
250 observed a clear *AR* amplification (Fig. 4e). This highlights the potential efficacy of cfDNA to
251 reveal clinically relevant changes in MPC, even in cases of very low or undetectable tumor
252 burden. Broadly, these sequencing results illustrate the feasibility of identifying relevant
253 genomic and evolutionary alterations from both archival tumor tissue and donated blood samples
254 irrespective of geographical source site, enabling patient-partners to participate in genomic
255 research at no cost and with little effort.

DISCUSSION

256 Here we describe the MPCproject, a patient-driven framework for partnering with MPC
257 patients in the U.S. and Canada to increase access to genomics research and strengthen our
258 understanding of this disease. The online enrollment process was jointly created with patient-
259 partners to emphasize simplicity, requiring only the completion of basic online consent and
260 survey forms, along with optional mailed saliva and blood kits. To our knowledge, no previous
261 effort in MPC has used patient partnership to integrated demographic, clinical, patient-reported,
262 and genomic data from patients at a national level.

263 To that end, we demonstrated the feasibility of working with over 700 patient-partners,
264 41% of whom live in rural, medically underserved, or health physician shortage areas. We found
265 that patient-partners living in rural areas in this study likely travel significantly farther for their
266 cancer care, which has been shown to independently predict worse outcomes and mortality for
267 cancer patients[47]. Furthermore, a recent study found that incomplete medical records are
268 associated with shorter overall survival for MPC patients, particularly for those with complicated
269 clinical histories or whose care is fragmented between institutions[48]. Our analysis of abstracted

270 medical record data revealed a strong overlap between clinical histories represented in medical
271 records and patient-reported data, even for patient-partners with complex treatment trajectories
272 or who had received treatment at multiple hospitals, supporting the use of patient surveys to
273 improve care in this disease.

274 We also demonstrated that tumor tissue collected from paraffin-embedded archival
275 samples and cfDNA from donated blood samples from across the U.S. and Canada, enriched for
276 samples not obtained from NCI cancer centers, accurately recapitulate known genomic findings
277 in MPC, including somatic alterations, mutational signatures, germline pathogenic variants, and
278 a rare kataegis event. There has been substantial effort in the field to identify molecular features
279 associated with selective response to therapies like PARP inhibition and immunotherapy,
280 including the use of mutational signatures to assess targetable HRD, MMR, and APOBEC
281 deficiencies in cases without a causative molecular alteration[33,49]. Our results strengthen previous
282 findings that such signatures can be detected using cfDNA and, combined with our ability to
283 obtain cfDNA from participants nationwide, demonstrate the scalability of a patient-partnered
284 approach to identify and validate such genomic findings within a 'real world' cohort[50,51].

285 Moreover, we used archival tumor tissue and cfDNA from donated blood to reconstruct
286 tumor phylogenetic profiles, revealing polyclonality between primary and metastatic diagnosis.
287 Despite well-known findings of heterogeneity in both primary and metastatic prostate cancer,
288 there is a paucity of matched primary-metastatic studies, owing mostly to the invasiveness and
289 logistical challenges of longitudinal biopsy studies[31,52]. Our project enables such studies paired
290 with comprehensive clinical histories with minimal patient effort. To that end, we also found
291 clinically relevant *AR* amplifications via low-pass WGS of cfDNA from donated blood, even at
292 very low or undetectable tumor fractions. This result provides additional inexpensive utility to

293 the suggested use of cfDNA tumor fraction as a clinically relevant biomarker in metastatic
294 prostate cancer[50,46]. We are working with patient-partners who continue to donate blood and have
295 been able to collect multiple secondary blood biopsy kits for future longitudinal analysis.
296 Through feedback from patient-partners and advocates, we continue to improve the
297 MPCproject's design and outreach. Despite the geographic diversity of our patient-partners, we
298 recognize that they do not reflect the racial diversity of MPC patients, a critical issue given
299 substantial disparities in both cancer care and genomics research by race and ethnicity[11,53,54]. In
300 light of structural racism and a well-founded mistrust of medical research by patients of color,
301 this unmet disparity demands that we rethink our models of outreach and patient engagement[55].
302 We continue to work with community-based advocacy partners to involve communities of color,
303 and we are building a campaign to amplify Black cancer patient voices and their lived
304 experiences. We are also working to translate enrollment and educational materials into Spanish.
305 In addition, a common request by our patient-partners is to enable return of clinically relevant
306 results to participants and their physicians. While the regulatory hurdles to accomplish this are
307 large, we recognize its importance to our patient-partners and are striving to institute return of
308 results under this project model prospectively.
309 Paired with open-access clinical trials, patient-driven studies hold great promise to
310 achieve equity and accelerate discovery in genomic research[56]. The MPCproject is part of a
311 wider 'Count Me In' patient-partnered initiative (joincountmein.org) that has already yielded
312 new findings in angiosarcoma and has expanded to metastatic breast cancer and osteosarcoma,
313 among others[57–59]. The success of the MPCproject is based entirely on the courage and altruism
314 of the men with whom we partner, who, in the words of one participant, hope that their
315 "participation will help other men… and lead eventually to a cure".

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AUTHOR CONTRIBUTIONS

N.W., C.A.P. and E.M.V.A. conceived and designed the MPCproject with support from E.S.L. J.C., S.B., L.S., and E.M.V.A. designed and prepared the study and interpreted the data. J.C. wrote the manuscript and performed the analyses. S.B. and L.S. led study operations including tumor sample and medical record acquisition, sample sequencing, and patient

337 coordination. L.S., B.S.T., M.D., E.A., S.S., A.L.D., R.R., D.M.S., I.K.S. oversaw medical
338 record abstraction. S.Y.C. provided feedback on various analyses of the study and completed
339 germline variant calling with oversight from S.H.A. S.B., L.S., J.C., B.T., M.D., M.M., and
340 P.S.C. coordinated data releases. M.M., P.S.C., A.D., B.Z. led recent project operations. M.D.
341 supervised early project operations. C.M.N. and E.A. led patient advocacy and outreach efforts.
342 A.T.M.C. and S.W. oversaw early project sequencing analyses. M.X.H. provided feedback of
343 study analyses. A.K.T. provided feedback on medical record abstractions and tissue sample
344 collection. D.K. enabled electronic medical record searching. J.N., J.M., Major I.H.G., B.O.
345 contributed to survey design, project development, assessment of patient criteria, and outreach
346 strategy.

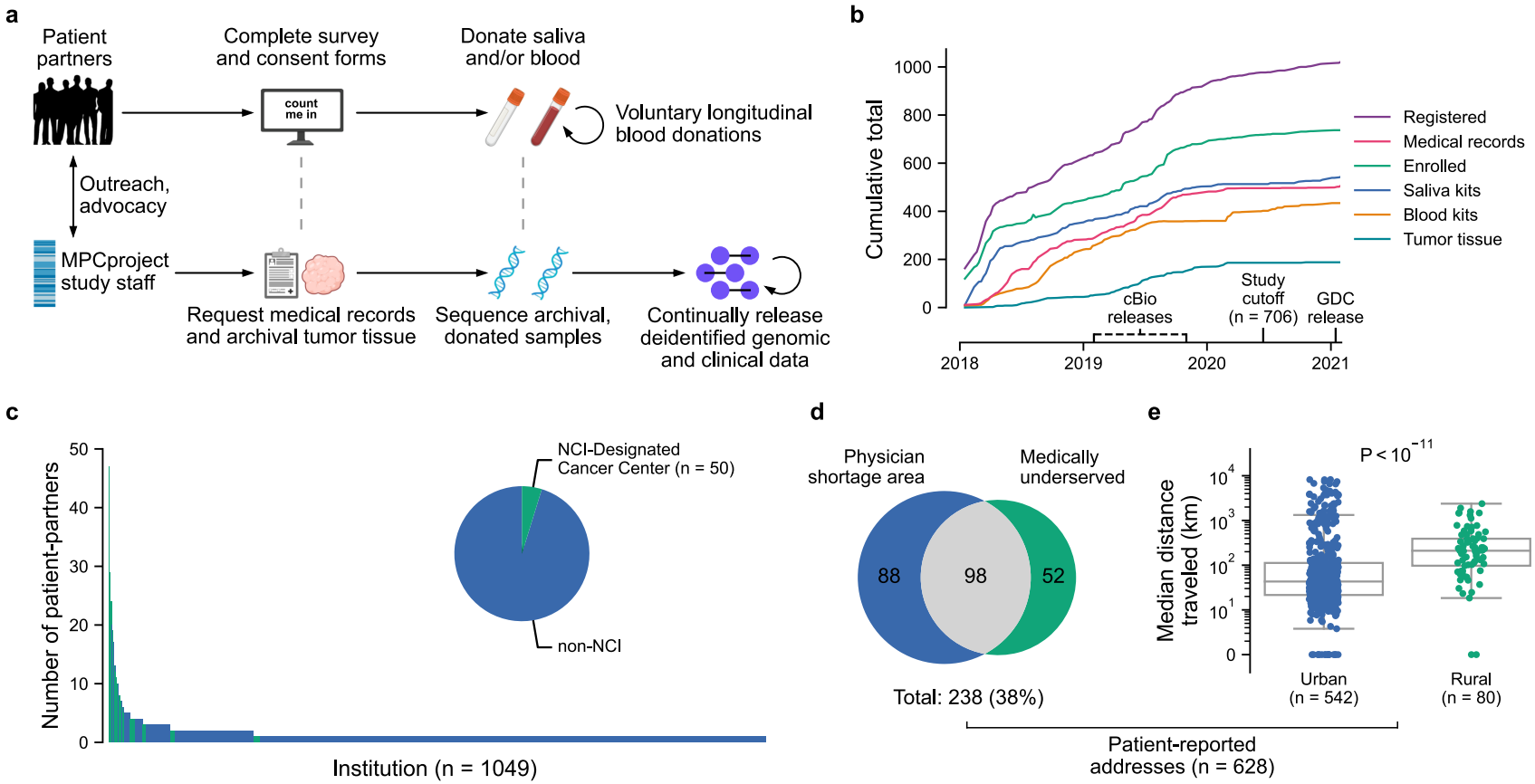
347 **COMPETING INTERESTS**

348 M.X.H. has been a consultant to Amplify Medicines and Ikena Oncology. E.S.L. is
349 currently in the process of divesting any relevant holdings. N.W. reports advisory relationships
350 and consulting with Eli Lilly and Co.; advising and stockholding interest in Relay Therapeutics;
351 and grant support from Puma Biotechnology. E.M.V.A. reports advisory relationships and
352 consulting with Tango Therapeutics, Genome Medical, Invitae, Illumina, Enara Bio, Manifold
353 Bio and Janssen; research support from Novartis and BMS; equity in Tango Therapeutics,
354 Genome Medical, Syapse, Manifold Bio and Enara Bio; and travel reimbursement from Roche
355 and Genentech, outside the submitted work.

356 **DATA AVAILABILITY**

357 Processed, deidentified data is available on cbioportal.org
358 (https://www.cbioportal.org/study/summary?id=prad_mpcproject_2018). Raw sequencing files
359 are available at the Genomic Data Commons (https://portal.gdc.cancer.gov/projects/CMI-MPC).

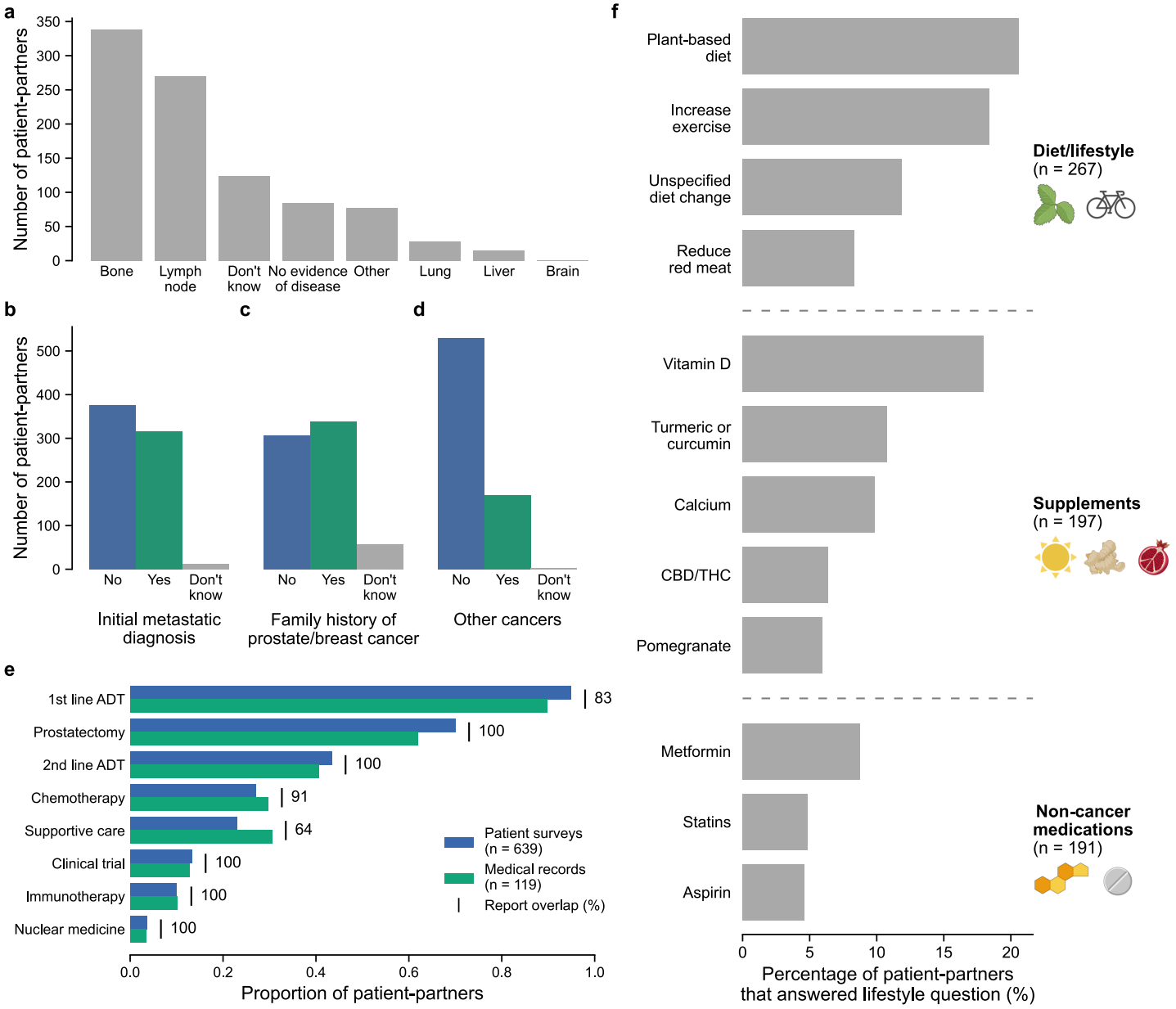
Please note that data is regularly being updated within these repositories and may not currently reflect all data generated from the project to date.


a
Patient partners
Complete survey and consent forms
Donate saliva and/or blood
count me in
Voluntary longitudinal blood donations
Outreach, advocacy
MPCproject study staff
Request medical records and archival tumor tissue
Sequence archival, donated samples
Continually release deidentified genomic and clinical data
b
Cumulative total
1000
800
600
400
200
0
Registered
Medical records
Enrolled
Saliva kits
Blood kits
Tumor tissue
cBio releases
Study cutoff (n = 706)
GDC release
2018
2019
2020
2021
c
Number of patient-partners
50
40
30
20
10
0
Institution (n = 1049)
NCI-Designated Cancer Center (n = 50)
non-NCI
d
Physician shortage area
Medically underserved
88
98
52
Total: 238 (38%)
e
Median distance traveled (km)
P < 10^-11
Urban (n = 542)
Rural (n = 80)
Patient-reported addresses (n = 628)

362

363 **Figure 1. Partnering with diverse patients to enhance our understanding of metastatic**
364 **prostate cancer**
365 **a)** Summary of MPCproject enrollment process. Patients learn about the project primarily
366 through outreach and partnered advocacy groups. If they register, patient-partners complete
367 online intake, consent, and medical release forms, then can opt into donating saliva via a mailed
368 kit and/or blood at routine blood draws at no charge. In parallel, MPCproject staff request
369 medical records and archival tumor samples from patients' medical institutions, then abstract
370 medical information from obtained records and sequence archival tumor tissue and/or donated
371 blood and saliva (Methods). Deidentified clinical, genomic, and patient-reported data are
372 released on a continual, prepublication basis and deposited in public repositories.
373 **b)** Enrollment statistics and timeline for the MPCproject. Depicted are the cumulative number of
374 patients that began the registration process (registered), patients that completed the survey and
375 consent forms (enrolled), patients with at least one medical record received (medical records),
376 and blood kits, saliva kits, and archival tumor tissue received at the Broad Institute for
377 sequencing (blood kits, saliva kits, tumor tissue, respectively). 706 patient-partners enrolled
378 before "Study cutoff", June 1, 2020, and are included in this study's analyses. cBioPortal
379 (cbioportal.org) releases include summary abstracted medical, genomic, and patient-reported
380 data; Genomic Data Commons (GDC) releases include raw sequencing files and demographic
381 data.
382 **c)** Represented medical institutions among patient-partners living in the U.S. and Canada. Shown
383 are the 1049 unique institutions (x-axis) where patient-partners report receiving care for their
384 prostate cancer, with the number of distinct patients at each institution (y-axis). NCI-designated

385 cancer centers are shown in green. Patient-partners that did not complete this survey question (n
386 = 36) and institutions outside the U.S. and Canada (n = 56) are not shown.
387 **d)** Access to medical care among patient-partners living in the U.S. Patient-reported U.S.
388 addresses were overlapped with primary care health physician shortage areas (HPSAs) and
389 medically underserved population/areas obtained from the Health Resources and Services
390 Administration (HRSA.gov). Patient-partners that live in Canada (n = 30), did not provide an
391 address (n = 40), or provided only a P.O. box (n = 8) are not shown.
392 **e)** Patient-partners in rural areas travel farther for clinical care. Using geographic census tract
393 information of self-reported home addresses along with USDA rural-urban continuum codes,
394 patient-partners were categorized as living in urban or rural areas. For each patient-partner, the
395 median Haversine round-trip distance between the zip code of their home address and that of
396 institutions they visited was calculated (Methods). Patient-partners that live in Canada (n = 30),
397 did not provide an address (n = 40), or provided only a P.O. box (n = 8) are not shown. *P*-value
398 calculated via two sided Mann-Whitney U test.


a
Number of patient-partners
350
300
250
200
150
100
50
0
Bone
Lymph node
Don't know
No evidence of disease
Other
Lung
Liver
Brain
b
c
d
Number of patient-partners
500
400
300
200
100
0
No
Yes
Don't know
Initial metastatic diagnosis
Family history of prostate/breast cancer
Other cancers
e
1st line ADT
Prostatectomy
2nd line ADT
Chemotherapy
Supportive care
Clinical trial
Immunotherapy
Nuclear medicine
83
100
100
91
64
100
100
100
Patient surveys (n = 639)
Medical records (n = 119)
Report overlap (%)
0.0
0.2
0.4
0.6
0.8
1.0
Proportion of patient-partners
f
Plant-based diet
Increase exercise
Unspecified diet change
Reduce red meat
Diet/lifestyle (n = 267)
Vitamin D
Turmeric or curcumin
Calcium
CBD/THC
Pomegranate
Supplements (n = 197)
Metformin
Statins
Aspirin
Non-cancer medications (n = 191)
0
5
10
15
20
Percentage of patient-partners that answered lifestyle question (%)

399

400 **Figure 2. Patient voices reveal the landscape of living with metastatic prostate cancer**

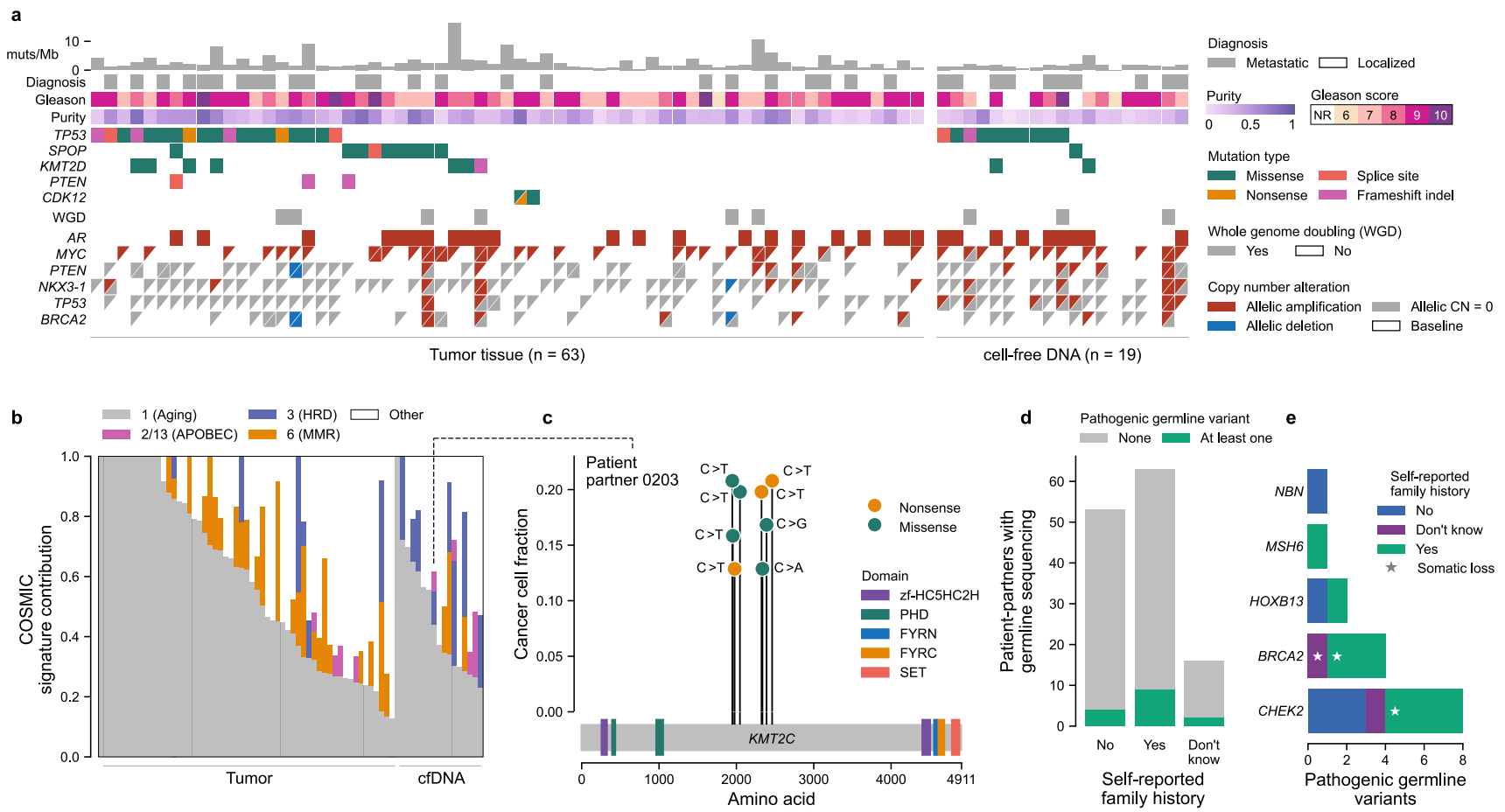
401 **a-d)** Self-reported data of 706 patient-partners related to their prostate cancer. In **a**, patient-
402 partners were asked for the current location of their cancer. Participants were free to choose
403 multiple if their cancer had metastasized to multiple locations. In **b-d**, responses were tabulated
404 from questions asking patient-partners if their initial prostate cancer diagnosis was metastatic
405 (**b**), if they have a family history of prostate/breast cancer (**c**), or if they have ever had another
406 cancer diagnosis (**d**). Patient-partners who did not complete these questions ($n < 5$) are not
407 shown.

408 **e)** Self-reported therapies show strong overlap with medical records. Drug categories are shown
409 on the y-axis, with the proportion of patient-partners from each data type (patient surveys and
410 medical records) receiving therapies of that category shown on the x-axis. In the online survey,
411 patient-partners selected therapies they received for their metastatic prostate cancer from a list.
412 639/706 patient-partners reported at least one therapy and are shown. 119 of these participants
413 also had abstracted therapy data from medical records. Report overlap refers to how often
414 patient-partners report receiving a therapy when their medical records show that they have
415 received that therapy, as a percentage. Only drugs available for selection in the patient survey
416 were used in this comparison (Supplementary Table 4).

417 **f)** Landscape of lifestyle changes for patient-partners. Participants were asked to list additional
418 medications, alternative medications, or lifestyle changes since their diagnosis of prostate cancer.
419 Free-text responses were manually abstracted and categorized into diet/lifestyle changes,
420 supplements, and non-cancer medications. The y-axis shows individual instances of diet/lifestyle
421 changes, supplements, or medications. The x-axis shows the percentage of patient-partners with
422 that lifestyle change or taking that supplement/drug out of all patient-partners that responded to

423 the lifestyle question (n = 456). CBD/THC: Cannabidiol/Tetrahydrocannabinol (oils, medical

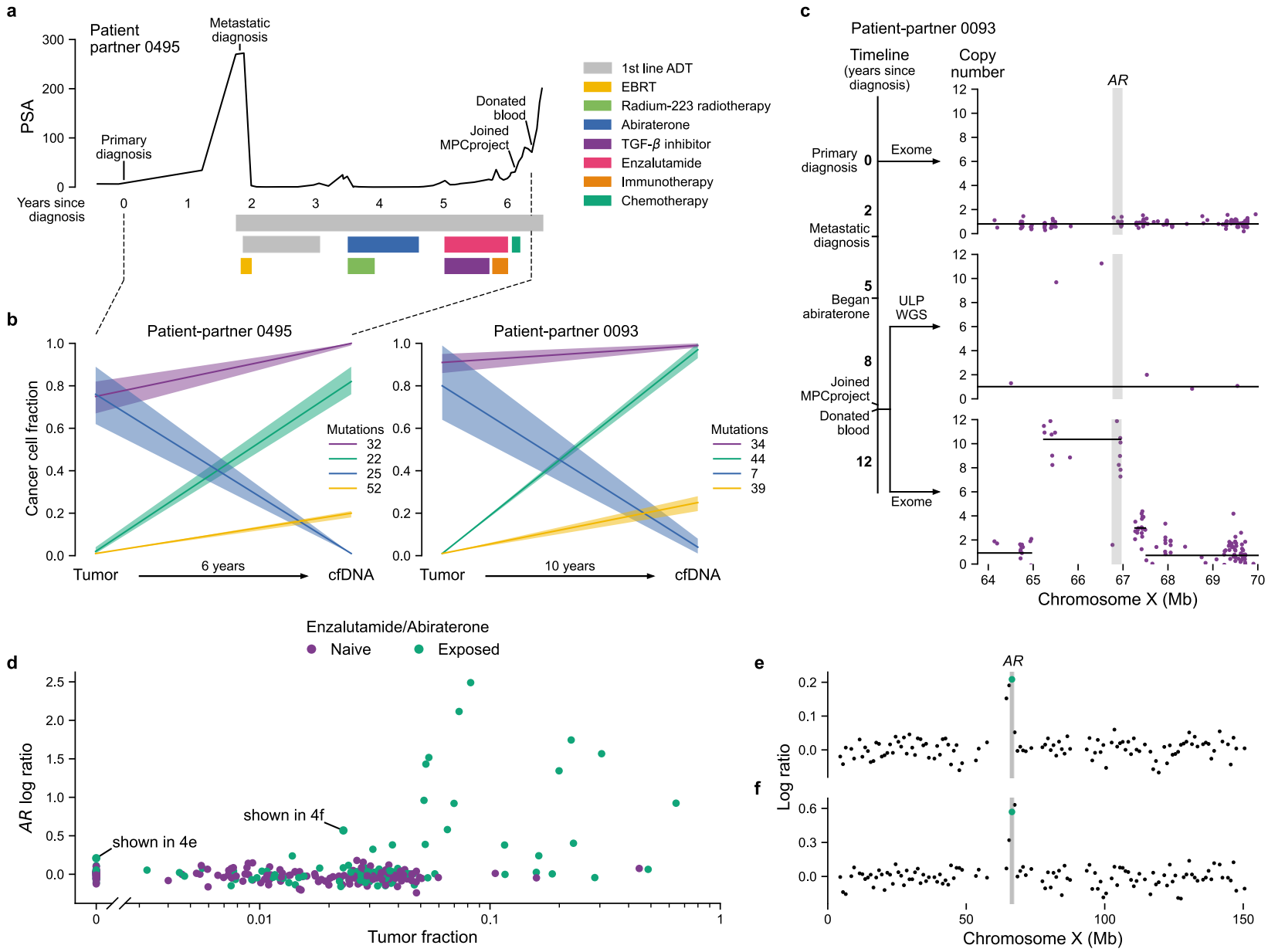
424 marijuana, etc).


a
muts/Mb
Diagnosis
Gleason
Purity
TP53
SPOP
KMT2D
PTEN
CDK12
WGD
AR
MYC
PTEN
NKX3-1
TP53
BRCA2
Tumor tissue (n = 63)
cell-free DNA (n = 19)
Diagnosis
Metastatic
Localized
Purity
Gleason score
NR 6 7 8 9 10
Mutation type
Missense
Splice site
Nonsense
Frameshift indel
Whole genome doubling (WGD)
Yes
No
Copy number alteration
Allelic amplification
Allelic CN = 0
Allelic deletion
Baseline
b
1 (Aging)
3 (HRD)
Other
2/13 (APOBEC)
6 (MMR)
COSMIC signature contribution
Tumor
cfDNA
c
Patient partner 0203
Cancer cell fraction
Nonsense
Missense
Domain
zf-HC5HC2H
PHD
FYRN
FYRC
SET
KMT2C
Amino acid
d
Pathogenic germline variant
None
At least one
Patient-partners with germline sequencing
No
Yes
Don't know
Self-reported family history
e
NBN
MSH6
HOXB13
BRCA2
CHEK2
Self-reported family history
No
Don't know
Yes
Somatic loss
Pathogenic germline variants

425

426 **Figure 3. Donated tumor and cell-free DNA samples obtained through patient partnership**
427 **recapitulate known genomic findings in metastatic prostate cancer**
428 **a)** Genomic and clinical landscape of 82 sequenced samples. Columns represent samples,
429 separated into tumor (prostate, left) and cfDNA (donated blood, right) samples, while rows
430 represent select clinical and genomic features. Gleason scores for tumor samples are taken from
431 the pathology report received with the sample (n = 58) or the patient-partner's medical records (n
432 = 5) if Gleason scores were not provided in the report. Gleason scores for cfDNA were taken
433 from pathology reports in the medical record, with NR representing cases where a Gleason score
434 was not reported in the medical record. Diagnosis refers to whether the initial diagnosis of
435 prostate cancer was localized or metastatic. Multiple mutations in the same gene are represented
436 as triangles. WGD refers to whole genome doubling. Copy number calls are allelic and defined
437 with respect to baseline allelic ploidy (2 for samples with WGD, 1 for those without), with calls
438 for the two alleles indicated by two triangles (except for *AR*, which has only one allele in men
439 and so is shown as a single box). Allelic CN = 0 refers to complete allelic deletions. Allelic
440 deletions that are not complete deletions are possible in samples with WGD. Figure created with
441 CoMut[60].
442 **b)** Mutational signature analysis of sequenced samples. The relative contribution of select
443 COSMIC v2.0 mutational signatures are shown, separated by tumor and cfDNA (donated blood)
444 sample type[30]. APOBEC refers to signatures associated with activity of APOBEC family of
445 cytidine deaminases (signature 2 and 13); MMR to the signature associated with deficient DNA
446 mismatch repair (signature 6); HRD to the signature associated with homologous recombination
447 deficiency (signature 3). Samples with too few mutations for signature analysis (< 50 mutations,
448 n = 5 samples) are not shown.

449 **c)** Instance of localized hypermutation (kataegis) of *KMT2C* in cfDNA from a donated blood
450 sample. The y-axis shows the cancer cell fraction of each mutation while the x-axis shows their
451 amino acid within *KMT2C*. Domains taken from Pfam[61]. The dotted line connects to this
452 sample's mutational signature profile.
453 **d)** Germline pathogenic DNA repair alterations and their overlap with patient reported family
454 history. Pathogenic germline alterations (as annotated by ClinVar) in genes from a select panel
455 of DNA repair genes implicated in prostate cancer were detected in patient-partners with
456 sequenced saliva or blood buffy coat (n = 132) (Methods; Supplementary Table 3)[62]. Survey
457 responses to a question asking about a family history of prostate or breast cancer were tabulated
458 and overlapped with this genomic data. Stars indicate instances where a somatic deletion also
459 affected that gene in a tumor or cfDNA sample from that patient-partner.


a
Patient partner 0495
Metastatic diagnosis
PSA
Primary diagnosis
Donated blood
Joined MPCproject
Years since diagnosis
1st line ADT
EBRT
Radium-223 radiotherapy
Abiraterone
TGF-β inhibitor
Enzalutamide
Immunotherapy
Chemotherapy
b
Patient-partner 0495
Patient-partner 0093
Cancer cell fraction
Mutations
32
22
25
52
34
44
7
39
Tumor
6 years
10 years
cfDNA
c
Patient-partner 0093
Timeline (years since diagnosis)
Copy number
AR
Primary diagnosis
Metastatic diagnosis
Began abiraterone
Joined MPCproject
Donated blood
Exome
ULP WGS
Chromosome X (Mb)
d
Enzalutamide/Abiraterone
Naive
Exposed
AR log ratio
shown in 4f
shown in 4e
Tumor fraction
e
f
Log ratio
Chromosome X (Mb)

460

461 **Figure 4. cfDNA from donated blood reveals patterns of clonal dynamics and clinically**
462 **relevant genomic changes**

463 **a)** Clinical trajectory of patient-partner 0495. This patient-partner's prostate specific antigen
464 (PSA) trajectory is shown on the y-axis, time in years since initial diagnosis is shown on the x-
465 axis, and bars denote the beginning and end of therapies. EBRT—external beam radiation
466 therapy; 1st line androgen deprivation therapy (ADT)—leuprolide and bicalutamide;
467 immunotherapy—nivolumab; chemotherapy—cisplatin and etoposide.

468 **b)** Tumor evolution from primary tumor to metastatic cfDNA samples. The y-axis shows the
469 cancer cell fraction (CCF) of clonal clusters identified between tumor and cfDNA samples (x-
470 axis). Time between samples shown on the x-axis. Colors indicate how many mutations were
471 identified in each clone, with a 95% confidence interval around the estimated CCF. Purple
472 represents the truncal/ancestral clone. Clusters with CCF < 0.10 across all biopsies are omitted.
473 The clinical trajectory of patient-partner 0495 (left) is shown in **a**, while the trajectory of patient-
474 partner 0093 (right) is shown in **c**.

475 **c)** Emergence of *AR* amplification in patient-partner 0093 induced by anti-androgen therapy. The
476 timeline depicts this patient's clinical trajectory, while the plots show the absolute copy number
477 (y-axis) of the genomic region around *AR* (x-axis, gene body shown in grey). The first plot
478 depicts exome sequencing from the patient's archival tumor tissue; the second and third plots
479 depict ultra-low pass whole-genome sequencing (ULP-WGS) and exome sequencing of cfDNA
480 from the patient's donated blood, respectively. Individual points represent copy number of target
481 regions (exome) or copy number of 1 Mb genomic windows (ULP-WGS). Black lines represent
482 discrete copy number segments.

483 **d – f)** ULP-WGS reveals clinically relevant *AR* amplifications even at low tumor fraction. Tumor
484 fraction of 318 cfDNA samples from donated blood of 300 patient-partners with ULP-WGS
485 sequencing is shown on the x-axis, while the log copy-ratio (logR) of the genomic interval
486 containing *AR* is shown on the y-axis. Points are colored by whether patient-partners self-
487 reported taking enzalutamide or abiraterone. 89 samples are shown with tumor fraction of 0
488 (undetectable), while 229 have nonzero tumor fractions. Two samples, one at a tumor fraction of
489 0 and another at a tumor fraction of 0.023, have chromosome X log copy-ratio profiles shown in
490 **e** and **f**, respectively. The green points represent the values shown in **d,** with the genomic interval
491 containing *AR* highlighted in grey.

METHODS

492 *Statistical computing*

493 Except where otherwise specified, analysis and data visualization were performed with
494 Python 3.8, SciPy v.1.5.2, Matplotlib v.3.3.2, seaborn v.0.11.0 and R v.3.5.1. All statistical tests
495 were two-sided unless otherwise specified. The code used to generate the main figures can be
496 found at https://github.com/vanallenlab/mpcproject-paper.

497 *MPCproject website*

498 The MPCproject utilizes a website (https://mpcproject.org/) to enroll patients through an
499 online consent and release form. The website provides information about the project and
500 advocacy groups that have partnered with the study. The website design, messaging, and
501 workflow were developed with direct input from patient-partners and advocates.

502 *Informed consent*

503 Patients who chose to enroll in this research study are provided informed consent using a
504 web-based consent form approved by the Dana-Farber/Harvard Cancer Center Institutional
505 Review Board (DF/HCC Protocol 15-057B). A link to the electronic informed consent document
506 for formal enrollment in the study (https://mpcproject.org/ConsentAndRelease.pdf) was sent to
507 registrant emails, and upon signing, a copy of the completed form was shared. At minimum,
508 informed consent enabled study staff to request and abstract medical records, send a saliva kit
509 directly to patients, perform sequencing on any returned saliva samples, and release de-identified
510 integrated clinical, genomic, and patient-reported data for research use. Patient-partners had the
511 additional option to consent to study staff obtaining a portion of archived tumor tissue and/or a
512 blood sample for further sequencing analysis.

513 *Patient-reported data*

514 After registering, patient-partners completed a 17-question survey asking them about
515 themselves and their disease (https://mpcproject.org/AboutYouSurvey.pdf). All questions were
516 optional. Information on how question responses were standardized and categorized can be
517 found in the Supplementary Methods.

518 *Acquisition of medical records*

519 Medical records were obtained for patient-partners from the U.S. and Canada who
520 completed the consent and medical release forms. Later in project development, a donated saliva
521 or blood sample was also required. Study staff submitted medical record requests to all
522 institutions and physician offices at which the patient reported receiving clinical care for their
523 prostate cancer. A detailed medical record request form, along with the consent and release
524 forms, were electronically faxed to each facility listed in a patient's release form. Medical
525 records were returned to the project via mail, fax, or secure online portals. If a record request was
526 not fulfilled in six months, study staff called the hospital, and a second request was submitted,
527 with up to three requests made. Patient-partners that communicated with study staff about
528 changes in their treatment could request a medical record update, in which case their current
529 hospital was again contacted for medical records. All medical records were saved in an
530 electronic format to a secure drive at the Broad Institute.

531 *Acquisition of patient samples*

532 All consented patient-partners living in the United States or Canada were mailed saliva
533 kits with appropriate instructions, a sample tube labeled with a unique barcode, and a prepaid
534 return box to send back the saliva sample. Samples were returned to the Broad Institute
535 Genomics Platform, logged, and stored at room temperature (25 °C) until further sequencing.

536 If a consented patient-partner opted into the blood biopsy component of the study, they
537 were sent a blood kit with instructions (https://mpcproject.org/BloodSampleInstructions.pdf,
538 Supplementary Figure 4). Participants could take this kit to their next blood draw and request a
539 courtesy draw by their medical provider; if a courtesy draw was not possible, patients could go to
540 Quest Diagnostics with a complimentary voucher to have their blood drawn. Blood kits were
541 returned free of charge to the Broad Institute Genomics Platform where they were fractionated
542 into plasma and buffy coats and stored at -80°C. If a patient-partner did not provide a saliva
543 sample, buffy coats were used to extract germline DNA for WES. Plasma samples continued to
544 WES if ultra-low pass WGS detected a tumor fraction of circulating tumor DNA greater than
545 0.03. Some patient-partners were selected to provide additional blood samples and were sent a
546 new consent form. If they agreed to submit another blood sample, a new blood kit was shipped.
547 For patient-partners that provided a germline sample and consented to the acquisition of
548 some of their archival tumor tissue, study staff reviewed each patient's medical records and
549 identified available tissue (Supplementary Methods). Patient-partners were screened by the study
550 staff to determine if they had metastatic or advanced prostate cancer based on the definition by
551 our study. If a patient-partner had a sample that met the project's strict requesting criteria, study
552 staff coordinated with that hospital's pathology department to fax a request for one H&E-stained
553 slide as well as either 5-20 5-µm unstained slides or one formalin-fixed paraffin-embedded tissue
554 block. Requests explicitly asked that the pathology department should not exhaust a sample to
555 fulfill the request. Samples were sent to the MPCproject by mail. Tissue samples received as
556 slides were labeled with unique barcode identifiers and submitted for whole exome sequencing.
557 Tissue samples received as blocks were cut into three 30-µm scrolls per block, labeled with
558 unique barcode identifiers, and then submitted for whole exome sequencing.

559 *Medical record abstraction*

560 A data dictionary comprising 60 clinical fields with possible options was curated by
561 trained study staff working with prostate oncologists. Electronic health records were converted to
562 searchable PDF files using the Optical Character Recognition (OCR) engine known as
563 Tesseract[63]. Three study staff abstractors were involved in the abstraction and QC process for
564 each record (Supplementary Methods). If a field had lack of concordance between abstractors or
565 there were outstanding questions, a prostate cancer oncologist reviewed the content. Whenever
566 possible, clinical data was abstracted directly from the records. For information that's not found,
567 it was abstracted as 'NOT FOUND IN RECORD'. In instances where ambiguity or incomplete
568 data was present, inferences were made considering the whole narrative of the medical record.
569 Incomplete dates missing the day or month are abstracted as the first day of the month or first
570 month of the year, respectively. While all medical records will eventually be abstracted, medical
571 records from patient-partners that received molecular sequencing of some form were prioritized
572 for this study, resulting in 125 patient-partners with medical record abstractions, 119 of which
573 had at least one therapy noted. In examining the overlap between patient surveys and medical
574 record therapies, we only considered therapies that were given for metastatic prostate cancer at
575 least one week before the patient enrolled.

576 *Geographic analysis*

577 Using secure Census Bureau geocoding, we identified the census tracts of patient
578 reported home addresses[64]. To identify patient-partners living in rural areas, this information was
579 overlapped with rural-area continuum (RUCA) codes from the United States Department of
580 Agriculture (USDA)[65]. Addresses with a secondary RUCA code greater than 3 were designated
581 as rural. For comparison, the proportion of metastatic prostate cancer patients within each RUCA

582 code from 2004 – 2017 was taken from Surveillance, Epidemiology, and End Results (SEER)
583 using SEER*stat with the following selection table: `{Site and Morphology.Site recode ICD-O-`
584 `3/WHO 2008} = 'Prostate' AND {Stage - Summary/Historic.SEER Combined Summary Stage`
585 `2000 (2004-2017)} != 'In situ', 'Localized only', 'Not applicable',`
586 `'Unknown/unstaged/unspecified/DCO', 'Blank(s)'`[23]. To identify patient-partners living in
587 medical shortage areas, the census tracts of home addresses were overlapped with primary care
588 health physician shortage areas (HPSA) and medically underserved areas (MUA) defined by the
589 Health Resources and Services Administration (HRSA)[25]. Addresses were labelled as existing
590 within a MUA if they were designated as within a medically underserved area or population and
591 as existing within a HPSA if they were designated as within a primary care HPSA. Published
592 geographic datasets of cancer patients (e.g., SEER, NPCR) do not contain census-tract resolved
593 data or summary results of MUA/HPSA status, so for comparison we instead used the total U.S.
594 population living in HPSAs and MUAs, taken from HRSA, divided by the entire U.S. population
595 taken from the U.S. Census[25,26]. To calculate appointment distances, we calculated the round-trip
596 Haversine distances between the zip code of home addresses and the zip code of reported
597 institutions.

598 *Whole exome sequencing analysis*

599 Whole exome sequences were captured using Illumina technology and the sequence data
600 processing and analysis was performed using Picard and FireCloud pipelines on Terra
601 (https://terra.bio/) (Supplementary Methods). The Picard pipeline (http://picard.sourceforge.net)
602 was used to produce a BAM file with aligned reads. This includes alignment to the GRCh37
603 human reference sequence using BWA[66] and estimation and recalibration of base quality score
604 with the Genome Analysis Toolkit (GATK)[67]. Somatic alterations for tumor samples were called

605 using a customized version of the Getz Lab CGA WES Characterization pipeline
606 (https://portal.firecloud.org/#methods/getzlab/CGA_WES_Characterization_Pipeline_v0.1_Dec2
607 018/) developed at the Broad Institute. Briefly, MuTect v1.1.6 algorithm was used to identify
608 somatic mutations[68]. Somatic mutation calls were filtered using a panel of normals (PoN), oxoG
609 filter and an FFPE filter to remove artifacts introduced during the sequencing or formalin
610 fixation process[69]. Small somatic insertions and deletions were detected using the Strelka
611 algorithm[70]. Somatic mutations were annotated using Oncotator[71]. Recurrently altered mutations
612 were identified using MutSig2CV[72]. To define somatic copy ratio profiles, we used GATK
613 CNV[67]. To generate allele-specific copy number profiles and assess tumor purity and ploidy, we
614 used ABSOLUTE and FACETS[73,74]. Final segmentation calls were taken from ABSOLUTE,
615 except for the X chromosome, which was taken from FACETS. We utilized GISTIC2.0 to
616 identify significantly recurrent amplification and deletion peaks[28]. For determining allele-specific
617 copy number alterations, we assessed the absolute allelic copy numbers of the segment
618 containing each gene. Mutation burden was calculated as the total number of mutations (non-
619 synonymous + synonymous) detected for a given sample divided by the length of the total
620 genomic target region captured with appropriate coverage from whole exome sequencing.
621 *Whole exome sequencing quality control*
622 Samples with average coverage below 55x in the tumor sample or below 30x in the
623 normal sample were excluded. Samples with purity < 0.10 from both ABSOLUTE and FACETS
624 were excluded. DeTiN was applied to samples to estimate the amount of tumor contamination in
625 the normal samples; samples with TiN (tumor in normal) > 0.25 were excluded[75]. ContEst was
626 applied to measure the amount of cross-sample contamination in samples; samples with
627 contamination > 0.04 were excluded[76]. The Picard task CrossCheckFingerprints was applied to

628 determine sample mixups; samples with Fingerprints LOD value < 0 were excluded[77]. Samples
629 which passed quality control were submitted to cBioPortal and GDC.

630 *Ultra-low pass whole genome sequencing analysis*

631 ichorCNA was used to assess the tumor fraction in cfDNA samples that completed ultra-
632 low pass whole genome sequencing[50]. The log copy ratio of *AR* was assessed by the log copy
633 ratio of the genomic interval containing *AR.* This value could not consistently be converted to
634 absolute copy number due to the low tumor fractions of many samples.

635 *Mutational signature analysis and kataegis*

636 Mutational processes in our cohort were determined using deconstructSigs with default
637 parameters applying COSMIC v2 signatures as the reference with a maximum number of
638 signatures of 6[29,30]. A signature was assessed as present if the signature contribution was greater
639 than 6%. Because tumor samples were formalin-fixed and paraffin embedded (FFPE), a process
640 known to introduce stranded mutational artifacts in specific nucleotide contexts, we used a filter
641 to remove likely FFPE artifacts according to nucleotide context and strand bias before using
642 deconstructSigs[78]. We also tried to assess the colocalization of the kataegis event with structural
643 variant breakpoints but were limited by targeted sequencing in exomes and low coverage in
644 ULP-WGS. *KMT2C* and its surrounding region were not copy number altered in the sample with
645 kataegis. Kataegis was not identified in any other sample.

646 *Association of DNA-repair alterations and presence of signature 3*

647 Alterations in a select list of genes previously implicated in DNA-repair in prostate
648 cancer were examined (Supplementary Table 3). An alteration was considered if there was a
649 somatic single-copy deletion, double deletion, nonsense mutation, missense mutation, frameshift

650 indel, or splice site mutation. An alteration was also considered if there was a pathogenic
651 germline alteration, denoted by "Pathogenic" in ClinVar[62].

652 *Germline variant discovery*

653 To call short germline single-nucleotide polymorphisms, insertions, and deletions from
654 germline WES data, we used DeepVariant (v0.8.0)[79,80]. Specifically, we used the publicly-
655 released WES model
656 (https://console.cloud.google.com/storage/browser/deepvariant/models/DeepVariant/0.8.0/Deep
657 Variant-inception_v3-0.8.0+data-wes_standard/) to generate single-sample germline variant call
658 files using the human genome reference GRCh37(b37). We filtered variants with bcftools v1.9 to
659 only keep high-quality variants annotated as "PASS" in the "FILTER" column. The high-quality
660 variants were merged into single-sample Variant Call Format (VCF) files using
661 CombineVariants from GATK 3.7 (https://github.com/broadinstitute/gatk/releases). To
662 decompose multiallelic variants and normalize variants, we used the computational package vt
663 v3.13 (https://github.com/atks/vt). Lastly, germline variants were annotated using the VEP v92
664 with the publicly-released GRCh37 cache file (https://github.com/Ensembl/ensembl-vep)[81].
665 Germline variants were denoted as pathogenic if they appeared as "Pathogenic" in ClinVar (Dec
666 2019 version)[62].

667 *Phylogenetic analysis*

668 To compare mutations between distinct samples (tumor and cfDNA) from the same
669 patient, we used a previously described method designed to recover evidence for mutations
670 called in one sample in all other samples derived from the same individual[82]. In brief, the 'force-
671 calling' method uses the strong prior of the mutation being present in at least one sample in the
672 patient to more sensitively detect and recover mutations that might otherwise be missed. A

673 mutation was deemed tumor/cfDNA specific if there were no force-called reads that supported
674 the mutation in the other sample, although this process underestimates the proportion of shared
675 mutations in low purity tumors. The cancer cell fraction (CCF) of mutations were defined using
676 ABSOLUTE, which calculates the CCF based on variant allele frequency, purity, and local
677 allelic copy number[73]. To reconstruct tumor phylogenies, we used PhylogicNDT, which clusters
678 mutations into subclones across multiple samples based on their underlying similar CCFs[42].

679 *Data releases*

680 The MPCproject releases de-identified clinical, patient-reported and research-grade
681 genomic data into public repositories, such as cBioPortal
682 (https://www.cbioportal.org/study/summary?id=prad_mpcproject_2018) and the Genomic Data
683 Commons (https://portal.gdc.cancer.gov/projects/CMI-MPC), at regular intervals and pre-
684 publication. Data is processed and formatted as required by each repository's guidelines. All
685 patient identifiers are stripped prior to data deposition to protect patient privacy. On the
686 MPCproject data release webpage (https://mpcproject.org/data-release), patients can access
687 project data, additional information about the data, list of common terms used in research,
688 methods used to generate the data, and an email address for any additional data-related questions.

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