

## Time to stop prophylactic platelet transfusion for adult dengue

Thrombocytopenia is a common manifestation of dengue infection.<sup>1</sup> The complex pathogenesis entails not only decreased platelet production due to bone marrow suppression and attenuation of megakaryocyte maturation, but also increased peripheral platelet destruction.<sup>2</sup> Thrombocytopenia in dengue fever is transient, and spontaneous recovery in platelet count is almost always observed.<sup>1</sup> The exact pathogenesis of bleeding in dengue is not fully understood and is thought to be multifactorial. Although low platelet count is considered a risk factor for bleeding in adult dengue,<sup>3</sup> a direct correlation between the degree of thrombocytopenia and risk of bleeding—as documented in other disorders such as acute leukaemia<sup>4</sup>—has not been observed.<sup>5</sup> The role of prophylactic platelet transfusion in the prevention of clinical bleeding in hypoproliferative thrombocytopenia seen in acute leukaemia, haemopoietic stem-cell transplantation, or chemotherapy has been well elucidated.<sup>6</sup> However, the usefulness of prophylactic platelet transfusion in dengue-related thrombocytopenia has remained questionable. The substantial heterogeneity in the use of prophylactic platelet transfusion for dengue fever worldwide reflects the poor evidence base in this area.<sup>7</sup>

Results from several retrospective studies<sup>5,8</sup> and a small randomised trial<sup>9</sup> have shown a lack of efficacy of prophylactic platelet transfusion in adult patients with dengue and thrombocytopenia. Patients who received platelet transfusion also had a higher incidence of adverse events than those who did not.<sup>8,9</sup> In *The Lancet*, David Lye and colleagues<sup>10</sup> report results of a multicentre, open-label, randomised, superiority trial of prophylactic platelet transfusion plus supportive care (transfusion group) versus supportive care alone (control group) in adult patients with dengue and thrombocytopenia. Of 372 enrolled patients, 188 were randomly assigned to receive four units of pooled platelets each day when their platelet count was 20 000 per  $\mu\text{L}$  or lower. The primary endpoint was clinical bleeding (excluding petechiae) by study day 7 or hospital discharge.

No significant difference was observed in the incidence of clinical bleeding between the transfusion group and the control group by day 7 (40 [21%] of 187 patients vs 46 [26%] of 182 patients;  $p=0.16$ ) and day 21 (42 [22%] vs 49 [27%];  $p=0.34$ ). The incidence

of severe bleeding was similar between the two groups: by day 21, severe bleeding occurred in three (2%) patients in the transfusion group and seven (4%) in the control group ( $p=0.21$ ). However, the overall incidence of severe bleeding was low, and the study was not powered to test the superiority of prophylactic platelet transfusion therapy in the prevention of severe bleeding. To find out whether transfusion can prevent bleeding at lower platelet counts, the investigators did a post-hoc subgroup analysis based on baseline platelet count. The incidence of clinical bleeding by day 7 was similar between the two groups in patients with a platelet count of less than 10 000 per  $\mu\text{L}$  and less than 5000 per  $\mu\text{L}$ , indicating a lack of efficacy at any degree of thrombocytopenia. Moreover, platelet transfusion did not expedite platelet recovery: the median time to a sustained platelet count of more than 50 000 per  $\mu\text{L}$  was similar between the two groups (4 days [IQR 3–5] in the transfusion group vs 4 days [3–4] in the control group;  $p=0.39$ ). The observation that platelet counts were higher in the transfusion group than in the control group on day 2 but similar on subsequent days is likely to reflect immune-mediated peripheral platelet destruction<sup>2</sup> and shortened survival time of transfused platelets.<sup>9</sup> The incidence of adverse events was significantly higher in the transfusion group than in the control group. Adverse events related to platelet transfusion included urticaria, anaphylactic reaction, transfusion-related acute lung injury, and fluid overload.

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However, this trial was not designed to study the efficacy of different doses of prophylactic platelet transfusion. Since all patients in the transfusion group received the same dose of platelets, nothing can be said about the efficacy of transfusion at doses higher than four units of pooled platelets per day. In a randomised trial of low, medium, and high doses of prophylactic transfusion in hypoproliferative thrombocytopenia,<sup>4</sup> platelet dose had no significant effect on the incidence of bleeding. Results from a randomised controlled trial<sup>9</sup> using high doses of transfused platelets ( $\geq 5 \times 10^{11}$  single-donor apheresis platelets) in adult dengue have also shown a lack of efficacy of prophylactic platelet transfusion with a significantly higher incidence of adverse events.

Lye and colleagues' study has generated robust data that strengthen the findings of previous studies showing a lack of efficacy of prophylactic platelet transfusion in dengue-related thrombocytopenia even when platelet count is lower than 5000 per  $\mu\text{L}$ . Prophylactic platelet transfusion in this setting not only increases financial costs with no added benefit to the patient, but also exposes patients to adverse events. Taken together, the evidence calls for prophylactic platelet transfusion to be stopped in the management of adult dengue.

Being the most rapidly spreading vector-borne viral disease, dengue is estimated to have affected nearly 390 million people in 2010, with 96 million having clinically apparent infection.<sup>11</sup> A strategy to withhold prophylactic platelet transfusion in adult dengue is expected to substantially reduce health-care costs, ease

demand on blood banks, and reduce harms related to platelet transfusion.

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I presented my research work at the National University of Singapore (NUS) to David C Lye and his colleagues in 2012, with travel and accommodation sponsored by NUS.

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# Prophylactic platelet transfusion plus supportive care versus supportive care alone in adults with dengue and thrombocytopenia: a multicentre, open-label, randomised, superiority trial



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## Summary

**Background** Dengue is the commonest vector-borne infection worldwide. It is often associated with thrombocytopenia, and prophylactic platelet transfusion is widely used despite the dearth of robust evidence. We aimed to assess the efficacy and safety of prophylactic platelet transfusion in the prevention of bleeding in adults with dengue and thrombocytopenia.

**Methods** We did an open-label, randomised, superiority trial in five hospitals in Singapore and Malaysia. We recruited patients aged at least 21 years who had laboratory-confirmed dengue (confirmed or probable) and thrombocytopenia ( $\leq 20\,000$  platelets per  $\mu\text{L}$ ), without persistent mild bleeding or any severe bleeding. Patients were assigned (1:1), with randomly permuted block sizes of four or six and stratified by centre, to receive prophylactic platelet transfusion in addition to supportive care (transfusion group) or supportive care alone (control group). In the transfusion group, 4 units of pooled platelets were given each day when platelet count was 20 000 per  $\mu\text{L}$  or lower; supportive care consisted of bed rest, fluid therapy, and fever and pain medications. The primary endpoint was clinical bleeding (excluding petechiae) by study day 7 or hospital discharge (whichever was earlier), analysed by intention to treat. Safety outcomes were analysed according to the actual treatment received. This study was registered with ClinicalTrials.gov, number NCT01030211, and is completed.

**Findings** Between April 29, 2010, and Dec 9, 2014, we randomly assigned 372 patients to the transfusion group ( $n=188$ ) or the control group ( $n=184$ ). The intention-to-treat analysis included 187 patients in the transfusion group (one patient was withdrawn immediately) and 182 in the control group (one was withdrawn immediately and one did not have confirmed or probable dengue). Clinical bleeding by day 7 or hospital discharge occurred in 40 (21%) patients in the transfusion group and 48 (26%) patients in the control group (risk difference  $-4\cdot98\%$  [95% CI  $-15\cdot08$  to  $5\cdot34$ ]; relative risk 0·81 [95% CI 0·56 to 1·17];  $p=0\cdot16$ ). 13 adverse events occurred in the transfusion group and two occurred in the control group (5·81% [ $-4\cdot42$  to  $16\cdot01$ ]; 6·26 [1·43 to 27·34];  $p=0\cdot0064$ ). Adverse events that were possibly, probably, or definitely related to transfusion included three cases of urticaria, one maculopapular rash, one pruritus, and one chest pain, as well as one case each of anaphylaxis, transfusion-related acute lung injury, and fluid overload that resulted in serious adverse events. No death was reported.

**Interpretation** In adult patients with dengue and thrombocytopenia, prophylactic platelet transfusion was not superior to supportive care in preventing bleeding, and might be associated with adverse events.

**Funding** National Medical Research Council, Singapore.

## Introduction

Dengue is a vector-borne viral infection estimated to affect 390 million people worldwide, with 96 million clinically apparent infections in 2010.<sup>1</sup> The global spread of dengue can be attributed to international trade and travel, urban crowding, and failure in vector control.<sup>2</sup> Climate change might increase geographical areas that are favourable for dengue transmission.<sup>3</sup> Autochthonous dengue transmission has occurred in non-endemic regions, including southern USA, mainland China, and Taiwan from travel-related importation and local *Aedes albopictus*.<sup>4–8</sup> Dengue has been associated with substantial cost to the health-care sector and national economy in endemic countries.<sup>9</sup>

Thrombocytopenia is reported in 79–100% of inpatients with dengue.<sup>10–12</sup> Platelet transfusion was administered to 22–50% of adult patients in various settings,<sup>11,13</sup> and could be inappropriate in 22–23% of patients who received transfusion.<sup>10,14</sup> Results from a survey of 306 doctors in 20 countries showed that 116 (38%) would give prophylactic platelet transfusion with varying platelet count thresholds.<sup>15</sup>

In a retrospective study of 106 children in Malaysia,<sup>16</sup> prophylactic platelet transfusion was associated with fluid overload and prolonged hospital stay without reduced bleeding or improved platelet recovery. No effect on bleeding or platelet recovery was found in 256 adults with dengue and thrombocytopenia ( $<20\,000$  platelets per  $\mu\text{L}$ ) in another retrospective study in Singapore.<sup>17</sup> In a

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### Research in context

#### Evidence before this study

We searched PubMed using the search terms “platelet” and “dengue” for English language publications of clinical studies that compared patients with dengue who did or did not receive prophylactic platelet transfusion. We excluded studies that examined platelet count as a risk factor for bleeding without any intervention. In a retrospective study in children with dengue shock syndrome, prophylactic platelet transfusion showed no benefit in platelet count recovery or the risk of bleeding, and increased risk of fluid overload and prolonged hospital stay. Results from a retrospective study of adult patients with uncomplicated dengue showed no difference in the risk of bleeding and platelet count recovery between those who received prophylactic platelet transfusion and those who did not. Findings from a similar, larger study in the same institution showed no difference in bleeding risk but increased time to platelet count recovery time and prolonged hospital stay. A pilot randomised trial of platelet transfusion in adult patients showed improved platelet increment and no difference in severe bleeding compared with those who did not receive transfusion; three severe transfusion reactions and two deaths occurred in transfused patients.

#### Added value of this study

To the best of our knowledge, this is the first rigorous, large, randomised trial of prophylactic platelet transfusion in adult dengue. We showed that, in adult patients with dengue and low platelet count (<20 000 per  $\mu\text{L}$ ), prophylactic platelet transfusion did not lead to any difference in clinical bleeding compared with supportive care alone. This finding is clinically significant because prophylactic platelet transfusion is widely used in patients with dengue and thrombocytopenia. Significantly more adverse events occurred in transfused patients, although all were resolved.

#### Implications of all the available evidence

In view of the scarcity and potential safety concerns of blood products in resource-limited settings, prophylactic platelet transfusion for patients with uncomplicated dengue is not recommended, since no benefit in reduction of clinical or severe bleeding or improvement in platelet count recovery was shown. Future studies should focus on children with dengue and patients with severe dengue.

small randomised trial in Pakistan ( $n=87$ ),<sup>18</sup> prophylactic platelet transfusion led to severe transfusion reactions and deaths, but without any benefit in bleeding reduction. Results from a large retrospective study of 788 adult patients with dengue<sup>19</sup> confirmed that transfusion did not reduce bleeding but led to slower platelet recovery and longer hospital stay compared with those who did not receive transfusion. Little evidence exists to guide decision on prophylactic platelet transfusion. To address this evidence gap, we designed a study comparing prophylactic platelet transfusion and supportive care with supportive care alone to test the hypothesis that prophylactic platelet transfusion was efficacious and safe in the prevention of bleeding in adult inpatients with dengue and thrombocytopenia ( $\leq 20\,000$  platelets per  $\mu\text{L}$ ).

### Methods

#### Study design

This open-label, randomised, superiority trial was done at four hospitals in Singapore and one hospital in Malaysia. This study was approved by the National Healthcare Group Domain Specific Review Board (E/2009/00235) and SingHealth Centralised Institutional Review Board (F/2009/565) in Singapore and by the University of Malaya Medical Centre Medical Ethics Committee in Malaysia. The full trial protocol can be requested from the corresponding author.

#### Patients

All patients with confirmed and probable dengue who were admitted to the participating hospitals were screened

by a trained research coordinator. Medical assessments were done by a study team doctor to determine whether patients met all inclusion and exclusion criteria. Patients were eligible for inclusion if they were aged at least 21 years; had a platelet count of 20 000 per  $\mu\text{L}$  or below; and were positive for dengue PCR or non-structural protein 1 in blood (ie, confirmed dengue), or positive for acute dengue serology with probable dengue criteria defined in WHO 1997<sup>20</sup> or 2009<sup>21</sup> dengue guidelines. Patients were excluded if they had persistent or recurrent epistaxis, haematemesis, melaena, menorrhagia, intermenstrual bleeding, haematochezia, or rectal bleeding; if they were pregnant or breastfeeding; if they had a history of severe adverse reaction to blood product transfusion; if they were likely to die within 48 h; if they had active peptic ulcer disease within 3 months, anticoagulant use within 4 weeks, chronic liver or kidney diseases or haemodialysis, or active haematological or autoimmune disorders; or if they had platelet transfusion within the same illness episode. All participants provided written informed consent.

#### Randomisation and masking

Patients were randomly assigned (1:1) to receive prophylactic platelet transfusion in addition to supportive care (transfusion group) or supportive care alone (control group). Randomisation was stratified by centre, and randomly permuted block sizes of four and six were used between the two groups over time. The block size was established by the randomisation statistician (who was involved in only the first interim analysis but not in any

subsequent analyses) and masked from the study team. Web-based randomisation, with back-up envelopes during web downtime, was used. Patients and study investigators were not masked to treatment allocation.

### Procedures

Patients in the transfusion group were given 4 units of pooled platelets each day if their platelet count was 20 000 per  $\mu\text{L}$  or lower. Supportive care consisted of bed rest, fluid therapy, and fever and pain medications. On the day of randomisation (day 1), medical history, physical examination, vital signs, full blood count, liver and renal function tests, coagulation profile (prothrombin time and partial thromboplastin time), and erect and decubitus chest x-ray were performed. From day 2 until day 7 or hospital discharge (whichever was earlier), and on day 21 (follow-up), medical history, physical examination, vital signs, and full blood count were performed. Liver panel, prothrombin time, and partial thromboplastin time were repeated on day 7 or hospital discharge and on day 21. Full blood count was assessed at 1 h, 12 h, and 24 h after transfusion. Patients were discharged from hospital if they showed defervescence, stable vital signs, rising platelet trend, and good oral intake.

### Outcomes

The primary endpoint was clinical bleeding, excluding petechiae, up to hospital discharge or 7 days after randomisation. Secondary efficacy endpoints were rate of change of platelet count at 1 h, 12 h, and 24 h post-transfusion; median time to sustained (ie, >2 days) platelet count greater than 50 000 per  $\mu\text{L}$ ; and clinical bleeding excluding petechiae within 21 days of randomisation. Secondary safety endpoints were plasma leakage, defined as an at least 20% change in serum haematocrit, development of pleural effusion, or ascites; dengue haemorrhagic fever or dengue shock syndrome (as defined in WHO 1997 dengue guidelines<sup>20</sup>), admission to intensive care unit, death, or secondary bacterial infection; median length of hospital stay; adverse events from platelet transfusion; and severe bleeding, defined as gastrointestinal bleeding, any internal bleeding (eg, retroperitoneal or intracranial), menorrhagia, or inter-menstrual bleeding not controlled by progesterone, or any clinical bleeding requiring endoscopy or surgery.

Adverse events were graded according to the US National Cancer Institute Common Terminology Criteria for Adverse Events (version 4). Serious adverse events were defined as untoward medical occurrences that result in death, persistent or significant disability, or congenital anomaly; are life-threatening or medically important; or prolong hospital stay. We reported warning signs and severe dengue (namely, severe plasma leakage, severe bleeding, and severe organ impairment) defined in the WHO 2009 dengue guidelines,<sup>21</sup> which were published after the trial protocol was written.

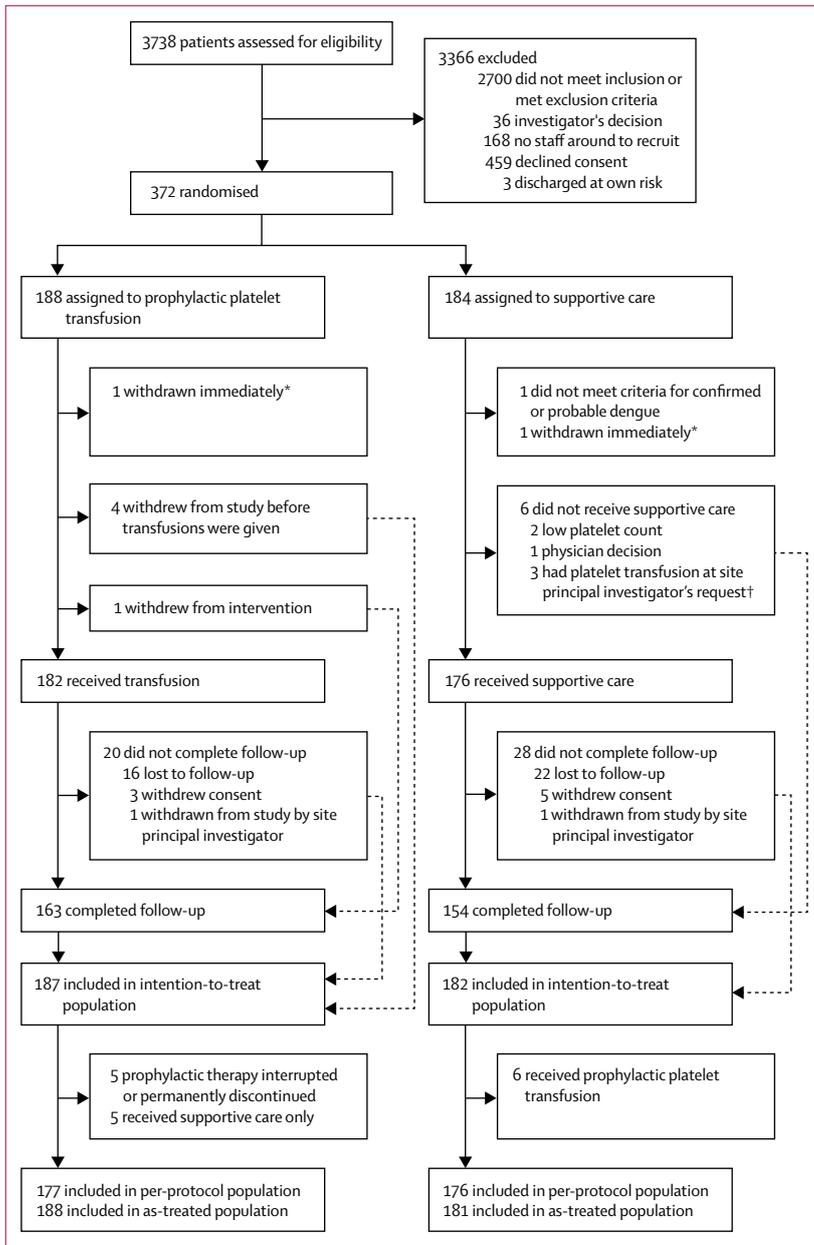
### Statistical analysis

We hypothesised that prophylactic platelet transfusion in patients with dengue and thrombocytopenia ( $\leq 20\,000$  per  $\mu\text{L}$ ) could reduce clinical bleeding by 50%. In a previous study<sup>17</sup> at Tan Tock Seng Hospital, Singapore, clinical bleeding occurred in 10% of patients with a platelet count of 20 000 per  $\mu\text{L}$  or below. With a power of 80% and one-sided alpha of 0.05, and assuming a 5% drop-out rate, 400 participants in each group were needed to reject the null hypothesis. Results from another study<sup>19</sup> at the same hospital in 2005–08 showed a higher clinical bleeding frequency of 20% in patients with a platelet count of 20 000 per  $\mu\text{L}$  or below. Using the same assumptions, the re-calculated sample size was 186 in each group.

An independent data safety monitoring board coordinated by the Singapore Clinical Research Institute reviewed primary endpoint and safety outcome data based on predefined stopping criteria after 50, 100, and 200 patients completed the study.

The primary and secondary efficacy endpoints were analysed by intention to treat. Analysis of platelet count at 1 h, 12 h, and 24 h after transfusion was done only in patients in the transfusion group. Sensitivity analyses of efficacy endpoints were done in the as-treated cohort (ie, according to actual treatment received) and per-protocol cohort (ie, all randomised patients whose actual treatment was the same as that allocated at randomisation without substantial protocol violations). Safety analyses were done in the as-treated cohort. Missing values were not imputed.

For the primary endpoint, we used Fisher's exact test and reported one-sided *p* values, risk difference (RD), relative risk (RR), and 95% CIs. For secondary endpoints, we reported mean and SD for platelet count at 1 h, 12 h, and 24 h post-transfusion. We reported difference in means, 95% CIs, and two-sided *p* values from two-sample *t* tests for daily platelet count until day 7 or discharge. Non-parametric Mann-Whitney *U* test was done if daily platelet count was not normally distributed. We reported RD, RR, 95% CIs, and two-sided *p* values from Fisher's exact tests for incidence of plasma leakage, dengue haemorrhagic fever or shock syndrome, admission to intensive care unit, death, secondary bacterial infection, severe bleeding, and clinical bleeding excluding petechiae by day 21. We produced Kaplan-Meier plots and reported *p* values from log-rank tests, and hazard ratio (HR) and 95% CI from Cox proportional hazards model for time to platelet count greater than 50 000 per  $\mu\text{L}$  and length of hospital stay. Proportional hazards assumption was verified. Patients who were withdrawn from the study before hospital discharge were censored. We did post-hoc analyses for warning signs, severe dengue, clinical bleeding excluding petechiae for baseline platelet count lower than 5000 per  $\mu\text{L}$  and lower than 10 000 per  $\mu\text{L}$  by study day 7 or discharge; RDs, RRs, 95% CIs, and two-sided *p* values from Fisher's exact tests



**Figure 1: Trial profile**

\*These patients did not receive any treatment and did not have any available baseline or outcome data, and were therefore excluded from the intention-to-treat analysis. †Because of epistaxis (n=2) and haematoma (n=1).

were reported in post-hoc analyses. Two-sided p values were reported and values less than 0.05 were considered significant, except for the primary endpoint, for which a one-sided p value was reported and a value less than 0.05 was considered significant. All statistical analyses were done with SAS version 9.3 (SAS Institute, Cary, NC, USA).

This study was registered with ClinicalTrials.gov, number NCT01030211.

### Role of the funding source

The funder of the study had no role in study design, data collection, data analysis, data interpretation, or writing of the report. The corresponding author had full access to all data in the study and had final responsibility for the decision to submit for publication.

### Results

From April 29, 2010, to Dec 9, 2014, we assessed 3738 patients for eligibility, of whom 372 patients across the five sequentially initiated study sites were randomly assigned to receive prophylactic platelet transfusion plus supportive care (n=188) or supportive care alone (n=184; figure 1). 187 patients in the transfusion group (one patient was withdrawn immediately) and 182 in the control group (one was withdrawn immediately and one did not have confirmed or probable dengue) were included in the intention-to-treat analysis. Five patients in the transfusion group received only supportive care and six patients in the control group received prophylactic platelet transfusion; therefore, the as-treated population comprised 188 patients in the transfusion group and 181 patients in the control group.

At study enrolment, age, sex, ethnic origin, comorbidities, method of dengue diagnosis, warning signs, dengue severity, and symptoms and signs were similar between the two groups (table 1). At baseline, history of mild clinical bleeding (mainly transient gum or non-persistent or recurrent nose bleed) was present in 25 (13%) patients in the transfusion group and 35 (19%) in the control group. All patients in the transfusion group received 4 units of pooled platelet transfusion, except for eight patients who received 1 unit of single donor-derived platelet transfusion because of a shortage of platelet transfusion at the time of randomisation. Of 182 patients receiving prophylactic platelet transfusion on day 1, 46 (25%) required a second platelet transfusion to maintain a platelet count above 20 000 per  $\mu\text{L}$ . The mean number of units of platelets given in the transfusion group was 4.71 (SD 2.17).

Clinical bleeding by day 7 or hospital discharge occurred in 40 (21%) patients in the transfusion group and 48 (26%) in the control group (RD -4.98% [95% CI -15.08 to 5.34]; RR 0.81 [95% CI 0.56 to 1.17]; p=0.16; table 2). Sensitivity analyses showed similar findings in the as-treated cohort (0.18% [-10.08 to 10.40]; 1.01 [0.70 to 1.45]; p=0.56) and in the per-protocol cohort (-1.83% [-12.08 to 8.71]; 0.93 [0.63 to 1.35]; p=0.39). In both groups, the commonest type of bleeding was gingival bleeding, followed by epistaxis (table 2). By day 21, clinically important bleeding was reported in 42 (22%) patients in the transfusion group and 49 (27%) patients in the control group (-4.46% [-14.59 to 5.86]; 0.83 [0.58 to 1.19]; p=0.34).

By day 21, severe bleeding occurred in three (2%) patients in the transfusion group and seven (4%) patients in the control group (RD -2.24%

	Transfusion group (n=187)	Control group (n=182)
Age (years)	44.3 (14.1)	45.2 (12.4)
Men	139 (74%)	140 (77%)
Ethnic origin		
Chinese	104 (56%)	116 (64%)
Malay	39 (21%)	25 (14%)
Indian	25 (13%)	26 (14%)
Others	19 (10%)	15 (8%)
Laboratory diagnosis*		
Dengue PCR positive	32 (17%)	28 (15%)
Dengue non-structural protein 1 positive	129 (69%)	142 (78%)
Acute serology with probable dengue criteria	107 (57%)	101 (55%)
Past history of dengue	22 (12%)	13 (7%)
Presence of seven warning signs	177 (95%)	177 (97%)
Severe dengue	15 (8%)	6 (3%)
Dengue haemorrhagic fever or shock syndrome	20 (11%)	27 (15%)
Comorbidities		
Hypertension	34 (18%)	35 (19%)
Hyperlipidaemia	19 (10%)	27 (15%)
Diabetes	18 (10%)	20 (11%)
Others	8 (4%)	10 (5%)
Symptoms		
Fever	187 (100%)	181 (99%)
Lethargy	157 (84%)	152 (84%)
Headache	122 (65%)	119 (65%)
Back pain	76 (41%)	77 (42%)
Eye pain	29 (16%)	16 (9%)
Myalgia	114 (61%)	108 (59%)
Arthralgia	73 (39%)	66 (36%)
Rash	82 (44%)	73 (40%)
Any bleeding	25 (13%)	35 (19%)
Gingival	22 (12%)	27 (15%)
Non-persistent or recurrent epistaxis	2 (1%)	7 (4%)
Others	3 (2%)	4 (2%)
Anorexia	138 (74%)	130 (71%)
Nausea	116 (62%)	105 (58%)
Vomiting	81 (43%)	71 (39%)
Diarrhoea	65 (35%)	58 (32%)
Abdominal pain	45 (24%)	43 (24%)

(Table 1 continues in next column)

[95% CI -12.4 to 8.06]; RR 0.42 [95% CI 0.11 to 1.59]; p=0.21; appendix). At the time of severe bleeding, one patient's platelet count was above 20 000 per  $\mu\text{L}$ , seven patients' platelet counts were between 20 000 and 10 000 per  $\mu\text{L}$ , and two patients' platelet counts were below 10 000 per  $\mu\text{L}$ .

	Transfusion group (n=187)	Control group (n=182)
(Continued from previous column)		
Laboratory test		
White cell count ( $\times 10^3$ per $\mu\text{L}$ )	4.2 (2.2)	4.3 (2.3)
Haematocrit (%)	43.7 (4.0)	44.4 (4.9)
Platelet count ( $\times 10^3$ per $\mu\text{L}$ )	14.1 (5.8)	13.1 (4.6)
Creatinine ( $\mu\text{mol/L}$ )	73.0 (22.3)	72.3 (17.3)
Alanine aminotransferase (IU)	153.2 (177.9)	128.1 (102.3)
Aspartate aminotransferase (IU)	243.0 (311.0)	209.2 (195.1)
Protein (g/L)	60.1 (6.0)	59.9 (6.0)
Prothrombin time (s)	12.1 (2.1)	12.0 (1.1)
Activated partial thromboplastin time (s)	41.8 (7.6)	43.0 (9.2)
Days of illness onset at enrolment	5.2 (1.3)	5.3 (1.1)

Data are mean (SD) or n (%). IU=international units. \*Some patients had more than one positive laboratory test.

**Table 1: Characteristics of patients at study enrolment**

In patients with platelet count below 10 000 per  $\mu\text{L}$  at baseline, clinical bleeding by day 7 occurred in ten (24%) of 42 patients in the transfusion group and ten (29%) of 35 patients in the control group (post-hoc RD -4.76% [95% CI -26.92 to 17.73]; RR 0.83 [95% CI 0.39 to 1.77]; p=0.41). In those with platelet counts below 5000 per  $\mu\text{L}$  at baseline, clinical bleeding by day 7 occurred in four (57%) of seven patients in the transfusion group and seven (50%) of 14 patients in the control group (post-hoc 7.14% [-39.97 to 52.25]; 1.14 [0.50 to 2.62]; p=0.78).

Plasma leakage occurred in ten (11%) patients in the transfusion group and eight (10%) patients in the control group (RD 1.11% [95% CI -13.97 to 16.17]; RR 1.11 [95% CI 0.46 to 2.67]; p=1.00), and dengue haemorrhagic fever or shock syndrome was reported in two (1%) patients in the transfusion group and two (1%) patients in the control group (-0.08% [-11.03 to 10.86]; 0.94 [0.13 to 6.59]; p=1.00). One (1%) patient in the transfusion group and one (1%) in the control group were admitted to intensive care unit (-0.02% [-10.25 to 10.22]; 0.96 [0.06 to 15.28]; p=1.00); both patients had no comorbidity and were discharged from intensive care after 3 days without intubation or inotropic support. The patient in the transfusion group was a 67-year-old man who developed fluid overload requiring non-invasive mechanical ventilation and diuresis. The patient in the control group was a 42-year-old man who had drowsiness, hypothermia, and hypotension requiring close monitoring. Only one patient in the control group developed secondary bacterial infection, and no death occurred. No patients required packed red blood cell or whole blood transfusion. The median length of hospital stay was 4 days (IQR 4-5) in the transfusion group and 5 days (4-6) in the control group (p=0.47).

See Online for appendix

	By day 7 or hospital discharge		By day 21	
	Transfusion group (n=187)	Control group (n=182)	Transfusion group (n=187)	Control group (n=182)
Total	40 (21%)	48 (26%)	42 (22%)	49 (27%)
Gingival	21 (11%)	32 (17%)	21 (11%)	33 (18%)
Epistaxis	8 (4%)	9 (5%)	8 (4%)	9 (5%)
Haemoptysis	3 (2%)	1 (1%)	3 (2%)	1 (1%)
Haematuria	0	1 (1%)	0	1 (1%)
Haematemesis	0	0	0	0
Melaena	0	4 (2%)	0	4 (2%)
Haematemesis or melaena not controlled by endoscopic procedure	0	0	0	0
Menorrhagia	0	2 (1%)	0	2 (1%)
Inter-menstrual bleed	0	1 (1%)	0	1 (1%)
Menorrhagia or inter-menstrual bleed not controlled by progesterone	0	0	0	1 (1%)
Haematoma	7 (4%)	8 (4%)	7 (4%)	8 (4%)
Usual menses	4 (2%)	4 (2%)	6 (3%)	4 (2%)
Others	4 (2%)	2 (1%)	4 (2%)	2 (1%)

Table 2: Clinical bleeding by day 7 or hospital discharge and by day 21

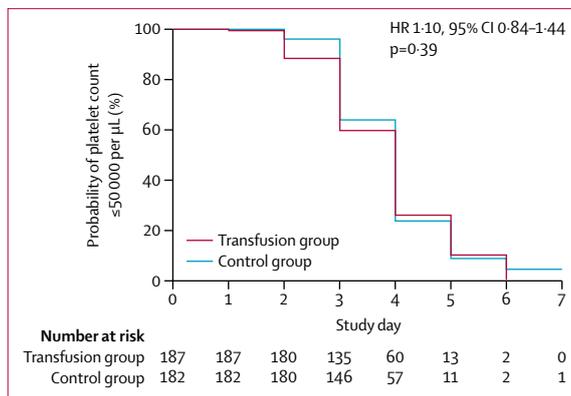


Figure 2: Kaplan-Meier estimates of probability of platelet count of 50 000 per µL or below by day 7  
HR=hazard ratio.

A post-hoc intention-to-treat analysis showed similar occurrence of warning signs by day 7 (178 [95%] in the transfusion group vs 179 [98%] in the control group; RD -3.16% [-13.42 to 7.03]; RR 0.97 [0.93 to 1.00]; p=0.14) and severe dengue (nine [5%] vs seven [4%]; 1.32% [-9.22 to 11.93]; 1.33 [0.51 to 3.49]; p=0.62).

The mean increase from baseline platelet count in transfused patients was 17.56 × 10<sup>3</sup> per µL (SD 13.76 × 10<sup>3</sup>) at 1 h, 16.68 × 10<sup>3</sup> per µL (17.37 × 10<sup>3</sup>) at 12 h, and 29.23 × 10<sup>3</sup> per µL (23.59 × 10<sup>3</sup>) at 24 h. The median time to sustained platelet count above 50 000 per µL was similar between the two groups (4 days [IQR 3–5] in the transfusion group vs 4 days [3–4] in the control group; figure 2). In a post-hoc analysis, no difference in mean daily platelet count was seen

between the two groups from day 1 to day 21, except on day 2 (30.49 × 10<sup>3</sup> per µL [SD 18.78 × 10<sup>3</sup>] vs 20.89 × 10<sup>3</sup> per µL [13.18 × 10<sup>3</sup>]; both parametric and non-parametric tests p<0.0001; figure 3).

13 adverse events occurred in the transfusion group and two occurred in the control group (RD 5.81% [95% CI -4.42 to 16.01]; RR 6.26 [95% CI 1.43 to 27.34]; p=0.0064; table 3); two serious adverse events occurred in the transfusion group and one occurred in the control group (0.51% [-9.72 to 10.76]; 1.93 [0.18 to 21.05]; p=1.00). Mild to moderate adverse events occurred in 12 patients in the transfusion group and one patient in the control group; four such events resulted in temporary or permanent interruption of transfusion, and nine were possibly, probably, or definitely related to transfusion. These nine adverse events included three urticaria, one maculopapular rash, one pruritus, and one chest pain; the remaining three resulted in serious adverse events: one each of anaphylaxis, transfusion-related acute lung injury, and fluid overload (appendix). Of the three serious adverse events, one resulted in permanent interruption of transfusion. All patients who had adverse events and serious adverse events fully recovered. No death was reported.

## Discussion

Results from our study show that prophylactic platelet transfusion was not superior to supportive care in the prevention of bleeding in adult patients with dengue. The rates of bleeding in our study were similar to those in the study of paediatric dengue shock syndrome (n=106, of whom 60 received platelet transfusion)<sup>16</sup> and in previous studies of uncomplicated adult dengue.<sup>17–19</sup> Compared with these studies, prophylactic platelet transfusion was associated with more adverse events in our study, although most were non-severe and all fully resolved. In the randomised trial in Pakistan,<sup>18</sup> three of 43 patients who received transfusion developed severe transfusion reactions and two died, highlighting potential hazards of transfusion; notably, single-donor filtered apheresis platelets with platelet dose fixed at 5 × 10<sup>11</sup> per µL or more was transfused. In the paediatric study,<sup>16</sup> prophylactic platelet transfusion resulted in fluid overload and increased length of hospital stay. In the large retrospective study,<sup>19</sup> patients who received prophylactic platelet transfusion stayed 1 day longer in hospital than did those who did not (p<0.0001).<sup>19</sup> In our study, no increased risk of severe dengue, dengue haemorrhagic fever or shock syndrome, and admission to intensive care unit was observed in non-transfused patients compared with transfused patients.

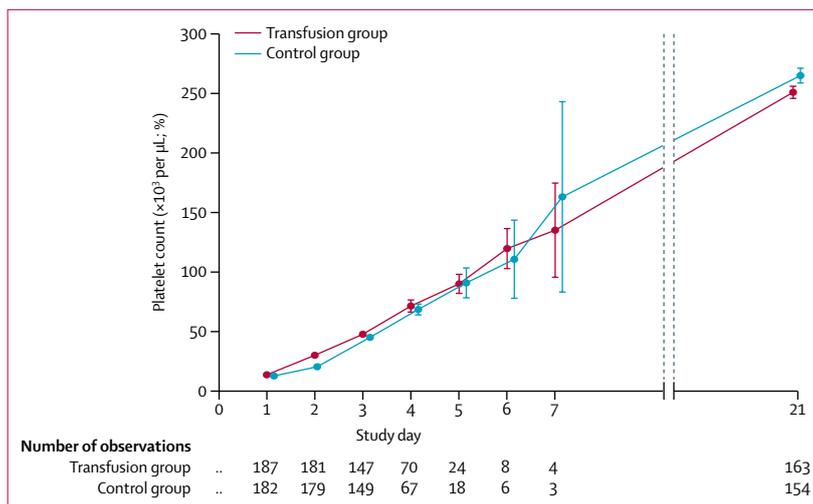
We found that the effect of prophylactic platelet transfusion on platelet recovery was transient, with similar platelet counts beyond day 2 post-transfusion. In our study, the duration of thrombocytopenia and time to platelet recovery were similar between the two treatment groups—a finding that is consistent with results from

the paediatric study of dengue shock syndrome<sup>16</sup> and the two studies of uncomplicated adult dengue.<sup>17,18</sup> With similar bleeding risk and platelet recovery, as well as potential harm, **prophylactic platelet transfusion should not be recommended**; such a policy will also ease demand on blood products in dengue-endemic and resource-limited countries. In patients with dengue and thrombocytopenia ( $\leq 20\,000$  platelets per  $\mu\text{L}$ ), severe bleeding was uncommon irrespective of the use of prophylactic platelet transfusion, confirming that careful observation is the preferred approach for these patients since most clinical bleeding was mild and self-resolving.

Thrombocytopenia in dengue might result from **reduced bone marrow function and decreased platelet production**; increased platelet destruction, evidenced by increased **megakaryocytes** and shortened platelet survival time; and increased platelet **consumption**, suggested by **immune complexes containing dengue antigen on platelets** and platelet adherence and lysis associated with **dengue-infected endothelium**.<sup>22</sup> In a paediatric study of dengue shock syndrome in Malaysia ( $n=114$ ),<sup>23</sup> shock and low haematocrit, but not platelet count, were identified as independent predictors of severe haemorrhage. Results from two retrospective studies in adults<sup>24,25</sup> showed that female sex, elevated lymphocyte count ( $>500$  per  $\mu\text{L}$ ), thrombocytopenia, use of antiplatelet drugs, activated partial thromboplastin time of more than 60 s, and international normalised ratio greater than 2 were independent predictors of bleeding risk. Hence, **bleeding risk in dengue—which can be due to a range of other factors—might not be improved by platelet transfusion alone**. Additionally, the effect of **platelet transfusion on platelet count lasted for less than 5 h** in the paediatric study.<sup>16</sup>

In a large retrospective study of adult patients with uncomplicated dengue who were given prophylactic platelet transfusion,<sup>19</sup> median time to platelet count above 50 000 per  $\mu\text{L}$  was significantly longer by 1 day. **Platelet transfusion might potentially depress serum thrombopoietin**, which is required for platelet production. In a study of 28 patients with dengue fever and seven patients with dengue haemorrhagic fever,<sup>26</sup> serum thrombopoietin was persistently elevated during febrile and critical phases and rapidly decreased on platelet recovery. However, in our randomised clinical trial, we did not observe slower platelet recovery in transfused patients than in non-transfused patients.

We primarily recruited adults in Singapore and Malaysia, and our findings might not be generalisable to children and patients in other geographical regions. Women were under-represented in our study (90 [24%] of 369 patients analysed), although the female sex is a risk factor for bleeding in dengue. The bleeding risk in our study was roughly 20%, and severe dengue occurred in 21 (6%) patients and dengue haemorrhagic fever and shock syndrome in 47 (13%) patients; these proportions might differ in different settings and limit comparison with our



**Figure 3: Mean daily platelet count**

Error bars represent SE. SEs for days 1–3 are too small to be shown.

	Transfusion group (n=188)	Control group (n=181)
<b>Severity</b>		
Mild	7 (4%)	1 (1%)
Moderate	5 (3%)	0
Severe	1 (1%)	1 (1%)
<b>Effect on study intervention</b>		
None	9 (5%)	2 (1%)
Temporarily interrupted	1 (1%)	0
Permanently discontinued	3 (2%)	0
<b>Relation to study intervention</b>		
Unrelated	4 (2%)	2 (1%)
Possibly related	3 (2%)	0
Probably related	4 (2%)	0
Definitely related	2 (1%)	0
Recovered	13 (7%)	2 (1%)

**Table 3: Adverse events**

findings. Because **severe bleeding** was a secondary outcome and **uncommon**, we could not exclude potential benefit of prophylactic platelet transfusion in the prevention of severe bleeding at lower platelet count thresholds; importantly, severe bleeding was an uncommon occurrence in our study. Although this study was open-label and not placebo controlled, we used clinical bleeding—an objective measurement—as the primary endpoint. Future clinical trials are needed in children with dengue and in settings where severe bleeding is more common to examine the effect of prophylactic platelet transfusion with severe bleeding as a primary outcome. The 4 units of pooled platelet transfusion in our trial were equivalent to medium-dose platelet transfusion ( $2 \cdot 2 \times 10^{11}$  platelets per  $\text{m}^2$  of body surface area) in a randomised clinical trial<sup>27</sup> examining

low-dose, medium-dose, and high-dose prophylactic platelet transfusion in patients with hypoproliferative thrombocytopenia, which found similar rates of bleeding across different doses of platelet transfusion. Hence, we cannot exclude potential benefit of higher-dose prophylactic platelet transfusion in our study population. Increment in platelet counts depends on the dose of platelet transfused and recipient's body surface area, but we did not have data on body surface area and were unable to report corrected count increment in this study.<sup>28</sup>

Results from our randomised trial provide evidence to support present WHO guidelines on dengue<sup>21</sup> in recommending against prophylactic platelet transfusion in patients with low platelet count. This conclusion is especially important in large dengue outbreaks where demand for platelet transfusion might increase<sup>29</sup> and where limited platelet products should be reserved for therapeutic use in severe bleeding.<sup>30</sup>

In conclusion, in adult patients with dengue and thrombocytopenia ( $\leq 20\,000$  platelets per  $\mu\text{L}$ ), prophylactic platelet transfusion was not superior to supportive care in the prevention of bleeding or improvement in platelet recovery, and might be associated with adverse events.

#### Contributors

DCL, JGL, EE-O, PAT, and Y-SL conceptualised and designed the study. DCL, SA, SFS-O, JGL, HMO, SSLP, LW, PAT, and Y-SL collected the data. YW analysed the data with inputs from DCL and Y-SL. DCL, SA, YW, DF, LKL, AK, LCL, PAT, and Y-SL interpreted the data. DCL drafted the manuscript, and all other authors revised the manuscript. All authors approved the final submitted version and agreed to be accountable for the manuscript.

#### Declaration of interests

We declare no competing interests.

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