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► **To cite this version:**

Morgane Verduyn, Guillaume Botto, Julien Jaubert, Clément Lier, Thomas Flament, et al..  
Serum IgG Concentrations in Adult Patients Experiencing Virus-Induced Severe Asthma Exac-  
erbations. *Journal of Allergy and Clinical Immunology: In Practice*, 2019, 7, pp.1507-1513.e1.  
10.1016/j.jaip.2018.12.028 . hal-03486266

**HAL Id: hal-03486266**

**<https://hal.science/hal-03486266v1>**

Submitted on 20 Dec 2021

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1 **Serum IgG concentrations in adult patients experiencing virus-induced severe asthma**  
2 **exacerbations**

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25 No funding.

26 All authors declare no conflict of interest.

27

28 Word count of the abstract: 234

29 Word count of the text: 2634

30 **Abstract:**

31 Background: Patients experiencing severe asthma exacerbations have a poorer quality of life  
32 and an increase in morbidity and mortality. Viruses are frequently involved in asthma  
33 exacerbations.

34 Objective: Determining the value of measuring serum immunoglobulin G (IgG)  
35 concentrations in asthma exacerbations and their link with viral infections in patients  
36 hospitalised for asthma has been assessed.

37 Methods: Patients hospitalised for asthma exacerbation were included in an observational  
38 study from the 1 January to 31 December 2015. Serum IgG concentrations on admission were  
39 compared between patients with a positive upper airway viral sample to those with a negative  
40 viral sample.

41 Results: Amongst the 82 patients included, those with positive viral nasopharyngeal samples  
42 (n=40) presented with lower serum IgG concentrations during exacerbation than those with a  
43 negative viral sample (n=42) ( $10.1 \pm 2.3$  g/l vs.  $11.5 \pm 3.6$  g/l,  $p < 0.05$ ). The median  
44 concentration of serum IgG was lower in patients hospitalised for more than three days  
45 compared to those hospitalised for less than three days ( $10.0$  g/l [8.2-12.4] vs.  $11.4$  g/l [10.1-  
46 12.8],  $p < 0.05$ ) and in patients who received oral corticosteroid therapy for more than five  
47 days compared to those treated with oral steroids for less than five days ( $10.1$  [8.3-12.2] vs.  
48  $11.6$  g/l [10.0-13.8],  $p < 0.05$ ).

49 Conclusion: Serum IgG was significantly lower when asthma exacerbations were associated  
50 with positive viral samples. The patients with lower serum IgG concentrations required longer  
51 hospitalisations and longer courses of steroids.

52

53 Highlights

54 What is already known about this topic? Many patients suffering from asthma experience  
55 virus-induced asthma exacerbations but the pathophysiology is not fully understood and no  
56 biomarkers have been found to predict severity.

57 What does this article add to our knowledge? Patients hospitalised for asthma exacerbations  
58 associated with a positive virus sample have lower serum IgG than their negative  
59 counterparts. Longer hospital stays and a longer duration of oral steroids were linked to lower  
60 serum IgG concentrations.

61 How does this study impact current management guidelines? Serum IgG quantification during  
62 asthma exacerbations may be lower in patients with positive virus samples but more data are  
63 needed.

64

65 Key words: asthma, immunoglobulin G, virus, exacerbation

66

67 Abbreviations:

68 ACT: Asthma control test

69 BMI: Body mass index

70 CVID: Common variable immune deficiency

71 COPD: Chronic obstructive pulmonary disease

72 FEV1: Forced expiratory volume in one second

73 FVC: Forced vital capacity

74 GINA: Global Initiative for Asthma

75 Ig: Immunoglobulin

76 IgG: Immunoglobulin G

77 IgE: Immunoglobulin E

78 PCR: Polymerase Chain Reaction

79 ROC curves: Receiver operating characteristics curves

80 RSV: Respiratory syncytial virus

81

## 82 **Introduction**

83 Asthma is a chronic respiratory disease associated with airway inflammation (1). This disease  
84 affects approximately 334 million people worldwide and its prevalence is increasing (2).  
85 According to the latest World Health Organization (WHO) estimates released in December  
86 2016, there were 383,000 asthma-related deaths in 2015 (3). The majority of these deaths  
87 occurs in low- and middle-income countries (3).

88 Exacerbations play an important role in asthma morbidity and mortality (4). An exacerbation  
89 is defined as acute or sub-acute episodes of progressive worsening respiratory symptoms (5).  
90 A severe exacerbation corresponds to an emergency consultation or hospitalisation (6).  
91 Asthma is an increasingly common cause of emergency department consultations (7). Twenty  
92 percent of asthma patients have experienced exacerbations requiring treatment within the  
93 emergency department or hospitalisation and these patients account for more than 80% of the  
94 total direct costs of asthma (8). Mortality amongst patients hospitalised for asthma  
95 exacerbations accounts for one-third of all asthma-related deaths (9).

96 Infections with pulmonary tropism are very frequently involved in triggering asthma  
97 exacerbations (40 to 80% of cases) (10) and are more often associated with failure of  
98 conventional treatments in asthmatic patients (11). Rhinovirus, human metapneumovirus,  
99 enterovirus, coronavirus and respiratory syncytial virus (RSV) are the most common viruses  
100 found in asthma exacerbations (12). Rhinovirus is particularly related to asthma exacerbations  
101 and is found in 60% of children experiencing asthma exacerbations (12). In adults, rhinovirus  
102 also seems to be frequently involved in asthma exacerbations (13).

103 Regarding virus-induced asthma exacerbations, the antiviral response is complex and involves  
104 immune cells leading to the release of inflammatory mediators (14, 15). Immunoglobulins  
105 (Ig) play a key role in the anti-viral response but knowledge remains limited about their

106 involvement in virus-induced asthma exacerbations. Interestingly, it has recently been shown  
107 that patients with primary hypogammaglobulinemia are more likely to suffer from viral  
108 infections, including rhinovirus, compared to healthy subjects, in spite of intravenous  
109 immunoglobulin replacement (16). The prevalence of asthma appears to be higher in patients  
110 with various immune deficiencies including hypogammaglobulinemia, with an estimated  
111 prevalence of 15% in the studies (17). The mechanism causing the increased prevalence of  
112 viral infections in patients with hypogammaglobulinemia is unknown. Secretory IgA  
113 deficiency in the bronchial mucosa has been suggested (16). To our knowledge, the link  
114 between viral infections and serum immunoglobulin in asthmatic patients is poorly  
115 understood.

116 The purpose of this project is to compare the serum immunoglobulin G (IgG) concentrations  
117 between patients with an upper airway specimen that tested positive for a virus to those with a  
118 negative viral specimen at the time of hospitalisation for asthma exacerbations.

119

## 120 **Population and Methods:**

121

### 122 Population

123 An observational study was conducted from 1 January to 31 December 2015. Patients with  
124 asthma were recruited from the Department of Respiratory Medicine of the University  
125 Hospital of Tours, located in the centre of France, as well as from the Department of  
126 Respiratory Medicine of the University Centre of Reunion, a French island located in the  
127 Indian Ocean. The socioeconomic statuses of the patients are very close because both  
128 locations are managed in the same way by the French government. Inclusion criteria were  
129 patients aged 18 or older and hospitalised for an asthma exacerbation. An exacerbation was

130 defined as the increase in respiratory symptoms for more than 24 hours or the use of oral  
131 corticosteroid therapy. Asthma diagnoses were established on a history of respiratory  
132 symptoms such as wheezing, shortness of breath, chest tightness and coughing associated  
133 with variable expiratory airflow limitation prior to hospitalisation according to the Global  
134 Initiative for Asthma (GINA) (1). Exclusion criteria were smoking more than ten packs of  
135 cigarettes a year, chronic obstructive pulmonary disease and pregnancy. The study was  
136 approved by the Institutional Review Board of the *Société de pneumologie de langue*  
137 *Française*. Each patient gave an informed written consent to take part in the study.

138

#### 139 Clinical and biological data during asthma exacerbation

140 Demographic data such as age, sex, body mass index, smoking status quantified in packs per  
141 year (P/A), inhaled treatment prescribed prior to exacerbation, as well as the use and duration  
142 of oral steroids during the exacerbation were collected. The total serum immunoglobulin E  
143 (IgE) concentration (immunoassay technique, ImmunoCAP IgE total, Thermo Fisher  
144 Scientific, Phadia AB, Uppsala, Sweden), blood eosinophilia (Beckman Coulter LH 780  
145 controlled analyser), and C-reactive protein (CRP) (Beckman Coulter LH 780 Controlled  
146 Analyser) were also collected. The severity of exacerbations was characterised by the length  
147 of the hospital stay including the time spent in the emergency room and if the patient needed  
148 to be treated in the intensive care unit. Controlled asthma was defined as an asthma control  
149 test (ACT) score greater than or equal to 20 points in the month preceding the consultation.

150

#### 151 Upper airway viral sampling



152 Viral carriage was assessed in the nasopharyngeal fluid obtained by nasopharyngeal aspiration  
153 performed as soon as possible by qualified nurses accustomed to the technique. The viral  
154 identification was performed by multiplex real time Polymerase Chain Reaction (PCR)  
155 (Tours: Anyplex II RV16, V1.1 detection, Eurobio, France and Thermal Cycler, C1000,  
156 BioRad, US Meeting; Reunion: FTD Respiratory pathogens 21, Fast track diagnostics,  
157 Luxembourg and Rotor-Gene Q instrument, Qiagen, France). At least 16 viruses (RSV A and  
158 B, Rhinovirus, metapneumovirus, myxovirus influenzae A, H1N1, B, parainfluenzae from 1  
159 to 4, coronavirus 229E, NL63 and OC43, adenovirus, enterovirus) were analysed in each  
160 centre. In Reunion, the panel of viruses also included coronavirus HKU1, bocavirus and  
161 parechovirus.

162

#### 163 Immunoglobulin G (IgG) serum concentration

164 Serum IgG concentrations and IgG subclasses were measured by the nephelometric technique  
165 on BN Prospect® (Siemens Healthcare Diagnostics, Marburg, Germany) at the time of  
166 admission to the hospital section.

167

#### 168 Follow-up visit

169 Patients received a follow-up visit three months after exacerbation. Functional respiratory  
170 tests were performed via flow-volume curves using a plethysmograph (SensorMedics Vmax  
171 Encore, Carefusion, San Diego, CA). The values of both forced vital capacity (FVC) and  
172 forced expiratory volume in one second (FEV 1) were expressed as a percentage of predicted  
173 values according to age, sex, weight and height. Airway obstruction was defined by a post-  
174 bronchodilator FEV1/FVC ratio of less than 70%. Measurements were taken according to

175 ATS/ERS 2006 recommendations using the predicted values published by the ERS (18, 19).  
176 Prick tests to aeroallergens (house dust mites (*Dermatophagoides pteronyssinus* and  
177 *Dermatophagoides farinae* at both sites and *Blomia tropicalis* in Reunion)), mould (*Alternaria*  
178 *alternata*), animal dander (cat and dog), cockroaches (*Blattella germanica*), tree pollens (olive  
179 tree, birch) and herbaceous plants (ragweed, ambrosia, wormwood and grasses) were also  
180 carried out.

181

## 182 Statistical analyses

183 Quantitative data, with a normal distribution, were expressed as mean and standard deviation  
184 (mean  $\pm$  SD) and quantitative data, with no normal distribution, were expressed as median  
185 deviation [interquartile range]. Qualitative data were expressed as frequencies (%). A  
186 Student's t-test (parametric) was used to compare continuous variables with normal  
187 distribution. The Mann-Whitney test was used for quantitative data and had no normal  
188 distribution. Statistical significance was defined as a p value of  $<0.05$ . Receiver operator  
189 characteristics (ROC) curves were used to assess serum IgG concentrations as a potential  
190 marker of a positive virus specimen. The best IgG cut-off was determined using the Youden's  
191 index. Sensitivity and specificity of IgG cut-off for a positive virus sample were also defined.  
192 A Spearman test was used to assess the correlation between serum IgG concentrations and  
193 other continuous variables. The linearity of these relations was then tested using linear  
194 regression. The statistical analysis was carried out using the Graph Pad Prism 5 software  
195 (license reference: GPW6-222441-RILS-2C3D1).

196

## 197 Results

198 Characteristics of patients and virus distribution:

199 Eighty-two patients were included in this study. The patients' characteristics are summarised  
200 in Table 1. The median age was 49.5 [28.8-63.3] years old. Patients were predominantly  
201 female (76.8%) and overweight with a median body mass index (BMI) of 26.3 [22.1-31.2]  
202 kg/m<sup>2</sup>. Inhaled corticosteroid treatment was used in 61% of patients before asthma  
203 exacerbation and 14.6% out of all the patients required intensive care at the time of  
204 exacerbation. The median duration of oral corticosteroid therapy and the median length of  
205 stay for the exacerbation were 7.0 days and 4.0 days, respectively.

206 Nasopharyngeal aspirations were positive for viruses in 48.8% of patients. The most common  
207 viruses were rhinovirus (38%), myxovirus influenza (18%), metapneumovirus (12%) and  
208 enterovirus (10%) (Figure 1).

209

210 Characteristics of patients according to the viral status of nasopharyngeal aspiration:

211 Patients with a positive or negative viral sample showed no statistically significant difference  
212 in terms of demographic and biological data (Table 2). Patients with positive nasopharyngeal  
213 aspiration for viruses were slightly younger than patients with negative nasopharyngeal  
214 aspiration for viruses (43.5 vs. 50.5 years, respectively) with no statistical significance ( $p$   
215  $>0.05$ ). Patients with positive viral specimens also seemed to be hospitalised in intensive care  
216 units more often compared to patients harbouring negative viral specimens (20% vs. 9.5%,  
217 respectively), however the difference was not significant.

218

219 Immunoglobulin blood concentration:

220 Patients with a positive viral sample had significantly lower serum IgG concentrations than  
221 patients hosting a negative viral specimen ( $10.1 \pm 2.3$  g/l vs.  $11.5 \pm 3.6$  g/l,  $p < 0.05$ ) (Table 3).  
222 No difference was found for serum IgA and IgM concentrations in both groups. IgG  
223 subclasses were similar between groups. An inverse correlation was observed between age  
224 and serum IgG concentrations ( $p = 0.016$ ). No other correlation was shown for the other  
225 demographic characteristics.

226

#### 227 Serum IgG concentrations and diagnosis of a positive virus sample.

228 ROC curve analysing the serum IgG concentrations as a marker for predicting a specimen  
229 positive for a virus is presented in Figure 2. According to Youden's index, the best cut-off  
230 value of serum IgG to predict a positive virus sample is 12.30 g/l, the optimal sensitivity and  
231 specificity were 80.0% and 40.5%, respectively.

232

#### 233 IgG concentration and characteristics of asthma exacerbation:

234 The median serum concentration of IgG was significantly lower in patients hospitalised for  
235 more than three days than in those whose hospital stay was less than or equal to three days  
236 ( $10.0$  g/l [ $8.2-12.4$ ] g/l vs.  $11.4$  g/l [ $10.1-12.8$ ] g/l,  $p < 0.05$ ) (Figure 3B). Patients receiving  
237 oral corticosteroids for more than five days had lower median serum IgG concentrations than  
238 patients who were given corticosteroid therapy for five days or less ( $10.1$  [ $8.3-12.2$ ] g/l vs.  
239  $11.6$  [ $10-13.8$ ] g/l respectively,  $p < 0.05$ ) (Figure 3C). The median serum concentration of IgG  
240 was not significantly different if patients were hospitalised in intensive care units or not  
241 (Figure 3A). An inverse correlation was observed between the length of stay and the serum  
242 IgG concentration ( $p = 0.022$  with the Spearman test) (Figure 4). No correlation was found  
243 between oral steroids and serum IgG concentrations. ROC curves have been created according  
244 to the length of stay and the steroid duration in order to know the reliability of serum IgG

245 concentrations as a predictive biomarker of exacerbation severity. For the length of stay  
246 prediction ( $\leq 3$  days or  $> 3$  days), the best sensitivity and specificity of IgG concentrations are  
247 80.00% and 50.00%, respectively for a cut-off value of 11.05 g/l (Figure E1a). Regarding the  
248 steroid duration prediction ( $\leq 5$  days or  $> 5$  days), for a cut-off value of 9.99 g/l, the best  
249 sensitivity and specificity of IgG concentrations are 62.07% and 66.67%, respectively (Figure  
250 E1b). If a positive virus sample is combined with the IgG serum concentrations, for a cut-off  
251 of 10.75 g/l, the best sensitivity and specificity of the length of stay prediction are 79.17% and  
252 61.11%. Regarding a cut-off of 11.05 g/l, the best sensitivity and specificity of the steroid  
253 duration prediction are 82.76% and 55.00% (Figure 5).

254

## 255 **Discussion:**

256

257 Asthma patients with a positive nasopharyngeal viral sample had a lower serum IgG  
258 concentration, at the time of exacerbation, than those with a negative viral sample. Moreover,  
259 the median serum IgG concentration was lower in patients hospitalised for more than three  
260 days as well as in those who were given oral steroids for more than five days compared to  
261 their counterparts.

262 In our study, the epidemiology of viruses found in patients hospitalised for asthma  
263 exacerbations was similar to data observed in the literature. Indeed, herein, rhinovirus was the  
264 most frequently observed pathogen (38%). This virus is known to be more often associated  
265 with asthma exacerbations in children (20-22), and some works suggest that it is also the most  
266 incriminated virus in severe asthma exacerbations in adults (13, 23, 24). Our patients also  
267 presented with other viruses such as myxovirus influenzae. Myxovirus influenza, particularly  
268 influenza A H1N1, is responsible for a significant morbidity and mortality in asthmatic  
269 patients (25-27). Enterovirus appears to be less frequently found in the literature (28). In our

270 cohort, enterovirus was always found in co-infection with rhinovirus and this represented five  
271 out of ten patients who had co-infections. The five remaining patients were co-infected with  
272 rhinovirus/myxovirus influenza A (n=3), metapneumovirus A and B/myxovirus influenza A  
273 H1N1 (n=1), adenovirus/myxovirus influenza A (n=1) and Coronavirus 229E/myxovirus  
274 influenza A (n=1). No relationship between co-infection and serum IgG concentrations or the  
275 severity of exacerbations was found.

276 No demographic differences were found between the patients with positive virus samples and  
277 those with negative virus samples. However, although the difference was not statistically  
278 significant, patients with a positive virus sample were slightly younger compared to their  
279 negative counterparts (43.5 vs. 50.5 years, respectively). The lack of significance is probably  
280 due to the small sample size. It is well known that asthma exacerbation in young people is  
281 associated with virus infections (29). BMI appears to be a bit higher in patients with positive  
282 virus samples compared to patients with negative virus samples (29 vs. 25 kg/m<sup>2</sup>,  
283 respectively) but the difference is also not significant. The risk of viral infections in asthma  
284 and obese patients has been identified elsewhere in the literature. Indeed asthma and obesity  
285 have been found as independent risk factors of pandemic influenza hospitalisation (30). In our  
286 study, more patients in the positive virus group required intensive care although there was no  
287 significant difference. Data from the literature suggest that respiratory viruses may trigger  
288 severe asthma exacerbations, particularly in young people (31).

289

290 The link between viral infections and asthma exacerbations is widely recognised and the  
291 impairment of viral immunity may partly explain the development of viral infections in  
292 asthma patients (32, 33). In influenza infections, IgG has been identified as being particularly  
293 involved in protecting the respiratory tract against viruses and its role seems to be superior to  
294 IgA (34). Rhinovirus-specific IgG appears to prevent and control re-infection and a high

295 concentration of specific IgG antibodies is correlated with the attenuation of respiratory  
296 symptoms and a reduction in viral shedding (35). A decrease in specific viral antibodies in  
297 patients with asthma has been suggested (36). However, in another study, higher antibody  
298 responses to rhinovirus have been shown in asthmatic patients compared to non-asthmatic  
299 controls (37). Given the study conducted by Johnston et al. on rhinovirus involvement in  
300 asthma exacerbations in children, we may also speculate that the admitted patients had less  
301 exposure to school-aged children in the past and therefore had not generated a sufficient  
302 antibody response to the current virus(es) (38). Other mechanisms beyond adaptive immunity  
303 dysfunction are involved in virus-induced asthma exacerbations. Indeed, an altered innate  
304 immune anti-viral response has been observed in asthmatic patients compared to non-  
305 asthmatic ones (39-41). Our study does not add a mechanistic explanation for the low level of  
306 serum IgG in patients with a positive virus sample.

307 Our results may suggest that serum IgG concentrations at the time of asthma exacerbation  
308 would be a marker of severity. Indeed, patients with a hospitalisation length of more than  
309 three days or a steroid course of more than five days show lower IgG concentrations at the  
310 time of exacerbation. Amongst patients with stable COPD, reduced total IgG levels have  
311 been linked to an increased risk of exacerbations and hospitalisations (42). In that study,  
312 patients with serum IgG <7 g/l had a higher risk of COPD exacerbations compared to patients  
313 with the highest quartile of serum IgG.. We assessed the predictive ability of serum IgG  
314 concentrations as a biomarker of exacerbation severity. However, it does not seem to be a  
315 useful predictor based on modest sensitivity and poor specificity. If we combine serum IgG  
316 concentrations and a positive virus sample, the prediction of exacerbation severity was not  
317 improved, and further studies are needed to specifically assess IgG level as a marker of  
318 severity in asthma exacerbations. The prediction of next hospitalisations with IgG levels  
319 would also be interesting but we cannot answer this question with our study as it was not

320 designed to answer this question. However, we may speculate that a low IgG level  
321 particularly at baseline would predispose patients to virus infection and consequently increase  
322 their risk of hospitalisation.

323 Our study does have several limitations. The difference in IgG concentrations between  
324 patients with positive and negative viral samples is statistically significant, but it remains  
325 uncertain whether this difference is clinically relevant or not. More robust studies on the role  
326 of IgG are needed. In addition, the change of IgG concentrations or other immunoglobulins  
327 over time was not assessed in our study. We did not measure IgG in patients in a stable  
328 condition. It would have been interesting to know if the IgG decline is transient or longer-  
329 lasting. Indeed, in patients with chronic obstructive pulmonary disease (COPD), low IgG  
330 serum concentrations at baseline seem to be associated with more severe exacerbations per  
331 year (42). However, in this study, no virus analysis was performed. Finally, our study is based  
332 on multiplex PCR for virus identification. Nevertheless, the list of viruses, though very  
333 extensive, is not exhaustive, therefore emerging viruses cannot be ruled out.

334

### 335 **Conclusion:**

336 The results of our study show that serum IgG was significantly lower when severe asthma  
337 exacerbations were associated with a positive viral sample. Serum IgG levels were lower in  
338 patients with a longer length of stay and when a course of oral steroids was prescribed for a  
339 longer duration. Further studies are requested to know the exact usefulness of IgG  
340 concentrations as a severity marker of severe asthma exacerbations. The understanding of  
341 immunological mechanisms of viral infection in severe asthma exacerbations may be an  
342 interesting lead for the development of new therapies in patients for whom severe  
343 exacerbations are a source of significant morbidity and mortality.



344

345 Figure 1: Details of viruses found in nasopharyngeal aspirations of hospitalised patients for  
346 asthma exacerbation.

347 Figure 2: Receiver operating characteristics (ROC) curve analysis of serum IgG  
348 concentrations for the prediction of a positive virus sample. At the cut-off value of 12.30 g/l,  
349 sensitivity and specificity are 80.0% and 40.5%, respectively.

350 Figure 3: Comparison of blood IgG concentrations a) between hospitalised patients in  
351 intensive care and those without ICU, b) between patients hospitalised for three days or less  
352 and those hospitalised for more than three days and c) between patients treated during five  
353 days or less with oral corticosteroids and those treated during more than five days, \* $p < 0.05$ .  
354 ICU: Intensive care unit.

355 Figure 4: Scatter plots with regression line showing the relationship between the serum IgG  
356 concentrations and the hospital length of stay. \* $p = 0.0058$  using linear regression.

357 Figure 5: Receiver operating characteristics (ROC) curve analysis of serum IgG  
358 concentrations combined with a positive virus sample for a) the prediction of a length of stay  
359 duration of more than three days (for a cut-off value of 10.75 g/l, sensitivity and specificity  
360 are respectively 80.0% and 40.5%) and b) the prediction of a steroid duration of more than  
361 five days (for a cut-off value of 11.05, the best sensitivity and specificity are 82.76% and  
362 55.00%).

363

364 Table 1: Characteristics of patients hospitalised due to asthma exacerbations.

	<b>n=82</b>
Age (years)	49.5 [28.8-63.3]
Female	63 (76.8)
Active or former smokers	21 (25.6)
BMI (kg/m <sup>2</sup> )	26.3 [22.1-31.2]
Allergen sensitisation	35 (42.7)
Use of an inhaled corticosteroid before hospitalisation	50 (61.0)
Inhaled corticosteroid dose (beclomethasone equivalent doses) (µg/d)	1600 [800-2000]
Exacerbation ≥2 12 months prior to exacerbation	15 (18.3)
Oral corticotherapy duration for exacerbation	7.0 [5.0-8.5]
Length of hospitalisation	4.0 [2.0-5.0]
Intensive care unit stay at the time of exacerbation	12 (14.6)
Positive viral sampling	40 (48.8)
FEV1 after hospitalisation (ml) (% predicted values)	76 [63-88]
FEV1/FVC after hospitalisation (%)	74 ± 15
Blood Eosinophil (G / l)	0.20 [0.03-0.68]
Total IgE (kIU / l)	339 [148-786]

365 Quantitative data, with normal distribution, are expressed as means and standard deviation  
 366 (mean ± SD) and quantitative data, without normal distribution, are expressed in median  
 367 [interquartile range]. The qualitative data are expressed as frequencies (%).  
 368

369 Table 2: Characteristics of patients hospitalised for asthma exacerbations with positive or  
 370 negative upper airway viral samples. FEV1: Forced expiratory volume in 1 second, FVC:  
 371 Forced vital capacity, IgE: Immunoglobulin E

	<b>Virus + n=40</b>	<b>Virus – n=42</b>	<b>p</b>
Age (years)	43.5 [24.3-60.3]	50.5 [36.0-68.3]	0.14
Female	33 (82.5)	30 (71.4)	0.30
Active or former smokers	11 (27.5)	10 (23.8)	0.80
BMI (kg/m <sup>2</sup> )	29.0 [22.0-33.2]	25.0 [22.3-30.4]	0.42
Allergen sensitisation	18 (45.0)	17 (40.5)	0.82
Use of an inhaled corticosteroid prior to hospitalisation	24 (60.0)	26 (61.9)	1.00
Inhaled corticosteroid dose (beclomethasone equivalent doses) (µg/d)	1600 [800-2000]	1600 [800-1600]	0.91
Exacerbation ≥2 12 months prior to exacerbation	5 (12.5)	10 (23.8)	0.39
Oral corticotherapy duration while exacerbation (days)	7.0 [5.0-9.5]	6.0 [5.0-8.5]	0.15
Length of hospitalisation	4.0 [2.0-5.0]	3.5 [2.0-5.0]	0.31
Intensive care unit stay at the time of exacerbation	8 (20.0)	4 (9.5)	0.22
FEV1 after hospitalisation (% predicted values)	80.5 [61.0-90.5]	75.5 [63.5-86.00]	0.50
FEV1/FVC after hospitalisation (%)	75 ± 12	66 ± 12	0.052
Blood Eosinophil (G/l)	0.2 [0.0-0.4]	0.2 [0.0-0.8]	0.39
Total IgE (kIU/l)	349 [185-1003]	310 [127-516]	0.29

372 Values are expressed as median and [interquartile range] or mean ± standard deviation for  
 373 quantitative data and raw values (%) for categorical data. A Student's t-test (parametric) is  
 374 used to compare continuous variables to a normal distribution. The Mann-Whitney test is used  
 375 for quantitative data that do not have a normal distribution.

376 Table 3: Comparison of serum Immunoglobulin G (IgG), Immunoglobulin A (IgA),  
 377 Immunoglobulin M (IgM) and IgG subclass levels between patients with positive viral  
 378 samples (Virus +) and negative (Virus -).

379

	<b>Virus + n=40</b>	<b>Virus – n=42</b>	<b>p</b>
IgG (g/l)	10.1 ± 2.3 10.10 [8.15-11.68]	11.5 ± 3.6 11.25 [9.09-13.43]	<0.05
IgA (g/l)	2.30 [1.77-3.06]	2.34 [1.98-2.86]	0.70
IgM (g/l)	0.88 [0.55-1.15]	0.75 [0.56-1.36]	0.91
IgG1 (g/l)	4.9 [3.5-5.8]	5.5 [4.5-7.0]	0.059
IgG2 (g/l)	3.7 ± 1.0	4.1 ± 1.4	0.12
IgG3 (g/l)	0.58 [0.30-0.80]	0.57 [0.43-0.86]	0.38
IgG4 (g/l)	0.27 [0.15-0.43]	0.27 [0.15-0.60]	0.85

380 Values are expressed as median and [interquartile range] or mean ± standard deviation for  
 381 quantitative data and raw values (%) for categorical data. Both median [interquartile range]  
 382 and mean ± standard deviation are given for IgG. A Student test (parametric) is used to  
 383 compare continuous variables to a normal distribution. The Mann-Whitney test is used for  
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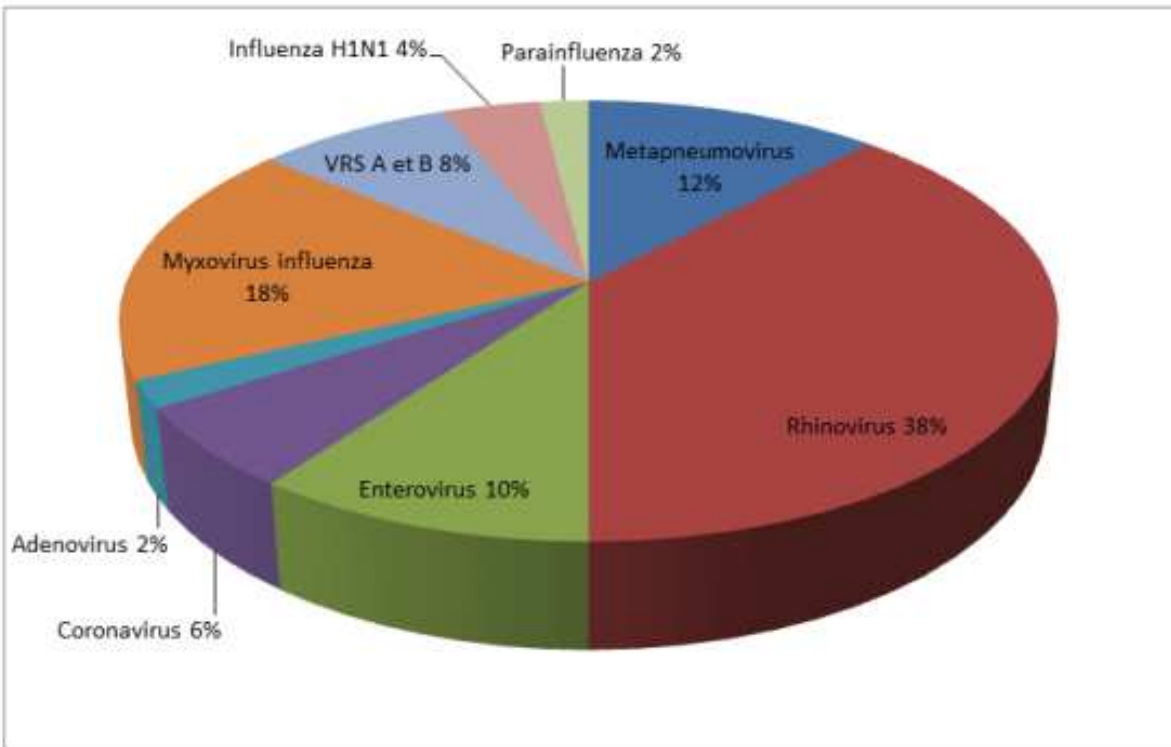
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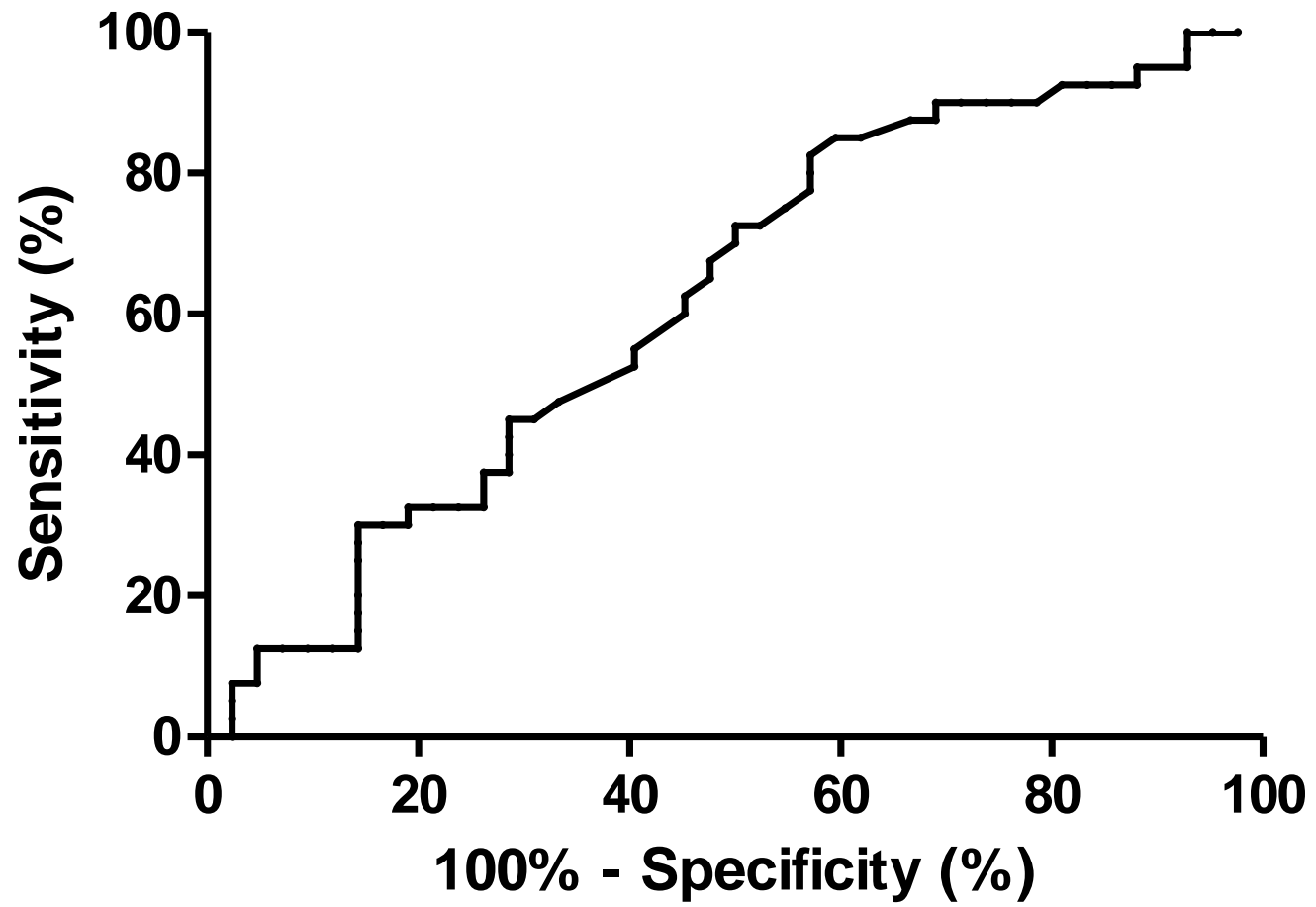
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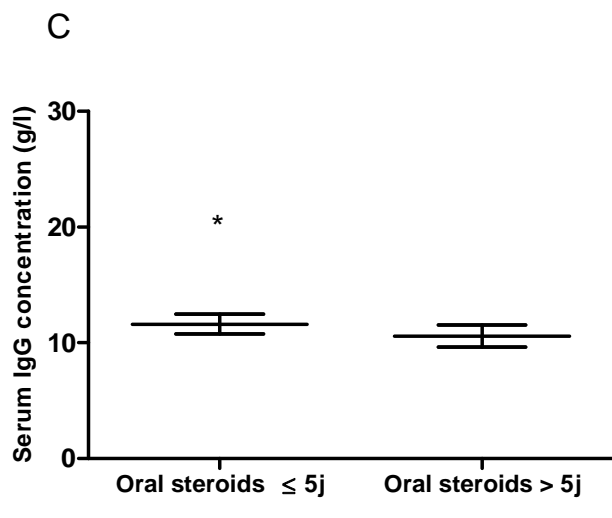
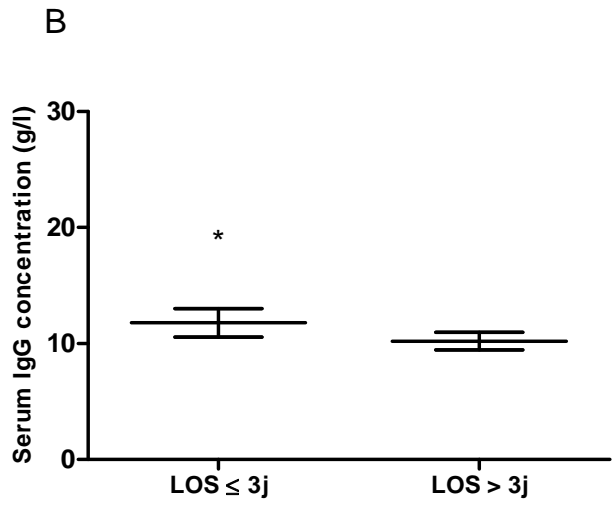
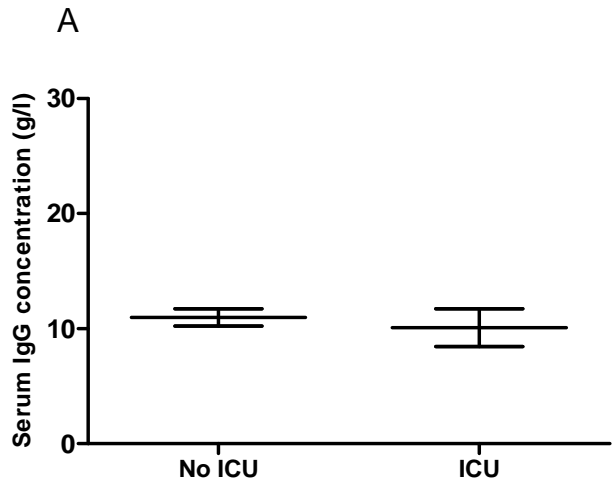
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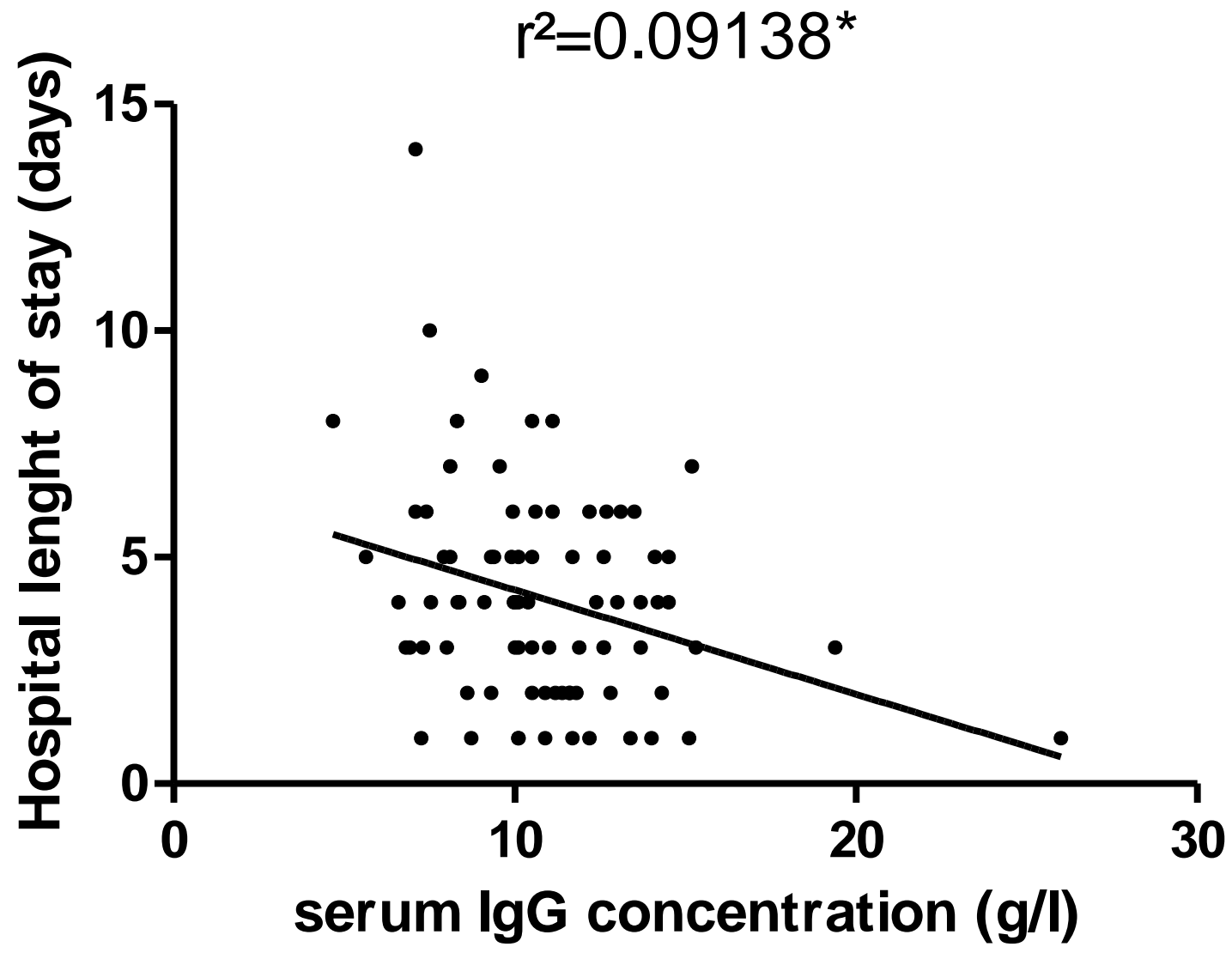
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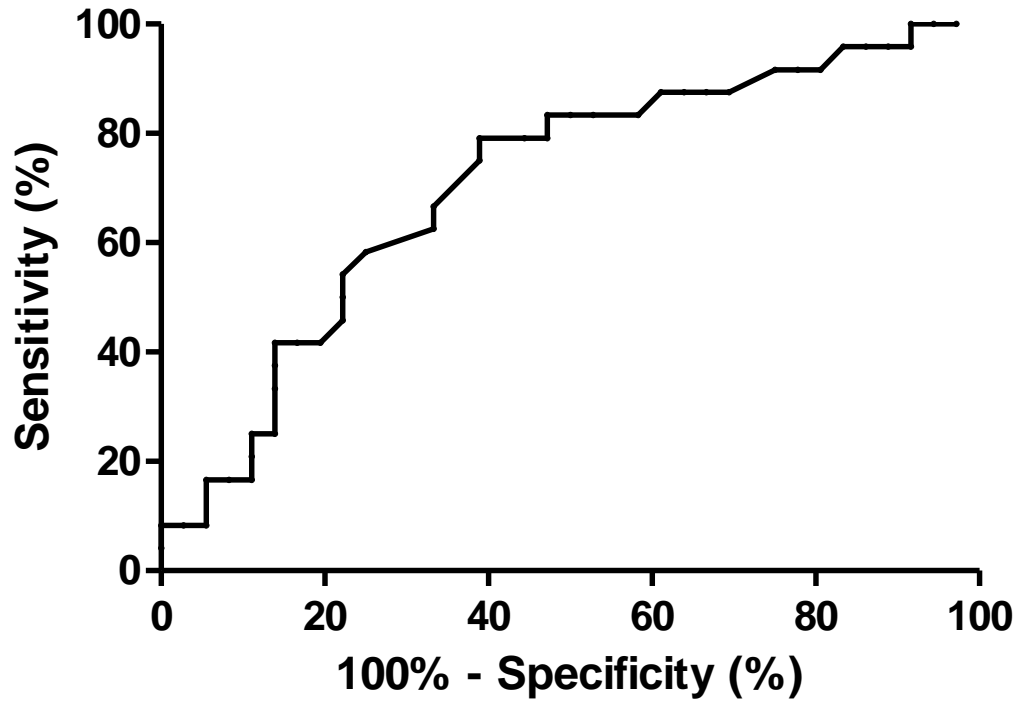








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