Case Report

Identification of a Novel *NRG1* Fusion with Targeted Therapeutic Implications in Locally Advanced Pediatric Cholangiocarcinoma: A Case Report

Sarah G. Mitchell[a, b]   Gargi D. Basu[c]   Bree Eaton[b]   Laurie J. Goodman[c]
Kelly C. Goldsmith[a, b]

[a]Aflac Cancer and Blood Disorders Center, Children's Healthcare of Atlanta, Atlanta, GA, USA;
[b]Emory University School of Medicine, Atlanta, GA, USA; [c]Ashion Analytics, Phoenix, AZ, USA

Keywords

Pediatric cholangiocarcinoma · *NRG1* fusion · Precision medicine · Case report

Abstract

Locally advanced cholangiocarcinoma has a poor prognosis, with long-term survival only for patients where complete surgical resection is achieved. Median overall survival with chemotherapy alone is less than 1 year. Novel strategies combining conventional chemotherapy and radiotherapy followed by targeted agents can lead to durable treatment responses and are applicable to cholangiocarcinoma management. Pediatric cholangiocarcinoma is exceedingly rare, with an estimate of 15–22 cases reported in the last 40 years. As such, no standard therapeutic regimen exists. We present a case of a 16-year-old previously healthy patient with unresectable cholangiocarcinoma whose tumor genetic sequencing revealed a novel, actionable neuregulin-1 (*NRG1*) gene translocation. The patient underwent standard systemic chemotherapy with gemcitabine and cisplatin followed by hypofractionated proton radiation therapy for local control. The patient then started an oral pan-*ERBB* (erythroblastic B receptor tyrosine kinases including *ErbB1/EGFR, ErbB2/HER2, ErbB3/HER3, ErbB4/HER4*) family inhibitor as a maintenance medication, remaining with stable disease and excellent quality of life for over 2 years. This case highlights a novel *NRG1* fusion in a rare clinical entity that provided an opportunity to utilize a multimodal therapeutic strategy in the pediatric setting.

© 2023 The Author(s).
Published by S. Karger AG, Basel

Correspondence to:
Kelly Goldsmith, kgoldsm @ emory.edu

Karger

Case Reports in Oncology

Case Rep Oncol 2023;16:249–255

DOI: 10.1159/000530164

© 2023 The Author(s). Published by S. Karger AG, Basel
www.karger.com/cro

250

Mitchell et al.: A Novel *NRG1* Fusion in Pediatric Cholangiocarcinoma

Introduction

Locally advanced cholangiocarcinoma has a generally poor prognosis, and complete surgical resection is crucial for long-term survival [1, 2]. Median progression-free survival and overall survival with chemotherapy alone have been documented as 8 months and 11.7 months, respectively [3]. Pediatric cholangiocarcinoma is exceedingly rare, with an estimate of 15–22 literature-reported cases in the last 40 years. Of these pediatric patients, the vast majority have an underlying gastrointestinal disorder predisposing them to malignancy [4]. We report a rare case of a previously healthy teenager who presented with obstructive jaundice and was subsequently diagnosed with locally advanced perihilar cholangiocarcinoma. Whole-exome and whole-transcriptome sequencing of tumor tissue and a paired normal sample identified a novel *NRG1* fusion, providing an opportunity for targeted oral therapy and clinical trial eligibility. The patient had an initial partial response to standard cytotoxic chemotherapy. Her tumor was unresectable, and she received hypofractionated proton beam radiotherapy for definitive local treatment and then maintained stable disease for 17 months while receiving oral *NRG1*-targeted therapy. She subsequently progressed with new liver metastases and enrolled in a clinical trial for patients with tumor *NRG1* alterations.

Case Presentation

A previously healthy 16-year-old Hispanic girl presented with abdominal pain and jaundice. She reported a 4-week history of intermittent, sharp, right upper quadrant pain that was worse after eating. When she developed scleral icterus, acholic stools, and dark urine, she presented for evaluation. Blood work revealed transaminitis (AST 322 U/L [normal 17–33 U/L], ALT 360 U/L [normal 8–24 U/L]) with a direct hyperbilirubinemia (total bilirubin 7.2 mg/dL [normal 0.2–1 mg/dL], direct bilirubin 5.2 mg/dL [normal <0.3 mg/dL]), and elevated GGT (1,346 U/L [normal 10–21 U/L]). Computed tomography of the abdomen and pelvis revealed a large, infiltrative central liver mass. Magnetic resonance imaging (MRI) confirmed a poorly defined, T1 hypointense, T2 hyperintense mass at the porta hepatis measuring 6 cm × 6.7 cm × 6.9 cm with distal biliary ductal dilatation. The left portal vein was not visualized in its course through the mass; the right portal vein demonstrated normal enhancement (shown in Fig. 1a, b). A chest computed tomography scan was notable only for a left lower lobe calcified granuloma, and a positron emission tomography scan revealed fluorodeoxyglucose avidity solely in the known hepatic mass. The patient underwent percutaneous biliary decompression and drain placement along with biopsy of the mass by interventional radiology. Pathologic evaluation of the biopsied tissue demonstrated immunohistochemical staining positive for CK7 (strong, diffuse), mucicarmine, CK19 (focal), CK56 (focal), and galectin-3 (focal) and staining negative for CK20, S100P, TTF-1, and CDX-2, which, in conjunction with morphologic features, confirmed the diagnosis of cholangiocarcinoma (shown in Fig. 1c, d). The liver tumor was deemed unresectable due to its locally advanced nature. The patient therefore received standard cytotoxic chemotherapy treatment for unresectable cholangiocarcinoma with eight cycles of cisplatin-gemcitabine (cisplatin 25 mg/m^2 followed by gemcitabine 1,000 mg/m^2 on days 1 and 8 every 3 weeks) [5]. A partial response to chemotherapy was documented. She then underwent hypofractionated proton beam radiation therapy for definitive treatment (58.05 Gy [RBE] in 3.87 Gy [RBE] per fraction for a total of 15 fractions (shown in Fig. 1e, f)). The patient tolerated the treatment well with only grade 1 nausea and fatigue that was transient. On follow-up imaging, her primary tumor remained stable.
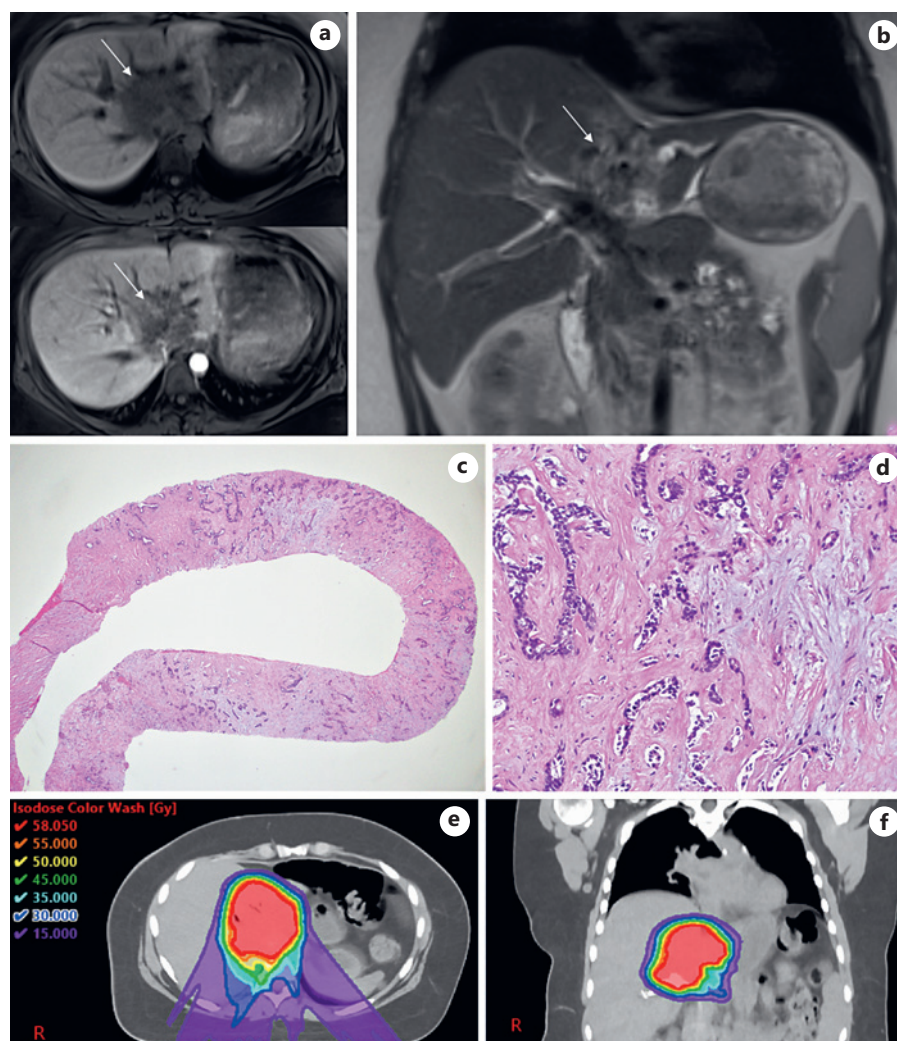
Karger

251

**Case Reports
in Oncology**

Mitchell et al.: A Novel *NRG1* Fusion in Pediatric Cholangiocarcinoma

Fig. 1. a, **b** Magnetic resonance imaging revealed a poorly defined, T1 hypointense, T2 hyperintense mass at the porta hepatis measuring 6 cm × 6.7 cm × 6.9 cm with distal biliary ductal dilatation. The left portal vein was not visualized in its course through the mass; the right portal vein demonstrated normal enhancement. **c**, **d** Immunohistochemical staining was positive for CK7 (strong, diffuse), mucicarmine, CK19 (focal), CK56 (focal), and galectin-3 (focal) and negative for CK20, S100P, TTF-1, and CDX-2. In combination with morphologic features, this confirmed cholangiocarcinoma. **e**, **f** Hypofractionated proton beam radiation therapy schematic for definitive treatment of unresectable cholangiocarcinoma, 58.05 Gy (RBE) in 3.87 Gy (RBE) per fraction for a total of 15 fractions.

The patient enrolled in our institution's precision medicine study. Tumor tissue from the diagnostic biopsy was sent for comprehensive genomic sequencing (whole exome/paired normal and transcriptome sequencing) utilizing the GEM ExTra® assay (Ashion Analytics, Phoenix, AZ, USA). Details of the test methodology and clinical reporting have been published previously [6]. Tumor tissue sequencing identified a novel, presumed oncogenic *AGRN-NRG1* fusion between chromosome 1 (*AGRN*) and chromosome 8 (*NRG1*) with breakpoints within exon 2 of *AGRN* and intron 1 of *NRG1* (shown in Fig. 2a). Based on our institution's molecular tumor board discussion, the patient was started on the oral pan-*ERBB* family inhibitor, afatinib, as a form of maintenance therapy. She continued on afatinib 40 mg daily with stable disease for 17 months with the main side effect being acneiform rash. The patient then
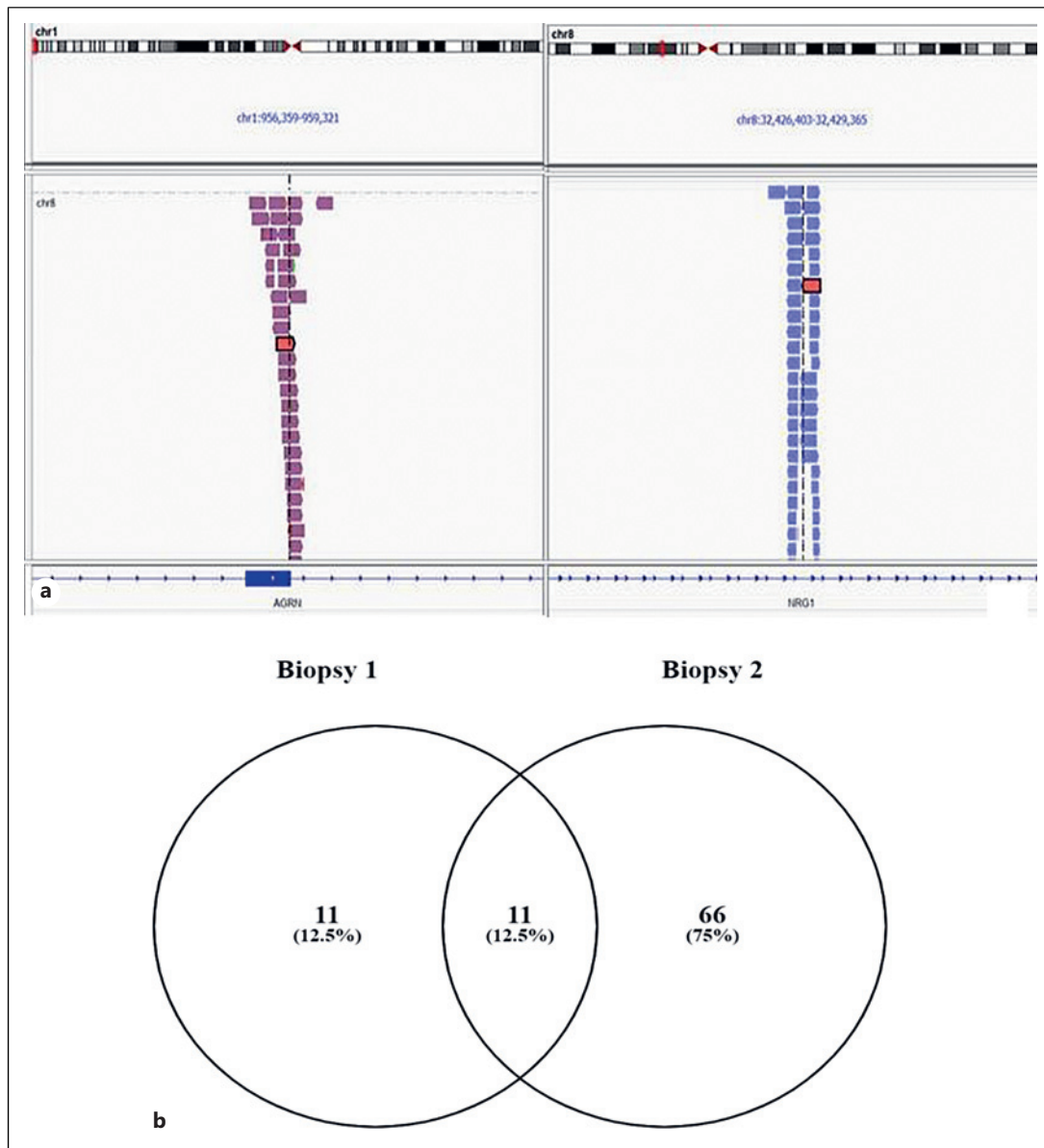
Case Reports in Oncology

Case Rep Oncol 2023;16:249–255

DOI: 10.1159/000530164

© 2023 The Author(s). Published by S. Karger AG, Basel
www.karger.com/cro

Mitchell et al.: A Novel *NRG1* Fusion in Pediatric Cholangiocarcinoma

252

Fig. 2. a Tissue sequencing identified a novel, presumed oncogenic AGRN-NRG1 fusion between chromosome 1 (AGRN) and chromosome 8 (NRG1) with breakpoints within exon 2 of AGRN and intron 1 of NRG1. **b** Initial tumor tissue sequencing identified 22 genomic alterations. Sequencing of progressive tumor tissue identified 66 new genomic alterations.

experienced disease progression with development of three new, subcentimeter, satellite liver lesions. Biopsy of the largest lesion confirmed cholangiocarcinoma, and sequencing identified persistence of the *AGRN-NRG1* fusion along with the development of a *MCL1* amplification. Despite detection of 66 new genomic alterations, none were classified as driver events (shown in Fig. 2b). The patient subsequently chose to enroll in a clinical trial utilizing an anti-*ERBB3* monoclonal antibody for *NRG1*-altered tumors.

Mitchell et al.: A Novel *NRG1* Fusion in Pediatric Cholangiocarcinoma

Discussion

Cholangiocarcinoma is a highly aggressive malignancy that arises from epithelial cells of the intrahepatic and extrahepatic bile duct system, often presenting in an advanced stage. Risk factors for the disease include chronic hepatitis and cirrhosis, biliary inflammatory diseases, and hepatobiliary fluke infection. However, in the majority of cases, no risk factor is identified [1]. In the pediatric population, cholangiocarcinoma is exceedingly rare, with only 15 cases identified from 1973 to 2013 based on SEER18 data and 22 cases reported in the literature. Ninety percent of these patients had an underlying gastrointestinal comorbidity, with a poor 3-year overall survival at 35.3% [4].

Surgery is the mainstay of curative therapy for cholangiocarcinoma, though only ~35% of patients present with early stage disease amenable to upfront resection [7]. Standard treatment for patients with unresectable cholangiocarcinoma includes eight cycles of cisplatin and gemcitabine. However, median progression-free survival and overall survival with chemotherapy alone have been documented as 8 months and 11.7 months, respectively [3]. The role of radiation in the treatment of liver tumors is evolving, and proton therapy is an attractive local control strategy, providing a theoretical clinical benefit over photon-based treatment by allowing for safer dose-escalation in large tumors [8]. A multi-institutional phase II study testing high-dose, hypofractionated proton therapy for unresectable hepatocellular carcinoma and intrahepatic cholangiocarcinoma in adults demonstrated excellent local control of 94% at 2 years with a median dose of 58 Gy in 15 fractions [8]. This proton treatment regimen was chosen for the patient presented in this study due to the large tumor with central location and allowed for effective dose delivery to the tumor with minimal radiation exposure to the remaining normal liver and adjacent stomach, small bowel, and kidneys, as illustrated in Figure 1e, f. To our knowledge, this is the first report of high-dose, hypofractionated proton therapy being used for definitive treatment of intrahepatic cholangiocarcinoma in a child.

Precision medicine and personalized therapies are becoming more common in the treatment of cholangiocarcinoma, with a goal of providing precise therapy with improved efficacy and safety profiles [9]. To date, a number of mutated oncogenes have been identified in cholangiocarcinoma, including *IDH1*, *KRAS, BRAF, ARID1A, PBRM1, EGFR/ERBB1,* and *HER2/ERBB2* among others [10, 11]. Inhibitors of key oncogenic pathways have been evaluated as single agents, combined targeted therapies, and in combination with standard chemotherapy [9]. Neuregulin-1 (*NRG1*) gene fusion events are rare, yet potentially actionable driver mutations that have been detected across multiple solid tumor types at a frequency of 0.2% with significant fusion partner heterogeneity [12]. They are largely mutually exclusive with other oncogenic drivers. In cholangiocarcinoma, *NRG1* fusions have been observed in approximately 0.8% of cases [12]. These fusions promote tumorigenesis via an *EGF*-like domain within *NRG1* that binds *ERBB3* in a para/juxtacrine or autocrine fashion, resulting in *ERBB2/ERBB3* heterodimerization and increased downstream signaling. Hence, targeted agents are often utilized. Clinical activity in *NRG1* fusion-positive malignancy has been reported with combined inhibition of *ERBB* and *ERBB2* with erlotinib and pertuzumab, other dual-inhibition of multiple *ERBB* receptors such as afatinib, and with anti-*ERBB3* monoclonal antibodies [13, 14]. Additional clinical trials are underway.

Conclusion

Pediatric cholangiocarcinoma is exceedingly rare, and in the case described here, identification of our patient's novel *AGRN-NRG1* fusion provided a molecular target for a

Karger

Case Reports in Oncology

Case Rep Oncol 2023;16:249–255

DOI: 10.1159/000530164

© 2023 The Author(s). Published by S. Karger AG, Basel
www.karger.com/cro

254

Mitchell et al.: A Novel *NRG1* Fusion in Pediatric Cholangiocarcinoma

personalized, multimodal therapeutic regimen as well as eligibility for clinical trial enrollment. She experienced stable disease for 17 months on afatinib monotherapy, and this period of prolonged stability may reflect the contribution of pan-*ERBB* inhibition in disease control. The CARE Checklist has been completed by the authors for this case report, attached as online supplementary material (for all online suppl. material, see www.karger.com/doi/10.1159/000530164).

Acknowledgments

Statement of Ethics

Ethical approval is not required for this study in accordance with local or national guidelines. Written informed consent was obtained from the parent of the patient for publication of the details of their medical case and any accompanying images.

Conflict of Interest Statement

The authors have no conflicts of interest to declare.

Funding Sources

Author Contributions

Dr. Sarah G. Mitchell primarily cared for the patient and drafted the manuscript. Dr. Bree Eaton cared for the patient and drafted pertinent portions of the manuscript. Dr. Gargi D. Basu, Dr. Laurie J. Goodman, and Dr. Kelly C. Goldsmith revised the manuscript critically for important content. All authors approved of the version of the manuscript to be published.

Data Availability Statement

Data are available within the article. Further inquiries can be directed to the corresponding author.

References

1   El-Diwany R, Pawlik TM, Ejaz A. Intrahepatic cholangiocarcinoma. Surg Oncol Clin N Am. 2019;28(4):587–99.
2   Banales JM, Cardinale V, Carpino G, Marzioni M, Andersen JB, Invernizzi P, et al. Expert consensus document: cholangiocarcinoma: current knowledge and future perspectives consensus statement from the European Network for the Study of Cholangiocarcinoma (ENS-CCA). Nat Rev Gastroenterol Hepatol. 2016;13(5):261–80.

Case Reports in Oncology

Case Rep Oncol 2023;16:249–255

DOI: 10.1159/000530164

© 2023 The Author(s). Published by S. Karger AG, Basel
www.karger.com/cro

255

3 Razumilava N, Gores GJ. Cholangiocarcinoma. Lancet. 2014;383(9935):2168–79.

4 Newsome JR, Venkatramani R, Heczey A, Danysh HE, Fishman DS, Miloh T. Cholangiocarcinoma among children and adolescents: a review of the literature and surveillance, epidemiology, and end results program database analysis. J Pediatr Gastroenterol Nutr. 2018;66(1):e12–8.

5 Valle J, Wasan H, Palmer DH, Cunningham D, Anthoney A, Maraveyas A, et al. Cisplatin plus gemcitabine versus gemcitabine for biliary tract cancer. N Engl J Med. 2010;362(14):1273–81.

6 Dizman N, Lyou Y, Salgia N, Bergerot PG, Hsu J, Enriquez D, et al. Correlates of clinical benefit from immunotherapy and targeted therapy in metastatic renal cell carcinoma: comprehensive genomic and transcriptomic analysis. J Immunother Cancer. 2020;8(2):e000953.

7 Rizvi S, Khan SA, Hallemeier CL, Kelley RK, Gores GJ. Cholangiocarcinoma: evolving concepts and therapeutic strategies. Nat Rev Clin Oncol. 2018;15(2):95–111.

8 Hong TS, Wo JY, Yeap BY, Ben-Josef E, McDonnell EI, Blaszkowsky LS, et al. Multi-institutional phase II study of high-dose hypofractionated proton beam therapy in patients with localized, unresectable hepatocellular carcinoma and intrahepatic cholangiocarcinoma. J Clin Oncol. 2016;34(5):460–8.

9 Geynisman DM, Catenacci DVT. Toward personalized treatment of advanced biliary tract cancers. Discov Med. 2012;14(74):41–57.

10 Hezel AF, Deshpande V, Zhu AX. Genetics of biliary tract cancers and emerging targeted therapies. J Clin Oncol. 2010;28(21):3531–40.

11 Banales JM, Marin JJG, Lamarca A, Rodrigues PM, Khan SA, Roberts LR, et al. Cholangiocarcinoma 2020: the next horizon in mechanisms and management. Nat Rev Gastroenterol Hepatol. 2020;17(9):557–88.

12 Jonna S, Feldman RA, Swensen J, Gatalica Z, Korn WM, Borghaei H, et al. Detection of NRG1 gene fusions in solid tumors. Clin Cancer Res. 2019;25(16):4966–72.

13 Heining C, Horak P, Uhrig S, Codo PL, Klink B, Hutter B, et al. NRG1 fusions in KRAS wild-type pancreatic cancer. Cancer Discov. 2018;8(9):1087–95.

14 Sequist LV, Gray JE, Harb WA, Lopez-Chavez A, Doebele RC, Modiano MR, et al. Randomized phase II trial of seribantumab in combination with erlotinib in patients with EGFR wild-type non-small cell lung cancer. Oncologist. 2019;24(8):1095–102.

KARGER