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Adjunctive effects of eltrombopag on immunosuppressive therapy for childhood aplastic anemia

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ABSTRACT

Eltrombopag is used with first-line immunosuppressive therapy for adult aplastic anemia, although its practical utility in childhood remains unclear. We retrospectively analyzed the outcomes of pediatric patients who received eltrombopag in Japan. Of the 27 eligible patients, 23 (85%) were previously treated, and 15 (56%) had severe or very-severe disease. Seventeen (63%) received eltrombopag with or after rabbit anti-thymocyte globulin plus cyclosporin-A. Within the first year of eltrombopag therapy, 12 patients showed a good or partial response, 15 showed no response, and 8 non-responders successfully underwent hematopoietic cell transplantation. Within the first 3 months after eltrombopag therapy, all but one of the transfusion-dependent responders became transfusion-independent. At 12 months, 6 of 12 responders were disease-free off-treatment. The one-year overall response rate was higher for severe or very-severe than non-severe cases (p=0.006). Multivariable analysis showed that very-severe disease at the start of eltrombopag therapy was a predictor of being disease-free off-treatment (p=0.03). No cytogenetic abnormalities developed, but myelofibrosis occurred 4 months after eltrombopag therapy in one non-responder with very-severe disease. The first 3 months' response to adjunctive eltrombopag may guide to the safe and effective use for the cure of disease, although prospective trials are needed to determine its long-term effects.

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INTRODUCTION

Acquired aplastic anemia (AA) is a life-threatening disease arising from the immunological depletion of hematopoietic stem cells (HSCs) [1,2]. Severe or very-severe cases are at high risk for mortality from bleeding or infection, although immunosuppressive therapy (IST) and/or allogeneic hematopoietic cell transplantation (HCT) have greatly improved the overall survival in a couple of decades [3]. HCT is a curable treatment for severe AA, while anti-thymocyte globulin (ATG) and/or cyclosporin-A (CSA)-based IST is the first-line treatment if no suitable donors are available. The overall response rate to IST was increased in childhood AA [4], but refractory very-severe cases have a poor prognosis during the early treatment and the early off-treatment of non-severe disease is not easy without HCT. The 2-year overall survival rate for the first HCT in patients under age 30 years has attained from 91% in related donors to 96% in unrelated donors [5]. However, alternative donor HCT may result in an early demise or acute and long-term complications associated with graft-versus-host disease (GVHD), regimen-related toxicity including growth retardation, infertility and subsequent neoplasms, compared with IST. The challenges with IST include unfavorable response, relapse, and clonal evolution, leading to a sustained remission in 40–60% of responders [6-9]. The occurrence of IST-resistant bone marrow failure syndromes (IBMFS) and IST-responsive hepatitis-associated AA also call the clinical dilemma to start the first-line treatment in pediatric patients.

Eltrombopag (EPAG) is an oral, non-peptide compound of low molecular weight that acts on the transmembrane domain of the thrombopoietin receptor, c-MPL. c-MPL is expressed on HSCs as well as megakaryocyte progenitor cells [10]. Desmond et al [11] reported that 40% of adult patients with relapsed or refractory severe AA who received EPAG after one or more horse ATG (hATG) achieved at least one lineage of blood cell recovery, including a few cases of complete response (CR). The prospective clinical trial of EPAG in combination with hATG and CSA for over 15 years patients with newly diagnosed severe AA reported a favorable response rate with 94% of the overall response rate including 58% CR cases [12]. The other trial indicating the favorable response allowed to use EPAG in practice for adult newly diagnosed or relapsed/refractory cases in Japan [13,14]. The Food and Drug Administration (FDA) provides the insurance coverage of EPAG for severe AA at age 2 years and older, but the reports on EPAG in pediatric cases are limited in number [15]. In a prospective, non-randomized clinical trial on EPAG in combination with hATG and CSA in 40 patients less than age 18 years with severe AA, the response rate at 6 months under

teens was 63% compared with 78% of a historical control [16]. Recently, the favorable effect of EPAG on IST response has been indicated in treatment-naïve cases with severe but not non-severe or very-severe disease [16-20]. Moreover, the time of off-treatment varies in patients who obtained CR because a cure of disease is hard to determine in pediatric cases.

To establish the HCT-free treatment for the cure of childhood AA, we retrospectively analyzed the outcomes of pediatric patients who received EPAG mainly adjunct to ATG-based IST in the Japanese registry, focusing on the effective application and safety over one-year course of treatment.

METHODS

Patients and treatments

Participants were patients age <15 years who received the diagnosis of acquired AA or refractory cytopenia of childhood (RCC) from January 2010 to December 2019 in the hospitals registered in the Japan Childhood Aplastic Anemia Study Group. The diagnosis and the severity of disease were determined according to the established Camitta's criteria [3]. Disease was considered "severe" if at least two of the following were present in complete blood counts: (i) absolute neutrophil count (ANC) $<0.5\times10^9$/L; (ii) platelet count $<20\times10^9$/L; and (iii) reticulocyte count $<20\times10^9$/L with a hypocellular bone marrow (BM). "Very-severe disease" was defined if the above criteria for severe disease were fulfilled and ANC was $<0.2\times10^9$/L. "Non-severe disease" was defined by at least two of the following: (i) ANC $<1.0\times10^9$/L, (ii) platelet count $<50\times10^9$/L; and (iii) reticulocyte count $<60\times10^9$/L with a hypocellular BM.

Oral EPAG was started to continue at the standard dose (up to 5 mg/kg/day) for treatment-naïve patients or patients who did not respond to any treatments including rATG, CSA, high-dose or conventional-dose corticosteroid, anabolic steroid, and/or granulocyte colony-stimulating factor (G-CSF). The dosage or the timing of the start and end of EPAG depended on the arbitrary decision by the attending doctors as a weaning from EPAG in responders or the withdrawal in non-responders. Crushed tablets of EPAG were given to children who could not swallow pills. The IST for very-severe, severe, or non-severe cases consisted of CSA with or without rabbit ATG (rATG, Thymoglobulin; Genzyme Co., Cambridge, MA, USA) (2.5-3.75 mg/kg per day, days 1 to 5) and methylprednisolone for prophylaxis against allergic reactions. G-CSF was given to very-severe, severe, or non-severe cases with infections. Allogeneic BM transplantation was recommended for patients with severe, very-severe, and

transfusion-dependent non-severe diseases who had a suitable donor in their relatives or the unrelated donor registry in the Japanese bone marrow (BM) or cord blood (CB) Bank Network. Newly diagnosed patients who received more than one month's administration of EPAG until age 15 were enrolled for the study. Clinical and laboratory information was obtained from the medical records by attending doctors. The last follow-up information was obtained March 31, 2023. The retrospective observational study was approved by the Ethic Committee of Kyushu University Hospital (#2019-594). The retrospective, observational, and questionnaire-based study was conducted based on the collected data from pediatric facilities throughout Japan to clarify the actual clinical practice for pediatric AA concerning the use of EPAG. The administration in 4 cases prior to the insurance approval in Japan was carefully started according to the regulation of each institution after the ethical considerations with the consents from patient's guardians.

Assessment of treatment response

Treatment response was evaluated in each patient at every 3 months from the start of EPAG for 12 months, and at the last visit until March 31, 2023. The treatment response was classified into "no response (NR)", "good response (GR)", "partial response (PR)", and "curable response (CuR)". NR is defined if either severity or transfusion dependency (TD) did not change. PR is defined if the severity did not change but the patient was weaned from blood transfusion. GR is defined if the severity changed from very-severe to severe, non-severe disease, subnormal complete blood counts, or no cytopenia. CuR is defined as having no cytopenia for more than 6 months. Because cytopenia means under the standard of complete blood counts for age, CuR is more strictly defined than the CR defined as ANC $>1.0\times10^9$/L, a platelet count $>100\times10^9$/L, and a hemoglobin concentration >10.0 g/dL [15]. A relapse of disease was defined by a decreased severity and/or the requirement for blood transfusions from the start of EPAG.

Statistical analyses

Statistical analysis was performed using the Kruskal-Wallis test for continuous variables and Fisher's exact test for dichotomous variables, supplemented with the Bonferroni correction when appropriate. Results with a 2-sided p–value <0.05 were considered significant. The univariate and multivariable analyses were statistically completed by using JMP 17.0 (SAS Institute, Inc, Cary, NC).

RESULTS

Diagnosis and pretreatment of EPAG

Patient characteristics before the start of EPAG are shown in Table 1. This observational study was performed on 27 patients including 3 treatment-naïve patients with 16 males and 11 females diagnosed at age of the median 9 years ranging from 1.5 to 14 years. The median observation time was 53 months ranging from 30 to 98 months. The final diagnoses were 23 idiopathic AA with one anorexia nervosa, 3 hepatitis-associated, and 1 RCC. The severity was severe or more severe in 15 patients and non-severe in 12 patients at diagnosis. The distributions of sex, severity, and choice of HCT until the last visit did not differ between the younger (age 1-8 years at diagnosis) and the older patients (9-14 years). Until the start of EPAG, 23 patients did not attain CuR after the conventional treatments including 13 rATG plus CSA, 9 CSA and/or danazol, and 1 prednisolone alone, and 4 received no treatment after the onset of disease. The time from the diagnosis to the start of pre-EPAG treatment and EPAG was the median 0 months (range 0-111) and 6 months (range 0-111), respectively. The baseline complete blood counts at the start of EPAG were as follows; the median neutrophils 0.76×10^9/L (range, 0.06-2.2×10^9/L), reticulocytes 40×10^9/L (range, 2-$10^9\times10^9$/L) and platelets 19×10^9/L (range, 4-79×10^9/L). TD at the start of EPAG was found for red cells in 18, for platelets in 17, and for both in 15. G-banding for the mitogen-unstimulated BM samples at diagnosis reported 47,XYY or 46,XX,inv(9)(p12q13) in each patient. The median starting dose was 1.0 mg/kg/day (range, 0.23-2.6 mg/kg/day), and the median final dose of EPAG was 2.1 mg/kg/day (range, 0.4-4.3 mg/kg/day).

Treatment outcomes

The flowchart of entire treatment course after the start of EPAG is shown in **Figure 1**. Until 12 months after EPAG, 4 transfusion-dependent patients and 4 non-responders without TD received HCT (**Supplementary figure 1**). During the period from 12 months after EPAG to the last visit, two of 19 patients underwent HCT because of prolonged NR. The overall response rates at 3 months, 6 months, and 12 months after EPAG therapy were 56%, 60%, and 63%, respectively, showing no significant difference (**Figure 2**). Among 19 patients who did not receive HCT for 12 months after EPAG, 13 patients showed 12 GR or 1 PR, and 6 showed NR within the first 3 months of EPAG (**Figure 3**). During the latter 9 months of EPAG therapy, 2 relapses and one late GR occurred. The final response rate at 12 months was significantly higher in

severe or very-severe patients than non-severe patients (91% *vs.* 25%, p=0.0063).

Details of 12 responders until 12 months after EPAG are shown in **Table 2**. They were all TD patients at the start of EPAG and all but one (UPN9) were weaned from transfusions within 3 months of EPAG therapy. All patients with the final CuR had severe or very-severe disease at diagnosis and underwent IST with rATG, 5 of them received the diagnosis less than 10 years. They had the time from first rATG to starting EPAG of median 2 months ranging 0-7, the duration of EPAG of median 10 months ranging 2-43, and the observation periods after the discontinuation of EPAG of median 42 months ranging 8-56. Multivariable analysis indicated that very-severe disease at the start of EPAG was a predicting factor for CuR at the last visit (p=0.03, Table 3) (Supplementary Table 1).

Adverse events

All 27 patients survived at the last visit. No clonal evolution, or progression to myelodysplastic syndromes / acute myeloid leukemia developed. Mild bilirubinemia and elevated serum creatinine levels appeared during the administration of EPAG in 6 and 2 patients, respectively. Herpes zoster and parotitis developed during EPAG therapy (Supplementary Table 2). Because all events were CTCAE grade 2, they continued the administration of EPAG. A 5-year-old girl with severe idiopathic AA and 46,XX,inv(9)(p12q13) received the diagnosis of myelofibrosis (WHO classification grade 2 according to the finding of bone marrow biopsy) 4 months after EPAG, that was started as a 6 months' non-responder to the 1st IST of rATG plus CSA. Successful HCT after the discontinuation of EPAG led to a cure of AA without myelofibrosis.

DISCUSSION

The retrospective study revelated that more than half of patients responded to adjunctive EPAG in previously treated and the first 3 month' weaning from blood transfusion were associated with the response one-year after EPAG therapy. The first 3 months' responses in very-severe cases were associated with no cytopenia and off-treatment for over 6 months at the latest observation. Notably, very-severe disease at the start of EPAG was indicated as an effective predictor for disease-free off-treatment.

The concurrent start of ATG and EPAG has been reported to have a favorable response in treatment-naïve adult SAA, suggesting an augment effect of EPAG on the IST response according to the severity [12]. This observational study included only 4

treatment-naïve patients; one received rATG plus CSA, one received CSA alone and 2 others. The majority of patients suffered from refractory diseases although 4 of 15 (27%) severe or very-severe diseases responded to responses. In a report of 11 pediatric patients with severe AA who received IST and EPAG, 4 patients started EPAG at the same time as ATG and 7 patients started EPAG within 2 months of starting ATG. Among the 7 patients with the later EPAG, only one was weaned from blood transfusions within 3 months but four others eventually achieved a CR [19]. The multivariable analysis did not significantly indicate that a shorter time from IST to the start of EPAG contributed to CuR at the last visit. The large-scale prospective study may be needed to demonstrate the favorable effect of early start of EPAG on the final comes over one year.

A prospective clinical trial in pediatric patients has recently reported the different treatment efficacy of IST with EPAG according to the severity of disease, focusing on the compared overall response rates [20]. This study showed a significant increase in the response rate in patients with severe AA compared with the conventional IST group (89% vs 57%; p=0.028), but not found in those with very-severe disease (52% vs 50%; p=0.902). On the other hand, the present study presented a conflicting result in patients with very-severe disease, who had a significant association with CuR (Table 2, 3). Although the time at the disease onset is unknown, the progressive speed of cytopenia depends on the interval from the onset to the diagnosis. At the start of EPAG, ANC in patients who receive IST might represent the depressed potential of hematopoietic stem cells as well as the decreased number of activated T cells originating from the BM T cell progenitors. The patient with the lowest number of ANC at the start of EPAG (0.006×10^9/L) showed NR within the first 3 month and then underwent alternative donor HCT (Supplementary figure 1). By contrast, 5 of 6 patients with CuR showed a median ANC of 0.11×10^9/L ranging from 0.044 to 0.14, although one patient with repeated rATG plus CSA showed an increasing ANC (2.2×10^9/L) at the start of EPAG not indicating the augmented effect of EPAG. In this context, the prospective trial might have included increased number of patients with very-severe AA with higher severity than this study population of very-severe disease. The dosage of EPAG for pediatric patients with AA was not specified and varies among previous reports (range, 1-2.5 mg/kg/day: maximum 150 mg/day) [15]. The median targeted dose of EPAG in this study was 2.1 mg/kg/day. There was no significant difference in the dosage between responders and non-responders (data not shown).

The other concern is the practical utility of EPAG in non-severe patients. This

study included many patients with non-severe disease although the proportion increased from 44% at diagnosis to 67% at the start of EPAG therapy. Several reports suggested the clinical utility of EPAG with IST in non-severe patients [13,14,21], while the long-term effect has not been established. In this study, no transfusion-dependent non-severe patients achieved the final CuR, even after they showed GR or PR up to 3 months after the start of EPAG. The prolonged use of EPAG in non-severe patients may be useful for the attainment of early transfusion independency but not the final CuR. In severe AA, higher amount of IFN-γ and activated effector memory $CD8^+$T cells inhibit the growth and differentiation of against hematopoietic stem cell progenitor cells. IFN-γ might act as a decoy circulating agent obstructing the physiological binding of growth factors, especially TPO, to their receptors [22]. These may explain the augmented IST response in severe cases in the setting of adjunctive EPAG.

Clonal evolution to MDS/AML is a noticeable complication in pediatric patients with AA during the long-term follow up [6]. There was no significant increase in cases of clonal evolution or MDS/AML transition in adult patients with EPAG compared with those with conventional IST [15,21]. However, there is a report of earlier development of clonal evolution in the EPAG group compared with the historical control group [23]. The NIH group reported that clonal evolution and MDS/AML transition in the EPAG group occurred in 5 pediatric patients (13%), with no significant difference from the historical control group (9%). Monosomy 7 developed in 3 patients including early onset diseases at 3 months, and all of them underwent HCT [16]. Goronkova et al. [20] reported only one transient chromosome aberration developed in 49 patients in another prospective study, with no significant expression compared with the IST group. During the long-term observation in the present study, there was no newly developed clonal evolution. However, one patient received the diagnosis of myelofibrosis 4 months after EPAG therapy and achieved CuR after successful HCT. There are no reports of myelofibrosis in pediatric or adult patients with AA due to concomitant use of EPAG, although the long-term use of thrombopoietin receptor-agonists may be considered as a potential cause of myelofibrosis in allogeneic HCT recipients [24]. Considering the early onset myelofibrosis, the first 3 months' BM study warrant to provide the response as well as serious complications to change the treatment strategy to an alternative HCT.

The observational study has several limitations. Because of a small number of patients, only 4 very-severe patients at the start of EPAG were associated with the final CuR. During the prolonged follow-up time, non-responders tended to undergo alternative HCT. In the study on treated patients, the treatment modality may affect the

subsequent response. Further studies are needed to stratify the patients with very-severe disease according to the low ANC at diagnosis in the prospective randomized trials.

In conclusion, the first 3 months' response to EPAG led to the weaning from blood transfusions in childhood AA and might also predict the cure of very-severe disease without HCT. Considering the risk of myelofibrosis and clonal disease, the first 3 months' response may guide to the safe and effective use of EPAG, although prospective trials are needed to determine the long-term effects.

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Data Availability Statement: The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

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Figure legends

Figure 1. The overall diagram of treatment course in 27 patients who received eltrombopag (EPAG). HCT, hematopoietic stem cell transplantation; TD, transfusion dependency.
HAAA, hepatitis-associated aplastic anemia; HCT, hematopoietic cell transplantation; MF, myelofibrosis

Figure 2. Treatment response by the severity at diagnosis.

Figure 3. Time-dependent treatment response after the start of EPAG in non-HCT patients. The solid and dashed lines stand for the one-year responder and non-responders after EPAG, respectively. Shaded areas show the grade of severity with or without transfusion dependency.
EPAG, eltrombopag; HCT, hematopoietic cell transplantation; IST, immunosuppressive therapy; rATG, rabbit anti-thymocyte globulin.

Table 1. Demographics and clinical profile of patients

	N = 27
Median age, range	9 years, 1.5−14
Sex (%)	
Female	11 (41)
Male	16 (59)
Median time of observation after EPAG, range	53 months, 30−98
Diagnosis (%)	
Idiopathic aplastic anemia	22 (81)
Hepatitis associated aplastic anemia	3 (11)
Refractory cytopenia of childhood	1 (4)
Aplastic anemia	1*(4)
Treatment before initiating EPAG (%)	
1st. rATG plus CSA	12 (44)
2nd. rATG plus CSA	1 (4)
CSA ± anabolic hormone	9 (33)
Prednisolone	1 (4)
No treatment	4 (15)
Cytogenetic abnormality	
47,XYY	1
46,XX,inv (9) (p12q13)	1
Severity (%) at diagnosis/start of EPAG	
Very-severe or severe	15/9 (56/33)
Non-severe	12/18 (44/67)
Transfusion dependency (%) at the start of EPAG	
Red blood cells	16 (59)
Platelet cells	15 (56)
Both	13 (48)
Median blood counts ($\times 10^9$/L), range at the start of EPAG	
Neutrophils	0.76, 0.006–2.2
Reticulocytes	40, 2−109
Platelets	19, 4−79

*The patient received the diagnosis of aplastic anemia and anorexia nervosa.
CSA, cyclosporin-A; EPAG, eltrombopag; rATG, rabbit antithymocyte globulin.

Table 2. Clinical profiles of all 12 responders within the first year of EPAG therapy

UPN	Diagnosis	Age (years) at diagnosis	Severity and transfusion at diagnosis/the start of EPAG	Time from the first rATG treatment	EPAG administration			Present state		
					dose, mg/kg/day	3-month-response	durations	treatment	final response	durations[#]
1	IAA*	1.5	vs / ns, TD	7/3 mo. after 1st/2nd rATG	4.2	GR	10 mo.	off	CuR	56 mo.
2	IAA	2	s / vs, TD	3 mo.	1.9	GR	43 mo.	off	CuR	18 mo.
3	HAAA	4	vs / vs, TD	0 mo.	2.6	GR	2 mo.	off	CuR	55 mo.
4	IAA**	7	vs / vs, TD	2 mo.	3.0	GR	9 mo.	off	CuR	42 mo.
5	IAA	9	vs / ns, TD	3 mo.	0.4	GR	22 mo.	off	CuR	8 mo.
6	HAAA	11	s / vs, TD	1 mo.	2.5	GR	7 mo.	off	CuR	47 mo.
7	IAA	14	s / ns, TD	1 mo.	1.5	GR	27 mo.	CSA	GR	12 mo.
8	IAA	4	s / ns, TD	no rATG	2.0	PR	56 mo.	EPAG	PR	56 mo.
9	IAA**	4	s / s, TD	6 mo.	2.9	NR	50 mo.	CSA, EPAG	GR	50 mo.
10	IAA	10	ns / ns, TD	2 mo.	2.9	PR	36 mo.	CSA, EPAG, DNZ	PR	36 mo.
11	IAA	12	ns / ns, TD	no rATG	2.5	PR	43 mo.	CSA, EPAG, DNZ	PR	43 mo.
12	AA with AN	13	s / s, TD	no rATG	1.5	GR	55 mo.	EPAG	GR	55 mo.

All responders were transfusion-dependent at the start of EPAG and all but one (UPN9) were weaned from transfusions within 3 months of EPAG therapy.
* This infant received the diagnosis of very severe AA because pancytopenia was noticed 3 months after the primary infection of cytomegalovirus.
** Theses two patients were reported in reference 25 (in Japanese).
# The time is calculated from the stop of EPAG to last observation in UPN1-7 and from the start of EPAG to last observation in UPN8-12.
Good response (GR), partial response (PR), and no response (NR) are defined by the Camitta's criteria. Curable response (CuR) means no cytopenias for more than 6 months. Cytopenia was defined as under the standard values of complete blood counts for age.
AA, aplastic anemia; AN, anorexia nervosa; CSA, cyclosporin-A; DNZ, danazol; EPAG, eltrombopag; HAAA, hepatitis-associated aplastic anemia; IAA, idiopathic aplastic anemia; mo., months; ns, non-severe; rATG, rabbit anti-thymocyte globulin; s, severe; TD, transfusion-dependent; UPN, unique patient number; vs, very-severe.

Table 3. Multivariable analysis for predicting the curable response (CuR)* at the last observation.

Variables	N	crude OR (95% CI)	p-value	adjusted OR† (95% CI)	p-value
Time from diagnosis to EPAG‡	27	0.82 (0.62−1.08)	0.15	0.75 (0.51−1.11)	0.15
Time from IST to EPAG‡	27	0.77 (0.55−1.06)	0.11	0.69 (0.44−1.09)	0.11
Very-severe at diagnosis	7	12 (1.48−97.2)	0.02	7.46 (0.80−69.5)	0.08
Very-severe at the start of EPAG	6	19 (2.03−177)	0.01	14.3 (1.37−149)	0.03
Wean from transfusions at 3 months after EPAG	12	1.38e8 (0.00−∞)	0.99	3.70e8 (0.00−∞)	0.99

* Curable response is defined as having normal complete blood counts without treatment.
† Adjusted for age at diagnosis.
‡ per one-month increase.
CI, confidence interval; EPAG, eltrombopag; IST, immunosuppressive therapy; OR, odds ratio.

Figure 1

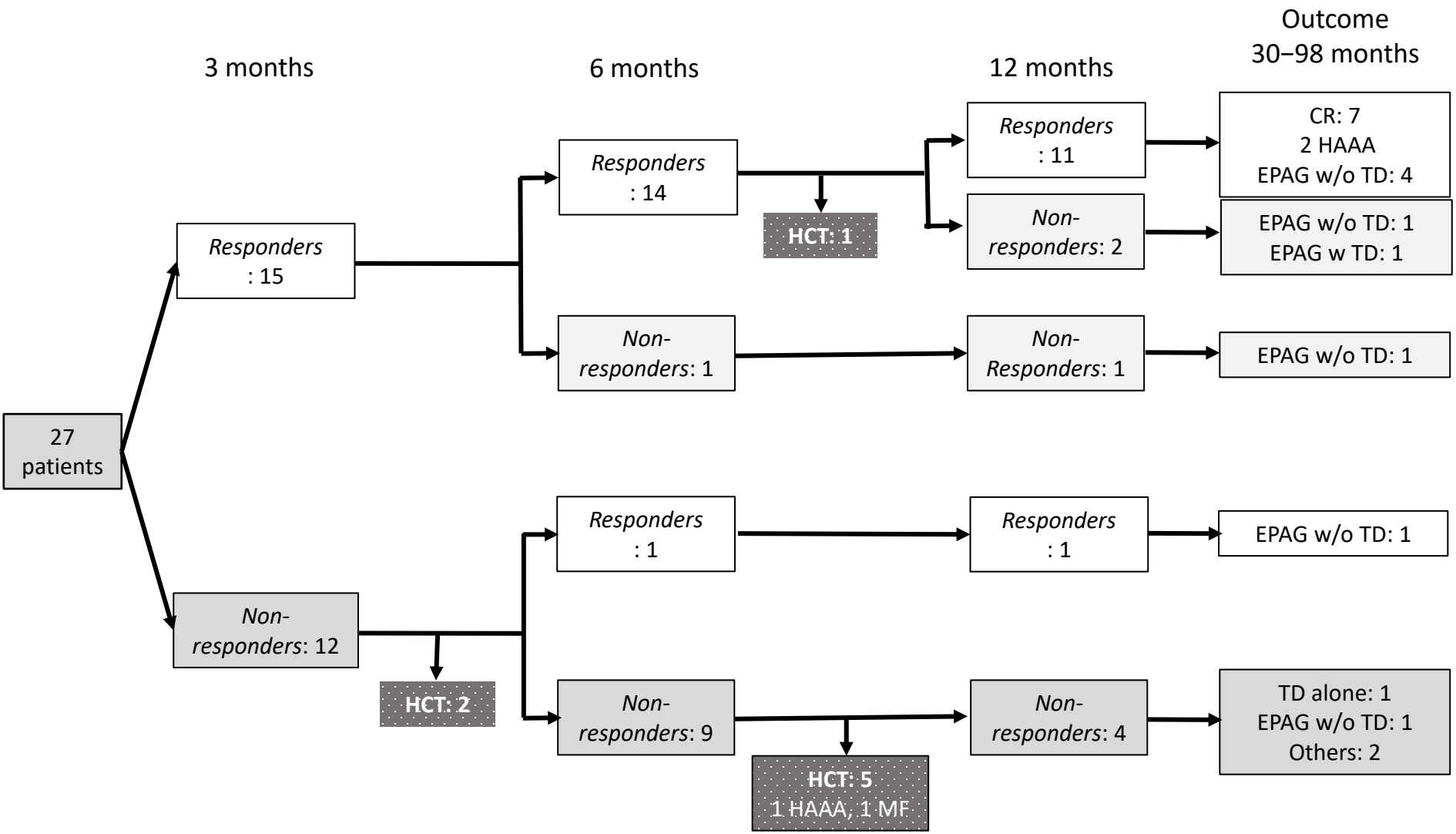

3 months
6 months
12 months
Outcome
30–98 months
27 patients
Responders : 15
Non-responders: 12
HCT: 2
Responders : 14
HCT: 1
Non-responders: 1
Responders : 1
Non-responders: 9
HCT: 5
1 HAAA, 1 MF
Responders : 11
Non-responders: 2
Non-Responders: 1
Responders : 1
Non-responders: 4
CR: 7
2 HAAA
EPAG w/o TD: 4
EPAG w/o TD: 1
EPAG w TD: 1
EPAG w/o TD: 1
EPAG w/o TD: 1
TD alone: 1
EPAG w/o TD: 1
Others: 2

Figure 2

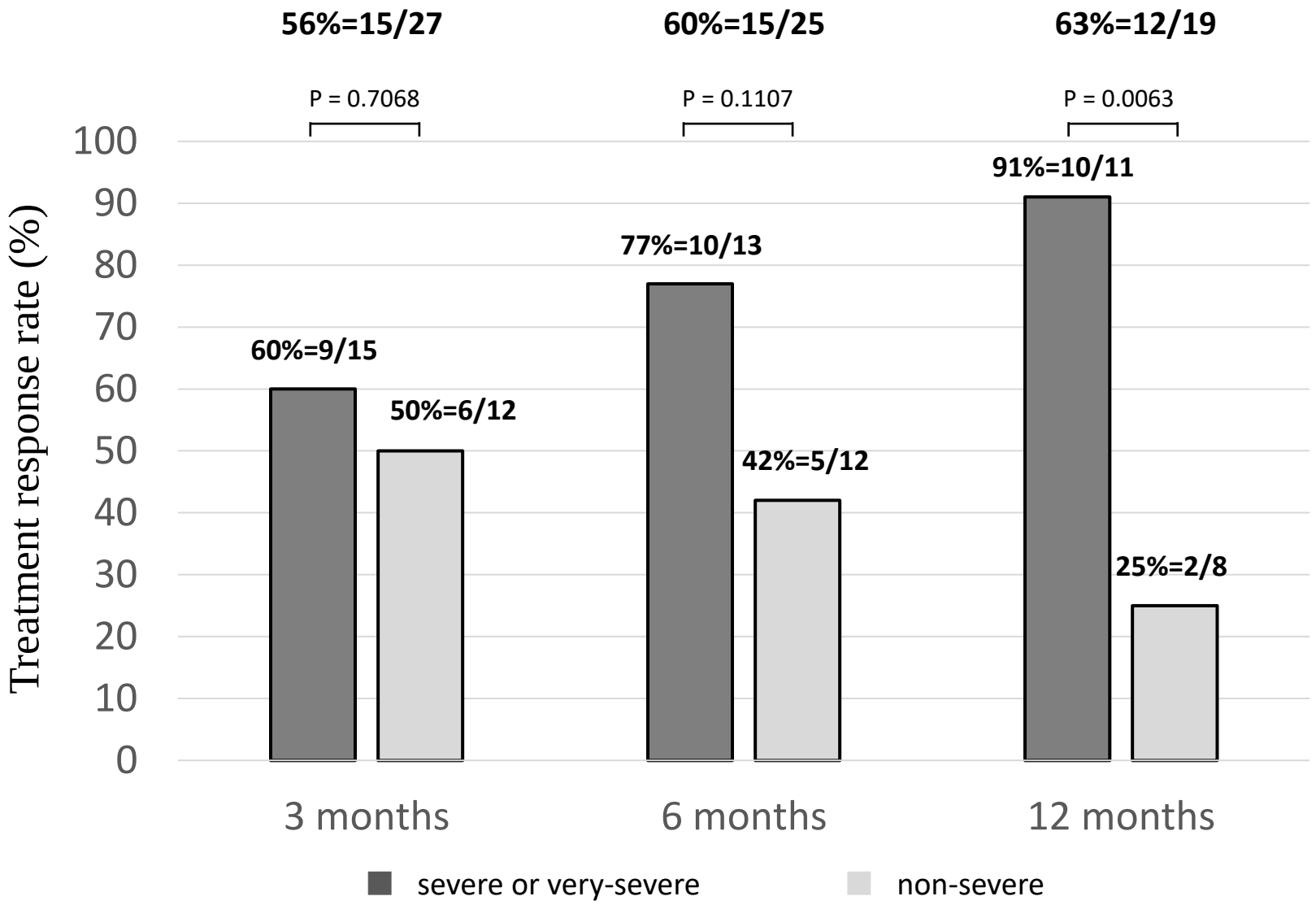
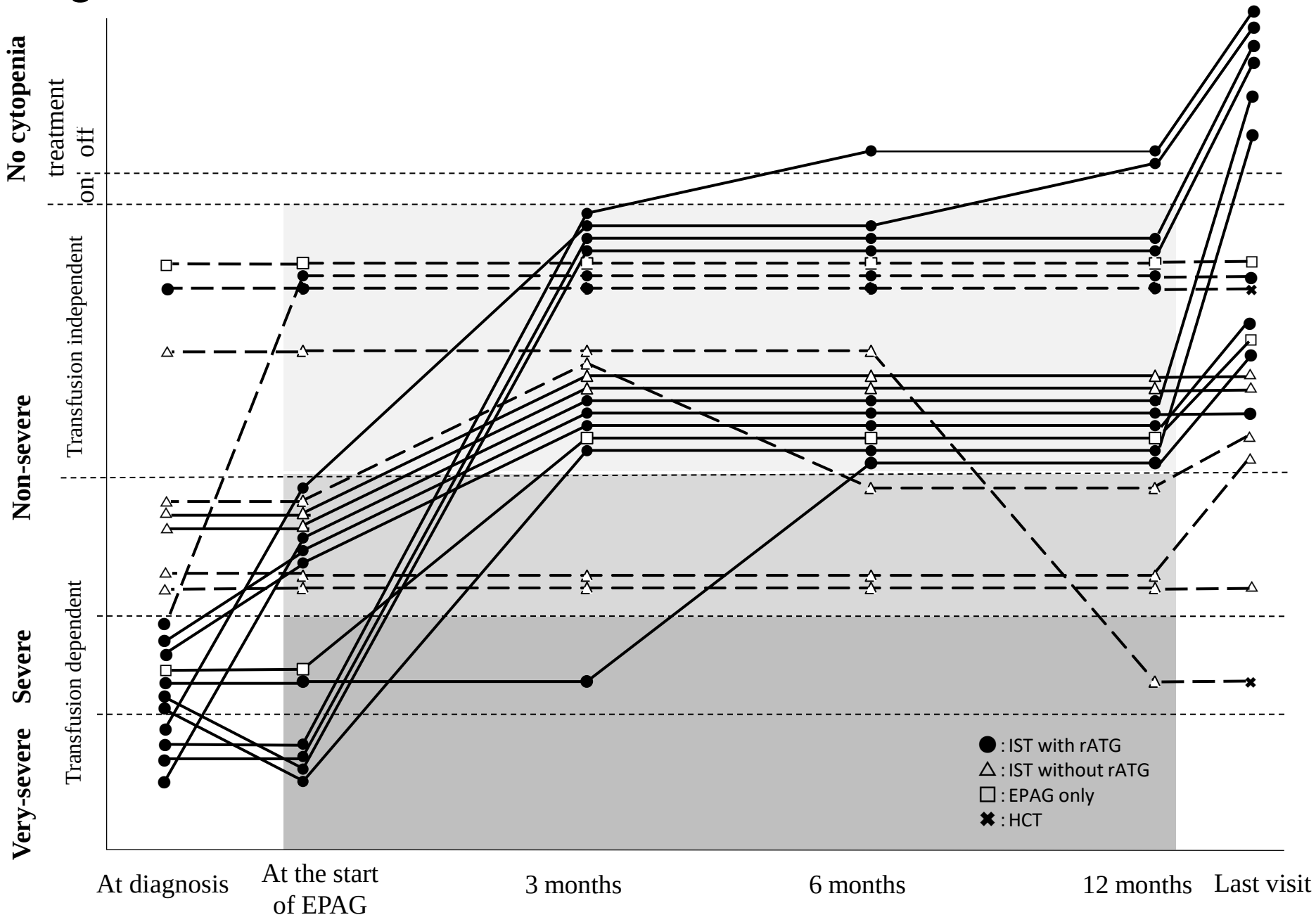

56%=15/27
60%=15/25
63%=12/19
P = 0.7068
P = 0.1107
P = 0.0063
Treatment response rate (%)
100
90
80
70
60
50
40
30
20
10
0
60%=9/15
50%=6/12
77%=10/13
42%=5/12
91%=10/11
25%=2/8
3 months
6 months
12 months
severe or very-severe
non-severe

Figure 3


No cytopenia
treatment off
treatment on
Non-severe
Transfusion independent
Severe
Transfusion dependent
Very-severe
: IST with rATG
: IST without rATG
: EPAG only
: HCT
At diagnosis
At the start of EPAG
3 months
6 months
12 months
Last visit

Supplementary table 1. Multivariable analysis for predicting the one-year complete response after EPAG

Variables	N	crude OR (95% CI)	p-value	adjusted OR† (95% CI)	p-value
Time from diagnosis to EPAG‡	27	0.36 (0.10–1.31)	0.12	0.20 (0.01–2.84)	0.23
Time from IST to EPAG‡	27	0.52 (0.19–1.40)	0.19	0.42 (0.13–1.38)	0.15
Very-severe at diagnosis	7	5.97e7 (0.00–∞)	0.99	2.22e8 (0.00–∞)	0.94
Very-severe at the start of EPAG	6	7.53e7 (0.00–∞)	0.99	2.45e8 (0.00–∞)	0.99
Wean from transfusions at 3 months after EPAG	12	2.85e7 (0.00–∞)	0.99	3.88e7 (0.00–∞)	0.99

† Adjusted for age at diagnosis.
‡ per one-month increase.

Supplementary table 2. Adverse events during EPAG therapy

	n = 27
Mild bilirubinemia (%)	6 (22)
Serum creatinine elevation (%)	2 (7)
Herpes zoster (%)	1 (4)
Parotitis (%)	1 (4)
Progression of myelofibrosis (%)	1 (4)

Supplementary figure 1

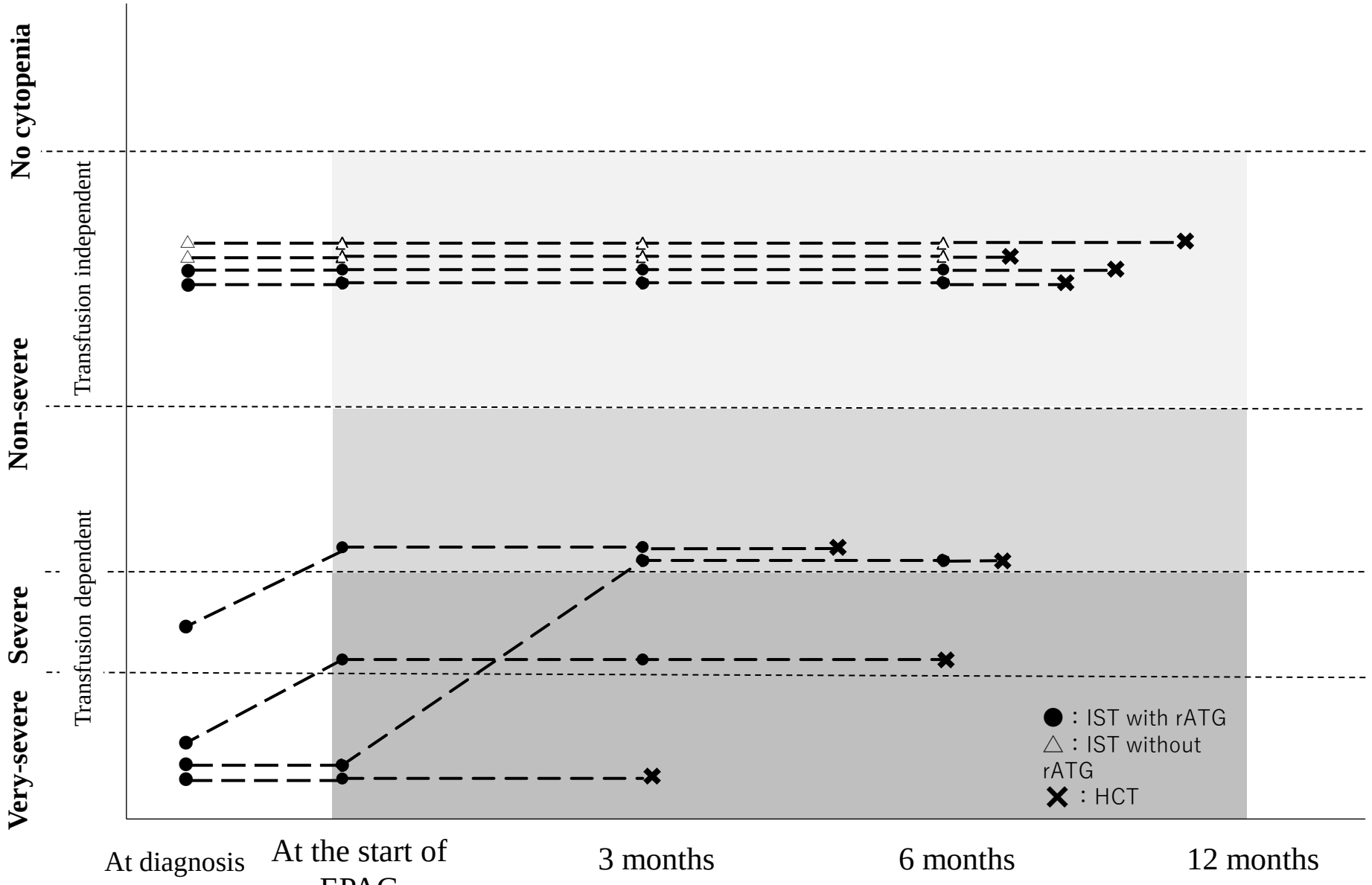


Figure legend. Time-dependent treatment response after the start of EPAG
in HCT patients