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1 **The role of IgG subclasses and platelets in experimental anaphylaxis**

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15 **Conflict of Interest Statement**

16 The authors declare no conflict of interest.

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46 **Keywords (5):** Experimental anaphylaxis; Mouse model; IgG subclasses; Platelets; FcγRs

47

## 48 **Introduction**

49         In 1902, Charles Richet coined the term “anaphylaxis” to describe a “state of heightened  
50 sensitivity of a subject to a substance induced by a first injection, that instead of protecting the  
51 organism, renders it more fragile and more susceptible”. Since this first description,  
52 experimental work led to identification of antibodies, receptors, cells and mediators in this  
53 severe allergic reaction, leading to the paradigm that anaphylaxis is an IgE-dependent affliction  
54 that is triggered when allergens aggregate cognate IgE antibodies bound to the high-affinity IgE  
55 receptor (FcεRI) on the surface of mast cells and basophils. Their activation leads to the release  
56 of diverse bioactive mediators, including histamine, which are responsible for the associated  
57 clinical signs (1). Seminal works from the Galli, Kinet and Finkelman labs revealed, however,  
58 that anaphylaxis can also occur in mice deficient for IgE, FcεRI or mast cells, and suggested  
59 that it could be driven by IgG antibodies engaging Fcγ-chain containing receptors (reviewed  
60 in (1)). Nowadays the international consensus on anaphylaxis defines anaphylaxis as “a serious,  
61 generalized or systemic, allergic or hypersensitivity reaction that can be life-threatening or  
62 fatal”, which is deliberately generic and excludes any precision on the pathophysiological  
63 mechanism involved.

64         In this manuscript we will discuss recent findings on IgG-dependent anaphylaxis with a  
65 focus on the role of IgG subclasses and platelets in these reactions.

66

## 67 **IgG-dependent anaphylaxis in wild-type mice**

68         IgG-dependent passive systemic anaphylaxis (IgG-PSA) can be elicited in mice by the  
69 transfer of specific IgG antibodies (of either IgG1, IgG2a/c or IgG2b subclass, but not IgG3)  
70 followed by injection of their cognate antigen, or by transfer of pre-complexed IgG (*i.e.* immune  
71 complexes (IgG-ICs) or heat-aggregated IgG). IgG-PSA depends on IgG receptor (FcγR)-  
72 transduced activation of myeloid cells, leading to mediator release, which notably include  
73 platelet-activating factor (PAF) (1, 2). Mice express three activating (FcγRI, FcγRIII and  
74 FcγRIV) and one inhibitory FcγRs (FcγRIIB) each having a specific expression profile and  
75 distinct affinities for the different IgG subclasses. IgG-PSA in mice is associated with  
76 vasodilation, augmented vascular permeability and a reduction in core temperature, motility  
77 and awareness (Figure 1). Animals usually return to normal activity and behavior within 1-2  
78 hours, but in rare cases cardiopulmonary failure results in death. (IgG)-anaphylaxis can also be  
79 triggered by antigen exposure of previously immunized mice that lack key players of the IgE-

80 dependent pathway. IgG-independent immune players may however contribute to the reaction,  
81 rendering the interpretation of results more complex. To efficiently engage activating IgG  
82 receptors, IgG generally need to be present as multivalent complexes. There is a large consensus  
83 that IgG-PSA relies mainly on the engagement of FcγRIII and, to a lesser extent, on FcγRIV  
84 and possibly on FcγRI (2, 3).

85 The relative importance of FcγR-bearing effector cells to IgG-PSA remains more debated  
86 and is likely to depend on the experimental conditions (1-3). Indeed, all cells of hematopoietic  
87 origin express at least one activating FcγR with the exception of T cells, B cells and platelets,  
88 and could hence contribute to the reaction. Their involvement in anaphylaxis is often assessed  
89 either using depleting antibodies, which is problematic in the context of an antibody-dependent  
90 reaction, or using inhibitors, which may not be specific. In a comparative study using mouse  
91 IgG1, IgG2a and IgG2b with the same specificity to induce IgG subclass-specific anaphylaxis,  
92 we found that IgG1 and IgG2b-PSA shared a common mechanism that involved all tested  
93 myeloid cells and in which histamine H1 receptor blockade showed a stronger beneficial effect  
94 on PSA-associated temperature drop than PAF receptor blockade (2). IgG1- and IgG2b-PSA  
95 were regulated by the inhibitory IgG receptor FcγRIIB present on all myeloid cells and B cells.  
96 In contrast in IgG2a-PSA, FcγRIIB-driven inhibition was negligible. IgG2a-PSA was  
97 significantly reduced through depletion of neutrophils or monocytes/macrophages and  
98 attenuated by both PAF-receptor and histamine H1 receptor antagonists (2). This particularity  
99 of IgG2a-PSA may be due to the overall higher affinity of IgG2a to FcγRs (2) that also may  
100 explain the relative resistance of IgG2a-PSA to changes in IgG/FcγR affinity induced by  
101 modification of IgG-glycosylation (i.e. terminal sialylation) compared to IgG1/IgG2b-PSA (4).  
102

### 103 **IgG-anaphylaxis in FcγR-humanized mice**

104 To approach human pathophysiology, anaphylaxis has been studied in mice carrying  
105 human FcγRs, either as a single transgene, as in the case of hFcγRIIA (5-7), or in more complex  
106 models expressing several FcγRs (6). Indeed, extrapolating results from IgG/FcγR-dependent  
107 reactions from mouse to human pathophysiology is challenging, because both species express  
108 very different sets of FcγRs (four in mice, six in humans) that each shows distinct interaction  
109 profile with the different IgG subclasses (IgG1-4 in human). Among human FcγRs, only  
110 FcγRIIB is inhibitory and expressed at much lower levels than in mice, suggesting that  
111 regulation of IgG-driven responses in humans though co-engagement of this inhibitory receptor  
112 is less effective than in mice. Furthermore, as an example, whereas IgG3 binds to all human

113 Fc $\gamma$ R<sub>s</sub>, its murine counterpart exclusively engages mouse Fc $\gamma$ RI. These differences extend  
114 through all IgG subclasses, their affinities for Fc $\gamma$ R<sub>s</sub> and their capacity to trigger Fc-dependent  
115 effector functions. Studies in Fc $\gamma$ R-humanized mice revealed that engagement of human Fc $\gamma$ R<sub>s</sub>  
116 by IgG-ICs is sufficient to trigger anaphylaxis (5). Among hFc $\gamma$ R<sub>s</sub>, hFc $\gamma$ R<sub>IIA</sub> appears to be the  
117 major contributor in IgG-PSA (6) and despite its expression on all myeloid cells, hFc $\gamma$ R<sub>IIA</sub>-  
118 expressing neutrophils and monocytes/macrophages, through their release of PAF, play a  
119 predominant role over mast cells, basophils and eosinophils (5). Unexpectedly, hFc $\gamma$ R<sub>IIA</sub>-  
120 transgenic mice also revealed the critical contribution of a blood component that was until then  
121 overlooked in the context of anaphylaxis.

122

### 123 **Role of platelets in IgG-anaphylaxis**

124 Mouse platelets are devoid of any Fc $\gamma$ R. Human platelets on the contrary express  
125 Fc $\gamma$ R<sub>IIA</sub>/CD32A and incubation with IgG-ICs can induce their activation, aggregation and  
126 release of granular content. Using hFc $\gamma$ R<sub>IIA</sub>-transgenic mice that confer IgG receptor  
127 expression to platelets, we and others demonstrated that IgG-induced platelet activation is  
128 critical for experimental anaphylaxis, and results in a rapid, severe and prolonged (24 h)  
129 thrombocytopenia (6, 7). Activated platelets released serotonin, which determined the severity  
130 of anaphylaxis (6, 7). Platelets also contributed to IgG-PSA in a more complex mouse model  
131 of cognate hFc $\gamma$ R expression (6). Recently, platelet-released PAF was similarly proposed to  
132 trigger a transient disruption of endothelial integrity and mast cell activation resulting in shock  
133 (8). Due to their abundance in blood, it is therefore conceivable that platelets are among the  
134 first players to become activated by circulating IgG-ICs triggering a cascade of events that  
135 drives the activation and mediator release from various cell types contributing to anaphylaxis  
136 (Figure 2).

137

### 138 **Relevance for human anaphylaxis and conclusion**

139 The fact that transgenic expression of a complete set of human Fc $\gamma$ R<sub>s</sub> reproducing  
140 mostly the original expression profiles on all hematopoietic cells stimulated with human  
141 aggregated IgG is sufficient to induce anaphylaxis in mice (6), is a strong indicator for the  
142 relevance of IgG anaphylaxis in humans. Indeed, several lines of evidence support the existence  
143 of IgG/Fc $\gamma$ R-, neutrophil- and PAF-dependent human anaphylaxis. Cases of anaphylaxis were  
144 reported after administration of different therapeutic IgG antibodies (1), and serum PAF  
145 concentrations correlate with anaphylaxis severity in humans (9). In a clinical study of

146 neuromuscular-blocking agent-induced anaphylaxis, concentrations of anti-drug IgG, markers  
147 of Fc $\gamma$ R engagement and neutrophil activation (upregulation of CD11b and CD18, elevated  
148 levels of elastase and DNA-MPO complexes in plasma), as well as reduced activity of PAF-  
149 acetyl hydrolase correlated with anaphylaxis severity (10). Notably, neutrophil activation could  
150 be observed in patients lacking evidence of classical IgE-anaphylaxis (10). Limited data from  
151 these patients further evidenced that platelet activation (upregulation of CD62P) was associated  
152 with anaphylaxis severity and that anaphylaxis occurrence was accompanied by a reduction in  
153 circulating platelet numbers (6). These findings open new perspectives for the understanding  
154 and management of IgE-independent anaphylaxis in humans. In addition to certain drugs that  
155 can directly activate mast cells (notably through the recently described MRGPRX2), IgG-  
156 dependent reactions may account for or contribute to anaphylaxis, in particular when large  
157 amounts of IgG-ICs can form in the circulation. Further clinical studies will allow to determine  
158 whether it could be beneficial for patients at risk of developing IgG-driven anaphylaxis (i.e.  
159 programmed intravenous administration of certain antibodies or drugs) to transiently receive  
160 treatments to block Fc $\gamma$ Rs (especially Fc $\gamma$ RIIA) or limit the biological effects of serotonin  
161 and/or PAF.

162

163

#### 164 **Author Contributions**

165 All authors have made a substantial intellectual contribution to the manuscript, and approved it  
166 for publication.

167

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174

#### 175 **References**

- 176 1. Reber LL, Hernandez JD, Galli SJ. The pathophysiology of anaphylaxis. *J Allergy*  
177 *Clin Immunol.* 2017;140(2):335-48.
- 178 2. Beutier H, Gillis CM, Iannascoli B, Godon O, England P, Sibilano R, et al. IgG  
179 subclasses determine pathways of anaphylaxis in mice. *J Allergy Clin Immunol.*  
180 2017;139(1):269-80 e7.

- 181 3. Khodoun MV, Kucuk ZY, Strait RT, Krishnamurthy D, Janek K, Clay CD, et al.  
182 Rapid desensitization of mice with anti-FcγRIIb/FcγRIII mAb safely prevents  
183 IgG-mediated anaphylaxis. *J Allergy Clin Immunol.* 2013;132(6):1375-87.
- 184 4. Epp A, Hobusch J, Bartsch YC, Petry J, Lilienthal GM, Koeleman CAM, et al.  
185 Sialylation of IgG antibodies inhibits IgG-mediated allergic reactions. *J Allergy Clin*  
186 *Immunol.* 2018;141(1):399-402 e8.
- 187 5. Jonsson F, Mancardi DA, Zhao W, Kita Y, Iannascoli B, Khun H, et al. Human  
188 FcγRIIA induces anaphylactic and allergic reactions. *Blood.* 2012;119(11):2533-44.
- 189 6. Beutier H, Hechler B, Godon O, Wang Y, Gillis CM, de Chaisemartin L, et al.  
190 Platelets expressing IgG receptor FcγRIIA/CD32A determine the severity of  
191 experimental anaphylaxis. *Sci Immunol.* 2018;3(22).
- 192 7. Cloutier N, Allaëys I, Marcoux G, Machlus KR, Mailhot B, Zufferey A, et al. Platelets  
193 release pathogenic serotonin and return to circulation after immune complex-mediated  
194 sequestration. *Proc Natl Acad Sci U S A.* 2018;115(7):E1550-E9.
- 195 8. Karhausen J, Choi HW, Maddipati KR, Mathew JP, Ma Q, Boulaftali Y, et al.  
196 Platelets trigger perivascular mast cell degranulation to cause inflammatory responses and  
197 tissue injury. *Sci Adv.* 2020;6(12):eaay6314.
- 198 9. Vadas P, Perelman B, Liss G. Platelet-activating factor, histamine, and tryptase levels  
199 in human anaphylaxis. *J Allergy Clin Immunol.* 2013;131(1):144-9.
- 200 10. Jonsson F, de Chaisemartin L, Granger V, Gouel-Cheron A, Gillis CM, Zhu Q, et al.  
201 An IgG-induced neutrophil activation pathway contributes to human drug-induced  
202 anaphylaxis. *Sci Transl Med.* 2019;11(500).

203

#### 204 **Figure Legends:**

205

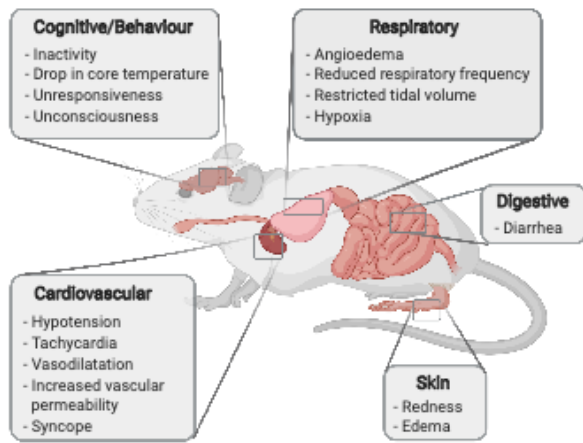
206 Figure 1. Pathophysiologic changes in experimental anaphylaxis in mice. Anaphylaxis is a  
207 systemic hypersensitivity reaction that affects multiple organs; the most common clinical signs  
208 of anaphylaxis in mice are indicated. Created with BioRender.com

209

210

211 Figure 2. Model of IgG-dependent experimental anaphylaxis in hFcγRIIA-expressing mice in  
212 the absence of mouse endogenous FcγRs. Human FcγRIIA expression is conserved in  
213 hFcγRIIA-transgenic mice, including its expression on all myeloid cells and platelets. Injected  
214 heat-aggregated (HA)-IgG, mimicking IgG-ICs forming inside the circulation, can engage  
215 hFcγRIIA on any of these cells, but will have a stochastically higher likelihood to encounter  
216 platelets>neutrophils>monocytes>>basophils, leading to their activation. As a consequence  
217 platelets will be activated, form aggregates, adhere to circulating leukocytes and degranulate.  
218 Platelet-released serotonin can directly trigger anaphylaxis-associated vascular leakage,  
219 vasodilation and bronchoconstriction. Platelet-released PAF, or PAF-release by other IgG-IC-  
220 activated myeloid cells can fuel the reaction through activation of perivascular mast cells  
221 leading to histamine release. PAF and histamine may contribute to clinical signs of hFcγRIIA -  
222 PSA in mice. Created with BioRender.com

223 **Figure 1**



224  
225 **Figure 2**

