·经验交流·

供者淋巴细胞鞘内注射治疗异基因造血干细胞移植后EB病毒相关中枢神经系统疾病三例报告并文献复习

桂瑞瑞 祖璎玲 张奘莉 韩利杰 赵慧芳 李珍 喻凤宽 王娟 赵娟娟
符粤文 宋永平 周健
郑州大学附属肿瘤医院(河南省肿瘤医院)血液科,河南省血液病研究所,郑州 450008
通信作者:周健,Email:zhoujiandoctor@163.com
DOI:10.3760/cma.j.issn.0253-2727.2019.04.014

Successful treatment of Epstein- Barr virus associated central nervous system diseases after allogeneic hematopoietic stem cell transplantation with intrathecal donor lymphocytes infusion: three cases report and literatures review

Gui Ruirui, Zu Yingling, Zhang Yanli, Han Lijie, Zhao Huifang, Li Zhen, Yu Fengkuan, Wang Juan, Zhao Juanjuan, Fu Yuewen, Song Yongping, Zhou Jian
Department of Hematology Affiliated Cancer Hospital Zhengzhou University, Henan Tumor Hospital, Institute of Hematology, Zhengzhou 450008, China
Corresponding author: Zhou Jian, Email: zhoujiandoctor@163.com

异基因造血干细胞移植(allo-HSCT)患者免疫功能低下,容易发生EB病毒(EBV)再激活。EBV感染可导致罕见且危及生命的中枢神经系统(CNS)疾病。尽管EBV相关CNS疾病的发病率较低,但其病死率很高,目前尚缺乏行之有效的治疗手段。近期,我们采用鞘内注射供者淋巴细胞(intrathecal donor lymphocyte infusion, IDLI)成功治疗3例allo-HSCT后EBV相关CNS疾病。

病例资料

例1,男,26岁,因"再生障碍性贫血"行allo-HSCT。供者为其胞弟(HLA配型6/6全相合)。预处理:全身放疗(TBI)+氟达拉滨(FLU)+环磷酰胺(CTX)+抗胸腺细胞球蛋白(ATG)。移植物抗宿主病(GVHD)预防:环孢素A(CsA)+霉酚酸酯(MMF)联合短程甲氨蝶呤(MTX)。回输胞弟G-CSF动员的骨髓和外周血造血干细胞[单个核细胞(MNC)13.40×10^8/kg、$CD34^+$细胞3.72×10^6/kg]。移植后14 d(+14 d)粒系重建,+20 d巨核系重建,无明显GVHD发生,+30 d检测骨髓细胞嵌合体为完全供者型。+37 d外周血巨细胞病毒(CMV)DNA(CMV-DNA)1.59×10^4拷贝/L,EBV-DNA阴性,更昔洛韦联合静脉丙种球蛋白抗病毒治疗后CMV-DNA转阴。+54 d出现高热(体温40.0 ℃),颈部双侧可触及多个花生米至枣样大小淋巴结(质韧、活动度差),扁桃体Ⅱ度肿大,血EBV-DNA 1.66×10^5拷贝/L;颈部淋巴结活检"考虑多形性移植后淋巴细胞增殖性疾病(post-transplantation lymphoproliferative diseases, PTLD)"。给予免疫抑制剂减量、抗病毒(阿昔洛韦、阿糖腺苷)、静脉丙种球蛋白、利妥昔单抗、供者淋巴细胞输注(DLI)及强力抗生素治疗。体温控制,颈部淋巴结明显缩小或消失,全身症状好转,PTLD控制。+60 d再次发热伴癫痫发作,头颅磁共振(MRI)示"小脑和脑室周围炎性改变",血CMV-DNA阴性,EBV-DNA 1.21×10^4拷贝/L;腰穿脑脊液:无色透明,未见凝块,潘氏试验阴性,未见异常细胞;流式细胞术免疫分型未见异常,细菌培养阴性,墨汁染色和抗酸染色均阴性,G试验和GM试验均阴性,EBV-DNA 1.20×10^4拷贝/L,CMV-DNA阴性。诊断为CNS-PTLD。予利妥昔单抗50 mg鞘内注射后神志恢复,继续减停免疫抑制剂和抗病毒治疗,予以IDLI每周2次×6次[MNC 4.67(3.82~6.02)×10^6,地塞米松2.5 mg],全部鞘注过程顺利,未发生不良反应;同时予DLI每周2次×6次,中位MNC 8.83(6.79~10.47)×10^7/kg。+68 d血CMV-DNA、EBV-DNA均阴性,脑脊液EBV-DNA阴性,神经系统症状消失,4周后复查头颅MRI未见异常。随访至移植后52个月,生存良好。

例2,女,13岁,因"再生障碍性贫血"行allo-HSCT。供者为其胞弟(HLA配型6/6全相合)。预处理方案:FLU+CTX+ATG。GVHD预防:CsA、MMF联合短程MTX。回输胞弟G-CSF动员的骨髓和外周血造血干细胞(MNC 19.7×10^8/kg,$CD34^+$细胞7.26×10^6/kg)。+13 d粒系重建,+16 d巨核系重建,无明显GVHD表现。+21 d血CMV-DNA 3.71×10^3拷贝/L,给予更昔洛韦抗病毒治疗。+28 d、+61 d骨髓细胞嵌合体为100%供者型。+63 d血CMV-DNA 1.51×10^4拷贝/L,EBV-DNA 7.41×10^4拷贝/L,给予更昔洛韦联合膦甲酸钠抗病毒治疗。+66 d发热(体温38.9 ℃),强力抗细菌治疗体温未降,C反应蛋白(CRP)33.5 mg/L,血小板计数较前下降。查体:颈部、腋窝及腹股沟浅表淋巴结肿大(质韧,活动

度可),胸部CT检查未见明显异常。+68 d行颈部淋巴结活检,病理和免疫组化"符合单形性PTLD(B细胞淋巴瘤)"。停用免疫抑制剂,给予阿昔洛韦、膦甲酸钠及输脉丙种球蛋白输注。每周3次输注低剂量DLI[MNC (1~3)×10^6/kg],输注5次后体温逐渐下降至37.5 ℃,血常规指标逐渐恢复,淋巴结逐渐缩小。+81 d复查血CMV-DNA和EBV-DNA均为阴性,CRP 10 mg/L。+116 d出现癫痫发作,血常规显示全血细胞减少,CRP正常,体温正常;查体:颈软,病理征阴性;颅脑MRI:脑内多发异常信号,松果体囊肿;血CMV-DNA 2.09×10^3拷贝/L,EBV-DNA阴性。腰椎穿刺脑脊液:微浊,细胞30×10^6/L,潘氏试验阴性;LDH 17 U/L,总蛋白0.537 g/L,未见异常细胞;流式细胞术免疫分型未见异常,墨汁染色和抗酸染色均阴性,G实验和GM实验均阴性,脑脊液EBV-DNA 2.75×10^3拷贝/L,CMV-DNA阴性。诊断为CNS-PTLD。+120 d开始予IDLI[MNC 5.14(4.32~6.47)×10^6,地塞米松2.5 mg]每周2次×7次,全部鞘注过程顺利,未发生不良反应。+125 d脑脊液EBV-DNA转阴,+133 d头颅MRI:原脑内多发异常信号基本消失。+165 d再次出现低热,伴食欲下降、恶心、间断呕吐,CRP 30 mg/L,抗细菌治疗无效,症状进行性加重,血象逐渐下降。颅脑增强MRI无异常,+168 d腰穿脑脊液EBV-DNA 5.49×10^3拷贝/L,CMV-DNA阴性,提示CNS PTLD复发,再次给予IDLI每周2次×5次并静脉滴注阿昔洛韦抗病毒治疗,1周后恶心、呕吐消失,体温正常。随访至移植后50个月,生存良好。

例3,男,42岁,因"急性髓系白血病-M_5(伴FLT3-ITD基因突变)"行allo-HSCT,入院评估病情为第3次完全缓解。供者为胞弟(HLA配型6/6全相合)。预处理采用Bu-Cy方案(白消安+环磷酰胺),回输MNC 15.54×10^8/kg,$CD34^+$细胞4.14×10^6/kg。给予CsA联合短程MTX预防GVHD。+14 d粒细胞重建。+22 d外周血CMV-DNA 1.71×10^4拷贝/L,EBV-DNA阴性,予阿糖腺苷联合更昔洛韦抗病毒治疗。+36 d外周血CMV-DNA 1.15×10^4拷贝/L,EBV-DNA阴性,予静脉丙种球蛋白输注后CMV-DNA转阴。+53 d出现轻微头痛、高血压,外周血EBV-DNA、CMV-DNA均阴性,腰椎穿刺脑脊液常规、生化、压力均正常,未见异常细胞,流式细胞术免疫分型未见异常,细菌培养阴性,墨汁染色和抗酸染色均阴性,G试验和GM试验均阴性;EBV-DNA 4.26×10^3拷贝/L,CMV-DNA阴性;MRI未见异常。诊断为"EBV脑炎"。予以减停免疫抑制剂、抗病毒治疗及给予IDLI(每周2次),IDLI 3次后脑脊液EBV-DNA转阴,头痛症状减轻。后续予IDLI巩固治疗4次(每周2次),全部7次IDLI的MNC为4.16(3.46~6.21)×10^6。随访至移植后46个月,生存良好。

讨论及文献复习

随着多种强效免疫抑制剂的出现和HLA配型技术的不断完善,替代供者allo-HSCT取得了快速进步[1]。强效免疫抑制剂可有效防治GVHD,极大推动了allo-HSCT的发展[2-3]。但患者免疫功能强烈抑制导致移植后EBV的感染率明显增加[4]。除了淋巴结,EBV亦可感染上皮细胞和神经细胞[5]。EBV感染CNS可引起EBV相关CNS疾病,包括EBV脑炎、EBV脊髓炎和EBV相关CNS-PTLD。移植后EBV脑炎发生率仅为0.23%,占所有病毒性脑炎的19%,但死亡率却高达83%[6]。EBV相关PTLD是allo-HSCT后致命的并发症,发生率为0.2%~22%[7-8],10%~15%的PTLD累及CNS[9]。

CNS-PTLD病情凶险,进展迅速,预后极差[10]。移植后EBV相关CNS疾病无特异性临床表现,很难早期发现和诊断[11]。例1主要表现为癫痫发作;例2表现为癫痫发作、恶心、呕吐;例3无典型CNS症状,仅表现为轻微头痛和高血压。确诊CNS-PTLD需经过病理证实,因血小板减少等原因,临床上开展脑组织活检极为困难,前2例病例均为临床诊断。此外早期诊断困难的另一原因为多样化的影像学改变[12]。例2头颅CT未发现异常,但MRI却发现异常信号,提示MRI比CT更敏感,这与文献[13]报道一致。例3颅脑MRI未见异常,可能与发病早期临床症状不明显有关。我们推测,EBV感染到EBV脑炎、脊髓炎,再到EBV相关CNS-PTLD是疾病由轻到重发展过程中的不同阶段,EBV相关CNS疾病的临床表现和所处阶段可能与受累部位和EBV负荷量有关,早期无明显CNS症状和相应体征时容易漏诊。

检测血EBV-DNA是诊断移植后EBV感染的常用方法。但外周血与脑脊液EBV-DNA检测结果分离现象。本研究例2和例3发生EBV相关CNS疾病时,外周血EBV-DNA均阴性;例2 EBV相关CNS疾病发生于系统性PTLD有效控制且外周血EBV-DNA转阴时。上述现象与文献[14-15]报道结果一致。Barberi等[16]报道在一些EBV脑炎患者的脑脊液中并未检出EBV-DNA。因此,在临床工作中,不能因为患者血EBV-DNA阴性即排除EBV相关CNS疾病,同样也不能因脑脊液EBV-DNA阴性即排除EBV相关CNS疾病。

利妥昔单抗治疗移植后EBV相关性疾病疗效较好,Coppoletta等[17]应用利妥昔单治疗allo-HSCT后EBV血症,91%的患者EBV-DNA转阴。由于利妥昔单抗不能透过血脑屏障,只能通过鞘内注射方式治疗EBV相关CNS疾病。尽管鞘内注射利妥昔单抗有效改善了allo-HSCT后EBV相关CNS疾病的预后[18],但其疗效依赖于感染EBV的B细胞表面CD20的表达水平,且复发率高。此外,昂贵的费用限制了其临床使用。本研究例1患者利妥昔单抗联合小剂量DLI有效控制了系统性EBV相关性PTLD,但并未能预防随后发生的EBV相关CNS疾病。

全身应用小剂量DLI不能在CNS中达到有效治疗浓度,而IDLI则能解决这一难题。IDLI治疗T淋巴细胞白血病/淋巴瘤allo-HSCT后CNS复发的安全性已得到验证[19-20]。本组3例患者在鞘注过程和后续随访中均未观察到不良反应,初步证实IDLI治疗EBV相关CNS疾病是安全的。

利妥昔单抗鞘内注射治疗EBV相关CNS疾病起效快但治疗效应持续时间短[21],IDLI起效慢但作用持久。IDLI联合利妥昔单抗鞘内注射治疗EBV相关CNS疾病值得探索。

参考文献

[1] Ljungman P, Urbano-Ispizua A, Cavazzana-Calvo M, et al. Allogeneic and autologous transplantation for haematological diseases, solid tumours and immune disorders: definitions and current practice in Europe[J]. Bone Marrow Transplant, 2006, 37(5): 439-449. DOI: 10.1038/sj.bmt.1705265.

[2] Messina C, Faraci M, de Fazio V, et al. Prevention and treatment of acute GVHD [J]. Bone Marrow Transplant, 2008, 41(2): 65-70.

[3] Locatelli F, Bernardo ME, Bertaina A, et al. Efficacy of two different doses of rabbit anti-T-lymphocyte globulin to prevent graft-versus-host disease in children with haematological malignancies transplanted from an unrelated donor: a multicentre, randomised, open-label, phase 3 trial[J]. Lancet Oncol, 2017, 18(8):1126-1136. DOI: 10.1016/S1470-2045(17)30417-5.

[4] Kawa K, Sawada A, Sato M, et al. Excellent outcome of allogeneic hematopoietic SCT with reduced-intensity conditioning for the treatment of chronic active EBV infection[J]. Bone Marrow Transplant, 2011, 46(1):77-83. DOI: 10.1038/bmt.2010.122.

[5] van Esser JW, van der Holt B, Meijer E, et al. Epstein-Barr virus (EBV) reactivation is a frequent event after allogeneic stem cell transplantation (SCT) and quantitatively predicts EBV-lymphoproliferative disease following T-cell-depleted SCT[J]. Blood, 2001, 98(4):972-978. DOI: https://doi.org/10.1182/blood.V98.4.972.

[6] Schmidt-Hieber M, Schwender J, Heinz WJ, et al. Viral encephalitis after allogeneic stem cell transplantation: a rare complication with distinct characteristics of different causative agents[J]. Haematologica, 2011, 96(1): 142-149. DOI: 10.3324/haematol.2010.029876.

[7] Styczynski J, Reusser P, Einsele H, et al. Management of HSV, VZV and EBV infections in patients with hematological malignancies and after SCT: guidelines from the Second European Conference on Infections in Leukemia [J]. Bone Marrow Transplant, 2009, 43(10):757-770. DOI: 10.1038/bmt.2008.386.

[8] Rouce RH, Louis CU, Heslop HE. Epstein-Barr virus lymphoproliferative disease after hematopoietic stem cell transplant[J]. Curr Opin Hematol, 2014, 21(6):476-481. DOI: 10.1097/MOH.0000000000000083.

[9] Evens AM, Roy R, Sterrenberg D, et al. Post-transplantation lymphoproliferative disorders: diagnosis, prognosis, and current approaches to therapy[J]. Curr Oncol Rep, 2010, 12(6):383-394. DOI: 10.1007/s11912-010-0132-1.

[10] Moosmann A, Bigalke I, Tischer J, et al. Effective and long-term control of EBV PTLD after transfer of peptide-selected T cells [J]. Blood, 2010, 115(14): 2960-2970. DOI: 10.1182/blood-2009-08-236356.

[11] Aimoto M, Yamane T, Inoue A, et al. Epstein-Barr virus-associated post-transplant lymphoproliferative disorder diagnosed by the episode of intestinal perforation following allogeneic hematopoietic stem cell transplantation[J]. Rinsho Ketsueki, 2010, 51(12):1775-1780.

[12] Ginat DT, Purakal A, Pytel P. Susceptibility-weighted imaging and diffusion-weighted imaging findings in central nervous system monomorphic B cell post-transplant lymphoproliferative disorder before and after treatment and comparison with primary B cell central nervous system lymphoma[J]. J Neurooncol, 2015, 125(2): 297-305. DOI: 10.1007/s11060-015-1903-1.

[13] Abul-Kasim K, Palm L, Maly P, et al. The neuroanatomic localization of Epstein-Barr virus encephalitis may be a predictive factor for its clinical outcome: a case report and review of 100 cases in 28 reports[J]. J Child Neurol, 2009, 24(6):720-726. DOI: 10.1177/0883073808327842.

[14] Shimizu H, Saitoh T, Koya H, et al. Discrepancy in EBV-DNA load between peripheral blood and cerebrospinal fluid in a patient with isolated CNS post-transplant lymphoproliferative disorder [J]. Int J Hematol, 2011, 94(5):495-498. DOI: 10.1007/s12185-011-0951-3.

[15] Terasawa T, Ohashi H, Tsushita K, et al. Failure to detect Epstein-Barr virus (EBV) DNA in plasma by real-time PCR in a case of EBV-associated posttransplantation lymphoproliferative disorder confined to the central nervous system[J]. Int J Hematol, 2002, 75(4):416-420.

[16] Barberi W, Perrone S, Iori AP, et al. Proven Epstein-Barr encephalitis with negative EBV-DNA load in cerebrospinal fluid after allogeneic hematopoietic stem cell transplantation in a child with acute lymphoblastic leukemia[J]. Pediatr Transplant, 2015, 19(1): E19-24. DOI: 10.1111/petr.12386.

[17] Coppoletta S, Tedone E, Galano B, et al. Rituximab treatment for Epstein-Barr virus DNAemia after alternative-donor hematopoietic stem cell transplantation[J]. Biol Blood Marrow Transplant, 2011, 17(6): 901-907. DOI: 10.1016/j.bbmt.2010.10.003.

[18] Czyzewski K, Styczynski J, Krenska A, et al. Intrathecal therapy with rituximab in central nervous system involvement of post-transplant lymphoproliferative disorder [J]. Leuk Lymphoma, 2013, 54(3):503-506. DOI: 10.3109/10428194.2012.718342.

[19] Meuleman N, Ahmad I, Duvillier H, et al. Intrathecal donor lymphocyte infusion for the treatment of suspected refractory lymphomatous meningitis: a case report[J]. Eur J Haematol, 2006, 77(6):523-526. DOI: 10.1111/j.0902-4441.2006.t01-1-EJH2498.x.

[20] Yanagisawa R, Nakazawa Y, Sakashita K, et al. Intrathecal donor lymphocyte infusion for isolated leukemia relapse in the central nervous system following allogeneic stem cell transplantation: a case report and literature review[J]. Int J Hematol, 2016, 103(1):107-111. DOI: 10.1007/s12185-015-1902-1.

[21] Sato E, Ohga S, Kuroda H, et al. Allogeneic hematopoietic stem cell transplantation for Epstein-Barr virus-associated T/natural killer-cell lymphoproliferative disease in Japan [J]. Am J Hematol, 2008, 83(9):721-727. DOI: 10.1002/ajh.21247.

（收稿日期：2018-08-22）
（本文编辑：徐茂强）