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SARS-CoV-2 spike protein potentiates platelet aggregation via upregulating integrin αIIbβ3 outside-in signaling pathway

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Abstract

Platelet hyperreactivity is one of the crucial causes of coagulative disorders in patients with COVID-19. Few studies have indicated that integrin αIIbβ3 may be a potential target for spike protein binding to platelets. This study aims to investigate whether spike protein interacts with platelet integrin αIIbβ3 and upregulates outside-in signaling to potentiate platelet aggregation. In this study, we found that spike protein significantly potentiated platelet aggregation induced by different agonists and platelet spreading *in vitro*. Mechanism studies revealed that spike protein upregulated the outside-in signaling, such as increased thrombin-induced phosphorylation of β3, c-Src. Moreover, using tirofiban to inhibit spike protein binding to αIIbβ3 or using PP2 to block outside-in signaling, we found that the potentiating effect of spike protein on platelet aggregation was abolished. These results demonstrate that SARS-CoV-2 spike protein directly enhances platelet aggregation via integrin αIIbβ3 outside-in signaling, and suggest a potential target for platelet hyperreactivity in patients with COVID-19.

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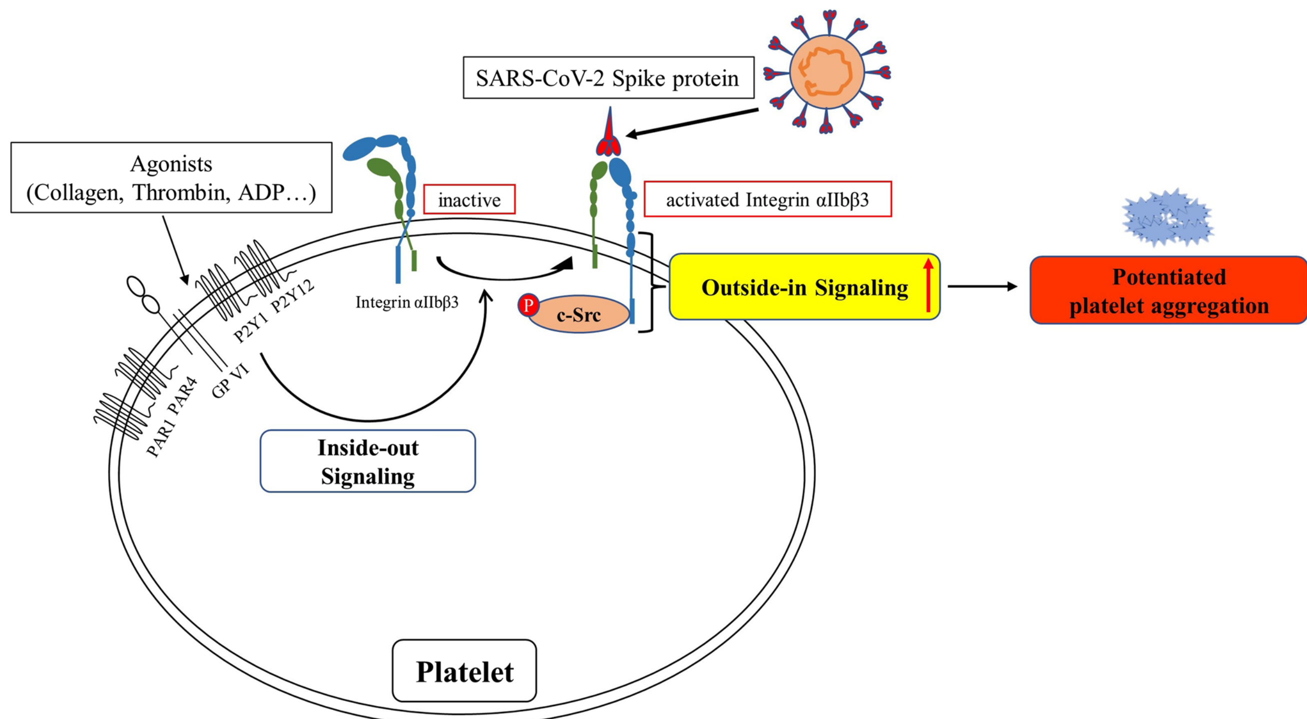
Graphical abstract



SARS-CoV-2 spike protein interacts with platelet integrin αIIbβ3, upregulating the outside-in signaling pathway and subsequently potentiating platelet aggregation.

Highlights

- Spike protein potentiates platelet aggregation and upregulates αIIbβ3 outside-in signaling.
- Spike protein interacts with integrin αIIbβ3 to potentiate platelet aggregation.
- Blocking outside-in signaling abolishes the effect of spike protein on platelets.

Keywords SARS-CoV-2 · spike protein · platelet · integrin αIIbβ3 · outside-in signaling

Introduction

The coronavirus disease 2019 (COVID-19), caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), results in significant morbidity and mortality [1, 2]. Platelet hyperreactivity is a hallmark of COVID-19 and contributes to an increased risk of thrombotic disorders observed in many COVID-19 patients [3, 4]. Platelets are small and anucleate blood cells that play crucial roles in hemostasis, thrombosis, and immune response [5]. Excessive platelet aggregation and activation contribute to occlusive thrombus formation and the development and progression of atherothrombosis [6]. As mounting evidence supports a link between COVID-19 and platelet hyperreactivity, the underlying mechanism for this association has multiple theories.

SARS-CoV-2 infects host cells via binding of spike protein to angiotensin-converting enzyme-2 (ACE2). However, it remains controversial whether platelets express ACE2 and if SARS-CoV-2 interacts with platelets through an ACE2-mediated mechanism. It is demonstrated that the SARS-CoV-2 spike protein has an RGD (arginine-glycine-aspartate) sequence in the receptor-binding domain, a motif that could bind to cell-surface integrins [7]. Integrin αIIbβ3, a highly abundant heterodimeric platelet receptor, plays a central role in platelet functions, hemostasis, and arterial thrombosis. Integrin αIIbβ3 can bind to several RGD-containing ligands, including fibrinogen and von Willebrand factor (vWF), which triggers various outside-in signaling events that further facilitate platelet spreading, stabilizes platelet aggregation, and so on [8]. Since Xiaoying Ma and their colleagues have found that SARS-CoV-2 spike protein RBD binds platelet β3 integrin [9], whether spike protein upregulates outside-in signaling to potentiate platelet function

is still unknown. In this study, we demonstrated that SARS-CoV-2 spike protein directly potentiated platelet aggregation and spreading by interacting with platelet integrin αIIbβ3 and upregulating the outside-in signaling pathway, suggesting a potential therapeutic target for the prevention and treatment of platelet hyperreactivity in subjects with COVID-19.

Materials and methods

Chemicals

Detailed descriptions of the reagents and antibodies are provided in Supplement material: Expanded Materials and Methods.

Platelet preparation, platelet functional assays and Western Blotting

This study was conducted in accordance with the Declaration of Helsinki and approved by the Ethics Committee of the School of public health (Shenzhen), Sun Yat-Sen University [[2020] No.013]. Healthy subjects aged between 18 to 30, recruited from Sun Yat-Sen University, participated in the study. Subjects who had self-reported cardiovascular disease or taken any antiplatelet drugs or nutrients within the previous 2 weeks were excluded. Whole human blood was drawn and gel-filtered platelets were prepared according to our previously described methods [10].

Detailed descriptions of the methods of platelet aggregation, activation, spearing on immobilized spike protein and western blotting are descripted in Supplement material: Expanded Materials and Methods.

Statistical analysis

Data were expressed as mean ± standard deviation of the means (SD) of at least three independent experiments. One-way analysis of variance followed by Tucky post hoc analysis was used for multiple comparisons and paired Student's t test was used for two paired sample comparisons. Statistical analysis was performed using SPSS 25.0. $P<0.05$ was considered statistically significant.

Results

Spike protein potentiates platelet aggregation and upregulates platelet integrin αIIbβ3 outside-in signaling *in vitro*

According to a previous study that has demonstrated that spike protein (0.001-1 μg/ml) induced platelet aggregation in a dose-dependent manner [11], we selected 1 μg/ml spike protein as the standard concentration for subsequent experiments and observed that 1 μg/ml spike protein significantly promoted platelets aggregation in response to thrombin, collagen, and ADP (Fig. 1A). However, we found no significant effects of spike protein on platelet granule secretion, such as the surface expression of P-selectin (Fig. 1B). The inside-out signaling to integrin αIIbβ3 leads to its activation and ligand binding [12]. Using PAC-1 binding assay, we observed that spike protein did not affect integrin αIIbβ3 inside-out signaling (Fig. 1C).

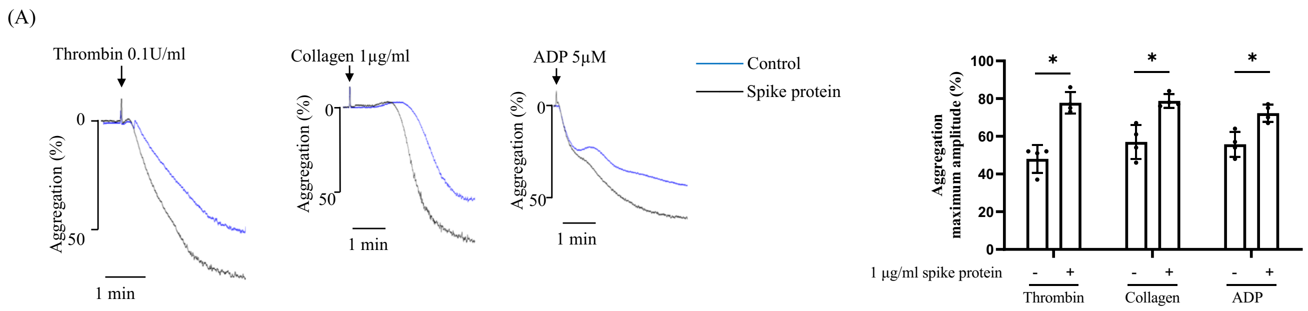
Platelet spreading is primarily regulated by integrin αIIbβ3 outside-in signaling [8]. Spike protein has an RGD-containing motif that enables it to bind with platelet integrin β3 [9], indicating platelets could adhere to immobilized spike protein similarly to immobilized fibrinogen and further inducing platelet spreading [13]. We found that platelets spreading on immobilized BSA (the control group) remained discoid, while the surface area of platelets spreading on immobilized spike protein was significantly greater (Fig. 1D). We further analyzed whether spike protein can directly affect platelet integrin αIIbβ3 outside-in signaling by measuring phosphorylation of β-integrin tail and c-Src in platelets. We further found that only spike protein could potentiate the phosphorylation of β-integrin tail and c-Src in resting platelets (Supplemental Figure 1), although spike protein cannot induce platelet activation without agonists [14]. Moreover, spike protein significantly potentiated the phosphorylation of β-integrin tail and c-Src (Fig. 1E) in thrombin-activated human platelets. These findings suggest that spike protein directly upregulates integrin αIIbβ3 outside-in signaling.

Spike protein potentiates platelet aggregation via interacting with platelet integrin αIIbβ3

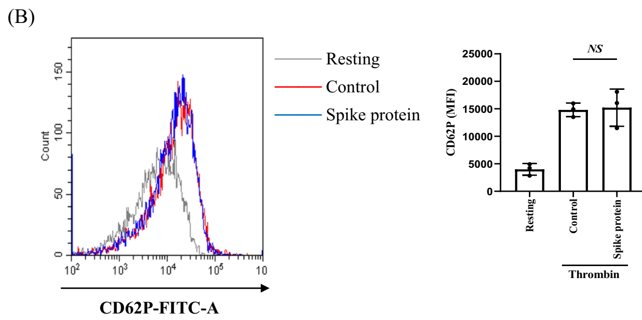
Firstly, whether ACE2 participants in the process of interaction between SARS-CoV-2 and platelet exits contradiction. Western blot assay showed that human platelets exhibited trace expression of ACE2 and TMPRSS2. These levels were far below those in PBMC (Supplemental Figure 2), indicating spike protein may use other receptors instead of ACE2 to interact with platelets.

As spike protein contains RGD sequence to bind cell-surface integrins and could bind to platelet β3 integrin [9], we further explore whether spike protein potentiates platelet aggregation via interacting with integrin αIIbβ3. To test whether spike protein blocks fibrinogen binding integrin αIIbβ3, we performed the fibrinogen-binding assay, in which platelets were incubated with labeled fibrinogen in the presence or absence of spike protein, to examine the formation of the platelet-fibrinogen complex. As shown in Fig. 2A, we found that 10 μg/ml spike protein significantly

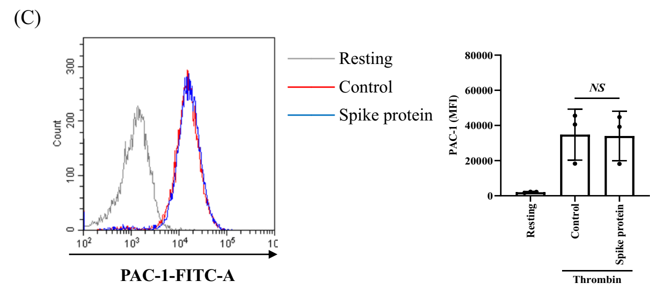
(A)



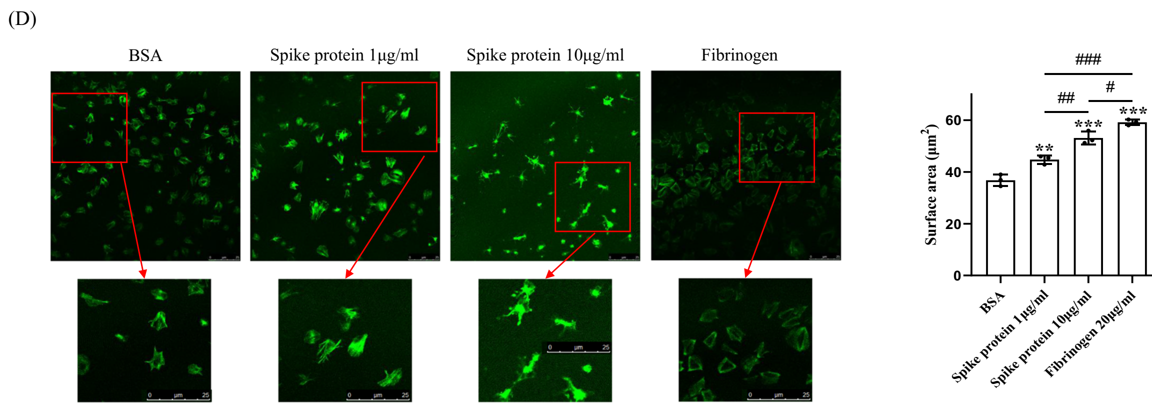
(B)



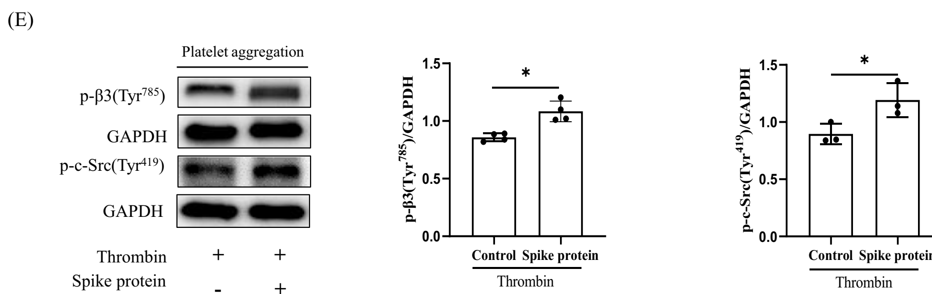
(C)



(D)



(E)



◂**Fig. 1** Spike protein potentiates platelet aggregation and upregulates platelet integrin αIIbβ3 outside-in signaling *in vitro*. (**A**) Platelets were preincubated with spike protein at 1 μg/mL for 10 min, and then gel-filtered platelets were stimulated with 0.1 U/mL thrombin or 1 μg/mL collagen, and PRP stimulated with 5 μM ADP. Aggregation was assessed under stirring at 1200 rpm (n=4). (B-C) Gel-filtered platelets were incubated with 1 μg/mL spike protein for 10 min and then stimulated with 0.1 U/ml thrombin, and then the expression of P-selectin (**B**) (n=3) or PAC-1 binding (**C**) (n=3) was analyzed using a flow cytometer. (**D**) Gel-filtered platelet was allowed to spread on the spike protein (1, 10 μg/ml) or positive control (100 μg/mL fibrinogen) coated surfaces (blocked with 1% BSA) for 60 min at 37 °C, and the representative images and summary data of the mean platelet surface area were shown (n=3). (**E**) After aggregation in (**A**), the platelets were collected and then lysed with lysis buffer. The cell lysates were prepared and Western Blot was performed to assess the phosphorylation levels of β3 (n=4) and c-Src (n=3). * P<0.05, ** P<0.01, *** P<0.001, indicate significant difference compared with control group. # P<0.05, ## P<0.01, ### P<0.001, indicate significant difference between the two group. *NS*: no significant difference between the two groups. Statistical analyses were performed using paired two-tailed Student's t test to compare the difference between control and spike protein groups in (**A**-**C**) and (**E**). One-way ANOVA with Tukey's post hoc test was performed in (**D**)

inhibited platelet fibrinogen binding and had no significant difference to 100 μg/ml fibrinogen from human plasma which directly interacts with integrin αIIbβ3. We next use tirofiban, the integrin αIIbβ3 specific antagonist, to prevent spike protein binding to αIIbβ3. We found that spike protein did not further potentiate platelet aggregation (Fig. 2B) and the phosphorylation of β3 (Fig. 2C) in the presence of tirofiban. These results suggest that spike protein potentiates platelet aggregation via interacting with platelet integrin αIIbβ3.

Spike protein potentiates platelet aggregation via upregulating outside-in signaling

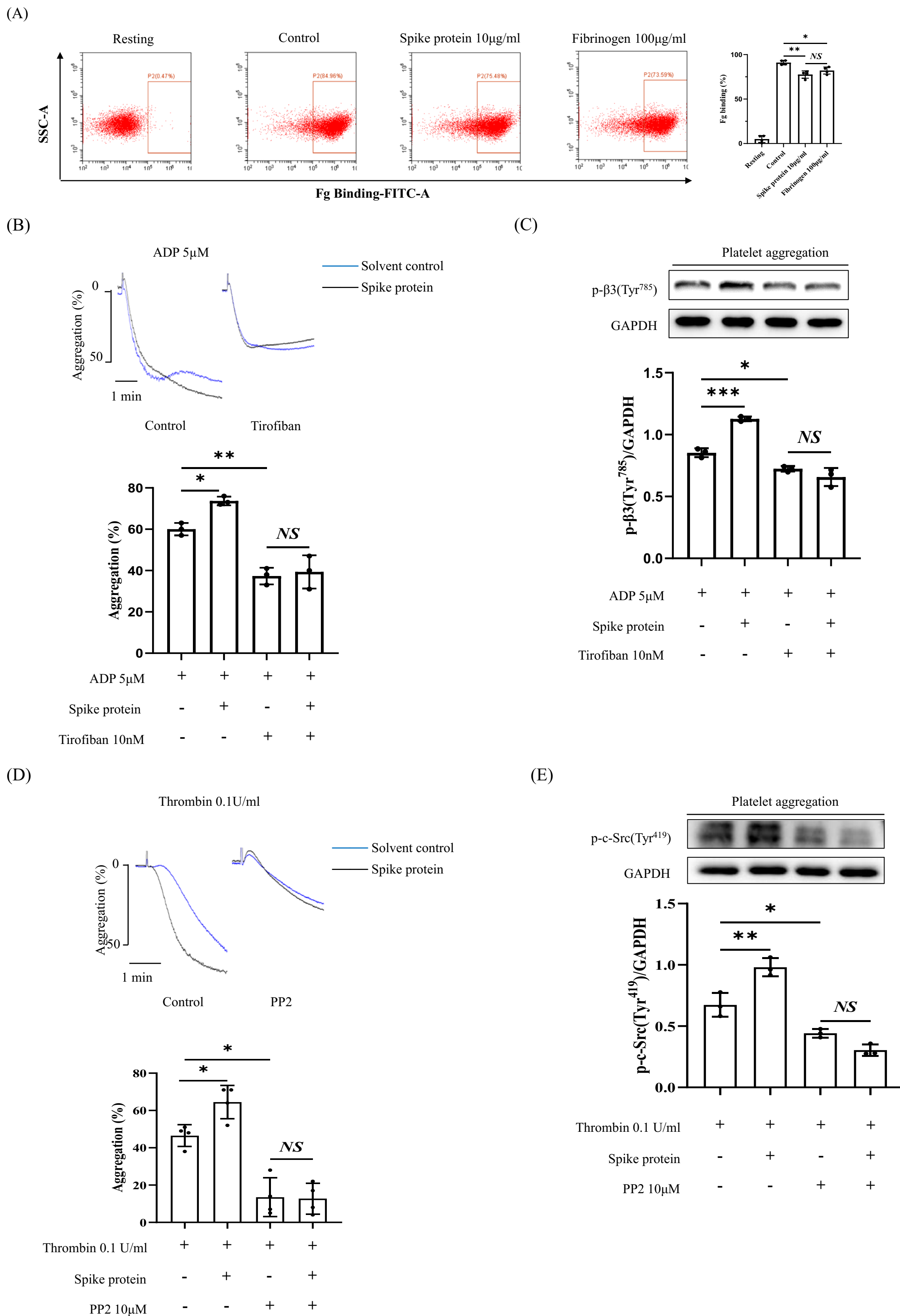
As the c-Src is proved to be the dominant effector of αIIbβ3 outside-in signaling [15, 16], we used 4-amino-5-(4-chlorophenyl)-7-(t-butyl)pyrazolo[3,4-d]pyrimidine (PP2), the Src inhibitor, to block c-Src activation and thus inhibiting outside-in signaling. We observed that spike protein did not further potentiate platelet aggregation (Fig. 2D) and the phosphorylation of c-Src (Fig. 2E) in the presence of PP2 (10 μM). These observations indicate that the potentiating effect of spike protein on platelet aggregation is predominantly mediated by upregulating integrin αIIbβ3 outside-in signaling.

Discussion

COVID-19 causes platelet hyperreactivity and severe thrombotic disorders, leading cause of mortality and morbidity worldwide. Multiple studies have found that platelets isolated from COVID-19 patients show increased platelet aggregation, integrin αIIbβ3 activation and P-selectin expression [4, 17]. Moreover, the pathogenicity of SARS-CoV-2 may be attributed to spike protein engaging with host cell receptors. In this study, we observed that spike protein potentiates platelet aggregation which plays a critical role in hemostatic plug and thrombus formation [18].

The integrins, a superfamily of cell adhesion receptors, play critical roles in cell adhesion and cell-extracellular matrix (ECM) interaction [19]. Integrin αIIbβ3 is the most abundant surface-expressed integrin in platelets and can bind to several RGD-containing ligands. Upon stimulation with agonists, the binding of activated αIIbβ3 and soluble ligands can be detected within seconds [20]. Since spike protein has an RGD sequence and could bind to platelet β3 integrin [9], we further perform Fg-binding assay to verify this viewpoint. While a low concentration of spike protein (2 μg/ml) did not affect platelet fibrinogen binding when stimulated with thrombin [14], we found that a high concentration of spike protein (10 μg/ml) attenuated platelet fibrinogen binding, similar to 100 μg/ml fibrinogen that directly binds to αIIbβ3. Additionally, we observed that the spike protein-potentiated platelet aggregation and the phosphorylation of β-integrin tail were abolished after being pre-incubated with tirofiban, a selective and reversible platelet integrin αIIbβ3 receptor antagonist. Consistent with this, another study also observed that RGD peptide, an αIIbβ3 inhibitor, blocked spike protein-induced platelet aggregation [11]. Moreover, subthreshold doses of GPIIb/IIIa blockers like eptifibatide and tirofiban could prevent thrombus formation in COVID-19 [21]. These results suggest that spike protein interacts directly with integrin αIIbβ3 to affect platelet aggregation, and indicate a potential target for SARS-CoV-2 interacting with platelet.

Platelet integrin αIIbβ3 outside-in signaling induces multiple events, such as stabilized platelet aggregation, spreading, and granule secretion, playing a critical role in the process of thrombosis [8]. We observed that spike protein upregulated platelet integrin αIIbβ3 outside-in signaling, like increasing exposure to immobilized spike protein spread and the phosphorylation of β-integrin tail, c-Src. Since spike protein could bind to platelet integrin αIIbβ3, we used immobilized spike protein to assess platelet spread function, which better reflects the effect of spike protein on platelet spread. Following ligand binding to integrin αIIbβ3, integrin αIIbβ3 clustering promotes Src activation, and the activated Src phosphorylates and supports the activation of numerous signaling proteins [8]. Therefore, using PP2, to inhibit c-Src activity, we observed that the spike protein did not further enhance platelet aggregation. The PP2 concentration (10 μM) chosen in blocked experiment was referred to previous study [22]. Thus, we conclude that the potentiated effect of spike


(A)
Resting
Control
Spike protein 10μg/ml
Fibrinogen 100μg/ml
SSC-A
Fg Binding-FITC-A
Fg binding (%)
Resting
Control
Spike protein 10μg/ml
Fibrinogen 100μg/ml
NS
(B)
ADP 5μM
Solvent control
Spike protein
Aggregation (%)
1 min
Control
Tirofiban
Aggregation (%)
NS
ADP 5μM
Spike protein
Tirofiban 10nM
(C)
Platelet aggregation
p-β3(Tyr785)
GAPDH
p-β3(Tyr785)/GAPDH
NS
ADP 5μM
Spike protein
Tirofiban 10nM
(D)
Thrombin 0.1U/ml
Solvent control
Spike protein
Aggregation (%)
1 min
Control
PP2
Aggregation (%)
NS
Thrombin 0.1 U/ml
Spike protein
PP2 10μM
(E)
Platelet aggregation
p-c-Src(Tyr419)
GAPDH
p-c-Src(Tyr419)/GAPDH
NS
Thrombin 0.1 U/ml
Spike protein
PP2 10μM

◂**Fig. 2** Spike protein potentiates platelet aggregation via interacting with platelet integrin αIIbβ3 and upregulating outside-in signaling. (**A**) Platelets were incubated with spike protein (10 μg/mL) or fibrinogen (100 μg/mL) for 10 min and stimulated with thrombin (0.1 U/mL) for 10 min. Platelet fibrinogen binding was analyzed using a flow cytometer (n=4). (**B**) Human PRP was incubated with spike protein at 1 μg/mL for 10 min in the presence of tirofiban (10 nM) for 1 min. Platelet aggregation assays were performed after stimulated with 5 μM ADP (n=3). (**C**) After aggregation in (**B**), the cell lysates were prepared and the phosphorylation levels of β3 were determined by Western Blot (n=3). (**D**-**E**) Platelets were preincubated with 10 μM PP2 for 10 min and incubated with spike protein at 1 μg/mL for 10 min. Platelet aggregation assays were performed after stimulated with 0.1 U/ml thrombin (n=4) (**D**). After platelet aggregation, the level of p-c-Src (**E**) was determined by Western blotting (n=3). * $P<0.05$, ** $P<0.01$, *** $P<0.001$, indicate significant difference between two groups. *NS*: no significant difference between the two groups. Statistical analyses were performed using One-way ANOVA with Tukey's post hoc test.

protein on platelet aggregation is mainly mediated by the upregulation of integrin αIIbβ3 outside-in signaling.

In the present study, our data shows that spike protein upregulates integrin αIIbβ3 outside-in signaling pathway to potentiate platelet aggregation. Other studies found that SARS-CoV-2 spike protein could potentiate platelet aggregation via ACE2 [17] or CD42b [11] -mediated signaling pathway, suggesting that spike protein may interact with platelets through a variety of targets. Subsequent experiments need to further investigate which target plays a more important role in this process. In conclusion, this integrin αIIbβ3-mediated outside-in signaling mechanism may be a potential therapeutic target to preventing and treating platelet hyperreactivity in COVID-19.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s11239-024-03008-8.

Institutional review board statement The study was conducted in accordance with the Declaration of Helsinki, and approved by the Ethics Committee of the School of public health (Shenzhen), Sun Yat-Sen University [[2020] No.013].

Author contributions RJW and YY designed the research. RJW, ZZT, MYZ, BYZ, YZL, YQZ, YHM and YMZ performed sample processing, experiments, and data analysis. RJW wrote the initial draft of the manuscript. YY critically revised the manuscript. All authors have read and agreed to the published version of the manuscript.

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Data availability Data described in the manuscript will be made available upon reasonable request.

Declarations

Conflicts of Interest The authors declare no conflict of interest.

References

1. Nguyen NT, Chinn J, Nahmias J, Yuen S, Kirby KA, Hohmann S, Amin A (2021) Outcomes and Mortality Among Adults Hospitalized With COVID-19 at US Medical Centers. Jama Netw Open 4. https://doi.org/10.1001/jamanetworkopen.2021.0417
2. Challen R, Brooks-Pollock E, Read JM, Dyson L, Tsaneva-Atanasova K, Danon L (2021) Risk of mortality in patients infected with SARS-CoV-2 variant of concern 202012/1: matched cohort study. BMJ 372:n579. https://doi.org/10.1136/bmj.n579
3. Zhang L, Feng X, Zhang D, Jiang C, Mei H, Wang J, Zhang C, Li H, Xia X, Kong S, Liao J, Jia H, Pang X, Song Y, Tian Y, Wang B, Wu C, Yuan H, Zhang Y et al (2020) Deep Vein Thrombosis in Hospitalized Patients With COVID-19 in Wuhan, China: Prevalence, Risk Factors, and Outcome. Circulation 142:114–128. https://doi.org/10.1161/CIRCULATIONAHA.120.046702
4. Manne BK, Denorme F, Middleton EA, Portier I, Rowley JW, Stubben C, Petrey AC, Tolley ND, Guo L, Cody M, Weyrich AS, Yost CC, Rondina MT, Campbell RA (2020) Platelet gene expression and function in patients with COVID-19. Blood 136:1317–1329. https://doi.org/10.1182/blood.2020007214
5. Xu XR, Zhang D, Oswald BE, Carrim N, Wang X, Hou Y, Zhang Q, Lavalle C, McKeown T, Marshall AH, Ni H (2016) Platelets are versatile cells: New discoveries in hemostasis, thrombosis, immune responses, tumor metastasis and beyond. Crit Rev Clin Lab Sci 53:409–430. https://doi.org/10.1080/10408363.2016.1200008
6. Yeung J, Li W, Holinstat M (2018) Platelet Signaling and Disease: Targeted Therapy for Thrombosis and Other Related Diseases. Pharmacol Rev 70:526–548. https://doi.org/10.1124/pr.117.014530
7. Makowski L, Olson-Sidford W, Weisel JW (2021) Biological and Clinical Consequences of Integrin Binding via a Rogue RGD Motif in the SARS CoV-2 Spike Protein. Viruses 13. https://doi.org/10.3390/v13020146
8. Huang J, Li X, Shi X, Zhu M, Wang J, Huang S, Huang X, Wang H, Li L, Deng H, Zhou Y, Mao J, Long Z, Ma Z, Ye W, Pan J, Xi X, Jin J (2019) Platelet integrin alphaIIbbeta3: signal transduction, regulation, and its therapeutic targeting. J Hematol Oncol 12:26. https://doi.org/10.1186/s13045-019-0709-6
9. Ma X, Liang J, Zhu G, Bhoria P, Shoara AA, MacKeigan DT, Khoury CJ, Slavkovic S, Lin L, Karakas D, Chen Z, Prifti V, Liu Z, Shen C, Li Y, Zhang C, Dou J, Rousseau Z, Zhang J et al (2023) SARS-CoV-2 RBD and Its Variants Can Induce Platelet Activation and Clearance: Implications for Antibody Therapy and Vaccinations against COVID-19. Research (Wash D C) 6:0124. https://doi.org/10.34133/research.0124
10. Ya F, Xu XR, Shi Y, Gallant RC, Song F, Zuo X, Zhao Y, Tian Z, Zhang C, Xu X, Ling W, Ni H, Yang Y (2019) Coenzyme Q10 Upregulates Platelet cAMP/PKA Pathway and Attenuates Integrin alphaIIbbeta3 Signaling and Thrombus Growth. Mol Nutr Food Res 63:e1900662. https://doi.org/10.1002/mnfr.201900662
11. Li T, Yang Y, Li Y, Wang Z, Ma F, Luo R, Xu X, Zhou G, Wang J, Niu J, Lv G, Crispe IN, Tu Z (2022) Platelets mediate

inflammatory monocyte activation by SARS-CoV-2 spike protein. J Clin Invest 132. https://doi.org/10.1172/JCI150101
12. Li Z, Delaney MK, O'Brien KA, Du X (2010) Signaling during platelet adhesion and activation. Arterioscler Thromb Vasc Biol 30:2341–2349. https://doi.org/10.1161/ATVBAHA.110.207522
13. Litvinov RI, Farrell DH, Weisel JW, Bennett JS (2016) The Platelet Integrin alphaIIbbeta3 Differentially Interacts with Fibrin Versus Fibrinogen. J Biol Chem 291:7858–7867. https://doi.org/10.1074/jbc.M115.706861
14. Wang R, Chen Y, Tian Z, Zhu M, Zhang B, Du S, Li Y, Liu Z, Hou S, Yang Y (2022) Coenzyme Q10 Attenuates Human Platelet Aggregation Induced by SARS-CoV-2 Spike Protein via Reducing Oxidative Stress In Vitro. Int J Mol Sci 23. https://doi.org/10.3390/ijms232012345
15. Senis YA, Mazharian A, Mori J (2014) Src family kinases: at the forefront of platelet activation. Blood 124:2013–2024. https://doi.org/10.1182/blood-2014-01-453134
16. Severin S, Nash CA, Mori J, Zhao Y, Abram C, Lowell CA, Senis YA, Watson SP (2012) Distinct and overlapping functional roles of Src family kinases in mouse platelets. J Thromb Haemost 10:1631–1645. https://doi.org/10.1111/j.1538-7836.2012.04814.x
17. Zhang S, Liu Y, Wang X, Yang L, Li H, Wang Y, Liu M, Zhao X, Xie Y, Yang Y, Zhang S, Fan Z, Dong J, Yuan Z, Ding Z, Zhang Y, Hu L (2020) SARS-CoV-2 binds platelet ACE2 to enhance thrombosis in COVID-19. J Hematol Oncol 13:120. https://doi.org/10.1186/s13045-020-00954-7
18. Jackson SP (2007) The growing complexity of platelet aggregation. Blood 109:5087–5095. https://doi.org/10.1182/blood-2006-12-027698
19. Takada Y, Ye X, Simon S (2007) The integrins. Genome Biol 8:215. https://doi.org/10.1186/gb-2007-8-5-215
20. Shattil SJ, Kashiwagi H, Pampori N (1998) Integrin signaling: the platelet paradigm. Blood 91:2645–2657
21. Weiss LJ, Drayss M, Manukjan G, Zeitlhöefler MJ, Kleiss J, Weigel ML, Herrmann J, Mott K, Beck S, Burkard P, Lam TT, Althaus K, Bakchoul T, Frantz S, Meybohm P, Nieswandt B, Weismann D, Schulze H (2022) Uncoupling of platelet granule release and integrin activation suggests GPIIb/IIIa as therapeutic target in COVID-19. Blood Adv. https://doi.org/10.1182/bloodadvances.2022008666
22. Hughan SC, Hughes CE, McCarty OJT, Schweighoffer E, Soultanova I, Ware J, Tybulewicz VLJ, Watson SP (2007) GPVI potentiation of platelet activation by thrombin and adhesion molecules independent of Src kinases and Syk. Arterioscl Throm Vas 27:422–429. https://doi.org/10.1161/01.Atv.0000252826.96134.21

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