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Rabbit Antithymocyte Globulin Versus Alemtuzumab in Allogeneic Stem Cell Transplantation： A Meta-Analysis

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***Abstract*— Rabbit antithymocyte globulin (ATG) and alemtuzumab have been used for graft-versus-host disease(GVHD) prophylaxis in allogeneic haematopoietic stem cell transplantation(allo-HSCT), but which is more efficient remain unclear.we perform a meta-analysis of all studies comparing rabbit ATG and alemtuzumab for GVHD prophylaxis in allogeneic haematopoietic stem cell transplantation（HSCT） to evaluate their benefits and drawbacks. There are 7 studies (one prospective and six retrospective)for comparing rabbit ATG vs alemtuzumab in GVHD prophylaxis with 622 patients. Our results showed that the incidence of grade II-IV acute GVHD (aGVHD) (RR 1.51, 95% CI 0.97–2.34, P = 0.07), incidence of grade III-IV acute GVHD (RR 1.48, 95% CI 0.63–3.47, P = 0.37) had a statistically non-significant reduction in alemtuzumab group, however, alemtuzumab significantly impaired OS (HR 0.61 (95% CI 0.41–0.90, P = 0.01) compared with ATG. The incidence of overall chronic GVHD(RR 0.97, 95% CI 0.67–1.40, P = 0.87) and the incidence of relapse(RR 1.03, 95% CI 0.72–1.47, P = 0.88) were similar in the two groups. We propose that using alemtuzumab for GVHD prophylaxis is effective for allogeneic stem cell transplantation due to the efficacy in grade III/IV acute GVHD, but OS is impaired compared with ATG group.**

***Index Terms*— Haematopoietic Stem Cell Transplantation, ATG, Alemtuzumab, GVHD, Meta-Analysis.**

I. INTRODUCTION

Haematopoietic stem cell transplantation (HSCT) is a potentially curative treatment for patients with haematological malignancies, bone marrow failure syndrome, and some inherited metabolic disorders. However, GVHD is still a major cause of morbidity and mortality[1] 、[2]. Serotherapy, which consists of antithymocyte globulin (ATG), a polyclonal antibody, or the monoclonal antibody alemtuzumab, is normally used for GVHD prophylaxis. ATG is a polyclonal immunoglobulin preparation obtained by immunization of rabbits or horses with human thymocytes or T cell lines. Alemtuzumab is a humanized monoclonal antibody specific for CD52. In the HSCT setting, both types of serotherapy are used to eliminate T cells, but they also target other cell types, e.g., B cells and natural killer cells [3]-[7]. ATG and alemtuzumab are considered to be instrumental in reducing the risk of rejection by suppressing the reaction of host T cells against the graft. However, due to their long half-lives, the antibodies will usually remain present after transplantation and eliminate the donor T cells infused with the graft as well. In this manner, these antibodies not only reduce the occurrence of GVHD [4]、[8]、[10]-[12], but they may also have a negative impact on the occurrence of a graft-versus leukemia effect [9]、12]. Alemtuzumab has a longer half-life (15 to 21 days) than that of ATG (4 to 14 days). This loner half-life leads to a more prolonged effect on lymphocyte recovery [13]、[14]-[16]. We conducted a systematic review, which included all studies available that compared rabbit ATG and alemtuzumab in GVHD prophylaxis before allo-HSCT. The objective of this study was to compare the effects of rabbit ATG with those of alemtuzumab, as part of the conditioning regimen after allo-HSCT, on various outcome parameters after allo-HSCT, i.e., GVHD, relapse and survival.

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II. METHODS

A. Inclusion criteria

The inclusion criteria consisted of (i)participants without the restrictions of special, irrespective of age, disease, graft source and donor; (ii) studies that compared the prophylactic use of rabbit ATG and alemtuzumab in patients; (iii) allo-HSCT for haematological diseases (irrespective of disease stage); (iv) use antithymocyte or alemtuzumab in any dose or given duration; (v) studies that reported data and included details regarding demographic, transplant, treatment, and outcome variables.

B. Literature search

Two authors independently conducted an electronic literature search in Pubmed, EMBASE and MEDLINE, we used a broad search strategy to identify both prospective and retrospective clinical studies evaluating the outcome of prophylactic rabbit ATG and alemtuzumab use in allo-HSCT from January 2000 to March 2015. Studies earlier than 2000 were not included because considerable progress in supportive care and antibody manufacturing has been made during the recent 10 years. The search was conducted in March 2015. We performed manual selection by reading the abstracts, and further selection by reading full texts. The experts in the field and the authors of published studies were not contacted. To reduce potential language bias, language restriction was not applied .

C. Outcomes

The outcomes were extracted regarding the benefits and drawbacks associated with the treatments. Their included (1) OS; (2) GVHD incidence; and (3) relapse.

D. Study selection, quality assessment, and data extraction

Two reviewers independently readed the titles and abstracts of all studies identified through the literature search to determine whether they met the predefined inclusion criteria. The same set of reviewers extracted data regarding the benefits (OS, GVHD incidence and relapse) associated with the compared treatments using a standardized data extraction form, according to the guidelines proposed by The Cochrane Collaboration. The full search strategy and data selection are described in Fig.1. The quality assessment strongly depended upon information regarding the design, performance and analysis of the trial. The quality of the study was evaluated using ratings on the Newcastle-Ottawa Scale [17] .

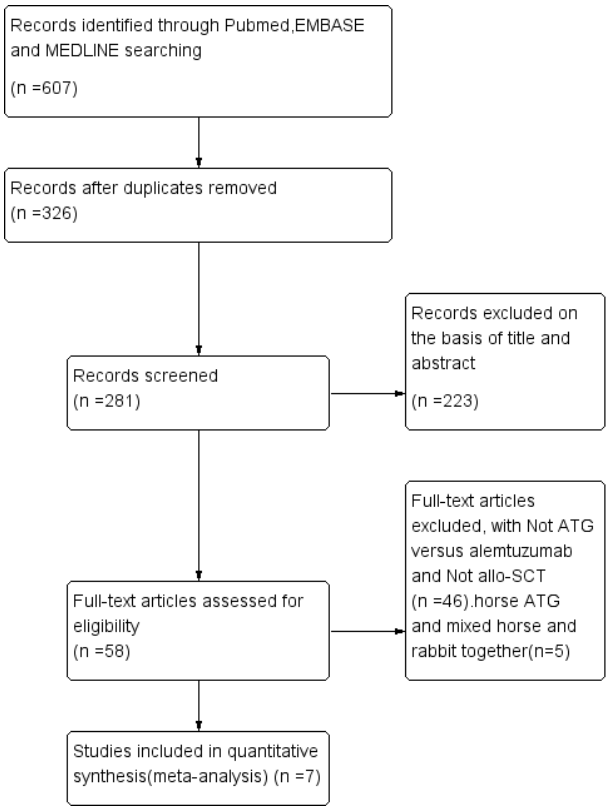

Fig.1. Flow Diagram Showing the Process of Identifying and Selecting Relevant Studies

E. Data analysis and statistical methods

Time-to-event data(OS) was pooled and was reported as hazard ratios (HR), meanwhile, dichotomous data (incidence of GVHD, relapse were pooled and are reported as risk ratios (RR)[18], with 95% confidence intervals (CI) under a random effects model,respectively[19]. We extracted these data according to the method described by Tierney et al[20] if time-to-event were unavailable for direct extraction. This method needs calculation of the hazard ratio from different parameters using indirect calculation of the variance and the number of observed events minus the number of expected events. A formal statistical test for heterogeneity using the chi square [19] and I2 [21] tests was conducted（I2 >30%, moderate heterogeneity; I2 >75%, severe heterogeneity）. This meta-analysis was performed using Review Manager Software (Review Manager, Version 5.3, The Cochrane Collaboration 2014, Copenhagen).

III.RESULTS

Table 1:Characteristics of the studies included into the meta-analysis

Author year	No.of pts	Median age(range)	Primary diagnosis	Graft source	Donor status	Dose
	ATG/Ale	ATG/Ale	ATG(%)/Ale(%)	ATG/Ale	ATG/Ale	ATG or Ale
Willemsen 2015[22]	110/38	7.8(0.4-18.6)/13.3(3.9-19.0)	Malignant disese 58%/71% nonmalignant-disease 42%/29%	85%BM+15%PBSC/79%BM+21%PBSC	RD18%/26% UD 82%/74%	ATG-T:cumulative dose of 10mg/kg Ale:cumulative dose of 1mg/kg
Busemann 2013[23]	20/19	47(20-67)/52(20-70)	Acute leukemia 50%/50%	100%PBSC/100%PBSC	UD100%100%	ATG-F:30mg/kg or ATG-T:6mg/kg Ale:10mg or 20mg
Marsh 2014[24]	55/100	21(2-57/18(1-67)	SAA 100%/100%	65%BM+27%PBSC/75%BM+20%PBSC	RD43%/87% UD55%/13%	ATG-T:median total dose 11.25mg/kg Ale:median total dose 50mg
Park 2009[25]	18/12	31(16-52)/37.5(17-68)	ALL,AML 50%/91.7% MDS,CML 50%/8.7%	NR/NR	RD50%/75% UD50%/25%	Rabbit-ATG:dose 2.5to 10mg/kg for 3 days Ale:total dose 10mg to 100mg
Juliusson 2006[26]	29/40	54(44-59)/52.6(40-58)	malignant 93.1%/100% nonmalignant 6.9%/0%	NR/NR	RD65.5%/47.5% UD34.5%/52.5%	ATG-F:30mg/kg or ATG-T:6mg/kg Ale:10mg or 20mg
Kroger 2005[27]	48/25	50(32-62)/47(33-60)	MM 100%/100%	19%BM+81%PBSC/72%BM+28%PBSC	NR/NR	ATG-F:median dose 60mg/kg Ale:total dose 100mg
Norlin 2011[28]	72/36	52(2-67)/55(1-67)	malignant 93.1%/91.7 nonmalignant 6.9%/8.3%	20.8%BM+79.2%PBSC/11%BM+81%PBSC	RD26.4%/36.1% UD73.6%/63.9%	ATG-T:total dose 6mg/kg or 8mg/kg Ale:total dose 30mg or 60mg or 90mg

Ale:alemtuzumab;NR: not report;RD:related donor;UD:unrelated donor;BM:bone marrow;PBSC:peripheral blood stem cells ATG-F:Fresenius,ATG-T:Thymoglobin

Seven studies that enrolled a total of 622 patients from 2005 to 2015 were included in this meta-analysis. Table 1 summarizes the details related to patient characteristics. The assessment of study quality is shown in table 2.

A. Overall survival (OS)

Kaplan–Meier plots of overall survival were available in 6 out of the 7 studies. A total of 553 patients were analysed, and the pooled HR showed a statistically significant benefit for OS with the use of rabbit ATG for the prevention of GVHD (fig.2.). The pooled hazard ratio for the comparison of rabbit ATG versus alemtuzumab was 0.61 (95% CI 0.41–0.90, P = 0.01). The heterogeneity was low among the studies (I2 =0%).

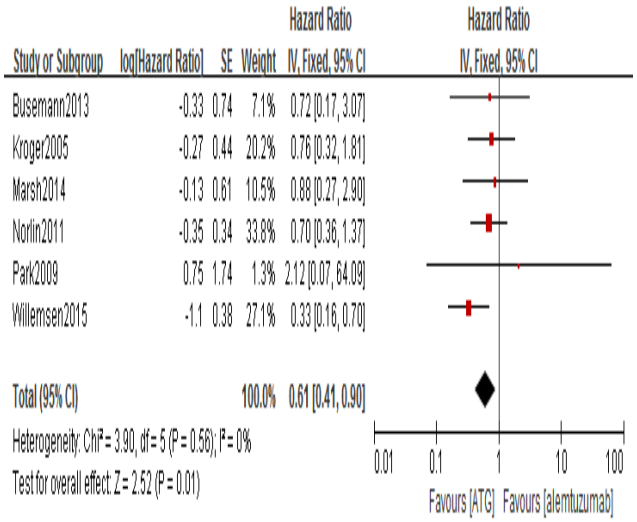

Fig.2:Forest plot of overall survival.

B. Incidence of aGVHD and chronic GVHD

Regarding the incidence of grade II-IV acute GVHD(aGVHD) (Fig.3.a), 398 patients from 4 studies, showed a statistically non-significant reduction in the alemtuzumab arm (RR 1.51, 95% CI 0.97–2.34, P = 0.07). The heterogeneity was low among the studies (I2 =0%). Regarding the incidence of grade III-IV aGVHD (Fig.3.b), 435 patients from 5 studies showed a statistically non-significant reduction in the alemtuzumab arm (RR 1.48, 95% CI 0.63–3.47, P = 0.37). The heterogeneity was low among the studies (I2 =0%). Regarding the incidence of overall chronic GVHD (Fig.3.c), 514 patients from 5 studies showed a statistically non-significant effect with rabbit ATG versus alemtuzumab (RR 0.97, 95% CI 0.67–1.40, P = 0.87). The heterogeneity was low among the studies (I2 =26%).

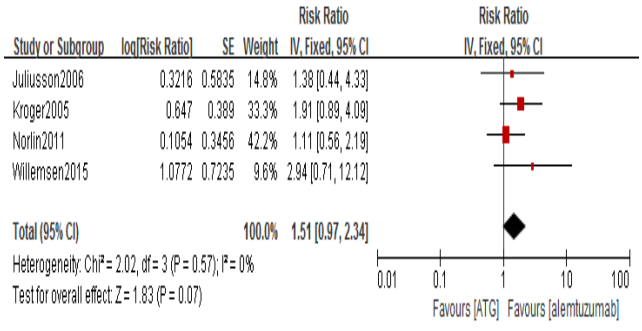

Fig.3(a):Forest plot of acute GVHD gradeII-IV

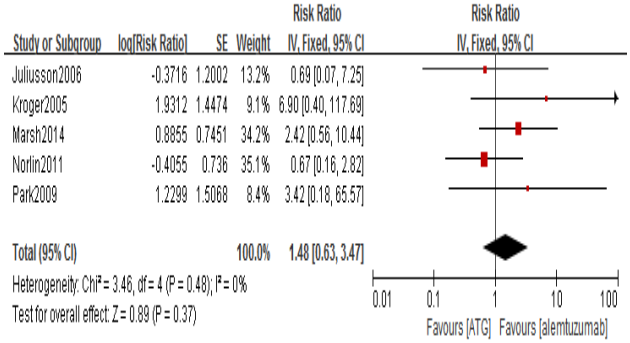

Fig.3(b):Forest plot of acute GVHD gradeIII-IV.

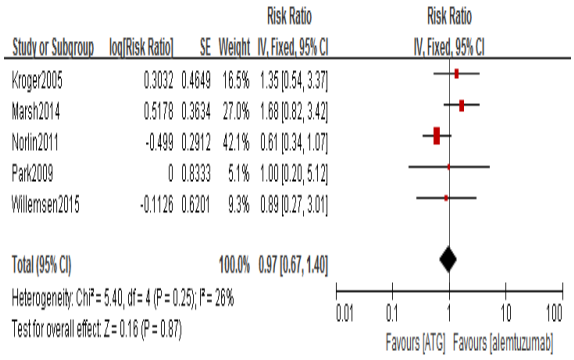

Fig.3(c): Forest plot of chronic GVHD

C. Relapse

Regarding the incidence of relapse (Fig.4.), 359 patients from 4 studies showed a statistically non-significant effect with rabbit ATG versus alemtuzumab (RR 1.03, 95% CI 0.72–1.47, P = 0.88). The statistically non-significant heterogeneity was low among the studies (I2 =30%).

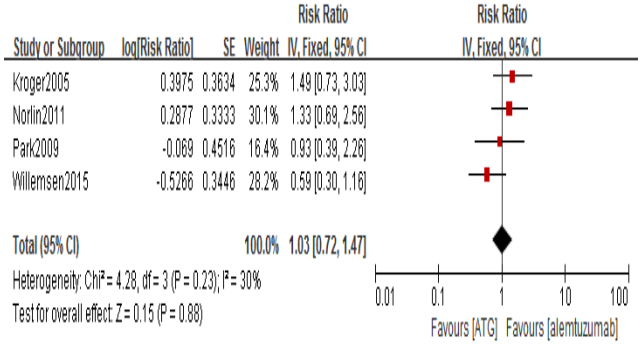

Fig.4: Forest plot of relapse

Table 2:Assessment of Study Quality

Author year	Quality Indicators From Newcastle-Ottawa Scale[17]								
	1	2	3	4	5A	5B	6	7	8
Willemsen2015[22]	Yes	No	No	Yes	Yes	Yes	Yes	Yes	No
Busemann2013[23]	Yes	Yes	No	Yes	Yes	Yes	Yes	Yes	No
Marsh2014[24]	Yes	No	No	Yes	Yes	Yes	Yes	Yes	No
Park2009[25]	Yes	No	No	Yes	Yes	Yes	Yes	Yes	No
Juliusson2006[26]	Yes	Yes	Yes	No	Yes	Yes	Yes	Yes	Yes
Kroger2005[27]	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No
Norlin2011[28]	Yes	Yes	No	Yes	Yes	Yes	Yes	Yes	No

*For case-control studies[22]-[25] · [27]-[28],1,indicates cases independently validated;2,consecutive or obviously representative series of cases;3, community controls;4, disease control;5A,study control for age;5B,study controls for additional factor(s);6, Ascertainment of exposure by secure record;7,same method of ascertainment used for cases and controls;8,Non-Response rate the same for the same cases and controls.For cohort studies[26],1,indicates exposed cohort truly representative;2,nonexposed cohort drawn from the same community;3 Ascertainment of exposure;4,outcome of interest not present at start;5A,cohorts comparable on basis of age;5B,cohorts comparable on other factor(s);6,quality of outcome assessment;7,follow-up long enough for outcomes to occur;8,complete accounting for cohort

IV.DISCUSSION

GVHD remains a serious clinical complication for patients undergoing allo-HSCT for haematological diseases. This systematic review and meta-analysis is the first to evaluate the efficacy of rabbit ATG versus alemtuzumab in GVHD prophylaxis before infusion of stem cells. Seven studies met the inclusion criteria. The pretreatment, basic diseases, stem cell source, donor type, age, follow-up times, doses of rabbit ATG, and dose of alemtuzumab were different between studies, but the rabbit ATG and alemtuzumab groups in each study were roughly consistent in several aspects. The baselines were comparable, therefore, the results are relatively reliable.

Our outcomes showed no obvious advantage of alemtuzumab in reducing grade III-IV aGVHD and grade II-IV aGVHD, and OS has not been improved in alemtuzumab group, these outcomes might be related to slow immune reconstitution and high mixed donor chimerism in alemtuzumab group, so fungal infections and fatal bacterial infections were more frequent, OS was impaired. We just chose rabbit ATG and did not horse ATG or mixed ATG to reduce heterogeneity, and use the incidence of initial aGVHD for statistical analysis because we thinked aGVHD after donor lymphocyte infusion(DLI) was induced by treating residual disease and/or mixed chimaerism and when the antibody has lose efficacy in human body in either sero-therapies. Busemann et al[23] studied alemtuzumab compared with the rabbit ATG/no AB, and used rabbit ATG in unrelate or mismatched transplantation but no antibody in HLA-identical sibling transplantation, so we only could extract OS between unrelated donor in two sero-therapies.and Juliusson et al[26] analyzed rabbit ATG, alemtuzumab-30mg and alemtuzumab-90mg, we could not extract OS between rabbit ATG and alemtuzumab, in this study, OS was excellent in the rabbit ATG-treated group, and the same in alemtuzumab-30mg, but impaired in the alemtuzumab-90mg group.

In conclusion, our results demonstrate that, whereas alemtuzumab may be effective in preventing grade III-IV aGVHD and grade II-IV aGVHD.The incidence of overall chronic GVHD and the incidence of relapse were similar in two group. However, OS has been impaired in alemtuzumab. Our meta-analysis is mainly based on retrospective studies with a lack of internal validity, and the sample size is relatively small. Therefore, the conclusions should be interpreted with caution; more sufficient evidence is needed from future prospective randomized trials based on the type of transplant, dose of antibidy, pretreatment, age of patient and disease types. Nevertheless, we propose that using alemtuzumab for GVHD prophylaxis is effective for allo-HSCT due to the efficacy in grade III/IV aGVHD and grade II-IV aGVHD, but OS is impaired compared with rabbit ATG group, how to speed up immune reconstitution, increase full donor chimerism rate and decrease infection rate still need great efforts.

CONFLICTS OF INTEREST

Authors declare that they have no conflicts of interests.

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REFERENCES

[1] Hansen JA, Gooley TA, Martin PJ, Appelbaum F, Chauncey TR, Clift RA,et al. Bone marrow transplants from unrelated donors for patients with chronic myeloid leukemia. N Engl J Med. 1998; 338：962-968..

[2] Sierra J, Storer B, Hansen JA, Martin PJ, Petersdorf EW, Woolfrey A, et al. Unrelated donor marrow transplantation for acute myeloid leukemia: an update of the Seattle experience. Bone Marrow Transplant. 2000; 26：397-404

[3] Bosch M, Dhadda M, Hoegh-Petersen M, Liu Y, Hagel LM, Podgorny P, et al. Immune reconstitution after anti-thymocyte globulin-conditioned hematopoietic cell transplantation. Cytotherapy. 2012; 14：1258-1275.

[4] Hoegh-Petersen M, Amin MA, Liu Y, Ugarte-Torres A, Williamson TS, Podgorny PJ, et al. Anti-thymocyte globulins capable of binding to T and B cells reduce graft-vs-host disease without increasing relapse. Bone Marrow Transplant. 2013; 48：105-114..

[5] Mohty M. Mechanisms of action of antithymocyte globulin: T cell depletion and beyond. Leukemia. 2007;21:1387-1394.

[6] Na IK, Wittenbecher F, Dziubianau M, Herholz A, Mensen A, Kunkel D, et al. Rabbit antithymocyte globulin (thymoglobulin) impairs the thymic output of both conventional and regulatory CD4+ T cells after allogeneic hematopoietic stem cell transplantation in adult patients. Haematologica. 2013; 98：23-30.

[7] Stauch D, Dernier A, Sarmiento ME, Kunert K, Volk HD, Pratschke J et al. Targeting of natural killer cells by rabbit antithymocyte globulin and campath-1H: similar effects independent of specificity. PLoS One. 2009; 4：e4709.

[8] Veys P, Wynn RF, Ahn KW, Samarasinghe S, He W, Bonney D, et al. Impact of immune modulation with in vivo T-cell depletion and myleoablative total body irradiation conditioning on outcomes after unrelated donor transplantation for childhood acute lymphoblastic leukemia. Blood. 2012; 119：6155-6161.

[9] Copelan EA. Hematopoietic stem-cell transplantation. N Engl J Med. 2006;354:1813-1826.

[10] Ayuk F, Diyachenko G, Zabelina T, Wolschke C, Fehse B, Bacher U, et al. Comparison of two doses of antithymocyte globulin in patients undergoing matched unrelated donor allogeneic stem cell transplantation. Biol Blood Marrow Transplant. 2008; 14：913-919.

[11] Jol-van DZC, Bredius RG, Jansen-Hoogendijk AM, Raaijmakers S, Egeler RM, Lankester AC et al. IgG antibodies to ATG early after pediatric hematopoietic SCT increase the risk of acute GVHD. Bone Marrow Transplant. 2012; 47：360-368.

[12] Shah AJ, Kapoor N, Crooks GM, Weinberg KI, Azim HA, Killen R, et al. The effects of Campath 1H upon graft-versus-host disease, infection, relapse, and immune reconstitution in recipients of pediatric unrelated transplants. Biol Blood Marrow Transplant. 2007; 13：584-593.

[13] Schmidt-Hieber M, Schwarck S, Stroux A, Ganepola S, Reinke P, Thiel E, et al. Immune reconstitution and cytomegalovirus infection after allogeneic stem cell transplantation: the important impact of in vivo T cell depletion. Int J Hematol. 2010; 91：877-885.

[14] Juliusson G, Theorin N, Karlsson K, Frodin U and Malm C. Subcutaneous alemtuzumab vs ATG in adjusted conditioning for allogeneic transplantation: influence of Campath dose on lymphoid recovery, mixed chimerism and survival. Bone Marrow Transplant. 2006; 37：503-510.

[15] Rebello P, Cwynarski K, Varughese M, Eades A, Apperley JF and Hale G. Pharmacokinetics of CAMPATH-1H in BMT patients. Cytotherapy. 2001; 3：261-267.

[16] Waller EK, Langston AA, Lonial S, Cherry J, Somani J, Allen AJ, et al. Pharmacokinetics and pharmacodynamics of anti-thymocyte globulin in recipients of partially HLA-matched blood hematopoietic progenitor cell transplantation. Biol Blood Marrow Transplant. 2003; 9：460-471.

[17] Wells GA, Shea B, O'Connell D, Peterson J, Welch V, Losos M, Tugwell P. The Newcastle-Ottawa Scale (NOS) for assessing the quality of nonrandomised studies in meta-analyses. http://www.ohri.ca/programs/clinical_epidemiology/oxford .htm. Accessed April 29, 2004.

[18] Egger MSD, Altman D. Systematic reviews in health care. Metaanalysis in context, 2nd edn. BMJ, London 2001.
[19] DerSimonian R, Laird N. Meta-analysis in clinical trials. Controlled clinical trials 1986; 7: 177–188
[20] Tierney J, Stewart L, Ghersi D, Burdett S, Sydes M. Practical methods for incorporating summary time-to-event data into metaanalysis. Trials 2007; 8: 16.
[21] Higgins JPT, Thompson SG, Deeks JJ, Altman DG. Measuring inconsistency in meta-analyses. BMJ 2003; 327: 557– 560.
[22] Willemsen L, Jol-van DZC, Admiraal R, Putter H, Jansen-Hoogendijk AM, Ostaijen-Ten DM, et al. Impact of serotherapy on immune reconstitution and survival outcomes after stem cell transplantations in children: thymoglobulin versus alemtuzumab. Biol Blood Marrow Transplant. 2015; 21：473-482.
[23] Busemann C, Neumann T, Schulze M, Klenner A, Thiele T, Greinacher A, et al. Low-dose alemtuzumab vs. standard policy for prevention of graft-versus-host disease in unrelated and related allogeneic stem cell transplantation-a matched pair analysis. Ann Hematol. 2013; 92：945-952.
[24] Marsh JC, Pearce RM, Koh MB, Lim Z, Pagliuca A, Mufti GJ, et al. Retrospective study of alemtuzumab vs ATG-based conditioning without irradiation for unrelated and matched sibling donor transplants in acquired severe aplastic anemia: a study from the British Society for Blood and Marrow Transplantation. Bone Marrow Transplant. 2014; 49：42-48.
[25] Park SH, Choi SM, Lee DG, Choi JH, Yoo JH, Kim SH, et al. Infectious complications associated with alemtuzumab use for allogeneic hematopoietic stem cell transplantation: comparison with anti-thymocyte globulin. Transpl Infect Dis. 2009; 11：413-423.
[26] Juliusson G, Theorin N, Karlsson K, Frodin U and Malm C. Subcutaneous alemtuzumab vs ATG in adjusted conditioning for allogeneic transplantation: influence of Campath dose on lymphoid recovery, mixed chimerism and survival. Bone Marrow Transplant. 2006; 37：503-510.
[27] Kroger N, Shaw B, Iacobelli S, Zabelina T, Peggs K, Shimoni A, et al. Comparison between antithymocyte globulin and alemtuzumab and the possible impact of KIR-ligand mismatch after dose-reduced conditioning and unrelated stem cell transplantation in patients with multiple myeloma. Br J Haematol. 2005; 129：631-643.
[28] Norlin AC and Remberger M. A comparison of Campath and Thymoglobulin as part of the conditioning before allogeneic hematopoietic stem cell transplantation. Eur J Haematol. 2011; 86：57-66.c

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