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REVIEW

The diverse heterogeneity of molecular alterations in prostate cancer identified through next-generation sequencing

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Prostate cancer is a leading cause of global cancer-related death but attempts to improve diagnoses and develop novel therapies have been confounded by significant patient heterogeneity. In recent years, the application of next-generation sequencing to hundreds of prostate tumours has defined novel molecular subtypes and characterized extensive genomic aberration underlying disease initiation and progression. It is now clear that the heterogeneity observed in the clinic is underpinned by a molecular landscape rife with complexity, where genomic rearrangements and rare mutations combine to amplify transcriptomic diversity. This review dissects our current understanding of prostate cancer 'omics', including the sentinel role of copy number variation, the growing spectrum of oncogenic fusion genes, the potential influence of chromothripsis, and breakthroughs in defining mutation-associated subtypes. Increasing evidence suggests that genomic lesions frequently converge on specific cellular functions and signalling pathways, yet recurrent gene aberration appears rare. Therefore, it is critical that we continue to define individual tumour genomes, especially in the context of their expressed transcriptome. Only through improved characterisation of tumour to tumour variability can we advance to an age of precision therapy and personalized oncology.
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INTRODUCTION

Prostate cancer is the most common cancer affecting men in the Western world and in 2012 accounted for 28 000 deaths in the United States alone.[1] In contrast to many other malignancies, there are few clear histopathological subtypes on which to base patient diagnoses and prognoses, and the majority of diagnostic weight falls on the differentiation status of tumour cells (Gleason grading and score).[2] Unfortunately, even when combined with predictive nomograms, Gleason score does not provide sufficient information to accurately stratify tumours, and patients receiving identical diagnoses can exhibit markedly different clinical outcomes. This is particularly pertinent when considering tumours of a low Gleason grade, which are now enriched in the clinic due to early screening for elevated serum-prostate-specific antigen (PSA). A significant proportion of low-grade tumours will remain comparatively indolent, whereas a small fraction will progress and require aggressive intervention (reviewed in Ref. 3). Since distinguishing these outcomes with high confidence is beyond Gleason grade and clinical nomograms, treatment policies tend toward intervention, incurring overtreatment costs (both financial and morbidity-related).

Clinical heterogeneity is not merely a problem complicating diagnoses: after surgery and/or radiation a proportion of patients will relapse and require further treatment, but response to standard therapies is highly mixed. For years, the primary therapy for relapse has been interference of the androgen-signalling axis, a growth and differentiation-inducing pathway mediated by the androgen receptor (AR). Initially, almost all tumours respond to androgen deprivation therapies, whether they were the relatively crude orchiectomies of the 1960s, or anti-androgens that bind to and inhibit the AR protein itself.[4,5] However, tumours eventually develop resistance to androgen deprivation, and are considered castrate-resistant prostate cancer (CRPC), although the latency period before CRPC development is highly variable. Tumours in this setting are technically androgen-independent, since they are no longer dependent on circulating androgens for growth, but frequently remain addicted to the androgen signalling axis, through reactivation of the AR by various mechanisms.[6] However, clinical heterogeneity is abundant even in CRPC, with a fraction of tumours exhibiting epithelial plasticity and transforming to a lethal form of the disease known as neuroendocrine prostate cancer (NEPC).[7–9] NEPC is AR- and PSA-negative, and there are currently no targeted treatments, although concentrated efforts are beginning to provide promising leads.[10,11]

In the context of such significant clinical challenges, an enormous variety of tools and technologies have been applied to all stages of

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prostate tumour development, with the ultimate goal of identifying molecularly defined subtypes which may influence patient diagnoses or prognoses and guide therapeutic strategies. In recent years, wide-scale application of microarray technology and next-generation sequencing has led to tremendous progress in identifying molecular events which underlie cancer initiation, progression, metastasis and resistance to therapy. This success has defined molecular subtypes of prostate cancer and described extensive genomic aberration, but the urgently required links to clinical outcome have remained largely elusive. However, there are few highly recurrent events, and the molecular landscape appears to be one of great biological heterogeneity, where each tumour develops a unique combination of somatic changes, some of which drive tumour development. Since for any given patient, it is this unique combination of somatic changes which will determine ultimate clinical outcome, it is relatively simple to understand how massive molecular heterogeneity has for so long confounded attempts to develop diagnostic, prognostic and therapeutic breakthroughs.

There is hope on the horizon. Despite the apparent molecular uniqueness of each tumour–patient combination, it is becoming increasingly clear that the overall functional or downstream effects of differing combinations of somatic alterations may be recurrent. Furthermore, with the age of personalized therapy rapidly approaching, unique changes themselves should not be ignored, and may not be as 'undruggable' as previously thought, especially in the context of developments in small molecule and antisense technologies.[12,13] Continuing definition and refinement of individual molecular landscapes will help our understanding of prostate cancer progress to a stage where specific pathway activation can be recognized from unique constellations of somatic changes, and (if possible) therapeutically targeted. This review provides a brief dissection of the complex molecular heterogeneity of prostate cancer, drawing on the growing wealth of data generated through next-generation sequencing.

GENOME REARRANGEMENT

Copy number aberration is common but highly heterogeneous

Prostate cancer is characterized by high levels of genome rearrangement disrupting tumour suppressor genes and activating oncogenic pathways. Genome rearrangements frequently manifest as alterations in the copy number state of chromosomal regions, and for years copy number variation has been recognized as a sentinel feature of prostate tumour genomes. Technological advances, from basic cytogenetics, through microarrays to high-coverage genome and exome sequencing have allowed fine-scale mapping of recurrently variable regions with increasing resolution. Several regions are aberrant at high frequency, including gains at 8q (MYC at 8q24) and losses at 3p, 8p (NKX3.1 at 8p21), 10q (PTEN at 10q23), 13q (RB1) and 17p (TP53 at 17p).[14–20] However, these commonly altered regions belie substantial heterogeneity. For example, a recent study compiled 372 prostate cancer genomes from published data and used systematic method called genomic identification of significant targets in cancer to define a staggering 90 regions which are recurrently altered in prostate cancer (73 amplifications and 17 deletions; see **Figure 1**).[21,22] Clearly, the potential combinations of 90 regions are extremely high, not even accounting for tumour-specific (non-recurrent) alterations.

Oncogenes or tumour suppressors often map to copy number aberrations, for example among the 73 recurrently amplified regions in prostate cancer identified by Kim *et al.*[21] were 18 cancer consensus genes including *FGFR3*, *STK11*, *NCOA2*, *HRAS*, *MLLT11* and *BRAF*.[23] However, attempts to link specific copy number alterations with outcome have had only limited success in adding to the predictive power of Gleason grading and clinical nomograms. For example, we defined 39 genomic markers associated with metastatic potential, while others have linked copy number aberrations of specific genes (e.g. *MYC*, *PTEN*) to poor prognosis.[24–26] On the other hand, it may simply be the degree of copy number aberration in a given genome which confers prognosis. Credence for this theory comes from the observation that a subset of localized prostate tumours resemble advanced prostate cancers and CRPCs (which typically exhibit high levels of CN aberration).[17,27]

Analyzing copy number aberration in the context of congruent gene expression alterations has provided some positive results, notably that there appears to be convergent alteration of particular growth mechanisms such as the AR, RB, PI3K and RAS/RAF signalling pathways.[17,27] Some of these pathways are therapeutically exploitable (e.g. with PI3K inhibitors), although stringent patient selection is likely to be important.[28] Furthermore, Beltran *et al.*[10] demonstrated concurrent amplifications and overexpression of MYCN and AURKA in NEPC, suggestive of a link to disease etiology.

Although there are few homozygous losses or high gains observed in primary tumours, CRPCs which have undergone significant evolution in response to therapies tend to exhibit several, including frequent amplifications of the AR loci, as well as those of AR cofactors.[6,20,29] Although gene amplification can demonstrate pathway dependence and offer therapeutic targets (e.g. *ERBB2* in breast cancer), genes residing within homozygous deletions can also be exploited. For example, we recently identified a homozygous deletion of the *MTAP* gene (methylthioadenosine phosphorylase) in an advanced prostate tumour.[11] Subsequently, in a high-fidelity patient-derived xenograft, we demonstrated that treatment with methylthioadenosine and high dose 6-thioguanine caused tumour growth inhibition, while protecting the host from 6-thiguanine toxicity. Since homozygous MTAP deletions or hypermethylation of promoter regions exhibit pan-cancer recurrence, MTAP may represent a viable therapeutic target in a wide range of tumours.

Genome breakpoints are an important mutational mechanism

Despite the oncogenic implications of recurrent regions of copy number aberration, the specific edges of copy number changes (the 'breakpoints') are also relevant, particularly when considering broad gains or losses where it can be difficult to determine the 'target' of the aberration. Hypothetically, an intact gene in the middle of a broad single copy loss can respond to feedback mechanisms, become more transcriptionally active, and compensate for the deletion of one allele. However, a breakpoint gene (i.e. not intact) is more akin to a heterozygous mutation, with upregulating feedback mechanisms potentially resulting in an admixture of both wild-type and mutant (broken) transcripts. Therefore, a breakpoint gene may be more likely to result in haploinsufficiency, or exert a dominant-negative effect. Indeed, breakpoint genes derived from aCGH screens of prostate tumours were significantly enriched with tumour suppressor genes, including p53, PTEN, BRCA1 and BRCA2.[30,31] Similar to copy number alterations themselves, the recurrence rates of specific genes were relatively low (highest $<10\%$ in Mao *et al.* study[30]), albeit with the proviso that a screen based on aCGH data alone is blind to copy number neutral rearrangements. Given that prostate tumour genomes harbour tens to hundreds of genome rearrangements,[32] DNA breakpoint genes should assume high priority when investigating mutational mechanisms.

Private fusion genes are tools for personalized oncology

Rearrangements can also generate fusion genes, the consequences of which include the disruption of the normal function of one or both

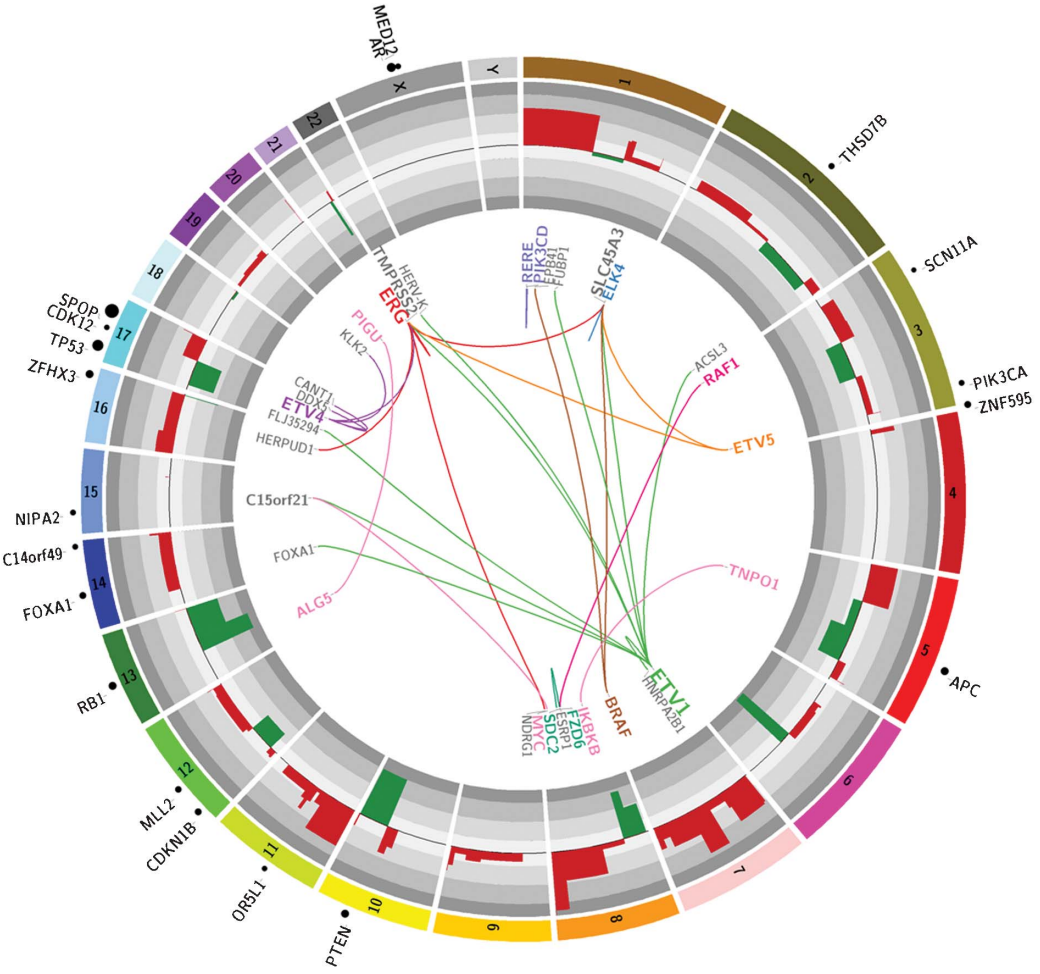

Figure 1 Significantly recurrent molecular alterations in prostate cancer. Circos plot showing the 90 significantly aberrant copy number aberrations from an analysis of 372 prostate tumours (green=loss; red=gain).[21] Significantly mutated genes from studies of large tumour cohorts are annotated by black dots around the outside, with dot size proportional to the level of significance.[29,55] Recurrent fusion genes involving ETS transcription factors are shown in the centre of the plot, as are recently identified non-ETS fusions genes.[36,40–43]

partners (e.g. PTEN[33]), the upregulation of an oncogene (e.g. ERG[34]), or the generation of a novel or truncated protein. Advances in methodology for transcriptome sequencing data analysis now permit resolution of a wide spectrum of fusion transcripts including those expressed at relatively low levels.[35] Prostate tumours frequently express fusion transcripts involving ETS transcription factors (**Figure 1**), the most common being TMPRSS2-ERG (reviewed by Rubin *et al.*[36] and Clark *et al.*[37]). However, links to prognosis are conflicting, and although ETS fusions are integral to prostate cancer biology and potentially both diagnostically useful (e.g. urine TMPRSS2-ERG detection[38]) and therapeutically exploitable (e.g. PARP1 inhibition[39]), focus therein alone masks the enormous number of 'private' fusion genes that are unique to individual tumours.

We and others have reported high numbers of non-ETS fusion genes.[40–43] Given that DNA breakpoints and rearrangements occur preferentially in transcriptionally active regions of the genome,[44–46] the fusion gene profile of an individual tumour can be a window into gene expression history, and therefore disease etiology. For example NEPC frequently harbours the *TMPRSS2-ERG* genome rearrangement (indicating the adenocarcinoma origins of NEPC), but can also accrue rearrangements involving neuronal-specific genes.[7,41] Private fusion transcripts are likely to be highly relevant for personalized oncology, and the recent discovery that a small fraction of prostate genomes harbour rearrangements in the RAF kinase pathway (e.g. *RAF1* and *BRAF* fusion genes; **Figure 1**) offers hope that fusion genes may offer precision targets.[47] Furthermore, their detection will aid molecular pathology, as highlighted by our identification of a subclinical metastasis in a patient's histologically benign lymph node.[41] In this patient, transcriptome sequencing allowed concurrent detection of identical fusion transcripts in both the lymph node and the primary tumour. Furthermore, the expression of one particular fusion gene (*FZD6-SDC2*) was markedly enriched in the lymph node metastasis and may have been responsible for some of the clone's metastatic properties. Nevertheless, since rearrangement is a major class of mutation in prostate cancer most fusion genes will represent tumour suppressor mutation or simply passenger events.

It was recently observed that balanced chromosomal translocations (without loss of chromosomal material loss) can occur in chains, forming closed loops of fusion genes which are presumably created more or less simultaneously.[32] The first chains identified involved *TMPRSS2-ERG* but they can also occur in non-ETS tumours. For example, we identified a closed chain of 4 fusion events including a

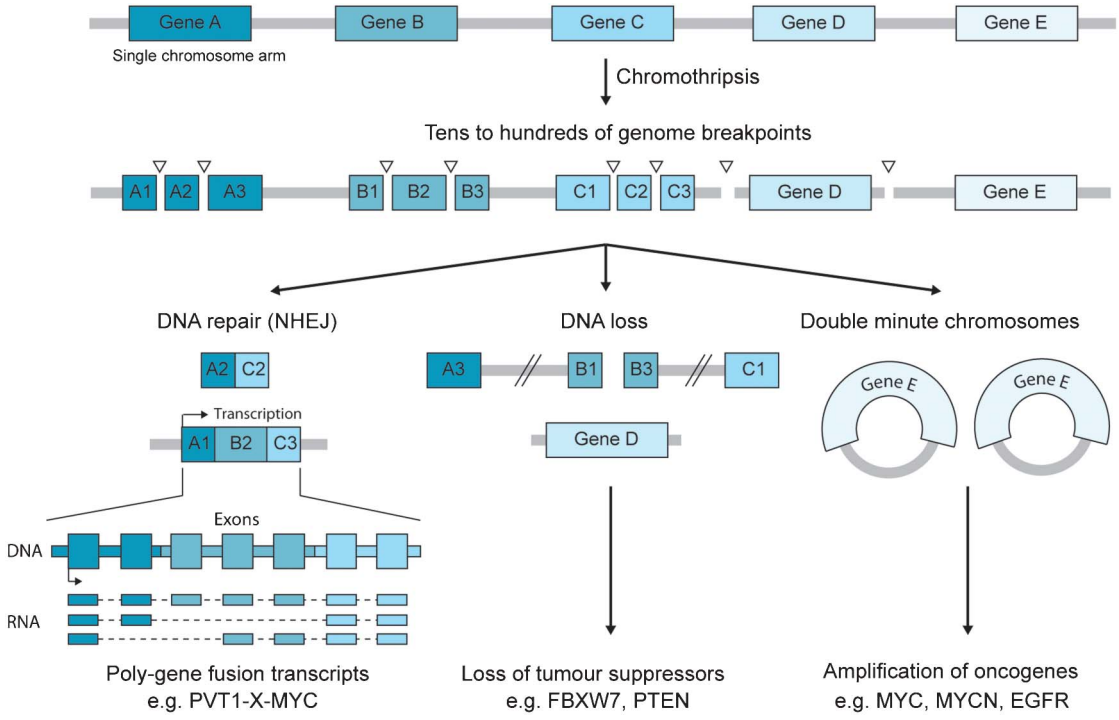

Figure 2 Chromothripsis generates complex genomic rearrangements which can be oncogenic. During chromothripsis tens to hundreds of breakpoints (indicated by arrowheads) can form in a single chromosome arm, generating fragments of DNA. These fragments are then incorrectly repaired through NHEJ, resulting in the fusion of multiple genes and the loss of genes including tumour suppressors. Double minute chromosomes containing oncogenes can also be formed and subsequently amplified. Transcription across reconstituted fragments of genes can result in poly-gene fusion transcripts, which may be a transcriptomic signature of chromothripsis. NHEJ, non-homologous end joining.

driver *C15orf21-MYC* fusion.[35,42] Although individual chains are likely to be unique, understanding the nature of their underlying genetic signature, including the average number of genes in a chain and homology of breakpoints will help understand their genesis.[48] It is tempting to speculate that they occur early in tumour development (especially given they often involve *TMPRSS2-ERG*, which is associated with early tumourgenesis[37]), before significant damage to DNA repair pathways have been accumulated. Indeed, they may represent a significant mutational mechanism in the minority of tumours exhibiting few copy number alterations.

Chromothripsis and poly-gene fusion transcripts

Genome rearrangements are frequently not balanced and can result in loss or gain of genetic material (overtly manifesting as copy number aberration). It was recently been reported that tens to hundreds of unbalanced genome rearrangements can occur in a single cell cycle.[49] This phenomenon is known as chromothripsis (or chromoangenesis[50]) and is the equivalent to the shattering and reassembly of one or more chromosome arms. Although it is a relatively rare phenomenon (estimated 2%–3% pan-cancer frequency), chromothripsis can promote the development of cancer through simultaneous deletion of tumour suppressors and creation of oncogenic rearrangements (e.g. involving MYCN or EGFR).[21,49] We recently reported the first cases of chromothripsis in prostate cancer[41,43] and demonstrated that the complex unbalanced rearrangements generated chains of genomic fragments comprised of small 'shards' from across the affected chromosome(s). Transcription across these chains resulted in 'poly-gene' fusion transcripts: transcripts containing genetic material from >2 genes (**Figure 2**). Furthermore, *de novo* detection of a poly-gene fusion transcript expressed by the LNCaP cell line suggests that this form of complexity in the transcriptome may be wide spread. Although we were not able to assign oncogenic potential to any of our detected transcripts, a more recent study identified a chromothripsis-driven subtype of medulloblastoma that recurrently expressed driver poly-gene fusion transcripts involving PVT1 and MYC.[51]

In a comprehensive survey using published aCGH data of 17 different cancers, prostate cancer had the highest incidence of chromothripsis, at 5.6%.[21] However, although reports have linked chromothripsis in some tumour types to poor patient outcome (e.g. multiple myeloma, acute myeloid leukemia, neuroblastoma[52–54]), the clinical implications of chromothripsis in prostate cancer are far from clear and should be addressed in future studies of large patient cohorts. It has been proposed that deregulation of DNA repair mechanisms is a major contributing factor for chromothripsis[50] and this appeared to be likely in at least one of the prostate tumours we examined, which had multiple mutations in the TP53 pathway. Although chromothripsis etiology is likely to be heterogeneous, if there is a unifying inability to repair DNA breaks, then an opportunity for rational therapy design may exist. Furthermore, the significant number of passenger events and massive genome aberrations may also provide exploitable weaknesses.

MUTATIONS

In context of the dominant role of genome rearrangement in prostate cancer, and the protracted natural history of the disease, it has traditionally been challenging to define the role mutations play in tumour development. However, the sequencing of hundreds of

tumour genomes and exomes in the last two years has shed considerable light on the complex and heterogeneous mutational landscape of prostate cancer.[29,32,55–59] Although several genes are recurrently mutated (reviewed here[60,61]), individual gene mutation rates are low (**Figure 1**). For prominent tumour suppressors (e.g. *TP53*, *PTEN*, *CDKN1B*, *RB1* and *ZFHX3*) the frequency of mutational disruption appears inferior to that of genomic loss through rearrangements. Nevertheless, genes that physically interact with the AR (e.g. *FOXA1*) are mutated in both primary tumours and CRPC, although direct mutation of the AR appears confined to the latter.[29,58] Furthermore, CRPC harbours recurrent mutations in the WNT signalling pathway (e.g. *APC*, *CTNNB1*); in chromatin and histone modification genes (including histone methyltransferases of the MLL family); and in several polycomb group genes (e.g. *ASXL2*).

SPOP mutations define a distinct subtype of prostate tumours

Although several studies have linked SPOP mutations to prostate cancer,[32,55] Barbieri *et al.*[58] recently reported that SPOP mutations define a class of ETS-fusion-negative prostate tumours. The SPOP protein forms part of an E3 ubiquitin ligase complex involved in transcriptional regulation is mutated in 6%–15% prostate tumours.[58] Interestingly, SPOP and other ubiquitin ligase complex genes (including FBXW7, which is deleted in ~4% of prostate tumours[21]) are also recurrently mutated in serous endometrial tumours.[62] In both prostate and serous endometrial tumours, SPOP mutations exclusively affect highly conserved amino acids within the MATH substrate recognition domain, but assigning downstream function to these mutations is challenging, especially since SPOP copy number aberration is rare in prostate cancer. However, in serous endometrial tumours a known SPOP substrate called NCOA3 can be oncogenic when amplified, potentially indicating SPOP loss of function as a dominant disease mechanism.

Prostate tumours with SPOP mutations harboured a distinct copy number profile, demonstrating enrichment for deletions of 5q21 and 6q21.[58] The chromatin remodelling factor CHD1 is located at 5q21, and rearrangements and mutations in CHD1 are also significantly associated with ETS-fusion negative tumours.[29] Although there is likely to be an overlap between SPOP and CHD1 disruption, their combined status can define a substantial fraction of ETS-fusion-negative tumours.

TRANSCRIPTOMIC COMPLEXITY

Unsurprisingly, given the genetic diversity of prostate tumours, deconstruction of transcriptomic complexity across different tumours has proven challenging. Stratifying cohorts of tumours by their gene expression profiles has had limited success,[63–67] perhaps most notably that tumours expressing a stem-cell like signature and exhibiting deregulation of TP53, PTEN and MYC, have a poor prognosis.[68] However, results have not proven sufficiently robust to affect clinical practice. Certain molecular subtypes, such as tumours with ETS rearrangements, or those overexpressing SPINK1, can be resolved through gene expression signatures alone, but do not reproducibly associate with patient outcome.

Transcriptome sequencing provides insight into mechanisms of disease

The wealth of detailed and accurate information afforded by deep transcriptome sequencing will be critical for further elucidation of transcriptome complexity and development of novel therapeutic strategies. In a notable example of the latter, transcriptome sequencing of NEPC coupled with *in vivo* and *in vitro* experiments implicated the MYCN and AURKA genes in disease development and demonstrated that inhibition of AURKA in NEPC may prove efficacious.[10] As primary treatment strategies for adenocarcinoma converge to force AR extinction, it is expected that the incidence of NEPC will increase, creating renewed urgency for targeted therapeutics. More recently, also using transcriptome sequencing of prostate tumours, we discovered that downregulation of REST, a key transcriptional repressor, results in upregulation of a spectrum of neuroendocrine genes (including CHGA and SYP). Further evidence for the relevance of REST in NEPC development can be observed in a cohort of 50 CRPC samples where the only tumour to exhibit a homozygous deletion of REST is also the only tumour with concurrent serum-PSA of 0 and histological neuroendocrine features.[29] Beyond the neuroendocrine component of prostate tumours, transcriptome sequencing also allows characterisation of the relative contribution of stromal and basal cells, and infiltration of lymphocytes.[41] In the context of elegant work by Sun *et al.*,[69] illustrating the role the prostate microenvironment 'secretome' plays in enhancing the therapy resistance of tumour cells, it will be particularly important to fully define the stromal and immune compartments of individual tumours.

Over the coming years, we expect that deep transcriptome sequencing of extreme phenotypes, particularly in context of their genetic landscapes, will define rare phenotypes of prostate cancer. Recently, we used transcriptome sequencing to identify a novel form of aggressive prostate cancer in a 46-year-old patient with primary and metastatic tumours.[42] His tumours exhibited a dual gene expression pattern associated with both AR-positive adenocarcinoma and AR-negative NEPC. This duality was shared by expressed fusion genes which, for example, involved the androgen-regulated *C15orf21* as well as neuronal-associated NTNG2. Experience with advanced, heavily treated tumours proposed the relatively simple explanation of a tumour with partial neuroendocrine differentiation. However, the patient was hormone-naive, and detailed histological examination of the entire prostate tumour and two lymph node metastases revealed a uniform cell type, with each cell exhibiting protein expression of both AR and CHGA. Furthermore, we observed remarkably high conservation of genome copy number profiles across five independent sites with the primary tumour and the metastases. Several lines of evidence suggest prostatic luminal, basal and neuroendocrine cells share a common ancestor,[70–72] yielding speculation that our hybrid tumour may have originated from a progenitor-like cell (**Figure 3**). Furthermore, we identified high amplification and overexpression of MSI2, a gene required to maintain stem cell identity, high levels of which in chronic myelogeneous leukaemia result in the blast crisis phase. It is possible that MSI2 contributed to the apparent frozen state of the hybrid adenocarcinoma-neuroendocrine tumour cells (uniformity across primary and metastatic tumours), while an MYC fusion gene (the first reported in prostate cancer) drove tumour aggressiveness.[42] This tumour is a clear example of a seemingly typical adenocarcinoma, which under the spotlight of transcriptome sequencing yielded unusual results. It is probable that other uncharacterized phenotypes exist, each with specific ramifications for personalized disease management.

Splicing, non-coding RNA and epigenetic modification magnifies diversity

The expression of genes is just one element of a transcriptome where complexity is significantly amplified by alternative splicing, non-coding RNAs, microRNAs and distinct epigenetic regulatory mechanisms.

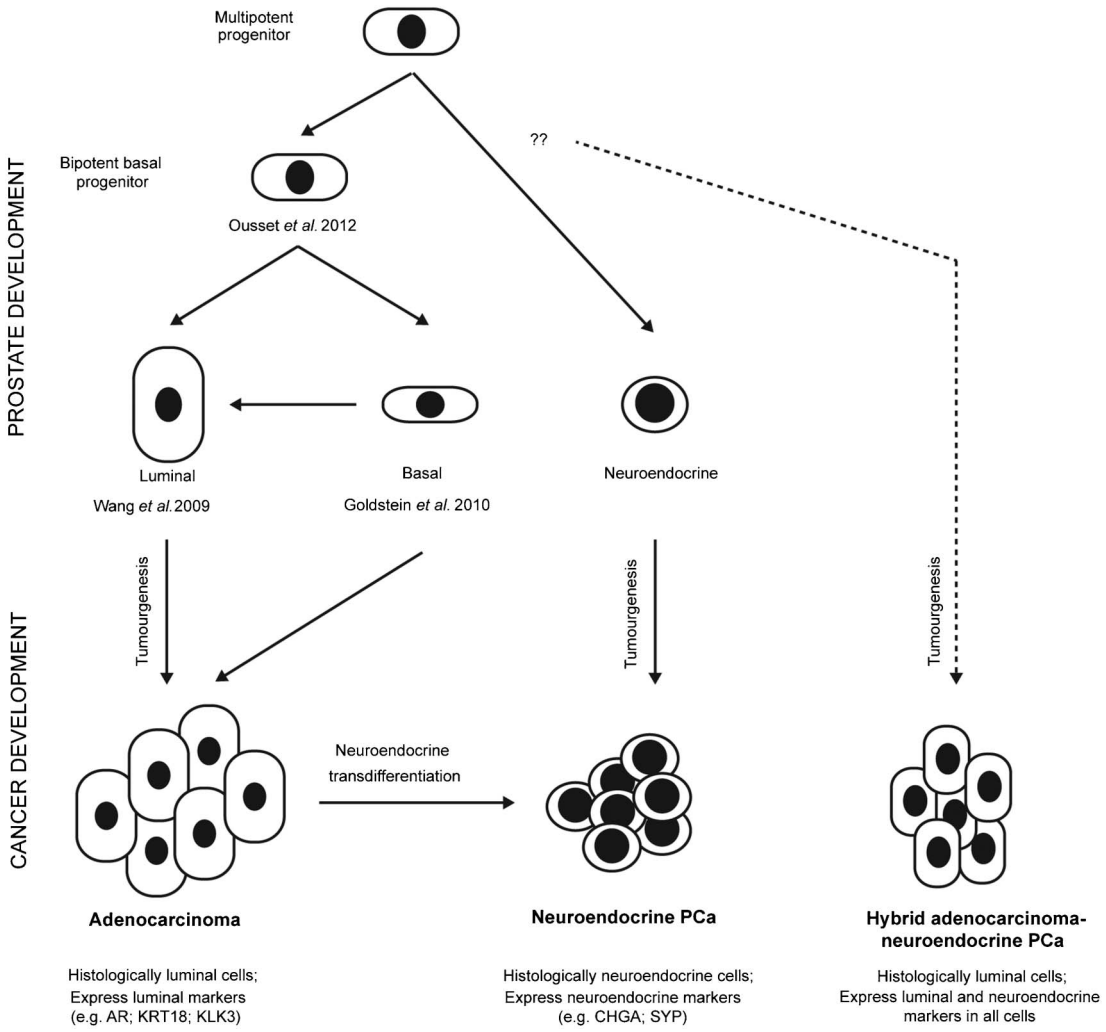

Figure 3 The hybrid adenocarcinoma-neuroendocrine prostate tumour hypothesis. A growing body of evidence suggests that the differentiated cells of the adult prostate (luminal, basal and neuroendocrine) share a common progenitor (top panel). It is traditionally thought that adenocarcinoma arises directly from mutation of luminal cells, although basal cell-of-origin models also exist (bottom panel). Neuroendocrine tumours can arise directly from mutation of neuroendocrine cells, or *via* a transdifferentiation mechanism from adenocarcinoma, induced by, e.g. androgen deprivation therapies. However, the novel hybrid adenocarcinoma-neuroendocrine tumour we identified is difficult to reconcile with any of these observations leading to the hypothesis that it arose from a progenitor-like cell that possesses properties of both luminal and neuroendocrine cells. PCa, prostate cancer.

In prostate cancer, splicing has only been extensively explored either in the context of splice site mutations or truncated variants of the AR which mediate anti-androgen resistance in CRPC.[73] However, compelling work by Scott Dehm and colleagues showed that the expression of AR variants in several CRPC models is linked to genomic rearrangements involving the AR loci.[74] Patient studies are now required to address the possibility that truncated AR variants are most biologically relevant in the context of genomic alterations, rather than aberrant splicing machinery. Future alternative splicing research in prostate tumours should draw lessons from lung cancer where targeted analyses revealed recurrent splicing events in oncogenes MET and RAC1, particularly relevant in the case of the latter where differential isoform usage associates with drug sensitivity.[75,76]

Early studies of non-coding RNA in prostate tumours have shown great promise with the identification of two long non-coding RNAs (PCAT-1, PCAT-2) whose expression was sufficient to stratify patients into molecular subtypes.[77] Furthermore, PCAT-1 is a target of the Polycomb Repressive Complex-2, suggesting cross-talk with epigenetic regulation: another burgeoning area of prostate cancer research, as different tumours exhibit distinct patterns of methylation.[78,79] MicroRNA networks have been shown in separate studies to regulate both Polycomb Repressive Complexes and PTEN expression in prostate cancer.[80,81] To date, there have been no detailed studies into the role of RNA editing in prostate cancer, but data from ENCODE suggests we should expect additional complexity.[82]

CONCLUSIONS

Several years of next-generation sequencing has unambiguously demonstrated that the clinical heterogeneity of prostate cancer is underlined by massive molecular heterogeneity. However, continuing to define individual molecular landscapes is imperative, if we are to decode the patient to patient variability which ultimately defines clinical outcome. In particular, we must strive to understand the transcriptional consequences of differing combinations of somatic alterations. In order to do this, there must be a focussed development of computational tools for integrated data analysis, in parallel with a

solution for the dearth of accurate model systems in which to assess therapeutic strategies. With respect to the next-generation of model systems, transplantable high-fidelity patient-derived xenografts appear increasingly promising, especially for personalized oncology applications. Finally, we believe that given the heterogeneity of prostate cancer, future studies that focus purely on recurrent molecular alterations will risk overlooking unique but biologically insightful events.

COMPETING FINANCIAL INTERESTS

The authors declare that they have no competing interests.

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