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Immunogenity of the pneumococcal polysaccharide vaccine in COPD patients. The effect of systemic steroids ☆

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Summary *Rationale*: To investigate if systemic steroids influence the antibody response to the 23-valent pneumococcal polysaccaride vaccine (23-PPV) in COPD patients.

Patients and methods: COPD patients on: (a) ⩾10 mg of prednisolone/day (SS, $n=30$); (b) inhalative steroids (IS, $n=30$); (c) controls without COPD (CG, $n=29$) were vaccinated with 23-PPV. The concentration (μg/ml) of capsular specific anti-pneumococcal IgG antibodies (AB) for the serotypes (PNC) 4,6B,9 V,14,18C,19F,23F were measured by Elisa technique before, 3 and 12 months (m) after vaccination. Non-responders were defined when AB-concentrations did neither doubled nor reach ⩾1 μg/ml.

Results: $N=24$ (CG), $n=29$ (IS), $n=18$ (SS) patients completed the study (mean age 64 yrs.). Serious adverse events were not observed. Geometric mean (GM) AB-concentration of all serotypes increased significantly (CG, IS, SS) 3 and 12 m after vaccination ($P<0.05$). The percentage of non-responders ranged between 16% (PNC 19F, IS) and 65% (PNC 4, SS) after 3 m and 21% (PNC 19F, IS) and 82% (PNC 4, CG) after 12 m. Neither post-vaccine AB-concentrations (3 and 12 m) nor the rate of non-responders differed significantly between patients on systemic steroids and the other groups (IS, CG).

Conclusions: Systemic steroids did not influence the AB-response. In all groups mean AB-concentration increased significantly after vaccination but an important percentage of subjects of all three groups were non-responders.

Introduction

COPD is a major public health problem and in recent projections COPD will be the third most frequent cause of death and the fifth cause of disability-adjusted life years lost worldwide in 2020.[1] *Streptococcus pneumoniae* is one of the main pathogens causing pulmonary infections in

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this patient group.[2] The 23-valent pneumococcal polysaccharide vaccine (23-PPV) is recommended for COPD patients by major health authorities in Europe and the US.[3–6] Despite this vaccination rates and acceptance by physicians varies widely.[5,7] Although the 23-PPV seems to be beneficial in large studies[8–10] the results of some publications addressing the effectiveness of the 23-PPV in preventing pneumococcal community-acquired pneumonia (CAP) in high risk groups are inconclusive.[10–13]

The immune response to the polysaccharide vaccine has been studied in several age groups and in subjects with co-morbidities. Data suggest that the protective effect of the pneumococcal vaccine diminishes with age.[10,14] In around 20% of elderly patients vaccinated with PPV antibody response and functionality of antibodies are deficient.[14,15] So far only few studies have addressed the issue of the immunogenity of the pneumococcal vaccine in COPD patients.[16–18] In this patient group systemic steroids are frequently prescribed during exacerbations and in the treatment of advanced stages of the disease. To the authors knowledge there are no data about a possible influence of systemic steroids, taken by COPD patients, to the immunogenity of the 23-valent polysaccharide vaccine. Thus the main objective of the present study was to examine whether a daily systemic steroid medication alters the immunogenity of the 23-PPV in this patient group.

Methods

The study period was from July 1999 until December 2001. Patients with COPD ($n = 60$) were recruited at our 250 bed chest city hospital and controls at a collaborating internal medicine doctors office ($n = 29$). The diagnosis of chronic obstructive bronchitis was made according to German and European guidelines.[19,20] We excluded patients previously vaccinated with 23-PPV, subjects under immunosuppressive medication other than prednisolone, patients with HIV, tuberculosis, insulin-dependent diabetes, known renal disease or malignancies. Every included patient fulfilled at least one criterion for pneumococcal vaccination according to the recommendations of the German health authorities.[3] All subjects were vaccinated with 0.5 ml of a standard, commercially available 23-PPV (Pneumopur®, Chiron Behring, Marburg, Germany). Concentration of capsular specific anti-pneumococcal IgG antibodies for the serotypes (PNC) 4,6B,9 V,14,18C,19F,23F at baseline, after 3 and 12 months were measured. Written informed consent was obtained from all subjects prior to inclusion to the study. The study protocol was approved by the Ethical Committee of the Medical Faculty, Free University Berlin.

Definition of groups

(1) *COPD patients with a systemic steroid therapy (SS)*: All subjects had a history of COPD and were medicated with 10 mg or more of prednisolone/day (or an equivalent dose of another steroid). Medication and dose had to be equal or over 10 mg/day (maximum 30 mg) prednisolone equivalent for at least 2 weeks prior to inclusion and had to be taken for at least 3 weeks after vaccination.
(2) *COPD patients on inhalative steroids (IS)*: Criteria for inclusion to this group was a documented history of COPD and a daily treatment with inhalative steroids (any dose of beclometasone, budenoside or fluticasone).
(3) *Control group (CG)*: At least one criterion for pneumococcal vaccination according to the recommendations of the German health authorities was applicable to all subjects in this group.[3] Patients with pulmonary co-morbidity (asthma, COPD, emphysema or bronchiectasis) were excluded.

Measurement of antibodies

Quantitative measurements of pneumococcal capsule-specific IgG were performed from patients serum collected before vaccination (1), after 3 months (2) and 12 months (3). Serotype-specific IgG-antibody concentration (PNC) was measured by a modified ELISA technique using Nunc CovaLink® microtiter plates (Nunc, Germany) and serum lot 89-SF as standard serum (FDA, Bethesda, MD). Serum samples were preincubated with 10 μg/ml pneumococcal polysaccharide C (CPS, Statens Seruminstitut, Denmark) for blocking of unspecific anti-CPS-antibodies. Reference serum 89-SF kindly provided by Dr. Frash (Rockville, USA) was used for assay standardisation. Sera with known high antibody titers were used as reference and quality control sera. Minimum antibody detection level was 0.1 μg/ml. Evaluation of the method and specific details about the proceedings of the Nunc CovaLink NH® ELISA method have been previously described.[21]

Performed comparisons and definition of non-responders

Mean pneumococcal capsule-specific IgG concentrations at baseline, 3 and 12 months after vaccination were compared. Patients were defined

as non-responders when after vaccination (3 and 12 months) baseline polysaccharide specific AB-concentration did neither increase two-fold nor the increment reached at least $\geqslant 1\,\mu g/ml$ (absolute). Patients with high pre-vaccination antibody levels to the tested serotypes ($\geqslant 4\,\mu g/ml$) were excluded from the analysis of non-responders. Further subjects were categorized in those who did respond (as previously defined) to less than two or more than five of the seven tested serotypes.

Statistical analysis

Results of antibody concentrations are expressed as geometric mean $\pm$ SD. Multiple comparisons of continuous variables were done by ANOVA with Bonferroni post hoc correction. In case of significant differences of mean serotype-specific IgG-antibody concentrations each patient group was opposed to each other group by an additional test. Therefore for non paired, continuous variables we used the Kruskal–Wallis or Mann–Whitney test, for paired variables the Wilcoxon signed rank test. Categorical variables were compared using the Chi-square test or Fisher exact test, when appropriate. The level of significance was set at $P<0.05$. Data analysis was performed with SPSS® version 10 on a standard personal computer with a Windows operating system (Windows 98®).

Results

Of 89 initially enrolled patients (CG $n=29$, IS $n=30$, SS $n=30$) $n=71$ completed the study (CG $n=24$, IS $n=29$, SS $n=18$; male 57%, female 43%) over the period of one year. Mean age in the studied groups was (SS) 64 yr ($\pm$10.4, range 45–84 yr), (IS) 62 yr ($\pm$12, 39–81 yr) and (CG) 64 yr ($\pm$18.5, 24–91 yr), respectively. Dropout rate was highest in SS (40%, $n=12$). 8 patients (27%) in the SS group died during the study period as a consequence of their advanced pulmonary disease. 97% ($n=29$) of IS and 83% ($n=24$) of CG-patients completed the study. Dropouts in IS and CG were due to missed follow up. The mean prednisolone doses at the moment of vaccination in the group on systemic steroids were as follows: (a) at the moment of vaccination 21.1 $\pm$ 7.5 mg; (b) 2 weeks prior to vaccination 17.1 $\pm$ 8.7 mg and (c) 3 weeks after vaccination 13.7 $\pm$ 5.3 mg.

Vaccine tolerability

The vaccine was well tolerated. No serious adverse events were observed. Dropouts due to death were in no relation to vaccination and occurred at least 3 months after vaccination (range 3–10 months). 16% of all patients reported local reactions and 5% fatigue for less than two days. In no case a physician had to be consulted because of vaccine related adverse event. Differences in side effects between the groups were not significant.

Antibody concentrations

Baseline pneumococcal capsule-specific IgG antibody concentrations of all tested serogroups showed no significant differences. Mean antibody concentrations increased significantly in all groups 3 months and one year after vaccination ($P<0.05$; see Table 1 and Fig. 1). Mean PNC 14 concentrations were significantly higher in (IS) compared to (CG) after 3 and 12 months (geometric mean ($\mu g/ml$) $\pm$ SD: 3 months (IS) 10.2 $\pm$ 40.5 vs. (CG) 4.4 $\pm$ 37.4, $P=0.03$; one year (IS) 9.4 $\pm$ 23 vs. (CG) 3.3 $\pm$ 22, $P=0.01$). (SS) showed no significant differences in mean antibody concentrations after vaccination when compared to the other groups. Response to the vaccine varied widely between all serotypes and all tested groups reflecting the wide range of antibody concentrations after vaccination (see Table 1).

Non-responders to vaccination

We found non-responders to vaccination in all groups and serotypes. The percentage of NR 3 months after vaccination ranged as follows in the different groups: CG 24% (PNC 18C, $n=4$) to 53% (PNC 9 V, $n=10$); IS 16% (PNC 19F, $n=3$) to 56% (PNC 18C, $n=13$); SS 17% (PNC 14, $n=2$) to 65% (PNC 4, $n=11$). After 12 months the percentages of non-responders increased: from CG 33% (PNC 18C, $n=6$) to 82% (PNC 4, $n=19$); IS 21% (PNC 19F, $n=4$) to 67% (PNC 23F, $n=16$); SS 39% (PNC 14, $n=5$) to 69% (PNC 23F, $n=11$) (see Fig. 2).

Analysis revealed the following significant differences: non-responders to PNC14 were more frequent in the control group when compared to subjects on inhalative steroids 3 months (47% vs. 17%, $P<0.05$) and 12 months (60% vs. 22%, $P<0.05$) after vaccination. The rate of non-responders to the tested serotypes in the SS group did not differ significantly when compared to the other groups (CG and IS). After 12 months non-responders to PNC18 were more frequent in patients on inhalative steroids when compared to controls (65% vs. 33%, $P<0.05$).

The percentage of subjects who responded to less than two serotypes ranged between 14% (CG)

Table 1 Geometric mean (GM), standard deviation (SD) and range of anticapsular pneumococcal antibodies of the tested serotypes (PNC, in μg/ml, m = months).

	CG			IS			SS		
	GM	SD	Range	GM	SD	Range	GM	SD	Range
PNC 4									
Baseline	0.6	1.3	0.1–6.6	0.8	1.4	0.1–5	0.6	3.1	0.1–17
3 m	2.2	5.6	0.1–18	2.3	5.1	0.3–29	1.1	3.6	0.1–19
12 m	1.6	5.6	0.2–25	1.6	2.8	0.3–13	1.2	3	0.1–12
PNC 6B									
Baseline	0.9	1.8	0.1–6	1.8	4.4	0.1–24	1.3	2.8	0.1–12
3 m	3.1	11.5	0.1–45	4.7	7.6	0.5–40	3.1	22.1	0.1–117
12 m	2.3	5	0.1–16	3.7	8	0.1–32	2.8	10.9	0.1–48
PNC 9									
Baseline	1.1	1.9	0.1–9.4	1.5	2.1	0.1–8.5	1	2.5	0.1–11
3 m	3.1	10.2	0.1–41	4.6	10.4	0.5–50	2.8	11.4	0.1–55
12 m	3.1	8.4	0.3–31	3.8	5.3	0.3–21	2.2	3.7	0.1–12
PNC 14									
Baseline	1.6	7.9	0.3–43	1.5	1.8	0.1–6.8	1.3	10.9	0.1–56
3 m	4.4*	37.5	0.2–145	10.2*	40.5	0.8–193	5	13.2	0.2–63
12 m	3.3*	22.2	0.3–87	9.3*	23.1	0.3–87	4.8	9.6	0.1–32
PNC 18C									
Baseline	1	1.9	0.1–5.9	1.4	1.9	0.2–8.1	1.3	6.5	0.1–36
3 m	5.1	26.5	0.4–136	4.5	9.9	0.5–40	3.9	10.1	0.1–37
12 m	3.8	10.5	0.3–37	3.5	8.3	0.2–31.1	2.6	5	0.1–20
PNC 19F									
Baseline	2	8.7	0.2–40	2.5	3.6	0.6–13.8	2	3.6	0.1–14
3 m	6.9	17.5	0.1–82	9.3	22.3	0.9–105	5.3	11.9	0.1–54
12 m	4.4	8.9	0.7–31	7.2	12.3	0.6–43	3.8	5.2	0.1–18
PNC 23F									
Baseline	2	8.7	0.2–40	2.5	3.6	0.6–13.8	2	3.6	0.1–14
3 m	6.9	17.5	0.1–82	9.3	22.3	0.9–105	5.3	11.9	0.1–54
12 m	4.4	8.9	0.7–31	7.2	12.3	0.6–43	3.8	5.2	0.1–18

CG = control group ($n = 29$), IS = inhalative steroids ($n = 30$), SS = systemic steroids ($n = 30$). Mean pneumococcal capsule-specific IgG did increase significantly after 3 and 12 months in all PNC and all groups (CG,IS,SS). The IS had higher mean PNC 14 concentration when compared to CG (3 and 12 months, $P < 0.05$). Significant differences are marked with "*" ($P < 0.05$).

and 21% (SS) after 3 months and 28% (both SS and IS) and 33% (CG) after 12 months. Responders to more than five serotypes ranged from 21% (SS) to 33% (IS) after 3 months and 17% (CG, SS) and 28% (IS) after 12 months. Differences were not significant (see Table 2).

Discussion

The main findings of our study are: (1) mean pneumococcal capsule-specific IgG antibodies increased significantly 3 and 12 months after vaccination in all groups; (2) the antibody concentrations after vaccination showed a high variability in all groups; (3) the hypothesis that systemic steroids would alter the immunogenity of the vaccine was not confirmed under the present experimental conditions. We observed differences between COPD-patients under inhalative steroids and controls, specially as regards PNC14; (4) We found an important percentage of non-responders to the seven tested serotypes after one year (21–83%). Non-responders to less than two serotypes ranged between 14–21% and 28–33% after 3 months and one year, respectively.

Previous studies have addressed the issue of magnitude, duration, quality and function of the 23-PPV. Rubins et al. compared the immunogenity

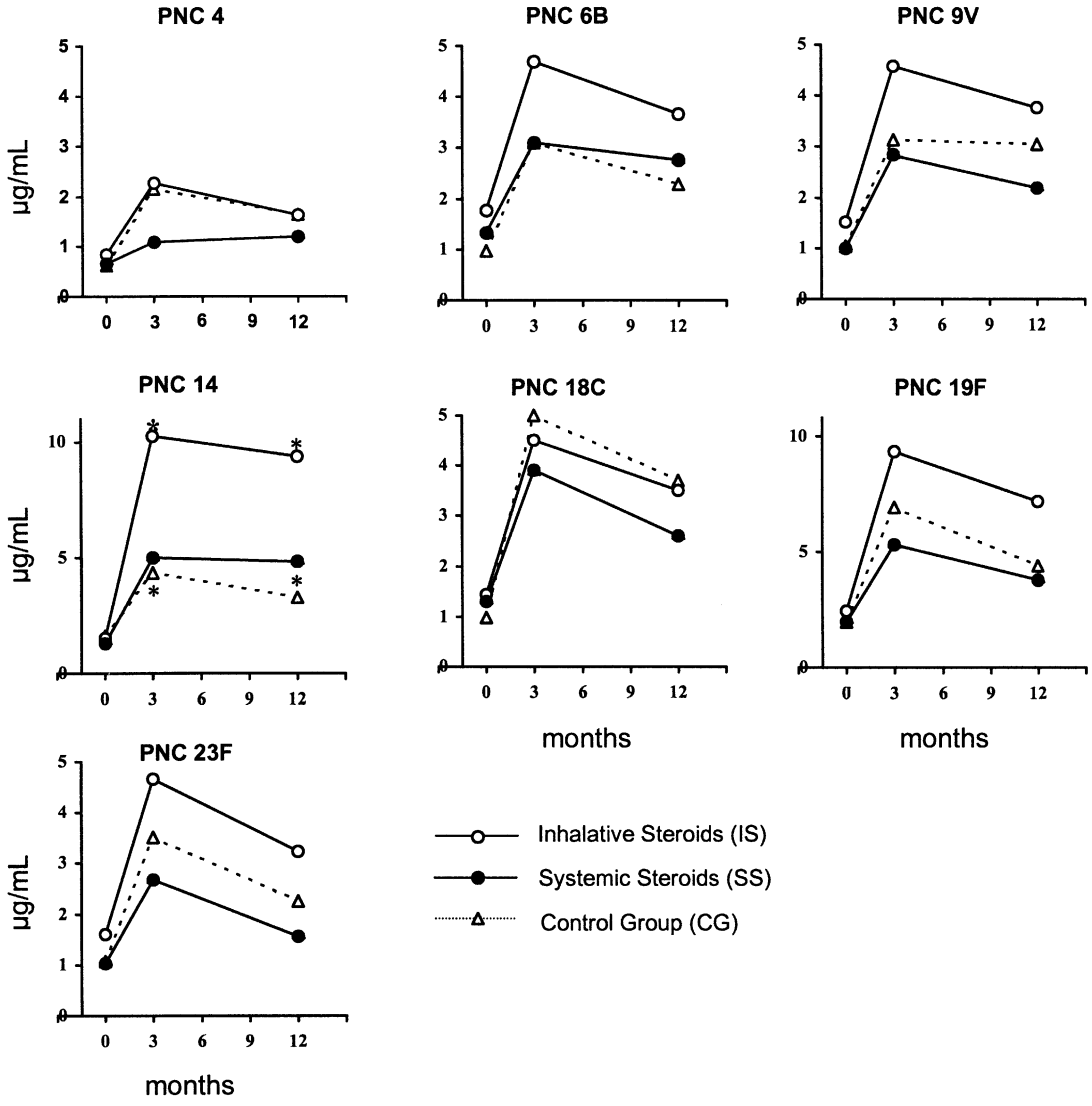

Figure 1 Geometric mean concentrations (µg/ml) of serotype-specific IgG antibodies to pneumococcal capsular polysaccharides for seven serotypes (µg/ml). Measurements were performed before, 3 and 12 months after vaccination with 23-valent PPV. Mean pneumococcal capsule-specific IgG did increase significantly after 3 and 12 months in all PNC and all groups (CG,JS,SS). Note that PNC 14 and 19F concentrations were higher (different scale). IS had higher mean PNC 14 concentration when compared to CG (3 and 12 months, $P<0.05$). Significant differences are marked with "*" ($P<0.05$).

of the vaccine (serotypes 2, 6B, 8, 12F, 14, 19A, 19F, 25) in elderly subjects (>65 years) and younger healthy adults (<35 years).[15] Both groups had comparable results as regards pre-vaccination titers and mean fold-rises of anti-pneumococcal antibodies 16 months after vaccination. However, in the elderly, serotype 8 was significantly lower and serotype 14 induced the highest antibody response showing an important variability of the immunogenity of the tested antibodies in the vaccine. A meta analysis showed high "antigenicity" of the serotypes 8, 3, 9 and 14 in normal subjects after vaccination.[22] Further serotypes 33F, 15B, 9N and 20 were reported to be the most immunogenic in elderly adults.[23] In our study we found PNC 14 and 19F to produce the highest immune response. PNC14 was significantly more immunogenic in COPD patients on inhalative steroids when compared to controls. A possible explanation for this finding can be co-morbidities or co-medications in the control group. The daily use of an angioten-sin-converting-enzyme inhibitor for e.g. has been described to be an independent risk factor for poor antibody response to the pneumococcal vaccine.[15] In the present study we did not control the patient groups for non-immunomodulating medication.

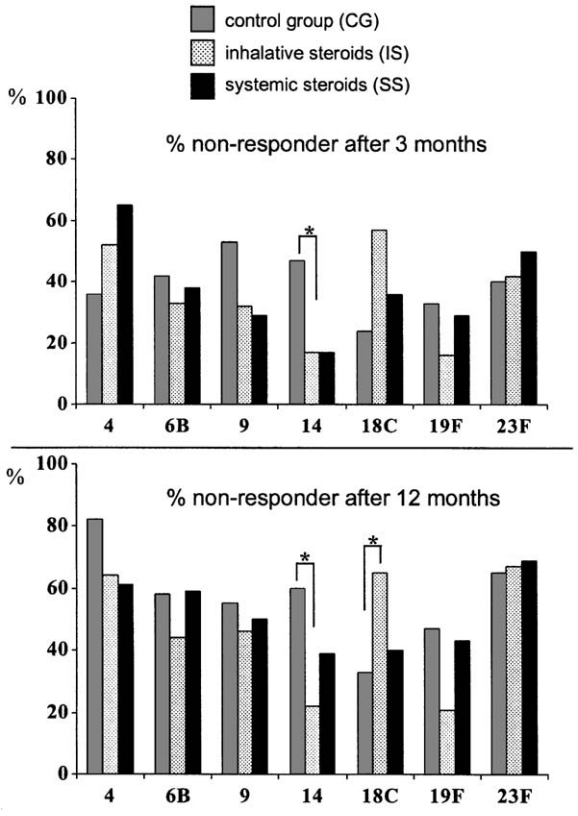

Figure 2 Percentages of non-responders (*Y* axis) 3 months (upper figure) and 12 months (lower figure) after vaccination with the 23-PPV. *X* axis = serotypes tested in the three groups. Only patients that completed the study are shown (CG $n=24$, IS $n=29$, SS $n=18$). Patients with high pre-vaccination antibody levels to the respective tested serotypes ($>4\,\mu g/ml$) were excluded (PNC 4 CG/SS/IS $n=1/1/4$, 6B $n=5/4/11$, 9 $n=5/3/7$, 14 $n=4/7/5$, 18C $n=6/6/6$, 19F $n=6/10/10$, 23F $n=3/4/5$). Non-responders were defined when after vaccination baseline polysaccharide specific AB-concentration did neither increase two-fold nor the increment reached at least $\geqslant 1\,\mu g/ml$. "*" indicates significant differences ($P<0.05$) between the groups.

Table 2 Number and percentage of subjects who responded to either <2 or >5 of the tested 7 serotypes.

	CG N(%)	IS N(%)	SS N(%)
3 m			
Response to <2PNC	4(14)	5(17)	6(21)
Response to >5PNC	9(32)	10(33)	6(21)
12 m			
Response to <2PNC	8(33)	8(28)	5(28)
Response to >5PNC	4(17)	8(28)	3(17)

Response is defined as a twofold increase of baseline antibody levels or an increase of at least $1\,\mu g/ml$. Differences between groups were not significant ($P>0.05$).

As regards mean antibody concentrations we found no significant differences between COPD patients on systemic steroids and the other groups. Despite this in 5 of the 7 tested serotypes (PNC 4, 9 V, 18C, 19F, 23F) mean antibody concentrations of the systemic steroid group were lower compared to both other groups. We cannot discharge that if the dose or duration of the systemic steroid treatment would have been higher this observation might have reached significance.

Rubins et al. reported that a significant proportion of the elderly ($\sim$20%) were "poor responders" defined as subjects with a less than a twofold increase of baseline titers in two of seven serotypes. The percentage of elderly with an >2 fold antibody titer ranged between 70% (PNC 2 after 3 months) and 10% (PNC 25 after 16 months). In the present study the rate of non-responders to the tested serotypes ranged between 16–65% after 3 months and 21–82% after one year. Further, approximately 20% of patients did respond to less than two of seven serotypes in the vaccine after 3 months. This rate increased to 30% after 12 months. For the evaluation of pneumococcal vaccination, it is important to consider the pre-immunization status and age in the studied patients. In the present study, there was no significant difference in the age distribution of groups nor an increased number of non-responders in the elderly patients >75 years.

The results of previous studies addressing the immune response after vaccination of COPD patients are controversial. In an early study Landesman et al. reported that antibody levels after vaccination with the 14-valent PPV induced antibody concentrations in elderly COPD patients for nine of 12 serotypes and concluded that antibody titers for at least one year after vaccination were sufficient.[17] In COPD patients antibody titers after vaccination were shown to decline faster when compared with healthy controls.[16] Neither of this studies did stratify patients according to their steroid medication. In the present study therapy with systemic steroids did neither reduce the immunogenity of the vaccine nor increase the rate of non-responders.

We recognize some possible limitations of the present study. First we did not test the opsonphagocytotic activity of the antibodies induced by the vaccine. Differences in the avidity of antibodies induced by 23-PPV have been strongly

suggested.[14,24] Despite this it is known that recurrence of pneumococcal pneumonia is related to a poor antibody response after vaccination with the 23-valent PPV as described by Hedlund et al.[25] Therefore the concentration of IgG specific antibodies reflects the level of protection against pneumococcal pneumonia.

A further possible limitation is our suggested definition of non-responders. We created this specification to identify those patients who respond poorly to the vaccine. It is frequently stated but not documented that a normal antibody response to vaccination should include at least a twofold increase in antibody titers. Former studies regarding the antibody response to pneumococcal vaccination have used this definition.[15,22] We choose the level of 1 μg/ml absolute increase in antibody concentration because some pre-vaccination antibody titers were very low and a twofold increase in antibody titers would not have reflected a sufficient (protective) immune response. Therefore we also did not include subjects with high pre-vaccine antibody levels (>4 μg/ml) in the analysis of non-responders as those patients have probably a sufficient AB-concentration to protect them against invasive pneumococcal disease. Although a protective antibody concentration has not been established most studies show a mean antibody response after vaccination of 2–10 μg/ml in healthy volunteers.[14,15,22]

We conclude that vaccination of COPD patients with or without a systemic steroid medication the 23-valent PPV is safe and produces a significant increase of mean antibody concentration. Therefore we agree with current vaccine recommendations, specially as recent data suggest that patients clearly benefit from a co-vaccination of the pneumococcal and influenza vaccine.[11] This measure has been proven to be safe in patients with chronic respiratory conditions.[26] But an important subset of our patients regardless of their steroid medication or concomitant COPD failed to respond sufficiently to the vaccine. If antibody concentrations neither double nor increase towards at least 1 μg/ml, or even decrease to pre-immunization levels after one year, a protection against pneumococcal disease is improbable regardless of the functionality of the antibodies. We are in need for new pneumococcal vaccines that provide longer-term protection and efficacy in high-risk groups as COPD patients. Conjugate vaccines have been proven to induce a sufficient antibody response in patients who are genetically incapable of responding to polysaccharide vaccines and might be an alternative.[27,28] There are also promising results in mice and in humans of a vaccine derived from pneumococcal surface proteins.[29,30]

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