

Supplemental appendix:

Supplemental Methods

Western blot of discordant samples

rPLA2R (300ng/track) was run on a 10% SDS PAGE gel under non reducing conditions. The gel was electroblotted onto immobilon membrane using semi dry conditions for 60 mins and blocked for 60 mins in Superblock solution. Each track was cut out and placed in a multiwell cassette for incubation with test serum at 1:10 dilution in Superblock containing 0.1% Tween 20. A positive serum (concordantly positive by both IFa and ELISA) and a negative serum (concordantly negative by both assays) and the 7 sera yielding discordant results by IFa and ELISA were tested. Following incubation for 2 hours with test sera, membrane strips were washed 3 times in PBS Tween, incubated with 1:25,000 dilution of biotin conjugated anti-IgG (Jackson Labs) for 2 hours, washed 3 times in PBS Tween, incubated with 1:1000 dilution of QDOT 625 Streptavidin for 2 hours and washed 3 times in PBS Tween. Strips were aligned on a glass plate and examined for fluorescence using an Alpha Imager (excitation 365nm, emission 625nm).

Supplemental Table S1

Table S1a. Outcome in different tertiles of IgG4 titer (ELISA)

Outcome:	aPLA2R_IgG4 0-63 (n=26)	aPLA2R_IgG4 64-133 (n= 25)	aPLA2R_IgG4 >133 (n=26)	p- value
Partial remission	10 (39%)	9 (36%)	10 (39%)	ns
Complete remission	5 (19%)	11 (44%)	8 (31%)	ns
Renal failure	5 (19%)	0	4 (15%)	ns
Persist. proteinuria	6 (23%)	5 (20%)	4 (15%)	ns
Spont. remission	6 (23%)	10 (40%)	2 (8%)	0.03

Table S1b. Outcome in different tertiles of IgG3 titer (ELISA)

Outcome:	aPLA2R_IgG3 0-4 (n=29)	aPLA2R_IgG3 5-13 (n= 24)	aPLA2R_IgG3 >13 (n=24)	p- value
Partial remission	6 (18%)	11 (46%)	12 (50%)	ns
Complete remission	12 (39%)	6 (25%)	6 (25%)	ns
Renal failure	3 (14%)	2 (8%)	4 (17%)	ns
Persist. proteinuria	8 (27%)	5 (21%)	2 (8%)	ns
Spont. remission	9 (31%)	4 (17%)	5 (21%)	ns

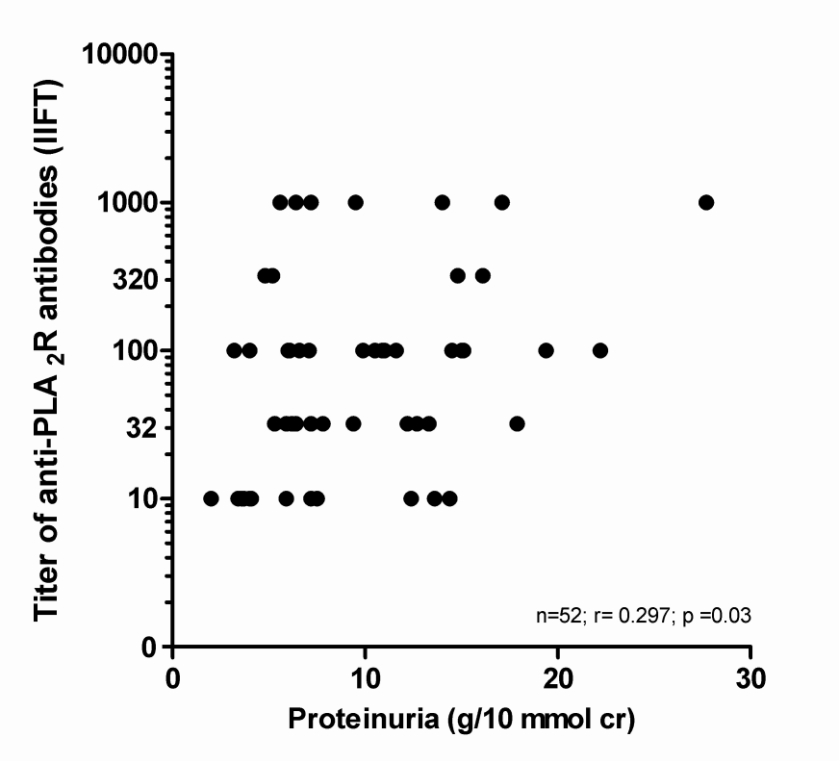
Table S1c. Outcome in different tertiles of IgG1 titer (ELISA)

Outcome:	aPLA2R_IgG1 0-9 (n=25)	aPLA2R_IgG1 10-26 (n= 27)	aPLA2R_IgG1 >26 (n=25)	p- value
Partial remission	8 (32%)	10 (37%)	11 (44%)	ns
Complete remission	7 (28%)	10 (37%)	7 (28%)	ns
Renal failure	2 (8%)	3 (11%)	4 (16%)	ns
Persist. proteinuria	8 (32%)	4 (15%)	3 (12%)	ns
Spont. remission	10 (40%)	5 (19%)	3 (12%)	ns

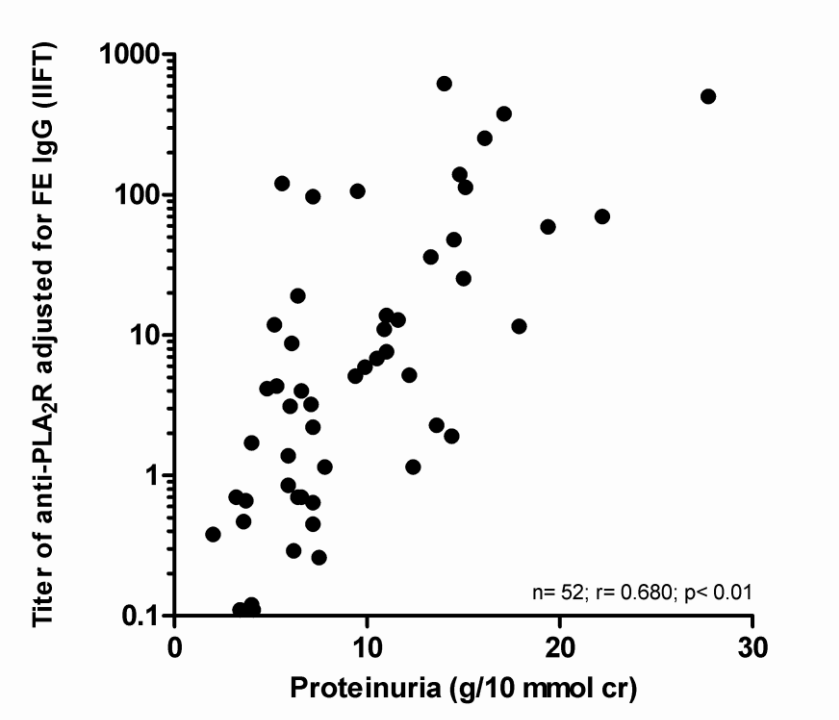
aPLA2R*Fract.IgG= anti-PLA2R antibody titer * fractional IgG excretion; Partial remission = proteinuria <3.5 g/day with Δ proteinuria >-50% from baseline and stable renal function; complete remission = proteinuria <0.2 g/day with stable renal function; renal failure = Δ serum creatinine >+50% from baseline; persist.proteinuria = persistent proteinuria >3.5 g/day or >50% of baseline proteinuria without Δ serum creatinine >+50%; Spont.remission = spontaneous remission (no treatment with immunosuppressive agents)

Supplemental Figures

Figure S1: Correlation between anti-PLA₂R titers measured with an indirect immunofluorescence technique and proteinuria in the patients of the Dutch cohort.



S1a: unadjusted analysis



S1b: anti-PLA₂R levels adjusted for fractional excretion of IgG

Figure S2: Western blot in 7 discordant patients
 WB =Western blot
 IFa = aPLA2R antibody titer by indirect immunofluorescence test
 ELISA = aPLA2R antibody titer by ELISA

