Response to Single Agent Cyclosporin in Patients with Non-Severe Aplastic Anaemia

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ABSTRACT

Objective: To determine the effectiveness of cyclosporin A (CSA) monotherapy in treating patients with non-severe aplastic anaemia (NSAA).

Study Design: A cross-sectional observational study.

Place and Duration of the Study: Department of Clinical Haematology, Armed Forces Bone Marrow Transplant Center, Rawalpindi, Pakistan, from January 2022 till December 2023**.**

Methodology: A total of 51 patients of NSAA, classified as aplastic anaemia not satisfying criteria for severe and very severe disease as per Modified Camitta Criteria, were included. Results were evaluated in terms of survival rate (OS) and responses. Responses were assessed as complete response (CR), partial response (PR), overall response (ORR), and no response (NR) by using standard British Committee for standard Haematology (BCSH) response criteria at 3, 6, and 12 months.

Results: Out of 51 patients, 34 (67%) were males and 17 (33%) were females. Median age at the time of diagnosis was 25 (IQR 26) years. At follow-up of 12 months, OS was 86.3%. Overall response rates to cyclosporin monotherapy at 3, 6, and 12 months were 49%, 57%, and 59%, respectively. Baseline haemoglobin was associated with responses at 6 and 12 months and a significant association was found between transfusion dependency at 3, 6, and 12 months with overall survival ($p = 0.01$, 0.005, and 0.04, respectively). Responses at time-defined points also had significant impact on OS (3 months $P_{log\text{-}rank} = 0.046$, 6 months $P_{log\text{-}rank} = 0.01$, and 12 months $P_{log\text{-}rank} = 0.008$).

Conclusion: Overall response rates at 3, 6, and 12 months indicate the potential of CSA as a viable treatment option, particularly in resource-constrained settings. Despite some patients experiencing treatment-related complications, CSA demonstrated a generally tolerable safety profile.

Key Words: *Cyclosporin A, Non-severe aplastic anaemia, Survival rate, Response rate, Complete response, Partial response.*

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INTRODUCTION

Acquired aplastic anaemia (AA) is a heterogeneous disorder characterised by pancytopenia with hypocellular marrow.[1] Peripheral blood cell counts indicate the degree or severity of acquired AA.[2] The incidence of AA is 2–3 per million per year in Europe.[3] Allogeneic haematopoietic stem cell transplantation (Allo-HSCT) is preferred treatment for patients with severe aplastic anaemia (SAA) or very severe aplastic anaemia (vSAA). However, for those with non-severe disease and who do not have human leukocyte antigen (HLA)-matched donors, immunosuppression remains the favoured treatment option.[4]

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For non-severe aplastic anaemia (NSAA), there are no standard or widely effective treatment approaches.[5] Treatment modalities for NSAA include immunosuppressive therapy (IST), horse antithymocyte globulin (h-ATG), androgens, eltrombopag, and HSCT. Various factors that influence the initial choice of treatment include clinical acuteness, degree of cytopenias, transfusion dependence, and other clinical factors.[6] The response rate is higher when cyclosporine-A (CSA) and horse antithymocyte antibody (ATG) are used together compared to when CSA is used alone.[6]

The incidence of AA is 2-3 times higher in Asia than in Europe.[7] The estimated incidence of AA in the Pakistani population is 3.5 patients / million population.[8] Limited availability and expense frequently make h-ATG less useful for patients who are not eligible for transplants.[9] In resource-constrained settings, CSA is a reasonable monotherapy option, given its lower cost and superior safety profile than ATG.[10] The aim of the study was to evaluate the efficacy and safety of CSA monotherapy in patients with NSAA.

METHODOLOGY

This study was carried out at the Department of Clinical Haematology, Armed Forces Bone Marrow Transplant Center, Rawalpindi, Pakistan, after approval by the Hospital's Ethical Committee (Ref: IRB-015/AFBMTC/Approval/2021) from January 2022 till December 2023 in compliance with the Declaration of Helsinki. Data retrieval was performed using medical records of patients with NSAA. The study design was cross-sectional observational. Patients of NSAA who presented to OPD from January 2020 till December 2022 were included by non-probability convenient sampling. A total of 51 patients with newly diagnosed NSAA who were given CSA monotherapy at the outset for at least 3 months were included in the study. Patients with SAA, vSAA, inherited bone marrow failure (IBMF), classical paroxysmal nocturnal haemoglobinuria (PNH), myelodysplastic syndrome (MDS), and not completing the initial 12 weeks of the treatment were excluded.

Results were evaluated in terms of overall survival (OS) and responsiveness to therapy at 3, 6, and 12 months. Aplastic anaemia (AA) was defined as per standard criteria. According to Modified Camitta Criteria, NSAA was classified as AA not satisfying the criteria for severe (i.e. marrow cellularity <25%, plus at least 2 of neutrophils <0.5 × 10^9/L, platelets <20 × 10^9/L, reticulocyte count <60 × 10^9/L) or very severe AA (with neutrophils<0.2× 10^9/L).[3]

All patients received cyclosporine at the initial dose of 3-5 mg/kg/day in two divided doses. Dose was titrated as per serum trough levels in affording patients (Target CSA trough levels = 150–200 ng/ml) and toxicity profile. Responses were assessed as per the British Committee for Standard Haematology (BCSH) response criteria. The terms complete response (CR) and partial response (PR) were used to describe responses that improved blood counts and transfusion dependency. CR was defined by the normalisation of counts (ANC > 1.5× 10^9/L, Hb >10g/dl, platelets >100 × 10^9/L) and transfusion independence. PR was defined as improvement in blood counts and transfusion dependency but not meeting the criteria of CR. The Overall response rate (ORR) incorporated both CR and PR. The term no-response (NR) was used if the cell counts and transfusion requirements remained unchanged. In non-responders, CSA was discontinued at 12 months. Periodically, the patients were checked for complications, side effects, infectious episodes, and need for transfusions, along with complete blood counts and tests for liver and kidney function.

SPSS version 25.0 software was used for statistical analysis. In descriptive analysis, percentage and frequency were calculated for categorical variables and mean ± standard deviation, or median and interquartile range (IQR) for all continuous variables. Chi-square test was applied to evaluate the association of age, gender, comorbidity, preceding cause, blood counts, and transfusion requirement, as well as the type of transfusion, cytogenetics, and presence of PNH clone on responses at 3, 6, and 12 months. One-way ANOVA was applied to look for the association of baseline Hb with responses. Kruskal-Wallis was applied to check the relationship of baseline ANC, ARC, and platelets with responses. Kaplan-Meier test was applied to calculate the median survival. Log-rank test was applied to check for the association of overall survival (OS) with various factors and baseline blood counts. A p-value of <0.05 was considered to be statistically significant.

RESULTS

A total of 51 patients were enroled in the study. Most of them 39 (77%), belonged to lower socioeconomic class. The most frequent comorbid were autoimmune illness 4 (8%), hypertension 4(8%), diabetes 2 (4%), and hepatitis 2 (4%). Multilineage cytopenias accounted for 53% of patients' initial presentations (Table I). Cytogenetic showed abnormalities in 2 (4%) patients, 1 (2%) patients had trisomy 20, while other 1(2%) patients had loss of Y chromosome. A PNH clone was detected by flow cytometry / FLAER in 8 (16%) patients, and was negative in 32 (63%), while it was not performed in 11 (22%) patients. Baseline characteristics of patients with CBC parameters are shown in Table I.

Overall response rates to CSA monotherapy at 3, 6, and 12 months were 49%, 57%, and 59%, respectively. Responses at 3, 6, and 12 months are shown in Table II. Transfusion dependency was seen in 28 (55%) *vs*. 21 (43%) *vs*. 16 (35%) patients at 3, 6, and 12 months. Six (12%) patients experienced major treatment-related problems; these included CSA-induced posterior reversible encephalopathy (PRES) in 1 (2%) at 7 months of treatment, which resulted in treatment cessation; other issues included azotaemia 2 (4%), hypertension 2 (4%), and hirsuitism 1 (2%).

Table I: Baseline characteristics of participants (Total = 51).

Parameters	p-value
Age in years; median (IQR)	25 (16 - 42)
Gender	
Male	34 (67)
Female	17 (33)
Initial Presentation	N, (%)
Pancytopenia	10 (19.5)
Anaemia with thrombocytopenia	12 (23.5)
Neutropenia with anaemia	2 (4)
Neutropenia with thrombocytopenia	3 (6)
Anaemia	18 (35)
Thrombocytopenia	2 (8)
Neutropenia	4 (4)
Aetiology	
Idiopathic	37 (72)
Autoimmunity	5 (10)
Drug / chemical exposure	4 (8)
Prior hepatitis	3 (6)
Recent infection	2 (4)
Transfusions	
RCC / whole blood	20 (39)
RCC and platelets	19 (37)
Platelets	3 (6)
No transfusions	9 (18)
Baseline CBC parameters	
Hb in g/dl;mean (Min- Max)	8.62 (4.60 - 12.00)
Absolute reticulocyte count x 10^9/l; median (IQR)	75.2 (45 - 141)
Absolute neutrophil count x10^9/l; median (IQR)	0.98 (0.70 - 1.60)
Platelet count x 10^9/l; median (IQR)	20 (10 - 36)
Number of RCC transfusion received; median (IQR)	4 (1 - 7)

Table II: Response to the treatment.

	Response outcomes n (%)		
	3 months	**6 months**	**12 months**
Complete response (CR)	4(8)	7(14)	12(24)
Partial response (PR)	21(41)	22(43)	18(35)
No response (NR)	26(51.0)	22(43)	21(41)
Over all response rate (ORR)			
Response (ORR)	25(49)	29(57)	30(59)
No response	26(51)	22(43)	21(41)

Table III: Association of overall survival with transfusion dependency and responses.

	No of events / total n (% survival)				
	Transfusion dependant			**Overall survival**	**$P_{log\text{-}rank}$**
	Yes		**No**		
3 months	7 / 28(75)		0 / 21 (100)	0 / 51(100%)	0.01
6 months	5 / 21(76)		0 / 28(100)	2 / 51(96%)	0.005
1 year	2 / 16(87.5)		0 / 30(100)	7 / 51(86%)	0.042
	No of events/total n (% survival)				
Responses	**CR**	**PR**	**NR**	**$P_{log\text{-}rank}$**	**ORR ($P_{log\text{-}rank}$)**
3 months	0 / 4(100)	1 / 21(95)	6 / 26(77)	0.13	0.046
6 months	0 / 7(100)	1 / 22(95.5)	6 / 22(73)	0.04	0.01
1 year	0 / 12(100)	1 / 18(94.4)	6 / 21(71)	0.02	0.008

Note: Log Rank test was applied.

Significant association of baseline haemoglobin was found with responses at 6 and 12 months (p = 0.003). However, no significant association was found between responses and other baseline parameters including age, gender, comorbidity, SES, preceding cause, baseline ANC, ARC, platelet counts, and number of RCC transfusions.

Out of the total patients, 44 were alive and 7 had died at the median follow-up of one year, yielding a survival rate (OS) of 86.3%. Significant association was found between transfusion dependency at 3, 6, and 12 months with OS ($P_{log\text{-}rank}$ = 0.01, 0.005, 0.04), and responses at 6 and 12 months with overall survival ($P_{log\text{-}rank}$ = 0.043, 0.02). No association was found between age groups, gender, baseline CBC parameters, comorbidities, SES, initial presentation, type and number of transfusions, cytogenetic abnormality, and presence of PNH clone with overall survival. While there was no association observed between the responses at three months and OS, when the overall response rates were computed, a significant association was found between the time-defined response rate and OS (3 months $P_{log\text{-}rank}$ = 0.046, 6 months $P_{log\text{-}rank}$ = 0.01, and 12 months $P_{log\text{-}rank}$ = 0.008, Table III).

DISCUSSION

The treatment landscape for NSAA remains challenging due to the paucity of novel therapeutic approaches. In resource-constrained settings, where access to specialised therapies such as horse ATG and TPO-RA may be restricted owing to the cost and availability, single-agent IST such as cyclosporine (CSA) is most frequently utilised. The authors demonstrate that the use of CSA alone in NSAA patients leads to a significant haematologic improvement (59% of cases). Major treatment-related problems, though relatively infrequent, necessitate careful monitoring and management to optimise patient outcomes.

The study's findings show that CSA monotherapy is notably effective in treating NSAA patients; after three, six, and 12 months, the overall response rates (ORR) were 49%, 57%, and 59%, respectively. Complete responses with transfusion independence were seen in 8%, 14%, and 24% at 3 , 6, and 12 months, respectively. Notably, the response rates observed in this study are comparable to those reported in previous studies evaluating combined CSA and ATG therapy.[5,10-12] Unlike previous studies that show a better and earlier response in the paediatric population[13] and older age as negative predictor of response,[14] there was no discernible variation in the response rate by age in this study's patient cohort.[13] Previous researches have demonstrated the predictive relevance of the PNH clone as an indicator of immunosuppressive treatment (IST) response in aplastic anaemia.[15,16] Nevertheless, the PNH clone's existence in this study had no impact on treatment responses. This study's findings concurred with those of Li *et al.* who found that the existence of a PNH clone had no impact on individuals' responses.[5]

This study highlighted responses at 6 and 12 months correlated with baseline haemoglobin. This study's finding is consistent with the results of the study done by of Fu *et. al*, in which they concluded that the effectiveness of CSA was related to the levels of RBC, Hb, and Platelet at the time of diagnosis.[17] In this study, the patient's age and baseline blood counts, including ANC, ALC, platelet count, and ARC, had no discernible effect on the response as also evaluated by Zhang *et al.*[18]

Importantly, the present study identified transfusion dependency as a significant predictor of overall survival (OS). This finding was also highlighted in the study done by Matsuda *et al.*, in which they observed improved transfusion-free survival among NSAA patients treated with CSA.[19] Patients who remained transfusion-dependant at 3, 6, and 12 months exhibited lower OS rates compared to those who achieved transfusion independence.

Furthermore, this analysis revealed a significant association between time-defined response rates and OS. Patients who demonstrated a favourable response to CSA at 6 and 12 months exhibited improved OS compared to those with inadequate responses. This dtudy's finding is consistent with the results of an international study conducted by Boddu *et al.* in which they also observed that the patients showing favourable responses to the treatment experienced better long-term outcomes.[20] Although CSA monotherapy was generally well-tolerated subset of patients who experienced treatment-related complications, including CSA-induced adverse events, such as posterior reversible encephalopathy syndrome (PRES), azotaemia, hypertension, and hirsutism. Comparatively, Matsuda *et al.*'s comparative analysis revealed similar incidence rates of adverse events related to renal toxicity associated with CSA therapy in NSAA patients.[19] Another study by Fu *et al.* assessed that the majority of side effects of CSA may be controlled by adjusting the dosage or changing the treatment.[17] In this study, one of the patient developed CSA induced PRES at 7 months of treatment with a partial response to treatment, however, it led to the discontinuation of therapy and switching to an alternate form of therapy.

Since a large portion of the patients had low incomes and were from lower socioeconomic classes, the CSA dosage was adjusted based on side effect profiles, and CSA trough levels were performed only for patients who could afford it.

CONCLUSION

This study highlights the efficacy and safety of CSA monotherapy in patients with NSAA. Overall response rates at 3, 6, and 12 months, indicating the potential of CSA as a viable treatment option, particularly in resource-constrained settings. Despite some patients experiencing treatment-related complications, CSA demonstrated a generally tolerable safety profile.

ETHICAL APPROVAL:
This study was carried out at the Department of Clinical Haematology, Armed Forces Bone Marrow Transplant Center, Rawalpindi, Pakistan, after approval by the Hospital Ethical Committee (Ref: IRB-015/AFBMTC/Approval/2021).

PATIENTS' CONSENT:
Data were retrieved from medical record of patients with informed consent.

COMPETING INTEREST:
The authors declared no conflict of interest.

AUTHORS' CONTRIBUTION:
MNA: Acquisition of the data, Drafting of the work, and agreement to be accountable for all aspects of the work.
NS: Revising the work critically for the important intellectual content.
IH: Acquisition of the data, conception, and design of the work.
AAK: Literature search, and data interpretation.
HK: Analysis of the data.
AS: Final approval of the version to be published.
All authors approved the final version of the manuscript to be published.

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