Khat abuse as risk factor for development of psychotic symptoms. A feasibility study for further genetic epidemiological studies on addiction of khat

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Methods

In our study, urine samples were collected randomly from khat chewers living in the Jimma zone of the GGFRC. During sample collection, samples were coded to facilitate ease of identification and the samples were transported to the study laboratory by a cold chain system preserving the integrity of the sample. After they had reached the study laboratory, samples were stored in refrigerators (2-8°C). To test the feasibility of a further genetic project, we collected blood samples as well. Blood samples were transported to the JU and stored in refrigerators (~80 °C) The average time from sample collection in the GGFRC until arrival at JU was 299.24 min. (5 h) (min. 95 min., max. 495 min). For DNA extraction a new method was established at the JU.

Results

Table 1. Laboratory Analyses

<table>
<thead>
<tr>
<th>Sample Types</th>
<th>Values</th>
<th>Interpretation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>~1.1 mg/ml</td>
<td>Test positive</td>
</tr>
<tr>
<td>Negative</td>
<td>~0.8 mg/ml</td>
<td>Test negative</td>
</tr>
</tbody>
</table>

Figure 1. Chromatogram of norephedrine (NE) extracted from Recatol capsules

Figure 2. Chromatogram of norephedrine (NE) extracted from urine sample

HPLC analyses of urine samples for norephedrine: As reference, we used standards like Recatol, which contains about 50 mg norephedrine per capsule. Urine sample preparations followed by using solid-phase extraction (SPE). Norephedrine elutes at an average retention time of 5.7 min with peak asymmetry As of 1.1 and retention factor k’ of 4.2. Method validation results indicated the fitness-for-use of the applied HPLC method. The 95% CI for the regression slope was 1.925 (95% CI: 1.796 to 2.053) and y-intercept was 3.476 (95% CI: 25.960 to 32.913) together with r2 value of 0.999 and ANDA F value of 21548 proved a strong positive linear relationship. The limit of detection (LOD) for this method was 0.04 μg/ml and limit of quantification 0.14 μg/ml. This shows that the method is suited for both qualitative and quantitative analysis.

Results laboratory analyses

Future plans

Our project can be seen as a pilot and feasibility study to prepare a comprehensive population-based genetico-epidemiological study on various gene-environment interactions that should be carried out in the very near future. The infrastructure of GGFRC offers us a unique opportunity to build a collective of multiple thousand people in a short period of time and to perform genetic studies. The project has not yet been taken in Africa in this form so far. The extensive epidemiological registration of a population of 50,000 people, the stable population structure, and the quite stable environment, such as the urban and rural way of life with all its characteristics of an African country provide ideal conditions for this. The population is ideally suited to study the impact of polygenic risk profiles of various psychiatric disorders on behavioral traits and their interaction with environment.