Maintenance With Hypomethylating Agents After Allogeneic Stem Cell Transplantation in Acute Myeloid Leukemia and Myelodysplastic Syndrome: A Systematic Review and Meta-Analysis

Smith Kungwankiattichai¹, Ben Ponvilawan², Claudie Roy³, Pattarapor Tunsing¹, Florian Kuchenbauer¹,⁴ and Weerapat Owattanapanich¹†

¹Division of Hematology, Department of Medicine, Faculty of Medicine Siriraj Hospital, Mahidol University, Bangkok, Thailand, ²Department of Pharmacology, Faculty of Medicine Siriraj Hospital, Mahidol University, Bangkok, Thailand, ³Vancouver General Hospital, L/BMT Program of British Columbia, Vancouver, BC, Canada, ⁴British Columbia Research Centre, Terry Fox Laboratory, Vancouver, BC, Canada

Introduction: Hypomethylating agents (HMAs) seem to have a range of properties favorable to post-allogeneic hematopoietic stem cell transplantation (allo-SCT) maintenance in acute myeloid leukemia (AML) patients.

Materials and Methods: The Embase, MEDLINE, and Cochrane Central Register of Controlled Trials databases were independently searched by two investigators to identify relevant studies published inception to 18 November 2021. These trials compared HMA maintenance to observation following allo-SCT for AML or myelodysplastic syndrome.

Results: The meta-analysis eligibility criteria were fulfilled by 14 studies. The overall survival and relapse-free survival of the HMA maintenance group were superior to the observation group, with a pooled risk ratio (RR) of 1.38 and 1.46, respectively. Moreover, the cumulative incidence of relapse was significantly lower in those who received HMAs. The HMA group also had lower non-relapse mortality compared with the observation group. Overall, the incidences of grades III–IV acute graft-vs.-host disease (GVHD) and chronic GVHD did not differ in both groups. However, when looking specifically at those receiving decitabine maintenance, the rate of chronic GVHD seemed to be lower compared with observation alone.

Conclusions: The current systematic review and meta-analysis illustrated that AML and MDS patients receiving HMA maintenance after allo-SCT had better outcomes in regards to OS, RFS, NRM, CIR as well as a reduced incidence of chronic GVHD.

Keywords: acute myeloid leukemia, azacitidine, decitabine, hypomethylating agent, maintenance, transplant
KEY MESSAGES

- Role of HMAs Maintenance After Allo-SCT in AML Has Been Extensively Studied in Recent Years in Order to Improve Clinical Outcome.
- This Meta-Analysis Demonstrated Favorable Outcome of HMAs Maintenance in Term of Relapse Rate, non-Relapse Mortality, Relapse-Free Survival and Overall Survival.
- Decitabine Maintenance Resulted in Lower Chronic GVHD Rate Compared With Observation Strategy.

INTRODUCTION

Allogeneic stem cell transplantation is the mainstay treatment for AML stratified as intermediate or unfavorable risk as well as for high-risk MDS. This therapy has demonstrated superior efficacy over non-alloSCT approaches in regards to long-term clinical outcomes (1, 2). Nevertheless, even after allo-SCT, 35–45% of patients suffer from disease relapse, leading to dismal outcomes (3, 4).

Several strategies have been adopted to prolong disease-free survival. Based on the time of intervention, they can be categorized into either preemptive approaches—those commenced at the time of detection of minimal residual disease (MRD)—or prophylactic approaches—those initiated in the absence of detectable leukemia. In the case of the prophylactic approaches, both cellular and pharmacological maintenance strategies have been reported, including prophylactic donor leukocyte infusion (DLI), hypomethylating agents (HMAs), histone deacetylase inhibitors, FMS-like tyrosine kinase 3 (FLT3) inhibitors, or isocitrate dehydrogenase inhibitors (5–7). Notably, HMAs have generated considerable research interest in recent years due to their favorable side effect profile.

HMAs exhibit several properties that make them suitable for post-allo-SCT maintenance. They mediate a direct anti-leukemic effect in AML and MDS, regardless of their molecular mutation profile. Moreover, their abilities to induce a CD8+ tumor-specific T cell response, together with the expansion of regulatory T cells, lead to an epigenetically enhanced Graft vs. Leukemia (GVL) effect that is not counterbalanced by an increased risk of GVHD (8–12). Lastly, they are safe and well-tolerated by AML patients in remission (13).

Many studies have examined the use of azacitidine and decitabine as maintenance after allo-SCT for AML and MDS. Although the majority of the studies supported consideration of HMA maintenance therapy, the remainder did not demonstrate clear benefits (14–29). A recent systematic review explored the safety and efficacy of maintenance treatment following allo-SCT in AML and MDS. It demonstrated rates of 65.6 and 56.2% for the 2-year overall survival (OS) and the relapse-free survival (RFS), respectively, of HMA-treated patients. In addition, acute and chronic GVHD were found in 39.9 and 44.4%, of patients respectively. These results suggest that HMA maintenance could be employed to prolong RFS and OS (30). Nonetheless, the benefit of HMA maintenance after allo-SCT is still uncertain.

This meta-analysis was performed to review all relevant studies to compare the outcomes of patients undergoing allo-SCT for AML or MDS receiving HMA maintenance therapy with observation only.

MATERIALS AND METHODS

Data Sources and Searches

The Embase, MEDLINE, and Cochrane Central Register of Controlled Trials databases were independently searched by two investigators (B.P., W.O.) to identify relevant studies published from inception to November 18, 2021. The search terms consisted of words associated with HMAs, acute myeloid leukemia, myelodysplastic syndrome, and stem cell transplantation. Supplementary Data 1 details the exhaustive search strategy lists. The study was conducted in accordance with the PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analysis) guidelines (Supplementary Data 2).

Selection Criteria and Data Extraction

The inclusion criteria were as follows: (1) studies had to be either randomized controlled trials (RCTs) or cohort studies (prospective or retrospective); (2) the patients underwent allo-SCT for AML or MDS; (3) the studies compared two patient groups: one receiving an HMA post-allo-SCT, and the other being an observational group; and (4) the studies needed to report at least one of our primary outcomes of interest (OS, RFS, grades III–IV acute GVHD, and chronic GVHD). The secondary outcomes of interest were the CIR and NRM. Study eligibility was individually assessed by three investigators (B.P., W.O, S.K.); disagreements were resolved by consensus.

Two investigators (B.P., W.O.) utilized a standardized collection form to extract the baseline characteristic data of the patients in each group, along with details of the primary and secondary outcomes of interest. The extracted data was cross-checked to confirm its accuracy.

Definitions of Outcomes

The OS rate was defined as the time between the stem cell infusion and the time of death or last follow-up, while RFS was defined as the time interval from the stem cell infusion to the date of relapse or death from any cause. All causes of death (other than death from a relapse) were used to calculate the NRM rate.

Quality Assessment

Two investigators (B.P., W.O.) assessed the quality of each study using the Jadad scale for RCTs and the Newcastle–Ottawa scale for cohort studies (31, 32).

Statistical Analysis

The Mantel–Haenszel method was used to combine the effect estimates and 95% confidence intervals (CIs) of each study, and to calculate the pooled odds ratio (OR) with 95% CI (33). A random-effects model was preferred over a fixed-effects model because it was more likely that high heterogeneity would be found among the studies. Statistical heterogeneity was calculated using Cochran’s Q test, estimated by the heterogeneity ($I^2$) statistic.
There were four heterogeneity levels: insignificant ($I^2$ values of 0–25%), low ($I^2$ values of 26–50%), moderate ($I^2$ values of 51–75%), and high ($I^2$ values of >75%) (34). The presence of a publication bias was visualized by a funnel plot along with Egger's regression test. Due to a lack of clinical studies, a subgroup analysis based on the types of HMAs could not be performed. All statistical analyses were performed using the Review Manager (RevMan) software (version 5.3; The Cochrane Collaboration, Oxford, UK) and “meta” package version 5.1-0. This study was registered at www.inplasy.com as #INPLASY2021110078.

RESULTS

Search Results

The systematic search of the Embase, MEDLINE and Cochrane Central Register of Controlled Trials databases identified 5,680 articles, from which 1,578 duplicates were removed. This resulted in 4,102 articles available for title and abstract review. Subsequently, 4,068 articles were excluded as the article type and study design did not fulfill the inclusion criteria, or there was no report on a primary outcome of interest. The remaining 34 articles underwent full-length review and 20 of those were excluded for the aforementioned reasons. Ultimately, the eligibility criteria for our meta-analysis were met by 14 studies: two RCTs, two prospective cohort study, and 10 retrospective cohort studies (14–22, 25, 27–29). Twelve of these compared azacitidine maintenance to observation, whereas two compared decitabine maintenance to observation. Figure 1 illustrates the full literature review and selection process.

Baseline Patient Characteristics

The 14 included studies were composed of 533 patients who received HMAs as maintenance, and another 784 patients who were observed post-allo-SCT. The age of the participants varied greatly (HMA group: 2 to 78 years; and observation group: 2 to 75 years). AML accounted for the largest proportion of the disease subtypes in the HMA group (84.8%), followed by MDS (14.6%) and mixed-phenotype leukemia (0.6%). These values were similar to the corresponding proportions found in the observation group (87.5, 11.2, and 1.3%, respectively). In both groups, matched unrelated donors and matched sibling donors were the most common donor sources for allo-SCT, accounting for 45.2 and 26.8%, respectively. In addition, myeloablative conditioning regimens (68.2%) were used more frequently compared to reduced-intensity conditioning regimens (31.8%). Details of the patient characteristics, such as disease status before allo-SCT, MRD status before allo-SCT cytogenetic risk, prior treatment and HMA before allo-SCT, comorbidities and performance status [Hematopoietic Cell Transplantation-Comorbidity Index (HCT-CI)], study period and quality assessment are summarized in Table 1. Details of the donor types, stem cell source, median dose of CD34+ stem cells, conditioning regimens, GVHD prophylaxis, MRD status after allo-SCT, maintenance protocols and prophylactic DLI are listed in Table 2.

HMA Maintenance Protocols After Allo-SCT

The median time of commencement of the HMAs varied between 56 and 154 days after allo-SCT. Twelve studies used azacitidine (14–22, 25, 27, 28), while two used decitabine (24, 29). The azacitidine dosage was 16–50 mg/m² on Day 1 to Day 5 every 6–12 weeks for 1–22 cycles. A study from Oshikawa et al. and Kaito et al. combined azacitidine with gemtuzumab ozogamicin for the maintenance protocol (15, 16). Decitabine was administered at 5–20 mg/m² on Day 1 to Day 5 every 6–12 weeks for 1–22 cycles. Some patients received prophylactic DLI in addition to HMA maintenance in five studies (14, 18–20, 22, 27).

Comparison of Clinical Outcomes of HMA and Observation Groups

The OS rates were reported as a 1-year rate in three studies (15–17), a 2-year rate in six studies (19–22, 28, 29), and a 3-year rate in three studies (14, 18, 24). The RFS rates were reported as a 1-year rate in two studies (15, 16), a 2-year rate in five studies (19–21, 27, 29), and a 3-year rate in two studies (18, 24). The OS of the HMA group was superior to that of the observation group, with a pooled RR of 1.38 (95% CI, 1.19–1.60; $I^2$, 50%; Figure 2A).

FIGURE 1 | Study identification and literature review process.
TABLE 1 | Patient’s baseline characteristics of studies included in the meta-analysis.

| References | Group | No. | Sex (M/F) | Median age (years, range) | Diseases | Cytogenetic risk | HCT-CI status before HSCT | Disease status before HSCT | MRD status before HSCT | Hypomethylating agents use before HSCT | Study period | Type | Quality assessment |
|------------|-------|-----|-----------|--------------------------|----------|-----------------|--------------------------|----------------------------|-------------------------|----------------------------------------|--------------|------|------------------|
| Oshikawa (15) | HMA | 10 | 8/2 | 49.5 (17–60) | AML | High | NA | 1: ≥CR2 2: relapse 5: PIF 2: relapse after HSCT | NA | NA | NA | PRO | S: 3 C: 1 O: 3 |
| Control | 30 | 20/10 | 50 (18–68) | AML | High | NA | 3: ≥CR2 6: relapse 15: PIF 6: relapse after HSCT | NA | NA | |
| Ovechkina (14) | HMA | 58 | 34/24 | 28 (2–68) | 51: AML 7: MDS | 14: High | NA | NA | 7: MRD+ 51: MRD- | NA | NA | RET | S: 2 C: 2 O: 3 |
| Control | 58 | 31/27 | 29 (2–60) | 51: AML 7: MDS | 10: High | NA | NA | 5: MRD+ 53: MRD- | NA | NA | |
| Kaito (16) | HMA | 23 | NA | 54 (17–67) | 21: AML 1: MDS 1: MPAL | High | NA | 6: CR 17: not in CR | NA | NA | RET | S: 3 C: 2 O: 3 |
| Control | 69 | NA | NA | NA | 63: AML 3: MDS 3: MPAL | High | NA | NA | NA | NA | |
| Américo (21) | HMA | 17 | NA | >18 | MDS/AML | NA | NA | NA | 7: MRD+ | NA | NA | RET | S: 3 C: 1 O: 3 |
| Control | 51 | NA | NA | NA | MDS/AML | NA | NA | NA | 13: MRD+ | NA | NA | |
| Maples (17) | HMA | 25 | 14/11 | 56 (24–72) | 18: AML 7: MDS | 1: Favorable 8: Intermediate 9: High 7: NA | NA | 14: CR1 4: CR2 7: MDS | 14: CR1 7: MDS 2: CR2 > CR3: 1 12: MDS | NA | |
| Control | 50 | 21/29 | 54 (20–70) | 38: AML 12: MDS | 3: Favorable 13: Intermediate 22: High 12: NA | NA | 15: CR1 2: CR2 > CR3: 1 12: MDS | 13: MRD+ | NA | |
### TABLE 1 | Continued

| References | Group | No. | Sex (M/F) | Median age (years, range) | Diseases | Cytogenetic risk | HCT-CI Disease status before HSCT | MRD status before HSCT | Hypomethylating agents use before HSCT | Study period | Type | Quality assessment |
|------------|-------|-----|-----------|---------------------------|----------|----------------|----------------------------------|-----------------------|---------------------------------------|--------------|------|-------------------|
| Danylesko (18) | HMA | 40 | 24/16 | 62 (25–74) | 32: AML 8: MDS | 21: High | NA | 19: CR1 37: NA | NA | NA | RET | S: 2 C: 2 O: 3 |
| Guillaume (19) | HMA | 30 | 13/17 | 58 (22–70) | 20: AML 10: MDS | 15: High | NA | 16: CR1 6: CR2 5: R/R 3: untreated MDS | NA | NA | RET | S: 3 C: 2 O: 3 |
| Joris (20) | HMA | 19 | NA | 52 (18–70) | 48: AML 5: MDS | High | NA | 21: CR 32: R/R | NA | NA | RET | S: 3 C: 1 O: 3 |
| Ali (22) | HMA | 34 | 36/23 | 62 (23–78) | 45: AML 14: MDS | NA | NA | 28: 0 19: 1–2 12: ≥3 | NA | NA | RET | S: 3 C: 2 O: 3 |
| Control | 90 | 57/33 | 60 (26–73) | 76: AML 14: MDS | 4: Favorable 37: Intermediate 25: High | NA | NA | 38: CR1 4: CR2 3: R/R 14: MDS | NA | NA | RET | S: 3 C: 2 O: 3 |
| Gao (29) | HMA | 100 | 56/44 | 30 (9–62) | High-risk AML 7: Favorable 19: Intermediate 74: High | NA | NA | 92: CR 1: PR 7: NR 24 MRD+CR 68 MRD- CR | NA | NA | RCT | R: 1 D: 0 W: 1 |

(Continued)
| References  | Group  | No. | Sex (M/F) | Median age (years, range) | Diseases | Cytogenetic risk | HCT-CI | Disease status before HSCT | MRD status before HSCT | Hypomethylating agents use before HSCT | Study period | Type  | Quality assessment |
|------------|--------|-----|-----------|---------------------------|----------|------------------|--------|---------------------------|-----------------------|---------------------------------------|-------------|-------|-------------------|
| Ma (24)    | HMA    | 21  | 13/8      | 28 (10–63)                | High-risk AML | 3: Favorable 20: Intermediate 79: High | NA     | 97: CR 5: NR              | 29 MRD+CR 88 MRD-CR | NA                                   | September 2015–October 2018 | RET   | S: 3 C: 2 O: 3   |
|            | Control | 63  | 37/26     | 29 (8–56)                 | AML 4: MPAL | High             | 26: 1 24: 2 8: 3 5: ≥4 | 41: CR1 10: CR2 12: NR | NA                                   | NA                                   |       |                   |
| Oran (25)  | HMA    | 87  | 51/36     | 57 (19–72)                | AML 4: MDS | 8: Favorable 33: Intermediate 48: High | 28: 0–1 22: 2–3 37: ≥4 | 54: CR1/2 33: Active disease | NA                                   | NA                                   | April 2009–January 2017 | RCT   | R: 1 D: 0 W: 1  |
|            | Control | 94  | 57/36     | 57.5 (20–75)              | AML 25: MDS | 15: Favorable 42: Intermediate 37: High | 37: 0–1 37: 2–3 20: ≥4 | 36: CR1/2 58: active disease | NA                                   | NA                                   |       |                   |
| Booth (27) | HMA    | 13  | NA        | 12.6                      | AML      | High-risk        | NA     | NA                        | NA                                   | NA                                   | January 2010–March 2020 | RET   | S: 3 C: 1 O: 3  |
| Keruakous (28) | Control | 28  | NA        | 7.0                       | AML      | High-risk        | NA     | NA                        | NA                                   | 7 MRD+ 24 MRD-     | September 2013–July 2018 | PRO   | S: 4 C: 2 O: 3  |
|            | HMA    | 31  | 16/15     | 47                        | Poor-risk AML | High             | NA     | 31: CR                    | NA                                   | NA                                   |                     |       |                   |
| Control    | 18     | 9/9 | 54        | Poor-risk AML             | High     | NA               | 18: CR 6 MRD+ 12 MRD- | NA                                   | NA                                   |                     |       |                   |

AML, acute myelogenous leukemia; C, compatibility; CR, complete remission; CR1, first complete remission; CR2, second complete remission; CR3, third complete remission; HCT-CI, Hematopoietic Cell Transplantation–Comorbidity Index; HMA, hypomethylating agent; HSCT, hematopoietic stem cell transplantation; M, male; MDS, myelodysplastic syndrome; MPAL, mixed phenotype acute leukemia; MRD, minimal residual disease; NA, not available; No., number; NR, no remission; O, outcome; PIF, primary induction failure; PR, partial remission; PRO, prospective cohort study; R, randomization; RCT, randomized controlled trial; RET, retrospective cohort study; R/R, relapsed/refractory; S, selection.
### TABLE 2 | Peri- and post-transplantation information of studies included in the meta-analysis.

| References     | Group | No. | Donor type       | Stem cell source | Median dose of CD34(×10^6/kg) | Conditioning regimen (MAC/RIC) | GVHD prophylaxis | MRD status after HSCT and timing | Maintenance therapy after HSCT | Prophylactic DLI |
|----------------|-------|-----|------------------|------------------|-------------------------------|---------------------------------|------------------|----------------------------------|-------------------------------|--------------------|
| Oshikawa (15)  | HMA   | 10  | 1: MSD 4; MUD 5; Haplo | 6: PB 4: BM      | NA                            | MAC (4: Bu/Cy, 1: Flu/Bu/ATG/TBI 2: Flu/Cy/ AraC/ATG/TBI/RIC (3: Flu/Mel/ATG/TBI) | MTX/CSA or MTX/TAC | NA                               | - AZA 30 mg/m² x 7 days combined with GO 3 mg/m² on Day 8 up to 4 cycles | -                 |
| Ovechkina (14) | Control | 30  | NA               | NA               | NA                            | NA                              | NA               | NA                               | Observation                     | 96% (21 patients) |
| Ovechkina (14) | HMA   | 58  | 12: MSD 35; MUD/ MMUD 11: Haplo | NA               | NA                            | NA                              | 16/42            | NA                               | - AZA 35 mg/m² x 5 days every 28 days - Median: 2.5 cycles (1–8 cycles) - Median time of starting AZA: 253 days (27–861 days) after HSCT | -                 |
| Control        | 58    | NA  | 22: MSD 28; MUD/ MMUD 8: Haplo | NA               | NA                            | 17/41                           | NA               | NA                               | Observation                     | 36% (21 patients) |
| Kaito (16)     | HMA   | 23  | 15: Matched donor 8: Haplo | NA               | NA                            | NA                              | 12/11            | NA                               | - AZA 30 mg/m² x 7 days combined with GO 3 mg/m² on Day 8 up to 4 cycles - Median time of starting AZA: 78 days (24–251 days) after HSCT | -                 |

(Continued)
| References | Group | No. | Donor type | Stem cell source | Median dose of CD34(x10^6/kg) | Conditioning regimen (MAC/RIC) | GVHD prophylaxis | MRD status after HSCT and timing | Maintenance therapy after HSCT | Prophylactic DLI |
|------------|-------|-----|------------|------------------|-----------------------------|--------------------------------|--------------------------|---------------------------------|-------------------------------|----------------|
| Américo (21) | Control | 69 | NA | NA | NA | NA | NA | NA | NA | Observation – |
| Américo (21) | HMA | 17 | NA | NA | NA | NA | NA | NA | NA | AZA |
| Américo (21) | Control | 51 | NA | NA | NA | NA | NA | NA | NA | observation – |
| Maples (17) | HMA | 25 | 6: MSD | 21: PB | 23/2 (MAC: 11 Bu/Cy, 11 Bu/Flu, 1 Cy/TBI, RIC: 2 Bu/Flu) | 13: TAC/MMF | 8: TAC/MMF | 13: CSA/MMF | - AZA 32 mg/m^2 x 5 days every 28 days for 4–6 cycles - Median time of starting AZA: 75 days (42–131 days) after HSCT |
| Maples (17) | Control | 50 | 15: MSD | 43: PB | 47/3 (MAC: 19 Bu/Cy, 15 Bu/Flu, 13 Cy/TBI, RIC: 1 Bu/Flu, 2 ATG/TBI) | 32: TAC/MMF | 5: TAC/MMF | 13: CSA/MMF | Observation – |
| Danylesko (19) | HMA | 40 | 8: MSD | NA | NA | NA | NA | 8: MRD + 32: MRD-Timing: NA | - AZA 32–50 mg/m^2 x 5 days every 28 days for 2 years - Median time of starting AZA: 2.2 months (1.2–6.9 months) after HSCT | 8% (3 patients) |
| Danylesko (19) | Control | 40 | NA | NA | NA | NA | NA | NA | Observation – |
| Guillaume (19) | HMA | 30 | 13: MSD | 12/18 | NA | NA | NA | NA | - AZA 32 mg/m^2 x 5 days every 28 days for 1 year starting after 8 weeks of HSCT |
| Guillaume (19) | Control | 58 | NA | NA | NA | NA | NA | NA | Observation – |

(Continued)
| References | Group | No. | Donor type | Stem cell source | Median dose of CD34 (x10^6/kg) | Conditioning regimen (MAC/RIC) | GVHD prophylaxis | MRD status after HSCT and timing | Maintenance therapy after HSCT | Prophylactic DLI |
|------------|-------|-----|------------|------------------|-------------------------------|--------------------------------|----------------|----------------------------------|-------------------------------|----------------|
| Joris (20) | HMA   | 19  | 13: MSD    | 48: PB 5: BM 5: CB | 7.9 0/53 (sequential RIC) | CSA/MMF | NA | - AZA 37.5 mg/m² x 5 days every 28 days for 1 year | - 3 cycles of DLI alternating with AZA | - |
|           |       |     | 27: MUD    | 76: PB 5: BM 9: CB | 5.4 18/41 | 33: CSA/MMF ± PTCy 6: Others | NA | 1: MRD + 45: MRD- 13: NA | Observation | - |
|           |       | 34  | 4: MMUD 9: Haplo | 77: PB 5: BM 4: CB | 5.4 42/48 | 71: CSA/MMF ± PTCy 3: Others | 6: MRD + 37: MRD- 14: NA | Timing: day +100 | Observation | - |
| Ali (22)  | HMA   | 59  | 11: MSD    | 50: PB 5: BM 4: CB | 5.4 18/41 | 33: CSA/MMF ± PTCy 6: Others | NA | 1: MRD + 45: MRD- 13: NA | Timing: day +100 | 7% (4 patients) |
|           |       |     | 32: MUD 12: Haplo | 78: PB 5: BM 9: CB | 5.4 42/48 | 71: CSA/MMF ± PTCy 3: Others | 6: MRD + 37: MRD- 14: NA | Timing: day +100 | Observation | - |
|           |       | 90  | 4: UCB     | 79: PB 5: BM 9: CB | 5.4 42/48 | 71: CSA/MMF ± PTCy 3: Others | 6: MRD + 37: MRD- 14: NA | Timing: day +100 | Observation | - |
| Gao (29)  | HMA   | 100 | 20: MSD    | 5: MUD 75: Haplo | 8.2 100/0 | NA | NA | - DAC 5 mg/m² x 5 days every 6–8 weeks up to 6 cycles combining with G-CSF 100 mcg/m² on Day 0–5 of DAC | NA | - |
|           |       |     | 5: MUD 75: Haplo | 8.2 100/0 | NA | NA | NA | NA | Observation | - |
|           |       | 102 | 2: MUD     | 5: PB 2: BM 14: PB+BM | 8.3 102/0 | NA | NA | - DAC 20 mg/m² x 5 days every 12 weeks for 4–6 cycles - Median numbers of cycles: 3 (2–8) - Median time of starting DAC: 154 days (55–358 days) after HSCT | NA | - |
| Ma (24)   | HMA   | 21  | 2: MUD 3: Haplo | 5: PB 2: BM 14: PB+BM | 8.3 102/0 | NA | NA | - DAC 20 mg/m² x 5 days every 12 weeks for 4–6 cycles - Median numbers of cycles: 3 (2–8) - Median time of starting DAC: 154 days (55–358 days) after HSCT | NA | - |

(Continued)
| References | Group | No. | Donor type | Stem cell source | Median dose of CD34 (x10^6/kg) | Conditioning regimen (MAC/RIC) | GVHD prophylaxis | MRD status after HSCT and timing | Maintenance therapy after HSCT | Prophylactic DLI |
|------------|-------|-----|------------|----------------|-------------------------------|---------------------------------|-----------------|-----------------------------------|------------------------------|----------------|
| Control    | 63    | 17: MSD 5: MUD 41: Haplo | 20: PB 9: BM 34: PB+BM | NA | 63/0 | NA | NA | Observation | – |
| Oran (25)  | HMA   | 87  | 33: MSD 44: MUD 4: Haplo 6: NA | 55: PB 31: BM 1: CB | NA | 73/14 | 4: PTCy 82: TAC/MTX 1: TAC/MMF | NA | - AZA 32 mg/m² x 5 days every 28 days for 12 cycles - Median time of starting AZA: 62 days (42–100 days) after HSCT Observation – |
| Control    | 94    | 31: MSD 53: MUD 5: Haplo 4: NA | 60: PB 32: BM 2: CB | NA | 75/18 | 9: PTCy 82: TAC/MTX 3: TAC/MMF | NA | Observation – |
| Booth (27) | HMA   | 13  | 2: MSD 6: MUD 1:MMUD 4: Haplo | NA | NA | 12/1 | NA | NA | AZA x 6 cycles starting on day+60 DLI x 3 doses after day +120 |
| Control    | 28    | 8: MSD 9: MUD 9:MMUD 2:Haplo | NA | NA | 25/3 | NA | NA | Observation |
| Kerakous (28) | HMA      | 31 | 6: MSD 23: MUD 2: Haplo | PB:11 BM:19 NA:1 | 3.64 | 24/7 | NA | NA | - AZA 32 mg/m² x 5 days every 28 days for 4 cycles starting after 8 weeks of HSCT Observation – |
| Control    | 18    | 6: MSD 10: MUD 2: Haplo | PB:11 BM:6 NA:1 | 3.55 | 8/10 | NA | NA | Observation – |

**ATG, antithymocyte globulin; AZA, azacitidine; BM, bone marrow; BU, busulfan; CSA, cyclosporin A; Cy, cyclophosphamide; DAC, decitabine; DLI, donor lymphocyte infusion; Flu, fludarabine; G-CSF, granulocyte colony-stimulating factor; GO, gemtuzumab ozogamicin; Haplo-, haploidentical; HMA, hypomethylating agent; HSCT, hematopoietic stem cell transplantation; MAC, myeloablative conditioning; MEL, melphalan; MMF, mycophenolate mofetil; MMUD, mismatched unrelated donor; MRD, minimal residual disease; MSD, matched sibling donor; MTX, methotrexate; MUD, matched unrelated donor; NA, not available; No., number; PB, peripheral blood; PTCy, post-transplant cyclophosphamide; RIC, reduced intensity consolidation; TAC, tacrolimus; TBI, total body irradiation; UCB, umbilical cord blood.**
FIGURE 2 | Forest plots of the meta-analysis of HMA maintenance compared with no HMA maintenance. (A) OS rate. (B) RFS rate.
Similarly, a pooled meta-analysis found that the RFS was significantly better in patients who received HMAs, with a pooled RR of 1.46 (95% CI, 1.31–1.62; $I^2$, 0%; Figure 2B) (15, 16, 18–21, 24, 27, 29). The patients receiving HMAs also had a lower NRM than those under observation (pooled RR, 0.36; 95% CI, 0.19–0.66; $I^2$, 0%; Figure 3A) (14, 16, 18, 19, 28). Furthermore, the CIR was significantly higher for the observed patients (pooled RR, 0.69; 95% CI, 0.50–0.95; $I^2$, 67%; Figure 3B) (14, 16, 18, 19, 21, 24, 25, 28, 29). However, the incidences of grades III–IV acute GVHD and chronic GVHD of the groups did not differ [pooled RR, 0.69; 95% CI, 0.50–0.95; $I^2$, 67%; Figure 3B] (14, 16, 17, 22, 24, 25); and pooled RR, 0.84; 95% CI, 0.58–1.23; $I^2$, 65% (22, 24, 25, 29); Figures 4A, B, respectively).

Because two studies combined gemtuzumab ozogamicin with HMA and five studies combined DLI with HMA, the efficacy of the HMAs was confirmed by conducting a sensitivity analysis that excluded those seven studies. As with the results of the full analysis, the OS, RFS, and NRM were found to be better for the HMA group than the observation group, whereas the CIR and the incidence of acute GVHD did not differ between the two groups (Supplementary Data 3). Nonetheless, the patients who received HMAs had a significantly lower incidence of chronic GVHD (pooled RR, 0.71; 95% CI, 0.55–0.91; $I^2$, 0%) (24, 25, 29).

Funnel plots of the OS, RFS, NRM, CIR, grades III–IV acute GVHD, and chronic GVHD outcomes of the HMA and observation groups did not show a publication bias (Figure 5). Egger's regression test confirmed this ($p = 0.1590, 0.2713, 0.8865, 0.1804, 0.3706, 0.8302$ for OS, RFS, NRM, CIR, grades III–IV acute GVHD, and chronic GVHD; respectively).

### Subgroup Analysis Based on Study Design

A subgroup analysis based on the study design was performed. There is a trend for a significantly lower risk of chronic GVHD in randomized studies but not in observational studies. The CIR
results appear to be similar between randomized studies and observational studies. In observational studies, other parameters which included OS, RFS were significantly better in HMAs arm. However, these parameters could not be analyzed in randomized studies subgroup due to limited number of studies (Supplementary Data 4).

Subgroup Analysis Based on Each HMA

Two HMAs were used in this meta-analysis: azacitidine and decitabine. The OS, RFS and NRM outcomes of the azacitidine group were significantly better than those of the observation group (Figures 2, 3) (14, 16–20, 22, 28). Likewise, the decitabine arm had superior OS, RFS and CIR outcomes to those of the observation arm (Figures 2, 3) (21, 24, 29). The incidence of grades III–IV acute GVHD and chronic GVHD in patients who received azacitidine were similar to those under observation (Figure 4) (14, 16, 22, 25). Interestingly, the rate of chronic GVHD in the decitabine group was significantly lower than in the observation group (pooled RR, 0.67; 95% CI, 0.50–0.90; $I^2$, 0%; Figure 4B) (24, 29).

Subgroup Analysis of Patients Who Received HMAs in Combination With DLI

The work by Guillaume et al., Joris et al., and Booth et al. compared HMAs in combination with DLI and observation arms. Although the OS and RFS outcomes tended to be better for HMAs combined with DLI, only RFS rates were statistically significant (pooled RR, 1.20; 95% CI, 0.86–1.67; $I^2$, 37%; and pooled RR, 1.33; 95% CI, 1.09–1.62; $I^2$, 0%; Figure 6) (19, 20, 27).

Sensitivity Analyses

In total, three sensitivity analyses were conducted. The first analysis was performed on studies which recruited only patients with AML. Only RFS and CIR were significantly better in HMAs arm. In addition, there was a trend of superior OS in HMAs group. Nevertheless, other parameters could not be evaluated due to limited number of studies (Supplementary Data 5). The second analysis was performed on studies with adult patients. The OS, RFS and NRM were significantly superior in HMAs arm while the other outcomes did not show significant superiority (Supplementary Data 6). The third analysis was performed on studies with low risk of bias by selecting only studies with a
Newcastle-Ottawa scale of at least eight or Jadad score of at least two. Similar results were obtained compared to the main results (Supplementary Data 7).

Quality of Evidence Using Grading of Recommendations Assessment, Development, and Evaluation Approach
The quality of evidence generated by the current systematic review and meta-analysis is moderate.

DISCUSSION
An earlier systemic review that focused on the safety and efficacy of HMAs as post-allo-SCT maintenance for AML and MDS found acceptable OS and RFS rates without a heightened GVHD rate (30). Unfortunately, a detailed analysis of the clinical outcomes of the control and treatment groups was not reported. A recent meta-analysis of HMAs and FLT3 Inhibitors as maintenance treatment for AML and MDS after allo-SCT showed a high percentage of OS and RFS (35). Due to the available limited studies comparing clinical outcomes between HMAs and observation arms, we focused on comparing the benefits of HMA maintenance following allo-SCTs with an observation approach and further analyzed the efficacy in each HMA subgroup. Notably, OS, RFS, NRM, and CIR were markedly improved with HMA maintenance. Our subgroup analysis demonstrated the advantages of both azacitidine and decitabine use in this setting. In terms of safety, the HMAs were not associated with a higher GVHD incidence. In the case of decitabine for post allo-SCT maintenance, the rate of chronic GVHD seemed to be lower than that of the observation arm. Previous studies showed that HMA maintenance had low rates of toxicities and infectious complications even if the treatment is given to elderly patients (36, 37). Furthermore, a prior study reported that 6.8% of post-allo-SCT patients experienced isolated extramedullary relapse which translate into dismal survival outcome (38). Accordingly, prophylaxis scheme post allo-SCT is a rational option to mitigate either isolated extramedullary or bone marrow relapse risk. Taken together, the use of HMAs is a feasible therapy for AML and MDS patients during the post-allo-SCT period, and it should be offered broadly to post allo-SCT patients.

Recently, an oral formulation of azacitidine (CC-486) was approved by the U.S. Food and Drug Administration for the continued treatment of adult AML. Based on data from the phase 3 QUAZAR AML-0001 clinical trial, the patients must have achieved first complete remission or must have an incomplete blood count recovery following intensive induction chemotherapy, and be unable to complete intensive curative therapy (39). The oral formulation of azacitidine may enhance patient convenience, eliminate injection-site reactions, and facilitate long-term administration. The application of this product in a post-allo-SCT setting has since been verified in a phase I/II study, which supports the promising clinical activity (40). A randomized, phase III trial to validate its efficacy is in development.
Although the present analysis confirms the usefulness of HMAs, several limitations are noted. First, variations in the disease status prior to transplantation, the difference in conditioning regimen, and the treatment and protocols of the studies (HMA dosage, number of cycles, and dates of administration) could lead to a diversity of clinical outcomes. Second, there was missing data on the European Group for Blood and Marrow Transplantation risk score and disease

![FIGURE 6](image)

**FIGURE 6** Subgroup analysis of studies with patients receiving HMA maintenance with DLI and without DLI (A) OS rate (B) RFS rate.
risk index. It is possible that older patients were selected to have less comorbidities, better performance status, and less prior treatment burden compared to the younger ones; which could be the confounding factors behind similar outcomes. Furthermore, the lack of comorbidity is an issue in identifying risk factors for NRM. Third, there is a recent trend in using MRD status in pre- and post-allo-SCT setting to classify MRD-positive patients who would benefit from HMAs maintenance after allo-SCT (41). However, MRD assessment data was scarce in published included trials precluding a subgroup analysis. In addition, the incorporation of other agents, such as gemtuzumab ozogamicin and G-CSF into each treatment protocol also impact outcomes of clinical trials. Large-scale randomized trials are warranted to clarify all of these unresolved issues.

CONCLUSION

The current systematic review and meta-analysis illustrated that the patients receiving HMA maintenance post-Allo-SCT had significantly better outcomes with regards to OS, RFS, NRM, and CIR. Furthermore, if decitabine was used for maintenance, the rate of chronic GVHD seemed to be lower than that of the observation arm. Further data, preferably from large prospective studies, is warranted to confirm the benefit of HMA-based maintenance after allo-SCT as well as describe the optimal agent, administration schedule, and the sub-groups of patients who benefit from such intervention.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/Supplementary Material, further inquiries can be directed to the corresponding authors.

ETHICS STATEMENT

The need for ethics approval by institutional board review was waived as this study did not directly involve human subjects.

AUTHOR CONTRIBUTIONS

BP and WO collected the data. BP performed the statistical analyses. WO and SK drafted the manuscript and prepared the final version. FK made critical revisions. All authors designed the study, read, and approved the final manuscript.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fmed.2022.801632/full#supplementary-material

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