Journal of Neuroimmunology 337 (2019) 577075

Contents lists available at ScienceDirect

Journal of Neuroimmunology

journal homepage: www.elsevier.com/locate/jneuroim

Short Communication

Lymphocytic ganglionitis leading to megacolon in lymphocyte-rich glioblastoma

Jared T. Ahrendsen[a], Kevin R. Anderson[a], Matthew P. Anderson[a,b,*]

[a] Departments of Pathology, Harvard Medical School, Boston, MA, USA
[b] Neurology, Beth Israel Deaconess Medical Center, Harvard Medical School, Boston, MA, USA

ARTICLE INFO

Keywords:
Glioblastoma
Lymphocytic
Ganglionitis
CD8
T-cell
Paraneoplastic

ABSTRACT

T-cell immune attack of cancer cells underlies the efficacy of immune checkpoint inhibitors in many cancer subtypes, but is not yet well established in the primary brain cancer glioblastoma. Immune checkpoint inhibitor treatments that disinhibit the immune system to enhance immune clearance of cancer have in rare cases resulted in T-cell attack of peripheral ganglia causing lymphocytic ganglionitis. In glioblastoma, lymphocytic ganglionitis has not been reported and checkpoint inhibitors are not routinely used. Here we report a case of glioblastoma not treated with checkpoint inhibitors in which the primary tumor and peripheral ganglia of the celiac and sympathetic chains, as well as myenteric plexus, are infiltrated by $CD8^+$ cytotoxic T-cells. In addition to the marked lymphocytic infiltrates, this case is also notable for an unusually long survival (8 years) after diagnosis with glioblastoma, but an ultimately fatal outcome due to ileus. The findings suggest T-cell immune attack of glioblastoma may prolong survival, but also suggest T-cell autoimmune diseases such as lymphocytic ganglionitis could become a risk with the future use of immune-targeted therapies for glioblastoma.

1.1. Case report

A 57-year-old man with a known history of right temporal lobe glioblastoma W.H.O. grade IV presented with increasing somnolence and confusion from a rehabilitation center. His glioblastoma was diagnosed 8 years prior and his treatment course included gross total resection along with multiple cycles of chemotherapy and radiation treatment. During his hospital course he developed increasing abdominal distention. Imaging at this time showed large intestine dilatation (megacolon) and was concerning for ileus and a sigmoid volvulus. Further intervention with a flexible sigmoidoscopy for decompression was deemed unsafe after the patient became hypotensive. He continued to decline and subsequently passed.

A full autopsy was performed according to institutional standard operating procedures. Gross examination showed marked colonic distention without evidence of perforation, as well as focal adhesions. The brain and spinal cord were removed and subsequently examined after fixation. The fresh brain weighed 1340 g. The brain was sectioned, which grossly showed tissue loss and encephalomalacia of the right temporal lobe and hippocampus, at the site of the patient's prior surgical resection.

Given the patient's extraordinarily long survival, the original tumor resection was evaluated further to confirm the diagnosis of glioblastoma and also to evaluate for an adaptive cell-mediated immune reaction. Sections showed a diffusely infiltrating, hypercellular lesion composed of severely atypical astroglial cells, numerous mitotic figures, prominent microvascular proliferation, and pseudopalisading necrosis supporting the histologic diagnosis of glioblastoma (W.H.O. grade IV) (Fig. 1A). Immunohistochemistry demonstrated that the tumor cells were diffusely positive for GFAP with a subset also positive for Olig2. Scattered tumor cells showed weak positivity for P53, consistent with a non-mutated wild-type pattern. Immunohistochemistry for the IDH1 R132H specific mutation was negative. Ki67 demonstrated a high proliferation index labeling 30–40% of tumor cells (data not shown). Together, these data support a diagnosis of glioblastoma, IDH1 (R132H) wild-type, W.H.O. grade IV. Importantly, prominent perivascular lymphocytic cuffs and tumor infiltrating lymphocytes were observed (Fig. 1B), composed of CD3-positive T-lymphocytes (Fig. 1C), including an admixed population of CD4-positive helper T-cells (Fig. 1D) and CD8-positive cytotoxic T-cells (Fig. 1E). Tumor infiltrating lymphocytes, particularly CD8-positive cytotoxic T-cells, were often observed in clusters surrounding tumor cells or with cytoplasmic processes directly touching tumor cells (Fig. 1E). Furthermore, granzyme positive polarized granules were observed in lymphocytes, indicating an active cytotoxic immune response against tumor cells (Fig. 1F). CD20-positive B-cells and CD138-positive plasma cells were

* Corresponding author at: Beth Israel Deaconess Medical Center, Harvard Medical School, 330 Brookline Ave, Boston, MA 02215, USA.
E-mail address: mpanders@bidmc.harvard.edu (M.P. Anderson).

https://doi.org/10.1016/j.jneuroim.2019.577075
Received 27 August 2019; Received in revised form 23 September 2019; Accepted 24 September 2019

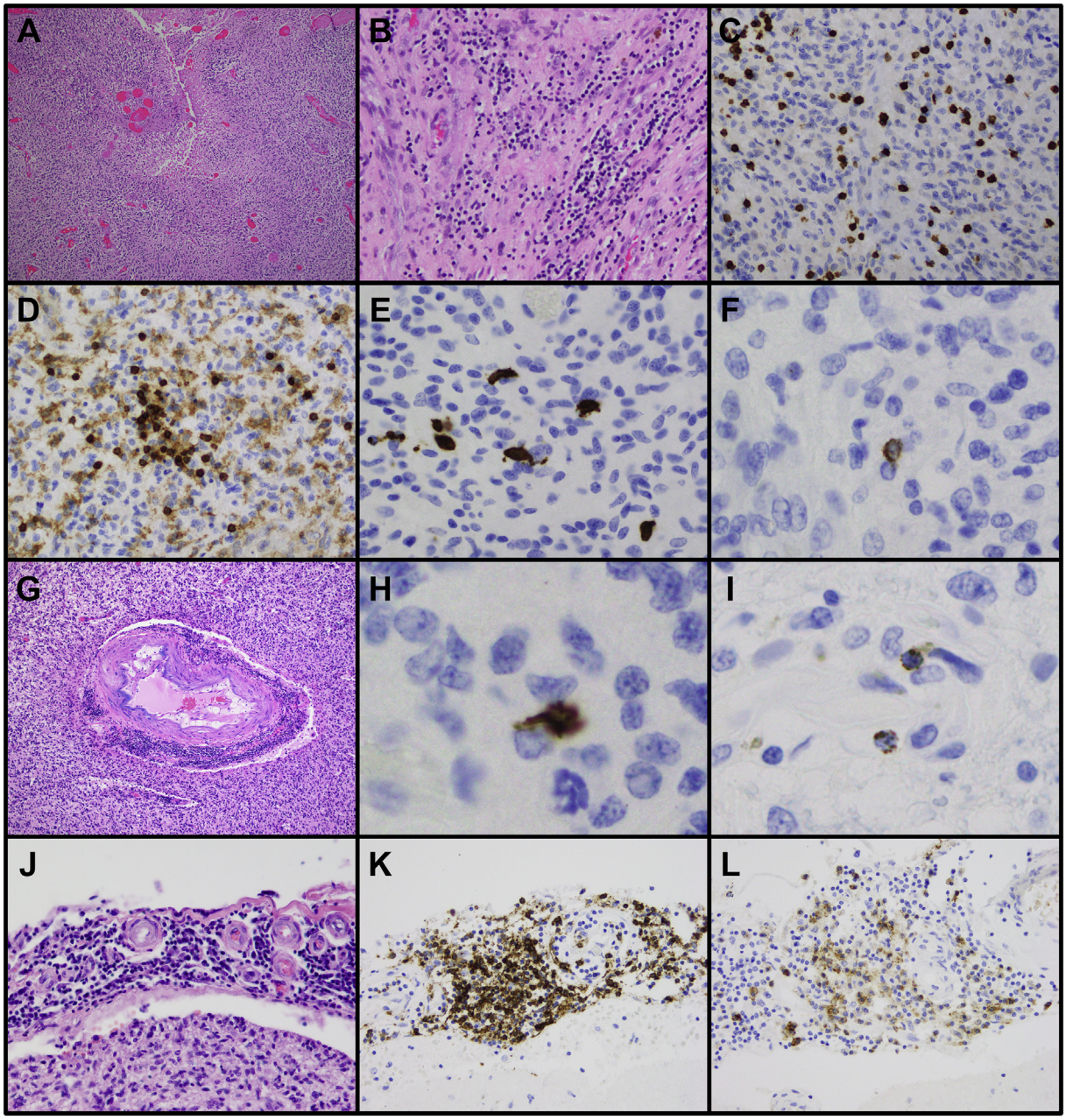

Fig. 1. Tumor-infiltrating lymphocytes. (A-F) Original tumor resection and (G-L) postmortem brain residual/recurrent glioblastoma contain a prominent perivascular and tumor-infiltrating lymphocyte population (B, G). Immunohistochemistry identifies numerous CD3-positive T-lymphocytes (C) composed of a mixture of CD4-positive (D) and CD8-positive (E, H) T-cells. Granzyme-positive granules polarized toward tumor cells (F, I). Leptomeningeal lymphocyte infiltrates (J) are composed of CD20-positive B-cells (K) and CD138-positive plasma cells (L). Magnification: A, G: 100×; B, C, D, J, K, L: 200×; F, H, I: 600×.

also observed within the tumor-infiltrated brain parenchyma, but were rare compared to CD3-positive T-lymphocytes (data not shown).

Microscopic examination of the autopsy-derived brain tissues identified recurrent/residual glioblastoma with features similar to those of the initial resection 8 years prior, including pseudopalisading necrosis, microvascular proliferation, and brisk mitotic activity, but also with large areas of radiation necrosis. Prominent perivascular lymphocytic cuffs and extensive lymphocytic infiltration within the tumor were again observed (Fig. 1G), composed predominantly of CD3-positive T-lymphocytes with an admixed population of CD4-positive helper T-cells and CD8-positive cytotoxic T-cells (Fig. 1H). Again, granzyme positive polarized granules were observed in lymphocytes (Fig. 1I). Interestingly, a significant leptomeningeal inflammatory infiltrate was observed adjacent to the recurrent/residual tumor (Fig. 1J), composed of many CD20-positive B-cells (Fig. 1K) and CD138-positive plasma cells (Fig. 1L). Notably, very few lymphocytes were observed in brain regions uninvolved by tumor (data not shown).

Based on the clinical evidence of megacolon, peripheral nerves and ganglia were sampled at the time of autopsy, including the sympathetic chain ganglia (Fig. 2A-E) and celiac ganglia (not shown). Samples from these peripheral ganglia showed a patchy dense CD3-positive lymphocytic infiltrate (Fig. 2B), consistent with lymphocytic ganglionitis. The infiltrating lymphocytes were a mixture of CD4- and CD8-positive cells (Fig. 2C, D, respectively). There was a notable absence of CD20-positve B-lymphocytes in the involved ganglia (Fig. 2E). Additionally, the myenteric plexus of the colon showed foci of lymphocytic infiltration of the ganglion cells (Fig. 2F) composed of CD3-positive T-lymphocytes (Fig. 2G).

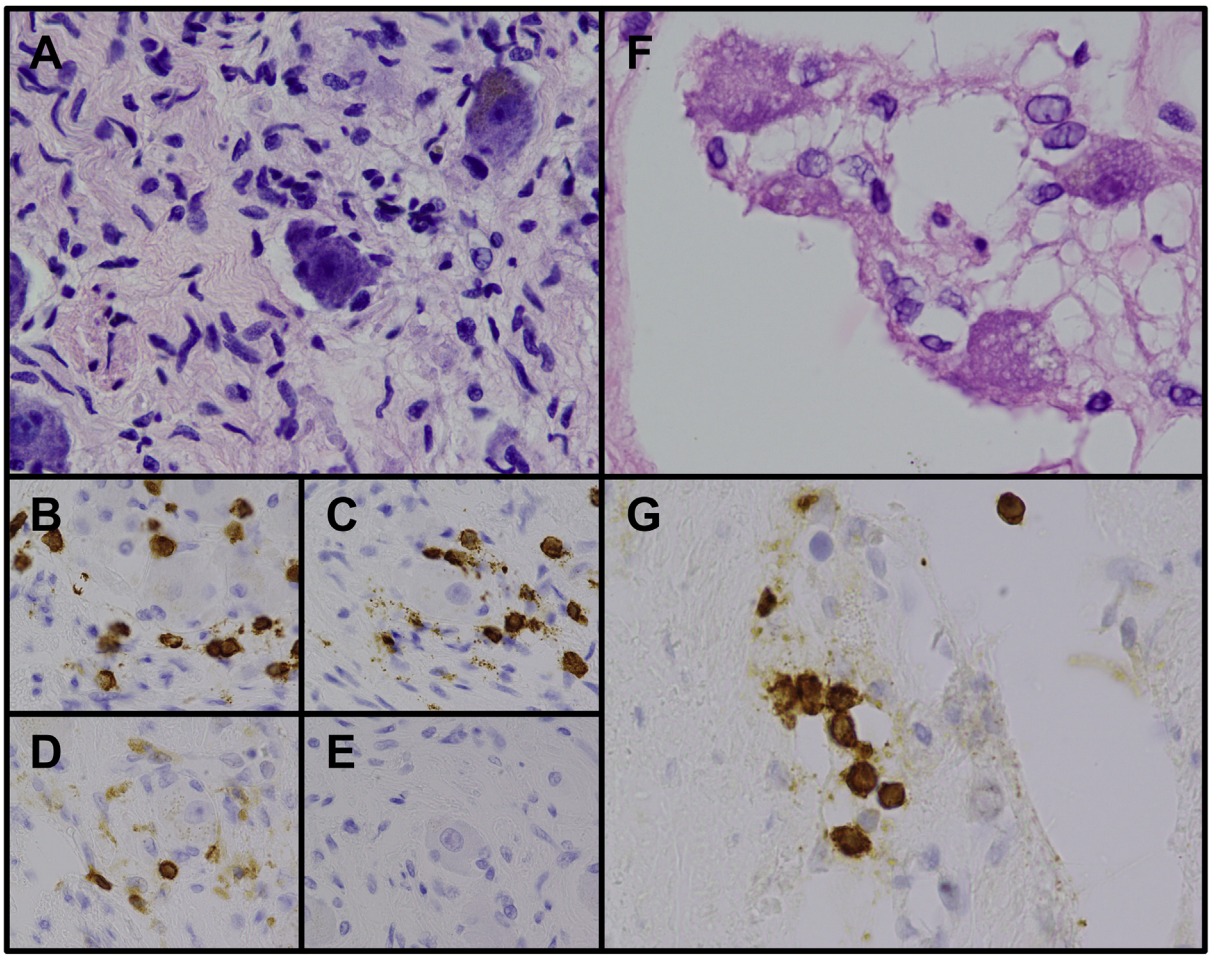

Fig. 2. Lymphocytic ganglionitis (A-E) Sympathetic chain ganglia and (F-G) myenteric plexus ganglia with abundant CD3-positive lymphocytes (B, G) composed of both CD4 and CD8-positive T-cells (C, D, respectively). CD20-positive B-cells were absent from these same lesions (E). Magnification: A, F, G: 600×; B-E: 400×.

One consideration is the remote possibility that the lymphocytic ganglionitis observed in this case is unrelated to the T-cell reaction to the glioblastoma and instead is caused by other etiologies. Therefore, we examined the tissue for other potential causes of lymphocytic ganglionitis. Case reports of herpes simplex virus (HSV) and varicella zoster virus (VZV) associated ganglionitis exist with prominent lymphocytic infiltrates and neuronophagia (Gilden et al., 2003; Mitchell et al., 2003; Valyi-Nagy et al., 2017). However, this patient's cerebrospinal fluid and serum prior to death were negative for various infectious entities, including syphilis (VDRL), HSV (PCR), enterovirus (PCR), and Cryptococcus (ELISA). Furthermore, CSF and blood cultures were negative for growth of bacteria, fungi, and mycobacteria. Additionally, VZV and HSV immunohistochemistry were negative in both the sympathetic and celiac ganglia of the autopsy-derived tissue (data not shown). Finally, this patient had no other known autoimmune diseases, such as Sjögren's Syndrome, or connective tissue disorders, both of which have been associated with sympathetic ganglionitis (DeSchryver-Kecskemeti and Clouse, 1989; Griffin et al., 1990; Oaklander, 2016).

Considering the robust T-lymphocyte immune cell infiltrate of the glioblastoma concurrent with the infiltrate of the sympathetic and myenteric ganglia, these findings support the diagnosis of T-lymphocyte autoimmune ganglionitis in the setting of glioblastoma (i.e. T-cell paraneoplastic ganglionitis) involving the celiac and sympathetic chains, as well as myenteric plexus ganglia.

1.2. Discussion

Glioblastoma has been associated with various manifestations of immune system dysregulation and autoimmunity (Chongsathidkiet et al., 2018; Rodríguez-Lobato et al., 2016; Woroniecka et al., 2018); however in our literature review, lymphocytic ganglionitis has not been previously described with this tumor type. Here we present a patient with known glioblastoma with an extraordinarily long survival of eight years. The host immune microenvironment is becoming increasingly linked to local tumor control and overall survival (Pagès et al., 2018) and has opened an armamentarium of cancer immunotherapeutics including immune-directed monoclonal antibodies, immune check point inhibitors, and chimeric antigen receptor T-cell infusion (Ottaviano et al., 2019; Sukari et al., 2019). This patient's prolonged survival may relate to the extensive lymphocytic infiltrates of the patient's glioblastoma. However, this ultimately led to an autoimmune lymphocytic ganglionitis that impaired function of the patient's gastrointestinal tract. As has been described previously, myenteric plexus ganglionitis can result in intestinal obstructive symptoms that result in death (Horoupian and Kim, 1982; Malhotra et al., 2011; Racalbuto et al., 2008).

It is unlikely that this patient's prolonged survival is due to misdiagnosis of the original tumor resection. Pleomorphic xanthoastrocytoma (PXA) is occasionally mistaken for glioblastoma; however, PXA typically arises in children/young adults and does not have a high Ki67 proliferation index or pseudopalisading necrosis, as observed in the patient's tumor (Shaikh et al., 2019). Other factors have been shown to impart longer survival in patients with glioblastoma, including IDH

mutations and MGMT promotor methylation (Czapski et al., 2018; Gately et al., 2019). Although the MGMT methylation status of this tumor is not known, the patient's tumor was negative for the most common IDH mutation (IDH1 R132H) by immunohistochemistry. While it is still possible that the tumor in this case harbors a different IDH mutation, none of these factors (IDH and MGMT status), even in combination, would be expected to impart the extraordinarily long survival observed in this case. Indeed, the median overall survival in patients with both IDH mutant and MGMT methylated glioblastoma was only 36 months in one study (Yang et al., 2015). Furthermore, in a large cohort of long-term glioblastoma survivors (defined as greater than 5 year survival), the molecular characteristics between long-term and short-term survivors were remarkably similar (Burgenske et al., 2019). Thus, there are tumor extrinsic factors that have a significant impact in long-term glioblastoma survivors.

Glioblastoma is remarkable in its ability to evade the immune system and it exerts this effects on many aspects of immune system function, particularly T-cells (Woroniecka et al., 2018). This ability of glioblastoma to hijack the host immune system likely underlies the challenges and limited success of immune mediated therapies for this tumor subtype (Tomaszewski et al., 2019). One recent study demonstrated that as many as 15% of treatment naïve glioblastoma patients have peripheral blood T-cell levels below 200 per µl, AIDS-defining levels in HIV-infected individuals. They go on to demonstrate that T-cells are sequestered in the bone marrow, disabling their ability to exert anti-tumor effects (Chongsathidkiet et al., 2018). The patient described in this report had high levels of peripheral CD4-positive T-cells throughout his prolonged course with the disease (not shown). Together, these results suggest enhancing the adaptive cellular immune response to glioblastoma may lengthen survival, but it might also increase the risk of lethal autoimmune disease.

1.3. Materials and methods

Consent for the autopsy was obtained from the decedent's next of kin, which included provisions to save tissue for diagnostic and research purposes. Autopsy was performed according to our department's standard operating procedure. The brain and spinal cord were removed and placed directly into 10% neutral buffered formalin and allowed to fix for 14 days before sectioning. Formalin fixed paraffin embedded tissue was sectioned and stained with H&E with or without luxol fast blue prior to examination. All antibodies used in this study (CD3, CD4, CD8, CD20, CD138, GFAP, Olig2, P53, Ki67, IDH1 R132H, VZV, HSV1/2) were obtained from Dako (Agilent Pathology Solutions, Santa Clara, CA). Immunohistochemistry staining was performed on a Dako Autostainer Link48 (Agilent Pathology Solutions, Santa Clara, CA), according to our institution's standard operating procedure.

Author Contributions

J.T.A., K.R.A and M.P.A. contributed to the conception and design of the study; J.T.A., K.R.A and M.P.A. contributed to the acquisition and analysis of data; J.T.A. and M.P.A. contributed to drafting the text and preparing the figures.

Acknowledgements

We thank the Boston Children's Hospital IDDRC (U54 HD090255, P30 HD18655) and other funding to M.P.A. from The National Institute of Mental Health (R01 MH114858, R01 MH112714), The National Institute of Neurological Disorders and Stroke (R01 NS08916), The Nancy Lurie Marks Family Foundation, The Landreth Foundation, Autism Speaks/National Alliance for Autism Research, and the Simons Foundation/Autism BrainNet (FA#345171).

Declaration of Competing Interest

The author has no conflict of interest.

References

Burgenske, D.M., Yang, J., Decker, P.A., Kollmeyer, T.M., Kosel, M.L., Mladek, A.C., Caron, A.A., Vaubel, R.A., Gupta, S.K., Kitange, G.J., Sicotte, H., Youland, R.S., Remonde, D., Voss, J.S., Fritcher, E.G.B., Kolsky, K.L., Ida, C.M., Meyer, F.B., Lachance, D.H., Parney, I.J., Kipp, B.R., Giannini, C., Sulman, E.P., Jenkins, R.B., Eckel-Passow, J.E., Sarkaria, J.N., 2019. Molecular Profiling of Long-Term IDH-wildtype Glioblastoma Survivors. Neuro Oncol. https://doi.org/10.1093/neuonc/noz129.

Chongsathidkiet, P., Jackson, C., Koyama, S., Loebel, F., Cui, X., Farber, S.H., Woroniecka, K., Elsamadicy, A.A., Dechant, C.A., Kemeny, H.R., Sanchez-Perez, L., Cheema, T.A., Souders, N.C., Herndon, J.E., Coumans, J.-V., Everitt, J.I., Nahed, B.V., Sampson, J.H., Gunn, M.D., Martuza, R.L., Dranoff, G., Curry, W.T., Fecci, P.E., 2018. Sequestration of T cells in bone marrow in the setting of glioblastoma and other intracranial tumors. Nat. Med. 24, 1459–1468. https://doi.org/10.1038/s41591-018-0135-2.

Czapski, B., Baluszek, S., Herold-Mende, C., Kaminska, B., 2018. Clinical and immunological correlates of long term survival in glioblastoma. Contemp Oncol (Pozn) 22, 81–85. https://doi.org/10.5114/wo.2018.73893.

DeSchryver-Kecskemeti, K., Clouse, R.E., 1989. Perineural and intraneural inflammatory infiltrates in the intestines of patients with systemic connective-tissue disease. Arch. Pathol. Lab. Med. 113, 394–398.

Gately, L., McLachlan, S.A., Philip, J., Rathi, V., Dowling, A., 2019. Molecular profile of long-term survivors of glioblastoma: a scoping review of the literature. J. Clin. Neurosci. 68, 1–8. https://doi.org/10.1016/j.jocn.2019.08.017.

Gilden, D.H., Cohrs, R.J., Hayward, A.R., Wellish, M., Mahalingam, R., 2003. Chronic varicella-zoster virus ganglionitis–a possible cause of postherpetic neuralgia. J. Neuro-Oncol. 9, 404–407. https://doi.org/10.1080/13550280390201722.

Griffin, J.W., Cornblath, D.R., Alexander, E., Campbell, J., Low, P.A., Bird, S., Feldman, E.L., 1990. Ataxic sensory neuropathy and dorsal root ganglionitis associated with Sjögren's syndrome. Ann. Neurol. 27, 304–315. https://doi.org/10.1002/ana.410270313.

Horoupian, D.S., Kim, Y., 1982. Encephalomyeloneuropathy with ganglionitis of the myenteric plexuses in the absence of cancer. Ann. Neurol. 11, 628–632. https://doi.org/10.1002/ana.410110613.

Malhotra, K., Georgiades, I.B., Davies, J., 2011. Functional colonic obstruction in an adult as a presenting feature of thymoma. Ann. R. Coll. Surg. Engl. 93, e61–e63. https://doi.org/10.1308/147870811X589588.

Mitchell, B.M., Bloom, D.C., Cohrs, R.J., Gilden, D.H., Kennedy, P.G.E., 2003. Herpes simplex virus-1 and varicella-zoster virus latency in ganglia. J. Neuro-Oncol. 9, 194–204. https://doi.org/10.1080/13550280390194000.

Oaklander, A.L., 2016. Immunotherapy prospects for painful small-fiber sensory neuropathies and Ganglionopathies. Neurotherapeutics 13, 108–117. https://doi.org/10.1007/s13311-015-0395-1.

Ottaviano, M., De Placido, S., Ascierto, P.A., 2019. Recent success and limitations of immune checkpoint inhibitors for cancer: a lesson from melanoma. Virchows Arch. 474, 421–432. https://doi.org/10.1007/s00428-019-02538-4.

Pagès, F., Mlecnik, B., Marliot, F., Bindea, G., Ou, F.-S., Bifulco, C., Lugli, A., Zlobec, I., Rau, T.T., Berger, M.D., Nagtegaal, I.D., Vink-Börger, E., Hartmann, A., Geppert, C., Kolwelter, J., Merkel, S., Grützmann, R., Van den Eynde, M., Jouret-Mourin, A., Kartheuser, A., Léonard, D., Remue, C., Wang, J.Y., Bavi, P., Roehrl, M.H.A., Ohashi, P.S., Nguyen, L.T., Han, S., MacGregor, H.L., Hafezi-Bakhtiari, S., Wouters, B.G., Masucci, G.V., Andersson, E.K., Zavadova, E., Vocka, M., Spacek, J., Petruzelka, L., Konopasek, B., Dundr, P., Skalova, H., Nemejcova, K., Botti, G., Tatangelo, F., Delrio, P., Ciliberto, G., Maio, M., Laghi, L., Grizzi, F., Fredriksen, T., Buttard, B., Angelova, M., Vasaturo, A., Maby, P., Church, S.E., Angell, H.K., Lafontaine, L., Bruni, D., Sissy, E.C., Haicheur, N., Kirilovsky, A., Berger, A., Lagorce, C., Meyers, J.P., Paustian, C., Feng, Z., Ballesteros-Merino, C., Dijkstra, J., Van de Water, C., van Lent-van Vliet, S., Knijn, N., Mușină, A.-M., Scripcariu, D.-V., Popivanova, B., Xu, M., Fujita, T., Hazama, S., Suzuki, N., Nagano, H., Okuno, K., Torigoe, T., Sato, N., Furuhata, T., Takemasa, I., Itoh, K., Patel, P.S., Vora, H.H., Shah, B., Patel, J.B., Rajvik, K.N., Pandya, S.J., Shukla, S.N., Wang, Y., Zhang, G., Kawakami, Y., Marincola, F.M., Ascierto, P.A., Sargent, D.J., Fox, B.A., Galon, J., 2018. International validation of the consensus Immunoscore for the classification of colon cancer: a prognostic and accuracy study. Lancet. 391, 2128–2139. https://doi.org/10.1016/S0140-6736(18)30789-X.

Racalbuto, A., Magro, G., Lanteri, R., Aliotta, I., Santangelo, M., Di Cataldo, A., 2008. Idiopathic myenteric ganglionitis underlying acute "dramatic" intestinal pseudoobstruction: report of an exceptional case. Case Rep Gastroenterol. 2, 461–468. https://doi.org/10.1159/000164309.

Rodríguez-Lobato, L.G., Martinez-Cibrian, N., Fernández-Avilés, F., Gaya, A., Pineda-Losada, E., Esteve, J., 2016. Romiplostim for the treatment of glioblastoma-related paraneoplastic autoimmune thrombocytopenia refractory to conventional therapy. Ann. Hematol. 95, 665–666. https://doi.org/10.1007/s00277-016-2600-7.

Shaikh, N., Brahmbhatt, N., Kruser, T.J., Kam, K.L., Appin, C.L., Wadhwani, N., Chandler, J., Kumthekar, P., Lukas, R.V., 2019. Pleomorphic xanthoastrocytoma: a brief review. CNS Oncol. https://doi.org/10.2217/cns-2019-0009. CNS39.

Sukari, A., Abdallah, N., Nagasaka, M., 2019. Unleash the power of the mighty T cells-basis of adoptive cellular therapy. Crit. Rev. Oncol. Hematol. 136, 1–12. https://doi.org/10.1016/j.critrevonc.2019.01.015.

Tomaszewski, W., Sanchez-Perez, L., Gajewski, T.F., Sampson, J.H., 2019. Brain Tumor Microenvironment and Host State: Implications for Immunotherapy. Clin. Cancer Res. https://doi.org/10.1158/1078-0432.CCR-18-1627.

Valyi-Nagy, T., Rathore, J.S., Rakic, A.M., Rathore, R.S., Jain, P., Slavin, K.V., 2017. Herpes Simplex Virus Type 1 Human Cervical Dorsal Root Ganglionitis. Case Rep Neurol. 9, 188–194. https://doi.org/10.1159/000479146.

Woroniecka, K.I., Rhodin, K.E., Chongsathidkiet, P., Keith, K.A., Fecci, P.E., 2018. T-cell dysfunction in glioblastoma: applying a new framework. Clin. Cancer Res. 24, 3792–3802. https://doi.org/10.1158/1078-0432.CCR-18-0047.

Yang, P., Zhang, W., Wang, Y., Peng, X., Chen, B., Qiu, X., Li, G., Li, S., Wu, C., Yao, K., Li, W., Yan, W., Li, J., You, Y., Chen, C.C., Jiang, T., 2015. IDH mutation and MGMT promoter methylation in glioblastoma: results of a prospective registry. Oncotarget 6, 40896–40906. https://doi.org/10.18632/oncotarget.5683.