Case Report

Genomic Integration and Molecular Dysregulation in Aggressive Stage IV Bladder Cancer Following COVID-19 mRNA Vaccination

John A. Catanzaro[1], Nicolas Hulscher[2], and Peter A. McCullough[2],

[1] Neo7Bioscience; john.catanzaro@neo7bioscience.com
[2] McCullough Foundation; nichulscher@gmail.com
* Correspondence: john.catanzaro@neo7bioscience.com

Abstract:

Background: Bladder cancer is rare in young women, and advanced presentations are exceptionally uncommon. We report a de-identified case of a previously healthy 31-year-old female who developed rapidly progressive stage IV bladder cancer within 12 months of completing a three-dose Moderna mRNA vaccination series (May 2021, June 2021, December 2021).

Case Findings: Comprehensive multi-omic profiling was performed using PBIMA (Molecular Surveillance and Individualized Targeted Immunotherapy Peptide Editing) and REViSS (Spike-associated Transcriptional/Translational Instability Surveillance), incorporating analyses of plasma-derived circulating tumor DNA, whole-blood RNA, and urine exosome proteomics. Dysregulated gene expressions were identified across oncogenic driver genes (KRAS, ATM, MAPK1, NRAS, CHD4, PIK3CA, and SF3B1), auxiliary tumor-promoting signals (TOP1, PSIP1, and ERBB2), and broad evidence of genome instability with impaired DNA repair (ATM, MSH2). Within circulating tumor DNA, a host–vector chimeric read mapped to chr19:55,482,637–55,482,674 (GRCh38), in cytoband 19q13.42, positioned ~367 kb downstream of the canonical AAVS1 safe harbor and ~158 kb upstream of ZNF580 at the proximal edge of the zinc-finger (ZNF) gene cluster. This sequence aligned with perfect 20/20 bp identity to a segment (bases 5905–5924) within the Spike open reading frame (ORF) coding region (bases 3674–7480) of the Pfizer BNT162b2 DNA plasmid reference (GenBank accession OR134577.1), despite the patient only receiving Moderna vaccinations. This apparent paradox is best explained by shared Spike ORF sequences within the expression cassette across both vaccine platforms; because Moderna's proprietary plasmid sequence has not been deposited in NCBI, BLAST defaults to Pfizer's published reference as the nearest available match. The integration site was located outside the canonical AAVS1 "safe harbor" and within a gene-dense, recombination-prone regulatory region, raising concern for transcriptional disruption, fusion transcript formation, and oncogenic potential. The probability of a random 20-base sequence perfectly matching a predefined target is approximately 1 in a trillion, making this alignment statistically compelling and highly unlikely to be an incidental artifact.

Conclusions: This sentinel case report provides the first documented evidence of genomic integration of mRNA vaccine-derived genetic material in a human subject, documenting a temporal association between COVID-19 mRNA vaccination and aggressive malignancy, reproducible multi-omic evidence of oncogenic signaling, and a non–safe harbor host–vector integration event. While causality cannot be established from a single case, the convergence of (i) close temporal proximity to vaccination, (ii) genomic integration of a vaccine plasmid–derived spike gene fragment, and (iii) consistent transcriptomic and proteomic instability across biospecimens represents a highly unusual and biologically plausible pattern. These findings highlight an urgent need for systematic genomic surveillance, orthogonal validation with long-read sequencing, and larger cohort studies to rigorously define the impact of synthetic mRNA vaccine platforms on genome integrity and cancer risk.

1. Introduction

Bladder cancer is predominantly a disease of older adults, and its occurrence in young women is exceptionally uncommon[1-8]. When advanced disease is present in this demographic, it is typically aggressive and carries an unfavorable prognosis[9-17]. Such atypical presentations demand close examination of potential molecular drivers and external contributors[18-25].

Synthetic mRNA vaccines introduce heavily modified RNA molecules and lipid nanoparticle carriers that differ fundamentally from endogenous cellular transcripts. Residual plasmid DNA, stabilizing nucleotide analogs, and potential for reverse transcription raise concerns about genomic disruption, transcriptional dysregulation, and oncogenic activation[26-29]. Reports of plasmid DNA contamination, enhancer sequences, and persistence of spike protein expression have intensified scrutiny regarding the role of mRNA vaccines in genomic instability and malignant transformation[30,31].

We describe a 31-year-old previously healthy woman who developed rapidly progressive stage IV bladder cancer within 12 months of completing a three-dose Moderna mRNA vaccination series (May 2021, June 2021, December 2021). Given the rarity of advanced bladder cancer in this demographic, her case warranted in-depth molecular investigation. Multi-omic profiling identified reproducible oncogenic driver activation, DNA repair impairment, transcriptomic instability, and evidence of a vaccine-derived sequence integrated outside a genomic safe harbor[26,32]. This case illustrates a potential association between synthetic mRNA vaccination and the development of aggressive malignancy, underscoring the urgent need for systematic genomic surveillance and independent investigation of the long-term oncogenic risks associated with mRNA vaccine platforms.

2. Case Presentation

A previously healthy 31-year-old female received three Moderna mRNA vaccinations (May 2021, June 2021, December 2021) and, within 12 months, was diagnosed with rapidly progressive stage IV bladder cancer—an unusual and aggressive presentation for this age. The patient underwent comprehensive multi-omic profiling using PBIMA (Molecular Surveillance and Individualized Targeted Immunotherapy Peptide Editing) and REViSS (Spike-associated Transcriptional/Translational Instability Surveillance). Analyses incorporated circulating tumor DNA sequencing from plasma, whole-blood RNA transcriptomics, and urine exosome proteomics, enabling a multidimensional evaluation of genomic and transcriptional dysregulation.

Multi-omic analyses revealed dysregulated oncogenic drivers (KRAS, ATM, MAPK1, NRAS, CHD4, PIK3CA, SF3B1) and auxiliary tumor-promoting pathways (TOP1, PSIP1, ERBB2) across circulating tumor DNA, blood RNA, and urine exosome proteomics[11,33-36]. DNA repair deficiencies (ATM, MSH2) were also identified, consistent with enhanced susceptibility to genomic instability. The specific oncogenic drivers, auxiliary promoters, and DNA repair deficiencies identified through PBIMA multi-omic profiling are summarized in **Table 1**, highlighting their biospecimen presence and functional consequences.

Gene Target	Classification	Biospecimen Presence	Functional Implications
KRAS	Oncogenic Driver	ctDNA, blood RNA, urine	Constitutive RAS–RAF–MEK–ERK activation → uncontrolled proliferation, therapy resistance
ATM	Oncogenic Driver & DNA Repair	ctDNA, blood RNA, urine	Impaired DNA double-strand break recognition and checkpoint control → genomic instability
MAPK1	Oncogenic Driver	ctDNA, blood RNA, urine	MAPK effector hyperactivation → invasion, survival signaling
NRAS	Oncogenic Driver	ctDNA, blood RNA, urine	MAPK/PI3K signaling amplification → RAF1 dependence, malignant transformation
SF3B1	Oncogenic Driver	ctDNA, blood RNA	Spliceosome disruption → aberrant splicing, transcriptomic remodeling
CHD4	Oncogenic Driver	ctDNA, blood RNA, urine	NuRD chromatin remodeling dysfunction → impaired DNA repair, angiogenesis, immune evasion
PIK3CA	Oncogenic Driver	ctDNA, blood RNA	PI3K–AKT pathway activation → metabolic rewiring, angiogenesis, immune suppression
TOP1	Auxiliary Promoter	ctDNA, blood RNA, urine (variable)	DNA topology dysregulation → replicative stress, transcriptional collapse
PSIP1	Auxiliary Promoter	ctDNA, blood RNA, urine (variable)	Chromatin co-activator upregulation → immune suppression, angiogenesis
ERBB2 (HER2)	Auxiliary Promoter	ctDNA, blood RNA, urine	Receptor tyrosine kinase amplification → proliferative signaling, invasion, epigenetic reprogramming
MSH2	DNA Repair Gene	ctDNA, blood RNA	Mismatch repair deficiency → microsatellite instability, mutational burden

Table 1. Dysregulated Oncogenic Drivers and Auxiliary Tumor-Promoting Pathways Identified by PBIMA Multi-Omic Profiling

A particularly striking finding was the detection of a host–vector chimeric read mapping to chr19:55,482,637–55,482,674 (GRCh38), within cytogenetic band 19q13.42, located ~367 kb downstream of the canonical AAVS1 safe harbor locus and ~158 kb upstream of ZNF580 at the proximal edge of the zinc-finger (ZNF) gene cluster[37–39]. This sequence aligned with 100% identity to a segment (bases 5905 – 5924) within the Spike Open Reading Frame (ORF) coding region (bases 3674 – 7480) of the Pfizer BNT162b2 DNA plasmid reference (GenBank accession OR134577.1), despite the patient only receiving Moderna vaccinations. The probability of a random 20-base sequence perfectly matching a predefined target is approximately 1 in a trillion, making this alignment statistically compelling and highly unlikely to be an incidental artifact. The integration site was located outside the canonical AAVS1 "safe harbor" and within a gene-dense, recombination-prone regulatory region, raising concern for transcriptional disruption, fusion transcript formation, and oncogenic potential[40,41].

Notably, the integrated fragment corresponded to a 20-nucleotide segment within the Spike ORF coding region of the engineered expression cassette. The apparent paradox is explained by (i) conservation of the Spike ORF across vaccine platforms and (ii) the absence of a deposited Moderna plasmid reference, which defaults BLAST alignment to Pfizer's published sequence. Thus, the event represents a vaccine-derived Spike cassette fragment integrated into chr19q13.42, at the proximal edge of the ZNF cluster, with plausible regulatory impact. Supporting evidence is summarized in **Table 2**, which documents the mapping parameters, alignment identity, genomic context, and functional implications of this integration event.

Parameter	Result
Sample Description	Aggressive bladder cancer (post–mRNA vaccination)
Vaccination History	Moderna (3 doses: May 2021, June 2021, December 2021)
Host Mapping	Human chromosome 19, positions 55,482,637 – 55,482,674 (GRCh38), cytoband 19q13.42
Viral Reference	OR134577.1 (Pfizer BNT162b2 expression vector)
Viral Alignment Range	Bases 5905 – 5924 (Spike ORF coding region of the engineered expression cassette)
Alignment Identity	20/20 bp (100%)
Overlap Type	Chimeric, "gap" alignment
Orientation	Host: forward (+); Viral: plus, orientation
Mapping Quality (MQ)	Host: 60 (high confidence), Viral: 6 (low confidence)
Edit Distance	Host: 4; Viral: 2; Total: 43
Integration Classification	Possible Host Translocation: TRUE; Vector Rearrangement: FALSE
Genomic Context	Located at 19q13.42, ~367 kb downstream of the AAVS1 safe harbor (chr19:55.09–55.12 Mb) and ~158 kb upstream of ZNF580.
Functional Relevance	Gene-dense, recombination-prone, transcriptionally active regulatory region; potential for transcriptional disruption, fusion transcripts, and genomic instability

Table 2. DNA Frag analysis of host–vector translocation in a Moderna-vaccinated patient, showing a chimeric read at chr19:55,482,637–55,482,674 (GRCh38), cytoband 19q13.42, with 100% alignment to a segment of the Pfizer BNT162b2 plasmid reference (OR134577.1). The integration site lies ~367 kb downstream of the canonical AAVS1 safe harbor and ~158 kb upstream of ZNF580, placing it within a gene-dense, transcriptionally active, recombination-prone region and raising concern for transcriptional disruption, fusion transcript generation, and genomic instability.

3. Discussion

This case raises several mechanistic concerns regarding the potential role of synthetic mRNA vaccination in malignant transformation. To our knowledge, it represents the first documented evidence of genomic integration of vaccine-derived genetic material in a human subject. Specifically, we identified a vaccine vector–derived sequence integrated at chr19:55,482,637–55,482,674 (GRCh38), within cytoband 19q13.42, positioned ~367 kb downstream of the canonical AAVS1 "safe harbor" and ~158 kb upstream of ZNF580 at the proximal edge of the zinc-finger (ZNF) gene cluster, providing direct molecular evidence of host–vector genomic interaction [42–44]. The integration site lies within a gene-dense, transcriptionally active, recombination-prone regulatory region, a context associated with increased risk of transcriptional disruption, fusion transcript formation, and genomic instability [45].

Second, the paradoxical alignment of the integrated sequence to the Pfizer BNT162b2 plasmid, despite the patient having received only Moderna vaccinations, highlights the issue of cross-platform plasmid homology and manufacturing commonalities. The 100% identity to Pfizer's deposited vector sequence suggests the presence of shared plasmid backbone elements or spike-encoding motifs across vaccine platforms. Given the absence of a deposited Moderna plasmid reference, BLAST alignment defaulted to Pfizer's sequence as the nearest available match, further underscoring the need for independent disclosure and validation of proprietary vaccine constructs.

Third, several plausible mechanisms could underlie the observed integration event [26,46,47]. These include (i) persistence and carry-over of residual plasmid DNA fragments from the in vitro transcription template, (ii) reverse

transcription of spike mRNA followed by insertion at double-strand DNA breaks, (iii) misrepair via non-homologous end joining (NHEJ) or microhomology-mediated end joining (MMEJ), (iv) homologous recombination when stretches of sequence similarity exist, (v) LINE-1 retrotransposon activity generating cDNA intermediates, and (vi) topoisomerase-mediated mis-ligation during DNA unwinding. Each of these routes is biologically feasible and consistent with the concurrent detection of DNA repair deficiencies (ATM, MSH2) in this patient, which would increase susceptibility to insertional mutagenesis. Such vulnerabilities may accelerate oncogenic transformation and malignant progression. Supporting this concern, Speicher et al. quantified billions of residual plasmid DNA fragments per vaccine dose, with levels in both bivalent and XBB.1.5 Pfizer-BioNTech and Moderna COVID-19 vaccine products exceeding regulatory safety thresholds by 36–627-fold, thereby providing a plausible source of template DNA for persistent chromosomal integration [28].

In addition to the host–vector integration event, the patient's multi-omic profile revealed a constellation of dysregulated oncogenic drivers and auxiliary tumor-promoting signals that together create a permissive landscape for aggressive malignancy[29,48,49]. Activating alterations in KRAS and NRAS converge on the MAPK signaling cascade, sustaining proliferative signaling and bypassing normal growth controls[9,50,51]. Concurrent dysregulation of MAPK1 amplifies downstream ERK-driven transcriptional programs that promote cell cycle progression and survival[1-3,35,52,53]. The chromatin remodeler CHD4 and the RNA splicing factor SF3B1 contribute to epigenetic reprogramming and aberrant transcript processing, fostering cellular plasticity and tumor adaptability[4,54-59]. PIK3CA activation drives PI3K/AKT signaling, enhancing metabolic fitness, invasion, and resistance to apoptosis, while auxiliary signals from TOP1**,** PSIP1, and ERBB2 further reinforce replication stress tolerance, transcriptional activation, and growth factor responsiveness[7,10,12,17,60,61]. Importantly, deficiencies in ATM and MSH2 compromise DNA repair fidelity, predisposing to genomic instability, accumulation of mutations, and chromosomal rearrangements[17,22,25,62]. Together, this pattern of multi-pathway dysregulation provides a mechanistic basis for the patient's unusually rapid disease evolution, linking vaccine-associated genomic perturbations with a molecular environment primed for malignant transformation.

Emerging evidence supports a link between mRNA vaccination and oncogenesis [63, 64]. A population-wide 30-month cohort study of nearly 300,000 residents of Pescara, Italy, found that receipt of ≥1 COVID-19 vaccine dose was associated with a 23% increased risk of cancer hospitalization overall (HR 1.23, 95% CI 1.11–1.37), with the strongest and statistically significant excess risks observed for breast cancer (HR 1.54), bladder cancer (HR 1.62), and colorectal cancer (HR 1.35) [61]. Complementing this epidemiologic evidence, Marik et al. formally defined the syndrome of COVID-19 mRNA vaccine-induced "turbo cancers," compiling clinical case reports, epidemiologic signals, and mechanistic pathways through which the spike protein may accelerate malignant transformation via metabolic reprogramming, apoptosis resistance, angiogenesis, and immune dysregulation [62]. Taken together, these reports provide convergent epidemiologic and mechanistic support for our present case, in which genomic integration and molecular dysregulation were directly documented.

Together, these observations support a biologically plausible framework in which synthetic mRNA vaccine exposure contributes to genomic instability, oncogenic signaling, and aggressive disease evolution. While causality cannot be inferred from a single case, the convergence of temporal proximity, vector sequence integration, and reproducible multi-omic dysregulation provides a strong hypothesis-generating signal. This warrants urgent systematic genomic surveillance, orthogonal validation with long-read sequencing, and independent investigation of the long-term oncogenic risks associated with mRNA vaccine technologies. A schematic overview of the clinical course, host–vector integration, and multi-omic dysregulation is shown in **Figure 1.**

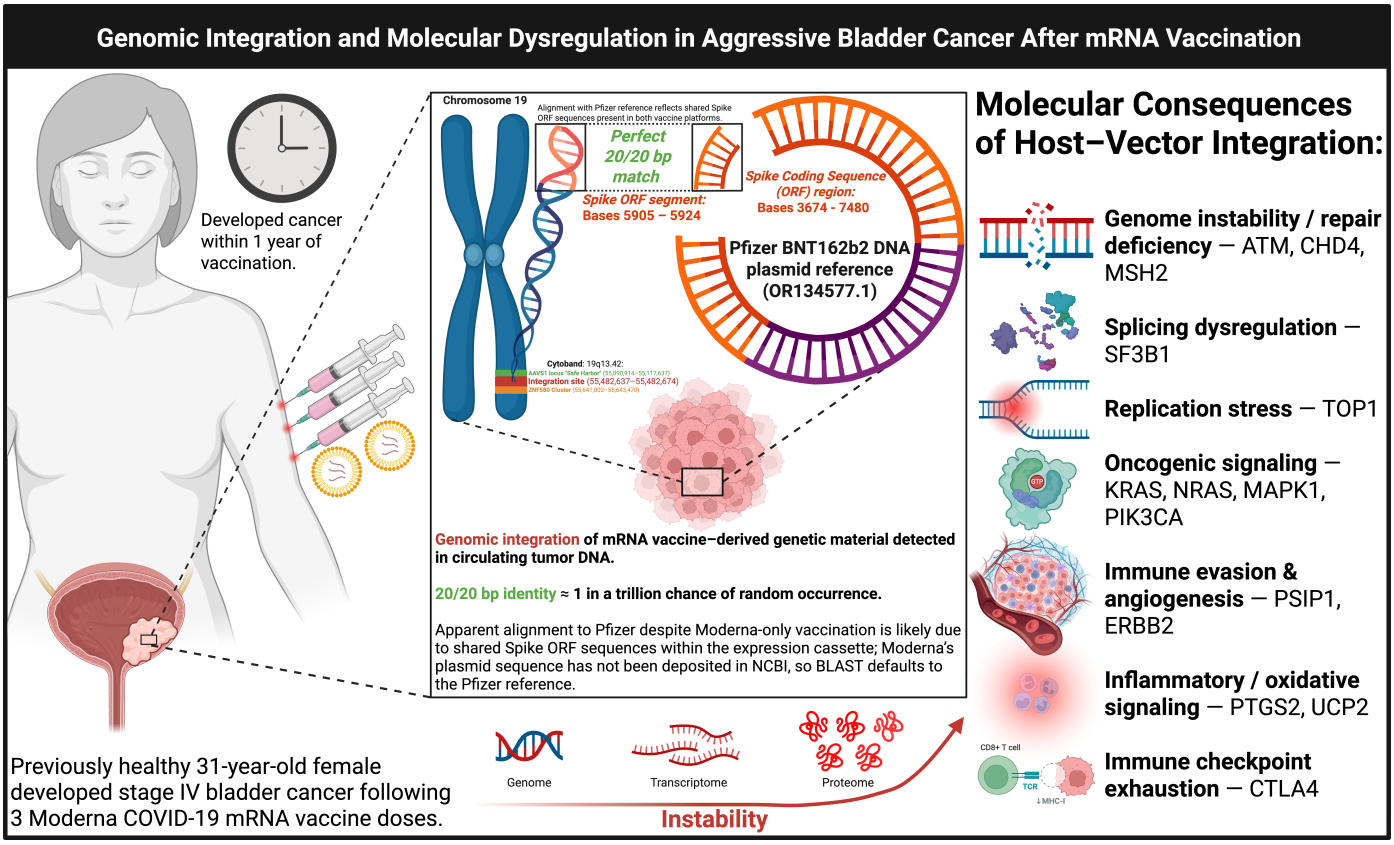

Figure 1. Genomic Integration and Molecular Dysregulation in Aggressive Bladder Cancer After mRNA Vaccination **Created with Biorender.com*

4. Conclusions

This sentinel case report describes the rapid onset of aggressive stage IV bladder cancer in a young woman after a Moderna mRNA vaccination series, marked by direct evidence of genomic integration of vaccine-derived genetic material. Multi-omic profiling revealed reproducible oncogenic driver activation, DNA repair deficiencies, transcriptomic instability, and integration of a vaccine vector–derived sequence outside a genomic safe harbor[8,30,31,33,38,39,41,58,65]. While causality cannot be established from a single case, the convergence of temporal proximity, integration evidence, and multi-system molecular disruption raises serious concern regarding the oncogenic potential of synthetic mRNA vaccine platforms. These findings highlight the urgent need for systematic genomic surveillance, independent validation using orthogonal sequencing methods, and rigorous investigation into the long-term genomic and oncologic risks of mRNA vaccination.

5. References

1. Bekele, R.; Samant, A.; Hanlon, T.; Mouw, K. Abstract 1709: MAPK pathway alterations are a targetable vulnerability in bladder cancer. *Cancer Research* **2023**, *83*, 1709-1709, doi:10.1158/1538-7445.am2023-1709.
2. Bekele, R.T.; Samant, A.S.; Nassar, A.H.; So, J.; Garcia, E.P.; Curran, C.R.; Hwang, J.H.; Mayhew, D.L.; Nag, A.; Thorner, A.R.; et al. RAF1 amplification drives a subset of bladder tumors and confers sensitivity to MAPK-directed therapeutics. *Journal of Clinical Investigation* **2021**, *131*, doi:10.1172/jci147849.
3. Cheng, G.; Zhou, Z.; Li, S.; Yang, S.; Wang, Y.; Ye, Z.; Ren, C. Predicting bladder cancer survival with high accuracy: insights from MAPK pathway-related genes. *Scientific Reports* **2024**, *14*, doi:10.1038/s41598-024-61302-0.
4. Columbres, R.C.; Chakraborty, A.; Apolo, A.; Banday, R. Abstract 5640: Unlocking the therapeutic potential of SF3B1 inhibitors for bladder cancer treatment. *Cancer Research* **2024**, *84*, 5640-5640, doi:10.1158/1538-7445.am2024-5640.
5. Dadhania, V.; Zhang, M.; Zhang, L.; Bondaruk, J.; Majewski, T.; Siefker-Radtke, A.; Guo, C.C.; Dinney, C.; Cogdell, D.E.; Zhang, S.; et al. Meta-Analysis of the Luminal and Basal Subtypes of Bladder Cancer and the Identification of Signature Immunohistochemical Markers for Clinical Use. *EBioMedicine* **2016**, *12*, 105-117, doi:10.1016/j.ebiom.2016.08.036.
6. Deng, Z.; Shen, D.; Yu, M.; Zhou, F.; Shan, D.; Fang, Y.; Jin, W.; Qian, K.; Li, S.; Wang, G.; et al. Pectolinarigenin inhibits bladder urothelial carcinoma cell proliferation by regulating DNA damage/autophagy pathways. *Cell Death Discovery* **2023**, *9*, doi:10.1038/s41420-023-01508-9.
7. Dueñas, M.; Martínez-Fernández, M.; García-Escudero, R.; Villacampa, F.; Marqués, M.; Saiz-Ladera, C.; Duarte, J.; Martínez, V.; Gómez, M.J.; Martín, M.L.; et al. PIK3CA gene alterations in bladder cancer are frequent and associate with reduced recurrence in non-muscle invasive tumors. *Molecular Carcinogenesis* **2013**, *54*, 566-576, doi:10.1002/mc.22125.
8. Hoffmann, M.J.; Schulz, W.A. Alterations of Chromatin Regulators in the Pathogenesis of Urinary Bladder Urothelial Carcinoma. *Cancers* **2021**, *13*, 6040, doi:10.3390/cancers13236040.
9. Hou, S.; Nieva, J.; Desai, N. KRAS G12C mutated NSCLC and bladder cancer xenografts treated with sotorasib and adagrasib in combination with mTOR inhibitors show improved antitumor activity of nab-sirolimus vs everolimus. *European Journal of Cancer* **2022**, *174*, S58-S59, doi:10.1016/S0959-8049(22)00957-1.
10. Kompier, L.C.; Lurkin, I.; van der Aa, M.N.M.; van Rhijn, B.W.G.; van der Kwast, T.H.; Zwarthoff, E.C. FGFR3, HRAS, KRAS, NRAS and PIK3CA Mutations in Bladder Cancer and Their Potential as Biomarkers for Surveillance and Therapy. *PLoS ONE* **2010**, *5*, e13821, doi:10.1371/journal.pone.0013821.
11. Lee, K.-H.; Ku, J.H. Targeting splicing factors as molecular non-muscle invasive bladder cancer predictors. *Translational Andrology and Urology* **2018**, *7*, S715-S717, doi:10.21037/tau.2018.10.09.
12. López-Knowles, E.; Hernández, S.; Malats, N.r.; Kogevinas, M.; Lloreta, J.; Carrato, A.; Tardón, A.; Serra, C.; Real, F.X. PIK3CA Mutations Are an Early Genetic Alteration Associated with FGFR3 Mutations in Superficial Papillary Bladder Tumors. *Cancer Research* **2006**, *66*, 7401-7404, doi:10.1158/0008-5472.can-06-1182.
13. Monnin, K.A.; Bronstein, I.B.; Gaffney, D.K.; Holden, J.A. Elevations of DNA topoisomerase I in transitional cell carcinoma of the urinary bladder: Correlation with DNA topoisomerase II-alpha and p53 expression. *Human Pathology* **1999**, *30*, 384-391, doi:10.1016/s0046-8177(99)90112-0.
14. Montero-Hidalgo, A.J.; Pérez-Gómez, J.M.; Martínez-Fuentes, A.J.; Gómez-Gómez, E.; Gahete, M.D.; Jiménez-Vacas, J.M.; Luque, R.M. Alternative splicing in bladder cancer: potential strategies for cancer diagnosis, prognosis, and treatment. *WIREs RNA* **2022**, *14*, doi:10.1002/wrna.1760.
15. Ouerhani, S.; Bougatef, K.; Soltani, I.; Elgaaied, A.B.A.; Abbes, S.; Menif, S. The prevalence and prognostic significance of KRAS mutation in bladder cancer, chronic myeloid leukemia and colorectal cancer. *Molecular Biology Reports* **2013**, *40*, 4109-4114, doi:10.1007/s11033-013-2512-8.
16. Ouerhani, S.; Elgaaied, A.B.A. The mutational spectrum of HRAS, KRAS, NRAS and FGFR3 genes in bladder cancer. *Cancer Biomarkers* **2012**, *10*, 259-266, doi:10.3233/cbm-2012-0254.

17. Pan, Y.-H.; Zhang, J.-X.; Chen, X.; Liu, F.; Cao, J.-Z.; Chen, Y.; Chen, W.; Luo, J.-H. Predictive Value of the TP53/PIK3CA/ATM Mutation Classifier for Patients With Bladder Cancer Responding to Immune Checkpoint Inhibitor Therapy. *Frontiers in Immunology* **2021**, *12*, doi:10.3389/fimmu.2021.643282.
18. Shuman, L.; Pham, J.; Wildermuth, T.; Wu, X.-R.; Walter, V.; Warrick, J.I.; DeGraff, D.J. Urothelium-Specific Expression of Mutationally Activated Pik3ca Initiates Early Lesions of Noninvasive Bladder Cancer. *The American Journal of Pathology* **2023**, *193*, 2133-2143, doi:10.1016/j.ajpath.2023.07.001.
19. Stenehjem, D.; Tran, D.; Nkrumah, M.; Gupta, S. PD1/PDL1 inhibitors for the treatment of advanced urothelial bladder cancer. *OncoTargets and Therapy* **2018**, *Volume 11*, 5973-5989, doi:10.2147/ott.s135157.
20. Wang, Z.; Shang, J.; Li, Z.; Li, H.; Zhang, C.; He, K.; Li, S.; Ju, W. PIK3CA Is Regulated by CUX1, Promotes Cell Growth and Metastasis in Bladder Cancer via Activating Epithelial-Mesenchymal Transition. *Frontiers in Oncology* **2020**, *10*, doi:10.3389/fonc.2020.536072.
21. Wu, T.-Z.; Lei, C.-Y.; Li, J.-M.; Guo, Y.-F. Sex discrepancy characterization revealed by somatic DNA alterations in muscle-invasive bladder carcinoma. *Chinese Medical Journal* **2019**, *132*, 2492-2494, doi:10.1097/cm9.0000000000000487.
22. Yi, R.; Lin, A.; Cao, M.; Xu, A.; Luo, P.; Zhang, J. ATM Mutations Benefit Bladder Cancer Patients Treated With Immune Checkpoint Inhibitors by Acting on the Tumor Immune Microenvironment. *Frontiers in Genetics* **2020**, *11*, doi:10.3389/fgene.2020.00933.
23. Zargar, P.; Koochakkhani, S.; Hassanzadeh, M.; Ashouri Taziani, Y.; Nasrollahi, H.; Eftekhar, E. Downregulation of topoisomerase 1 and 2 with acriflavine sensitizes bladder cancer cells to cisplatin-based chemotherapy. *Molecular Biology Reports* **2022**, *49*, 2755-2763, doi:10.1007/s11033-021-07087-1.
24. Zheng, Y.; Izumi, K.; Yao, J.L.; Miyamoto, H. Dihydrotestosterone upregulates the expression of epidermal growth factor receptor and ERBB2 in androgen receptor-positive bladder cancer cells. *Endocrine-Related Cancer* **2011**, *18*, 451-464, doi:10.1530/erc-11-0010.
25. Zhou, Y.; Börcsök, J.; Adib, E.; Kamran, S.C.; Neil, A.J.; Stawiski, K.; Freeman, D.; Stormoen, D.R.; Sztupinszki, Z.; Samant, A.; et al. ATM deficiency confers specific therapeutic vulnerabilities in bladder cancer. *Science Advances* **2023**, *9*, doi:10.1126/sciadv.adg2263.
26. Aldén, M.; Olofsson Falla, F.; Yang, D.; Barghouth, M.; Luan, C.; Rasmussen, M.; De Marinis, Y. Intracellular Reverse Transcription of Pfizer BioNTech COVID-19 mRNA Vaccine BNT162b2 In Vitro in Human Liver Cell Line. *Current Issues in Molecular Biology* **2022**, *44*, 1115-1126, doi:10.3390/cimb44030073.
27. Solomon, A.L.; Ratchford, E.V.; Armitage, K.B.; Kovacic, J.C. Vascular Disease Patient Information Page: Vascular considerations with COVID-19 vaccines. *Vascular Medicine* **2021**, *27*, 102-106, doi:10.1177/1358863X211066128.
28. Speicher, D.J.; Rose, J.; McKernan, K. Quantification of residual plasmid DNA and SV40 promoter-enhancer sequences in Pfizer/BioNTech and Moderna modRNA COVID-19 vaccines from Ontario, Canada. *Autoimmunity* **2025**, *58*, 2551517, doi:10.1080/08916934.2025.2551517.
29. von Ranke, N.; Zhang, W.; Anokin, P.; Hulscher, N.; McKernan, K.J.; McCullough, P.A.; Catanzaro, J.A. Synthetic mRNA Vaccines and Transcriptomic Dysregulation: Evidence from New-Onset Adverse Events and Cancers Post-Vaccination. *ResearchGate,* **2025.**https://www.researchgate.net/publication/394007467_Synthetic_mRNA_Vaccines_and_Transcriptomic_Dysregulation_Evidence_from_New-Onset_Adverse_Events_and_Cancers_Post-Vaccination
30. Li, Y.E.; Wang, S.; Reiter, R.J.; Ren, J. Clinical cardiovascular emergencies and the cellular basis of COVID-19 vaccination: from dream to reality? *International Journal of Infectious Diseases* **2022**, *124*, 1-10, doi:10.1016/j.ijid.2022.08.026.
31. Mohseni Afshar, Z.; Tavakoli Pirzaman, A.; Liang, J.J.; Sharma, A.; Pirzadeh, M.; Babazadeh, A.; Hashemi, E.; Deravi, N.; Abdi, S.; Allahgholipour, A.; et al. Do we miss rare adverse events induced by COVID-19 vaccination? *Frontiers in Medicine* **2022**, *Volume 9 - 2022*.

32. Bartolomucci, A.; Nobrega, M.; Ferrier, T.; Dickinson, K.; Kaorey, N.; Nadeau, A.; Castillo, A.; Burnier, J.V. Circulating tumor DNA to monitor treatment response in solid tumors and advance precision oncology. *npj Precision Oncology* **2025**, *9*, doi:10.1038/s41698-025-00876-y.
33. Hanahan, D.; Weinberg, Robert A. Hallmarks of Cancer: The Next Generation. *Cell* **2011**, *144*, 646-674, doi:10.1016/j.cell.2011.02.013.
34. Kim, H.; Park, K.U. Clinical Circulating Tumor DNA Testing for Precision Oncology. *Cancer Research and Treatment* **2023**, *55*, 351-366, doi:10.4143/crt.2022.1026.
35. Raghavan, S.; Kundumani-Sridharan, V.; Kumar, S.; White, C.W.; Das, K.C. Thioredoxin Prevents Loss of UCP2 in Hyperoxia via MKK4–p38 MAPK–PGC1α Signaling and Limits Oxygen Toxicity. *American Journal of Respiratory Cell and Molecular Biology* **2022**, *66*, 323-336, doi:10.1165/rcmb.2021-0219oc.
36. Torgovnick, A.; Schumacher, B. DNA repair mechanisms in cancer development and therapy. *Frontiers in Genetics* **2015**, *6*, doi:10.3389/fgene.2015.00157.
37. Cassandri, M.; Smirnov, A.; Novelli, F.; Pitolli, C.; Agostini, M.; Malewicz, M.; Melino, G.; Raschellà, G. Zinc-finger proteins in health and disease. *Cell Death Discovery* **2017**, *3*, doi:10.1038/cddiscovery.2017.71.
38. Kamaliyan, Z.; Clarke, T.L. Zinc finger proteins: guardians of genome stability. *Frontiers in Cell and Developmental Biology* **2024**, *12*, doi:10.3389/fcell.2024.1448789.
39. Li, X.; Han, M.; Zhang, H.; Liu, F.; Pan, Y.; Zhu, J.; Liao, Z.; Chen, X.; Zhang, B. Structures and biological functions of zinc finger proteins and their roles in hepatocellular carcinoma. *Biomarker Research* **2022**, *10*, doi:10.1186/s40364-021-00345-1.
40. Dutheil, N.; Yoon-Robarts, M.; Ward, P.; Henckaerts, E.; Skrabanek, L.; Berns, K.I.; Campagne, F.; Linden, R.M. Characterization of the Mouse Adeno-Associated Virus AAVS1 Ortholog. *Journal of Virology* **2004**, *78*, 8917-8921, doi:10.1128/jvi.78.16.8917-8921.2004.
41. Hamilton, H.; Gomos, J.; Berns, K.I.; Falck-Pedersen, E. Adeno-Associated Virus Site-Specific Integration and AAVS1 Disruption. *Journal of Virology* **2004**, *78*, 7874-7882, doi:10.1128/jvi.78.15.7874-7882.2004.
42. Cheng, Y.; Liang, P.; Geng, H.; Wang, Z.; Li, L.; Cheng, S.H.; Ying, J.; Su, X.; Ng, K.M.; Ng, M.H.L.; et al. A Novel 19q13 Nucleolar Zinc Finger Protein Suppresses Tumor Cell Growth through Inhibiting Ribosome Biogenesis and Inducing Apoptosis but Is Frequently Silenced in Multiple Carcinomas. *Molecular Cancer Research* **2012**, *10*, 925-936, doi:10.1158/1541-7786.mcr-11-0594.
43. Jen, J.; Wang, Y.-C. Zinc finger proteins in cancer progression. *Journal of Biomedical Science* **2016**, *23*, doi:10.1186/s12929-016-0269-9.
44. Petre, G.; Lorès, P.; Sartelet, H.; Truffot, A.; Poreau, B.; Brandeis, S.; Martinez, G.; Satre, V.; Harbuz, R.; Ray, P.F.; et al. Genomic duplication in the 19q13.42 imprinted region identified as a new genetic cause of intrauterine growth restriction. *Clinical Genetics* **2018**, *94*, 575-580, doi:10.1111/cge.13449.
45. Oceguera-Yanez, F.; Kim, S.-I.; Matsumoto, T.; Tan, G.W.; Xiang, L.; Hatani, T.; Kondo, T.; Ikeya, M.; Yoshida, Y.; Inoue, H.; et al. Engineering the AAVS1 locus for consistent and scalable transgene expression in human iPSCs and their differentiated derivatives. *Methods* **2016**, *101*, 43-55, doi:10.1016/j.ymeth.2015.12.012.
46. Chang HHY, Pannunzio NR, Adachi N, Lieber MR. Non-homologous DNA end joining and alternative pathways to double-strand break repair. *Nat Rev Mol Cell Biol*. 2017;18(8):495-506. doi:10.1038/nrm.2017.48
47. Huang R, Zhou PK. DNA damage repair: historical perspectives, mechanistic pathways and clinical translation for targeted cancer therapy. *Signal Transduct Target Ther*. 2021;6(1):254. Published 2021 Jul 9. doi:10.1038/s41392-021-00648-7
48. Schaafsma, E.; Fugle, C.M.; Wang, X.; Cheng, C.A.-O. Pan-cancer association of HLA gene expression with cancer prognosis and immunotherapy efficacy.
49. Szolek, A.; Schubert, B.; Mohr, C.; Sturm, M.; Feldhahn, M.; Kohlbacher, O. OptiType: precision HLA typing from next-generation sequencing data.

50. Bahar, M.E.; Kim, H.J.; Kim, D.R. Targeting the RAS/RAF/MAPK pathway for cancer therapy: from mechanism to clinical studies. *Signal Transduction and Targeted Therapy* **2023**, *8*, doi:10.1038/s41392-023-01705-z.
51. Kodaz, H. Frequency of RAS Mutations (KRAS, NRAS, HRAS) in Human Solid Cancer. *Eurasian Journal of Medicine and Oncology* **2017**, doi:10.14744/ejmo.2017.22931.
52. Braicu, C.; Buse, M.; Busuioc, C.; Drula, R.; Gulei, D.; Raduly, L.; Rusu, A.; Irimie, A.; Atanasov, A.G.; Slaby, O.; et al. A Comprehensive Review on MAPK: A Promising Therapeutic Target in Cancer. *Cancers* **2019**, *11*, 1618, doi:10.3390/cancers11101618.
53. Clark-Garvey, S.; Kim, W.Y. RAF1 amplification: an exemplar of MAPK pathway activation in urothelial carcinoma. *Journal of Clinical Investigation* **2021**, *131*, doi:10.1172/jci154095.
54. Novillo, A.; Fernández-Santander, A.; Gaibar, M.; Galán, M.; Romero-Lorca, A.; El Abdellaoui-Soussi, F.; Gómez-del Arco, P. Role of Chromodomain-Helicase-DNA-Binding Protein 4 (CHD4) in Breast Cancer. *Frontiers in Oncology* **2021**, *11*, doi:10.3389/fonc.2021.633233.
55. Wang, J.; Zhong, F.; Li, J.; Yue, H.; Li, W.; Lu, X. The epigenetic factor CHD4 contributes to metastasis by regulating the EZH2/β-catenin axis and acts as a therapeutic target in ovarian cancer. *Journal of Translational Medicine* **2023**, *21*, doi:10.1186/s12967-022-03854-1.
56. Liu, Z.; Yoshimi, A.; Wang, J.; Cho, H.; Chun-Wei Lee, S.; Ki, M.; Bitner, L.; Chu, T.; Shah, H.; Liu, B.; et al. Mutations in the RNA Splicing Factor SF3B1 Promote Tumorigenesis through MYC Stabilization. *Cancer Discovery* **2020**, *10*, 806-821, doi:10.1158/2159-8290.cd-19-1330.
57. Samy, A.; Ozdemir, M.K.; Alhajj, R. Studying the connection between SF3B1 and four types of cancer by analyzing networks constructed based on published research. *Scientific Reports* **2023**, *13*, doi:10.1038/s41598-023-29777-5.
58. Goswami, K.; Venkatachalam, K.; Singh, S.P.; Rao, C.V.; Madka, V. Chromatin Remodulator CHD4: A Potential Target for Cancer Interception. *Genes* **2025**, *16*, 225, doi:10.3390/genes16020225.
59. Xia, L.; Huang, W.; Bellani, M.; Seidman, M.M.; Wu, K.; Fan, D.; Nie, Y.; Cai, Y.; Zhang, Y.W.; Yu, L.-R.; et al. CHD4 Has Oncogenic Functions in Initiating and Maintaining Epigenetic Suppression of Multiple Tumor Suppressor Genes. *Cancer Cell* **2017**, *31*, 653-668.e657, doi:10.1016/j.ccell.2017.04.005.
60. Madsen, R.R.; Vanhaesebroeck, B.; Semple, R.K. Cancer-Associated PIK3CA Mutations in Overgrowth Disorders. *Trends in Molecular Medicine* **2018**, *24*, 856-870, doi:10.1016/j.molmed.2018.08.003.
61. Ross, R.L.; McPherson, H.R.; Kettlewell, L.; Shnyder, S.D.; Hurst, C.D.; Alder, O.; Knowles, M.A. PIK3CA dependence and sensitivity to therapeutic targeting in urothelial carcinoma. *BMC Cancer* **2016**, *16*, doi:10.1186/s12885-016-2570-0.
62. Lee, J.-H. Targeting the ATM pathway in cancer: Opportunities, challenges and personalized therapeutic strategies. *Cancer Treatment Reviews* **2024**, *129*, 102808, doi:10.1016/j.ctrv.2024.102808.
63. Acuti Martellucci, C.; Capodici, A.; Soldato, G.; et al. COVID-19 vaccination, all-cause mortality, and hospitalization for cancer: 30-month cohort study in an Italian province. *EXCLI Journal* 2025, 24, 690–707. doi:10.17179/excli2025-8400.
64. Marik, P.; Hope, J. COVID-19 mRNA-Induced "Turbo Cancers." *Journal of Independent Medicine* 2025, 1, 185–194. doi:10.71189/JIM/2025/V01N03A02.
65. Mills, A.A. The Chromodomain Helicase DNA-Binding Chromatin Remodelers: Family Traits that Protect from and Promote Cancer. *Cold Spring Harbor Perspectives in Medicine* **2017**, *7*, a026450, doi:10.1101/cshperspect.a026450.

6. Funding: Neo7Bioscience funded the case study and was involved in its design, data collection, analysis, and preparation of the manuscript**.**

7. Conflicts of Interest

This study was funded by Neo7Bioscience, which was involved in the study design, data collection, analysis, and manuscript preparation. John Catanzaro, who conceived the case study and served as Principal Investigator, is the CEO and a shareholder of Neo7Bioscience and also receives salary support from the company. Neo7Bioscience is a privately held biotechnology company specializing in the development of personalized therapeutic peptides. Nicolas Hulscher, who provided scientific input, receives salary support from the McCullough Foundation. Peter McCullough, who provided research oversight, is the founder of the McCullough Foundation but receives no compensation from the organization. The McCullough Foundation is a nonprofit entity dedicated to advancing independent public health research, medical ethics, and evidence-based policy.

8. Informed Consent: Informed consent was obtained from the subject involved in the study, and all data were de-identified before analysis.

9. Data Availability Statement: The sequencing datasets generated and analyzed in this study have been submitted to the NCBI Gene Expression Omnibus (GEO) and will be made publicly available under an accession number to be assigned. In the interim, the data are available from the corresponding author upon reasonable request.