Concordance between CRP and SAA in familial Mediterranean fever during attack-free period: A study of 218 patients

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Concordance between CRP and SAA in familial Mediterranean fever at steady state: a study of 218 patients

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Abstract

Introduction: monitoring SAA level in attack-free FMF patients is recommended in order to adjust colchicine dose, lower the SAA level under 10 mg/L and minimize the risk of AA amyloidosis. In countries where this test is not available, C-reactive protein (CRP), another acute phase reactant, is used instead. However, CRP is low and SAA is increased in some patients and vice versa.

Objectives: to determine the threshold of CRP corresponding to SAA < 10 mg/L in patients with FMF and to assess their concordance at the patient level.

Patients and Methods: consecutive FMF patients in attack-free period and no other cause of intermittent inflammation including infections were recruited during their regular visits in the French reference center for FMF. Demographic and genetic data were recorded, CRP and SAA were tested simultaneously. The threshold value of CRP corresponding to 10 mg/L for SAA was determined and the concordance between the two markers was assessed with Cohen's kappa index.

Results: 399 samples were obtained from 218 patients, mean age of 27 years (33% under 18 years old), 55% of female, from Sephardic Jewish origin in 71%, Arabic, Armenians and Turkish in 10%, 6% and 6% respectively. *MEFV* mutation was M694V homozygous or compound heterozygous in 52%, and simple heterozygous in 18%. Six patients had AA amyloidosis. The appropriate CRP threshold was found to be 5 mg/L in children and 8.75 mg/L in adults. Global agreement with SAA < 10 mg/L was 84% [95% confidence interval: 82 to 86%], leading to a kappa index at 0.62 [95% confidence interval: 0.57 to 0.68].

Conclusion: CRP < 5 mg/L in FMF children or 8.75 mg/L in FMF adults during attack-free periods might be a convenient substitute to guide therapeutic decisions when SAA is unavailable.

Key words: C reactive protein; serum amyloid A; familial Mediterranean fever

Introduction

Familial Mediterranean fever (FMF) is a genetic autosomal recessive disease associated with mutations in the *MEFV* gene, and resulting in recurrent inflammatory attacks starting in childhood. As with other chronic inflammatory diseases, a major complication is AA amyloidosis, a systemic disease involving the kidneys and leading to renal failure. Serum amyloid A (SAA) is a circulating acute-phase reactant. Its level increases substantially during inflammatory episodes of FMF and usually decreases in attack-free periods. It has been suggested that SAA level under 10 mg/L in attack-free period is associated with a decreased risk of AA amyloidosis in a series of patients with AA amyloidosis of various causes among which 5% were FMF [1]. Then, this threshold has been considered as a target for an efficient maintenance anti-inflammatory treatment. In FMF, colchicine dose should thus be adjusted in order to reach this target level. C reactive protein (CRP) is an acute phase reactant used in many countries where SAA is not routinely available. Discordances between CRP and SAA have been described in patients with FMF [2]. Our objectives were to establish a threshold value for CRP corresponding the most closely to a SAA value of 10 mg/L and to assess the concordance of these two inflammatory markers at the patient level during attack-free periods.

Patients and methods

Patients were recruited during outpatient visits at the adult and pediatric reference centers for FMF between July 2008 and January 2013. The diagnosis of FMF was based on expert opinion including the exclusion of other periodic fever syndromes; all adult patients met the FMF Tel Hashomer criteria [3] and pediatric patients met the FMF Yalçinkaya criteria [4]. Inclusion criteria were: patient in attack-free period (at least 15 days after the last attack); no other inflammatory disease or concomitant disorder that would enhance inflammation including infections; indication of blood tests for the regular follow-up of the disease. CRP

and SAA are routinely measured in these centers. Patient characteristics were recorded: age, sex, ethnic origin, genetic data, long-term treatment and its dose, presence of AA amyloidosis i.e proven AA amyloidosis by biopsy after Red Congo stain and immunohistochemical technique using anti-SAA antibody. To describe age and dose of long-term treatment, we took the data at the first visit recorded in the study. In order to test the reproducibility, some patients had several samples during the study period if they completed the inclusion criteria at each visit intervalle max entre 1er et dernier prélèvement pas patient = préciser maladies pédiatrique restés pédiatriques. The study complies with the French regulations regarding clinical research and respects the Declaration of Helsinki.

High sensitive (Hs)CRP and SAA assays were centralized in a single laboratory. HsCRP and SAA were measured by immunonephelometry on an IMMAGE® analyzer (Beckman-Coulter, Villepinte, France). The sensitivities of the assays were 0.11 mg/L for HsCRP and 3 mg/L for SAA. The CV intra-series and inter-series were 4.8% and 5.1% for SAA and 4.7% and 6.4% for HsCRP respectively.

The threshold value of CRP was determined to obtain as many upward discordances (CRP > threshold while SAA < 10 mg/L) than downward discordances (CRP < threshold while SAA ≥ 10 mg/L). Several patients had more than one blood test over the study period. To avoid bias due to the overinfluence of these patients, only one sample per patients could be analyzed simultaneously. We used within patient resampling to produce unbiased statistics [5]: 500 lots were made up by randomly drawing one blood test per patient, then results of all lots were combined to produce the final statistics. We looked for an interaction between age and the CRP threshold in order to derive different thresholds in children and adults if appropriate.

Once the CRP threshold was defined, we first assessed if the concordance between CRP < threshold and SAA < 10 mg/L was stable within patients who had more than one sample at the same colchicine dose (0 mg/d, ≤ 1 mg/d, ≤ 2 mg/d, > 2 mg/d). We then assessed the

overall concordance between CRP < threshold and SAA < 10 mg/L across all patients with Cohen's kappa index. Finally, we used logistic regression to assess if the concordance between CRP and SAA differed in several prespecified subgroups: with or without colchicine, M694V homozygosity or not, Sephardic origins or not. Further details are provided in the appendix.

All statistical analyses were performed with Stata 9.2 (StataCorp, College Station, TX).

Results

A total of 399 blood samples were taken from 218 patients during the study period; 94 patients had more than one sample drawn (median 2 samples; range from 2 to 42 samples). There were 252 clusters of samples from the same patient at the same colchicine dose; 75 clusters contained more than one sample (median 2 samples; range from 2 to 42 samples). Patient clinical and demographic characteristics are displayed in Table 1.

The CRP threshold producing as much upward and downward discordances with SAA < 10 mg/L was 7.5 mg/L in the whole population. However, there was a significant interaction with age and we therefore derived two different threshold for pediatric (< 18-year old) and adult (≥ 18-year old) patients. The resulting CRP threshold was 5 mg/L in pediatric patients and 8.75 mg/L in adult patients.

When two samples were randomly selected within each clusters from the same patient at the same colchicine dose, CRP and SAA were both over or both under their threshold in 81% of cases [95% confidence interval (CI): 79 to 84]. When one sample was randomly selected within each cluster, agreement between CRP and SAA was 84% [95% CI: 82 to 86] at the population level with a kappa index at 0.62 [95% CI: 0.57 to 0.68].

In multivariate logistic regression, a better concordance of CRP and SAA was associated with being of Sephardic origin (beta coefficient 0.79 [95% CI: 0.41 to 1.18]) and not being M694V

homozygous (beta coefficient 0.58 [95% CI: 0.18 to 0.98]). However, the magnitude of the difference the whole population and these subgroups was small, with an overall agreement of 85% [95% CI: 82 to 88] in patients of Sephardic origin, 87% [95% CI: 84 to 90] in those not M694V homozygous and 91% [95% CI: 88 to 94] in those of Sephardic origin and not M694V homozygous.

Discussion

The purpose of our study was to determine a CRP threshold corresponding to SAA < 10 mg/L and to assess their concordance at the patient level. We found that CRP < 5 mg/L in pediatric patients and < 8.75 mg/L in adult patients was in good agreement with SAA < 10 mg/L.

Various acute phase reactants have been tested to predict subclinical inflammation in FMF, such as SAA, CRP, ESR (erythrocyte sedimentation rate), ferritin, and fibrinogen. SAA appears to be the best marker of subclinical inflammation in FMF patients since it remains elevated in 30 to 100% of FMF patients in attack-free period [6,7,8], is more elevated in patients with AA amyloidosis, and decreases dramatically after increasing colchicine dose [6]. Several studies showed that other inflammatory markers were less sensitive since they were within normal ranges or slightly increased at steady state [6,9]. In fact, a recent systematic review concluded that no acute phase reactant has shown superiority over the other for the monitoring of FMF [10] and therapeutic decision should include clinical and biochemical data.

AA amyloidosis is due to the deposition of insoluble abnormal fibrils derived from the acute phase reactant SAA protein. It is the most severe complication of FMF, preferentially affecting the kidneys. The objective of long-term colchicine treatment in FMF is to prevent inflammatory attacks but also to prevent the occurrence of AA amyloidosis [11]. In a cohort of 374 patients with AA amyloidosis from various chronic inflammatory disorders, it has been

shown that SAA < 10 mg/L was associated with lower mortality and better renal prognosis [1]. From this study, it has been extrapolated that low levels of SAA at steady state in FMF should be a target to guide the colchicine dose [2]. This extrapolation should be viewed with caution because of the large dispersion of SAA results in the original study and because only 20 patients of this study had FMF [1]. Moreover, even if SAA > 10 mg/L is indeed a risk marker in patients with chronic inflammatory conditions at the population level, it does not automatically mean that lowering SAA will translate into better prognosis in individual patients. We kept the target threshold of 10 mg/L for SAA in our study. However, the evidence is too weak and indirect so far to take this marker as an established surrogate for the risk of long term amyloidosis in FMF patients. Due to the lack of validated gold standard to predict AA amyloidosis occurrence in FMF, we did not assess the prognostic value of CRP in this population (sensibility and specificity), but only its concordance with SAA at the appropriate thresholds.

In France as in some other countries, SAA is not a routine test and is not covered by health care insurance. Instead, CRP is used for monitoring the inflammatory activity in FMF in medical centers other than reference centers for FMF. However, it has been shown that in some FMF patients, CRP at steady-state can be normal with a concurrently elevated SAA [2]. In the retrospective study published by Berkun et al. [2], the objective was to evaluate the role of elevated SAA in diagnostic and in treatment decisions. They noticed that the CRP was < 0.5 mg/L in 34% of patients with SAA > 6 mg/L (corresponding to 23 patients). This result suggested that normal CRP was not able to guide therapeutic decisions. However, our study suggests that high sensitive CRP could be a liable substitute in countries without access to SAA dosage, provided that an appropriate threshold is used. Medical teams who have SAA as a routine test should use this marker for therapeutic guidance. However, those who do not have SAA dosage routinely can use high sensitive CRP as a substitute for the follow-up.

Conclusion

CRP < 5 mg/L in children and < 8.75 mg/L in adults appear to be in good agreement with SAA < 10 mg/L in attack-free FMF patients, especially if they have *MEFV* mutation other than M694V homozygous or are currently taking colchicine. This threshold could guide physicians in treatment decisions when SAA dosage is not available.

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