Correspondence

Acetylator phenotype prevalence in HIV-infected patients without previous trimethoprim-sulfamethoxazole hypersensitivity

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Summary – This trial was conducted to study the frequency of the slow acetylator phenotype in asymptomatic HIV patients having no previous reaction to sulfa-drugs, and to compare this frequency with the frequency found in healthy controls. Results show that HIV alone is not capable of modifying the acylating phenotype; the prevalence of slow acetylating phenotype is the same in immune competent subjects and HIV-positive patients. It is more common in HIV-positive patients with a CD4⁺ lymphocyte count of less than 200 mm⁻³.

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The increased frequency of hypersensitivity reactions to sulfa-drugs among HIV-infected patients is a well known problem, but its pathogenesis is still obscure. Some studies have suggested a toxic hypothesis related to the metabolism of sulfa-drugs [1-4]. In fact sulfamethoxazole is metabolized predominantly by hepatic acetylation to a stable N4-acetylsulfamethoxazole, and by cytochrome P450 to 5-hydroxysulfamethoxazole or to a highly reactive hydroxylamine metabolite. This pattern of metabolism is typical of subjects with ‘fast acetylator phenotype’. The pattern of the ‘slow acetylator phenotype’ is a shortage of transacetylase, in this case the sulfa-drugs are metabolized by cytochrome P450 with the overproduction of hydroxylamine [5, 6]. Several hydroxylamine metabolites may have cytotoxic effects or may activate the immune system. In a study on HIV-infected patients, Carr et al. found a 42% prevalence of the slow acetylator phenotype in those patients who had never shown hypersensitivity reactions to sulfa-drugs vs. 94% in those who had such reactions [7]. The aim of our study was to evaluate whether HIV infection alone or related to the degree of immune deficiency influences the prevalence of the slow acetylator phenotype in HIV-positive patients without ongoing acute infections, who had never had hypersensitivity reactions to sulfa-drugs.

Two groups of patients were evaluated for acetylator phenotype: a group of HIV-infected patients without previous hypersensitivity to sulfa-drugs, and a group of healthy subjects as a control. Patients with acute infections were excluded since these patients show a prevalence of slow acetylator phenotype similar to that observed in patients with hypersensitivity reactions [8]. A wash-out period of at least a week was mandatory for all HIV patients who were receiving trimethoprim/sulfamethoxazole for Pneumocystis carinii pneumonia prophylaxis before entering the study (i.e., those with a CD4⁺ count of less than 200 mm⁻³); a pentamidine aerosol was administrated to those patients. The concomitant use of drugs that might interfere with the P450 cytochrome pathway (i.e., cimetidine, rifampicin, azoles) was the main exclusion condition.

The data investigated in our study had never before been tested, therefore it was difficult to define in statistical terms the sample size required. Patients were not stratified by sex or age since no relationship between these factors and the acetylator phenotype has been demonstrated. The chi-square test was performed for comparison of the groups.

Since the patients in our study had never experienced hypersensitivity reactions to sulfa-drugs, we could employ a method using sulfametazine (SMZ) for the characterization of the acetylator phenotype. The acetylation phenotype was established using the modified Bratton-Marshall procedure [9] for the evaluation of unacetylated and total (acetylated and unacetylated)
Table I. Prevalence of slow and fast acetylators in health and HIV-infected patients.

<table>
<thead>
<tr>
<th></th>
<th>Slow acetylator</th>
<th>Fast acetylator</th>
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<tbody>
<tr>
<td>Healthy subjects</td>
<td>24 (60%)</td>
<td>16 (40%)</td>
</tr>
<tr>
<td>HIV-infected</td>
<td>50 (61%)</td>
<td>32 (39%)</td>
</tr>
<tr>
<td>Patients with CD4+ &gt; 200 mm⁻³</td>
<td>21 (50%)</td>
<td>21 (50%)</td>
</tr>
<tr>
<td>Patients with CD4+ &lt; 200 mm⁻³</td>
<td>29 (72.5%)</td>
<td>11 (27.5%)</td>
</tr>
</tbody>
</table>

sulfamethazine excretions in urine. Briefly, a single
dose of 10 mg/kg of SMZ was administered orally to
the fasting patient. Urine volumes were measured four
hours later and two quantities each of 10 mL were stored
at −20°C. Later on, 7 mL of urine was added to 2.1 mL
of 20% trichloroacetic acid (TCA) and mixed; this pro-
cedure was followed by centrifugation at 1,500 rpm for
10 min. Then a quantity of 1 mL was used for the deter-
mination of unacetylated sulfamethazine by adding
8.5 mL of water and 500 g/L of HCL (4N); a second
quantity of 200 mL was used for the determination of
total SMZ by adding 0.2 mL of 4 N HCL. Each tube
was then immersed in a boiling water for 1 h. After
this step the procedure was identical for both deter-
minations. Freshly prepared 0.1% sodium nitrite was
added to each tube and mixed; then 0.5% ammonium
sulphamate was added and mixed. Finally, 1 mL of
0.05% N-1 naphthylethylendiamine dihydrochloride
was added and the mixture was allowed to stand for
10 min for color development. Absorbance reading was
determined at 540 nm in a Gilford 300 N recording
spectrophotometer against water as a blank with the use
of an automatic sampling cuvette. Fast acetylators were
those with free sulphametazine concentrations greater
than 75%.

Eighty-two HIV positive patients, 56 males and
26 females with a mean age of 34 years, were enrolled:
50 IVDU, 23 heterosexuals, eight homosexuals and one
transfused. Forty patients (20 males and 20 females
with a mean age 32 years) were selected for the control
group. The slow acetylator phenotype was observed in
50 out of 82 patients with HIV infection (61%) and in
24 out of 40 healthy subjects (60%) as reported in
table I; this was without statistical significance.

We then stratified the HIV-infected group by immune
depression between those with a CD4⁺ count higher or
lower than 200 mm⁻³. Out of 40 patients with CD4⁺
counts less than 200 mm⁻³ (72.5%), and 21 out of
the 42 subjects with CD4⁺ cell counts greater than
200 (50%) were slow acetylators (table I). This differ-
ence did not reach statistical significance (P = 0.06),
but showed a certain relationship between the two
parameters.

Our results allowed us to conclude that the acetylator
phenotype, though playing an important role, is proba-
bly not the only cause of hypersensitivity reactions in
HIV subjects. Indeed, among asymptomatic patients,
the frequency of slow and fast acetylators was the same
as in healthy volunteers. We cannot also draw any con-
clusion regarding a possible concomitant role of the
level of immune depression, although it seems that in
the most immune deficient subjects the percentage of
slow acetylators was higher, though this was not statis-
tically significant.

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REFERENCES

1 Carr A, Swason C, Penny R, Cooper DA. Clinical and labo-
   ratory markers of hypersensibility to trimethoprim-
   sulfamethoxazole in patients with Pneumocystis carinii
2 Van der Ven AJ, Koopmans PP, Vree TB, Van der Men JW.
   Adverse reactions to cotrimoxazole in HIV infection. Lancet
3 Koopmans PP, Van der Ven AJ, Vree TB, Van der Men JWM.
   Pathogenesis of hypersensitivity reactions to drugs in patients
   with HIV infection – allergic or toxic? AIDS 1995 ; 9 :
   217-22.
4 Mathelier-Fusade P, Leynadier F. Intolérance aux sulfamides
   chez les sujets infectes per Le FHV. Press Med 1993 ; 22 :
   1363-5.
5 Lee BL, Delahunty T, Safrin F. The hydroxilamine of sul-
   famethoxazole and adverse reactions in patients with acquired
   184-90.
6 Van der Ven AI, Koopmans PP, Vree TB, Van der Meen JW.
   Adverse reactions to cotrimoxazole in HIV infection. Lancet
7 Carr A, Gross AS, Hoskins JM, Penny R, Cooper DA. Acety-
   lation phenotype and coetaneous hypersensitivity to trimetho-
   prim-sulfamethoxazole in HIV-infected patients. AIDS 1994 :
   8 : 333-7.
8 Lee BL, Wong D, Benovits NL, Sullam PM. Altered pat-
   terns of drug metabolism in patients with acquired immunode-
   ficency syndrome. Clin Pharmacol Ther 1993 ; 53 :
   529-35.
9 Olson W, Miceli J, Weber W. Dose-dependent changes in sul-
   phametazine kinetics in rapid and slow isoniazid acetylators.