RISK ASSESSMENT OF WORKERS EXPOSED TO FUMES OF BITUMEN: GENOTOXIC EFFECTS AND URINARY METABOLITES OF NAPHTHALENE, PHENANTHRENE, AND PYRENE

Marczynski B1, Raufi-Heimsoeth M1, Käfferlein HU1, Förster K2, Spickenheuer A1, Pesch B1, Breuer D3, Welge P1, Angerer J3, Bramer R1, Hahn J-U3, Mensing T1, Brünig T1

1 BGFA - Forschungsinstitut für Arbeitsmedizin der Deutschen Gesetzlichen Unfallversicherung, Institut der Ruhr-Universität Bochum, Bochum, Germany; 2 Institut und Poliklinik für Arbeits-, Sozial- und Umweltmedizin, Universität Erlangen-Nürnberg, Erlangen, Germany; 3 Institut für Arbeitsschutz der Deutschen Gesetzlichen Unfallversicherung (BGIA), St. Augustin, Germany

INTRODUCTION

BACKGROUND
- Bitumen is a complex mixture of hydrocarbons consisting of both aliphatic and aromatic compounds, e.g., polycyclic aromatic hydrocarbons (PAH).
- There is inadequate evidence of carcinogenicity of bitumen in humans. Previous studies cannot be interpreted due to major qualitative or quantitative limitations.
- Overall, bitumen is labelled as „high priority substance“ for future evaluation by the International Agency for Research on Cancer.

OBJECTIVE
- Determination of genotoxic properties of exposure to fumes of bitumen in humans.
- Generation of dose-response relationships which can help to establish health-based threshold limit values for exposed workers.

METHODS

STUDY DESIGN
- Cross-sectional and cross-shift study design.

STUDY SUBJECTS
- 202 bitumen-exposed mastic asphalt workers and 55 construction workers without exposure to bitumen were examined (Table 1).

Table 1: Characteristics of the study groups

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Reference group</th>
<th>Bitumen-exposed workers</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n = 55</td>
<td>n = 202</td>
</tr>
<tr>
<td>Age (years; median, range)</td>
<td>37 (19-61)</td>
<td>40 (17-63)</td>
</tr>
<tr>
<td>Current smoking (n, %)</td>
<td>23 (41.8)</td>
<td>132 (65.7)</td>
</tr>
<tr>
<td>German nationality (n, %)</td>
<td>48 (87.3)</td>
<td>136 (67.7)</td>
</tr>
<tr>
<td>Duration of exposure in the current company (years; median, interquartile range)</td>
<td>6.5 (3-10)</td>
<td>8 (3-14)</td>
</tr>
<tr>
<td>Exposure to bitumen fumes and aerosols (mg/m³; median, interquartile range)</td>
<td>-</td>
<td>3.7 (1.7-7.1)</td>
</tr>
</tbody>
</table>

ANALYTICAL PARAMETERS
- Exposure was assessed using urinary 1-hydroxypyrene (1-OHP), the sum of 1-, 2-, and 3-hydroxypyrene (OHPH) and 1- and 2-naphthol (NOL). Genotoxic effects in white blood cells were determined with non-specific DNA adduct levels of 8-oxo-7,8-dihydro-2′-deoxyguanosine (8-oxoGua) and the formation of DNA strand breaks as well as alkali-sensible sites.

STATISTICAL MODEL
- Linear mixed model with log-transformed study variables and control of confounders (SAS Software).
- Implemented in the model are independent fixed factors (time of measurements, smoking status, ethnicity) and a random factor (participants).
- Age is included in the model as a continuous independent variable.
- Results presented are means adjusted for the set of potential confounders and F-tests (P-values) for the exposure effect.

RESULTS

Personal air monitoring of exposed persons revealed a median shift concentration of fumes of bitumen of 3.7 mg/m³. Concentrations of fumes of bitumen showed a moderate association with 1- and 2-naphthol (r = 0.18, P = 0.01), 1-, 2-, 9-, and 4-hydroxypyrrenthrene (r = 0.36, P = 0.001) and 1-hydroxypyrene (r = 0.25, P = 0.001) after shift (Table 2). Significantly increased 8-oxoGua adduct levels were observed after shift in exposed and non-exposed persons (P = 0.0001, Figure 1). However, decreased DNA strand break frequencies were observed after shift in both groups (P = 0.05, Fig. 1). Significantly higher levels of DNA damage were found in exposed workers compared with reference subjects (Figure 1). DNA strand break frequencies after shift were only moderately associated with NOL (r = 0.20, P = 0.01) and 1-OHP (r = 0.19, P = 0.01) in urine after shift at the group level (Table 2).

CONCLUSIONS
- Exposure to fumes of bitumen results in excretion of 1-OHP, five isomers of OHPH and two isomers of NOL in urine after the shift.
- The excretion of urinary metabolites was dependent on concentrations of fumes of bitumen. Therefore, 1-OHP, the sum of five OHPH and the sum of NOL are suitable biomarkers to assess exposure to fumes of bitumen.
- Exposed mastic asphalt workers had higher levels of oxidative DNA damage (8-oxo-dGuo) and higher frequencies of DNA strand breaks in both pre shift and post shift blood samples compared to non-exposed individuals. Consequently these workers show increased biomarkers of genotoxicity.
- Increases in oxidative DNA damage during the shift were of statistical significance but did not depend on external exposure. Therefore, 8-oxo-dGuo is capable to assess oxidative DNA damage but it is not specific of exposure to fumes of bitumen.
- Decreases of DNA strand breaks (not significant) were observed after shift in both study groups. Additionally, associations between DNA strand break frequency and PAH metabolites of pyrene and naphthalene in urine after shift were observed.
- Due to an only weak association between 1-OHP and DNA strand breaks the reasons for increased DNA damage in workers exposed to fumes of bitumen remain unclear.

Table 2: Spearman rank correlations between exposure to fumes of bitumen, urinary metabolites, 8-oxodGuo and Olive tail moment

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Pre shift</th>
<th>Post shift</th>
<th>Shift difference</th>
</tr>
</thead>
<tbody>
<tr>
<td>1-OHP (ng/g creatinine)</td>
<td>185 0.40 0.06</td>
<td>190 0.45 0.06</td>
<td>189 0.40 0.04</td>
</tr>
<tr>
<td>8-OHdG (ng/L)</td>
<td>182 2.75 0.00</td>
<td>186 2.80 0.01</td>
<td>184 2.77 0.00</td>
</tr>
<tr>
<td>8-oxodGuo/106dGuo</td>
<td>0.01 0.12 0.01</td>
<td>0.06 0.01 0.05</td>
<td>0.03 0.06 0.01</td>
</tr>
<tr>
<td>Olive tail moment (median)</td>
<td>8-oxodGuo adduct level in white blood cells</td>
<td>DNA strand break frequencies in lymphocytes</td>
<td></td>
</tr>
</tbody>
</table>