Contents of Nitrates and Nitrites in some Greek and Imported Cheeses

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ABSTRACT

Fifty-four samples of various types of Greek cheese and 69 samples of imported cheese were analyzed for nitrates and nitrites. Domestic Gruyère from cow’s milk and processed cheese contained 1-10 and 35-104 mg of NO₃⁻ per kg, respectively. All other domestic cheeses were either free of or contained nitrates less than 1 mg/kg. Imported cheese contained nitrates as follows: Edam 1-11, Gouda 1-24, Teleme 13-81, Goudasana 27, processed cheese 0-8, cheese spread 19-30, bacon cheese 24-508 and others <4 mg/kg. All types of domestic and imported cheese were either negative for nitrites or contained <1 mg/kg.

Late blowing and gassy defects in cheese are usually caused by species of Clostridium, the spores of which survive milk pasteurization and cause butyric acid fermentation in cheese starting a few weeks after manufacture (9,11). Methods to control these defects include addition of nitrates to cheese milk, improvement of hygienic quality of milk, adequate salting of cheese, vigorous acid formation in the curd, bactofugation, use of hydrogen peroxide, changes in cheesemaking procedure, use of nisin-producing cultures and lysozyme (1,4,9,11,17,28).

Of the methods mentioned, the most convenient to apply in industry and the most successful in preventing late blowing of cheese seems to be addition of potassium or sodium nitrate to the cheese milk (12).

Nitrates are reduced to nitrites, which prevent growth of the Clostridium species (22). This is attributed to the activity of the milk enzyme xanthine oxidase (10) but other factors may also bring about nitrate reduction independently or in combination. Included are the amount of nitrate added and time after production (11), storage temperature (18), pH (27) and microflora (19).

Although nitrite accumulation in cheese is temporary as it is soon transformed to other products of reduction (11,27), its presence in cheese may lead to formation of N-nitrosamines many of which are carcinogenic (12,30). On the other hand, it has been reported that there does not appear to exist any relationship between N-nitrosamine content and either the initial or the residual nitrate in the cheese and that N-nitrosamines have also been found in cheese to which no nitrate was added (12). In any event, the practice of adding nitrate to cheese milk is either questioned (9,17,28) or considered inadvisable (1,4) as there are other methods to prevent late-blowing and gassy defects in cheese.

Nevertheless nitrate addition is considered indispensable in the manufacture of brine-salted cheeses like Gouda and Edam (II). Nitrates are also being used in a great number of other cheese varieties (3,4,5,16,23,24).

Therefore this study was undertaken to find out the extent of nitrate and nitrite present in domestic and imported cheeses and their compliance with Greek cheese regulations.

MATERIALS AND METHODS

All 123 cheese samples were randomly obtained from the market of northern Greece, and they were believed to be representative of the cheese available to the urban Greek consumer. The samples were purchased evenly throughout a year and were analyzed immediately.

Nitrates and nitrites were determined according to the International Dairy Federation standard method (15). This involves extraction of the cheese with warm water, precipitation of the fat and filtration. Nitrate is reduced to nitrite in a portion of the filtrate by means of copperized cadmium in a glass column. A red color is developed in portions of both reduced solution and unreduced filtrate by addition of sulfanilamide and N-1-naphthyl-ethylenediamine dihydrochloride. The color is measured by photometry at 538 nm and compared to standard nitrite solution. Thus the nitrite content of the sample and the total nitrite content after reduction of nitrate is determined. The nitrate content is calculated from the difference between these two values.
RESULTS AND DISCUSSION

Table 1 presents the nitrate and nitrite contents of 54 domestic and 69 imported cheese samples categorized by variety. Among the domestic varieties of cheese, Feta, Teleme, Kefalotyri and Kaseri, which are the most widely consumed, contained no nitrates. The rest of the natural cheeses contained less than 1 mg of NO\textsubscript{3} per kg, which is normal, as nitrates may be present in the cheese even when no nitrates have been used in their manufacture (3,11,12,24). Only Gruyère made from cow’s milk and manufactured by the large dairy factories contained 1-10 mg of NO\textsubscript{3} per kg. The variation could be due to the age of the cheese as nitrates decrease during ripening and storage (12). In contrast, processed cheese contained nitrates in the range of 35-104 mg/kg, with an average of 57 mg/kg, despite Greek government regulations that do not prescribe nitrate use in cheesemaking (13).

Of the imported cheeses only blue-veined cheese, Pecorino and Parmesan contained little or no nitrates while Edam and Gouda contained from 1-11 and 1-24 mg of NO\textsubscript{3} per kg, respectively. The wide variation might be due to their probable long storage time and to the possible variation in the amounts added initially to the cheese milk (11). It is worth noting that imported Teleme, contrary to similar domestic cheese, contained large amounts of nitrates ranging from 13-81 mg/kg. This cheese is packaged in sealed tin cans of about 16-kg capacity, and use of nitrates in Teleme manufacture is to prevent the tin cans from blowing during their storage rather than to protect the cheese itself from gas defects. The imported Teleme samples had a pH of 4.8-5.0, brine concentration\textsuperscript{1} of 6.5-7.5 and represented 5 lots of cheese stored in the refrigerator. These conditions, along with good manufacturing practices, could limit or eliminate use of nitrates (11,21,22). So the high recorded values of nitrates in Teleme could be attributed to not fully ripened cheese and undue addition of nitrates to the cheese milk. Most brands of the imported processed cheese examined were free of nitrate and the others had low values while cheese spreads contained an average of 21 mg of NO\textsubscript{3} per kg probably due to their vegetable constituents. One of the two bacon cheese samples was in the usual range of nitrates for cheese while the other sample contained 508

TABLE 1. Nitrate and nitrite content of various cheeses in mg/kg.

<table>
<thead>
<tr>
<th>Cheese</th>
<th>Number of samples</th>
<th>Nitrates\textsuperscript{a}</th>
<th>Nitrites\textsuperscript{a}</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Range</td>
<td>Average</td>
<td></td>
</tr>
<tr>
<td>Domestic</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1. Feta\textsuperscript{b}</td>
<td>5</td>
<td>Nd</td>
<td>N</td>
</tr>
<tr>
<td>2. Teleme\textsuperscript{c}</td>
<td>5</td>
<td>N</td>
<td>N</td>
</tr>
<tr>
<td>3. Kefalotyri</td>
<td>5</td>
<td>N</td>
<td>N</td>
</tr>
<tr>
<td>4. Kaseri</td>
<td>3</td>
<td>N</td>
<td>N</td>
</tr>
<tr>
<td>5. Myzithra</td>
<td>2</td>
<td>N</td>
<td>N</td>
</tr>
<tr>
<td>6. Gruyère from sheep’s milk</td>
<td>5</td>
<td>1-10</td>
<td>7</td>
</tr>
<tr>
<td>7. Gruyère from cow’s milk</td>
<td>5</td>
<td>&lt;1</td>
<td>&lt;1</td>
</tr>
<tr>
<td>8. Kefalograviera</td>
<td>5</td>
<td>&lt;1</td>
<td>&lt;1</td>
</tr>
<tr>
<td>9. Romano</td>
<td>5</td>
<td>&lt;1</td>
<td>&lt;1</td>
</tr>
<tr>
<td>10. Korfou</td>
<td>3</td>
<td>&lt;1</td>
<td>&lt;1</td>
</tr>
<tr>
<td>11. Processed cheese (3 brands)</td>
<td>11</td>
<td>35-104</td>
<td>57</td>
</tr>
<tr>
<td>Imported</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1. Blue-veined (2 brands)</td>
<td>6</td>
<td>N</td>
<td>N</td>
</tr>
<tr>
<td>2. Parmesan</td>
<td>2</td>
<td>&lt;1</td>
<td>&lt;1</td>
</tr>
<tr>
<td>3. Pecorino</td>
<td>2</td>
<td>1-4</td>
<td>2</td>
</tr>
<tr>
<td>4. Edam</td>
<td>12</td>
<td>1-11</td>
<td>4</td>
</tr>
<tr>
<td>5. Gouda</td>
<td>18</td>
<td>1-24</td>
<td>6</td>
</tr>
<tr>
<td>6. Goudasana\textsuperscript{e}</td>
<td>1</td>
<td>-</td>
<td>27</td>
</tr>
<tr>
<td>7. Teleme\textsuperscript{c}</td>
<td>5</td>
<td>13-81</td>
<td>45</td>
</tr>
<tr>
<td>8. Processed (6 brands)</td>
<td>13</td>
<td>0-8</td>
<td>3</td>
</tr>
<tr>
<td>9. Cheese spread (2 brands)</td>
<td>8</td>
<td>19-30</td>
<td>21</td>
</tr>
<tr>
<td>10. Bacon cheese</td>
<td>2</td>
<td>24-508</td>
<td>-</td>
</tr>
</tbody>
</table>

\textsuperscript{a}Corrected for column efficiency.

\textsuperscript{b}Feta: Dry salted white soft cheese made from sheep’s milk and stored in brine.

\textsuperscript{c}Teleme: Brine salted white soft cheese made from cow’s milk and stored in brine.

\textsuperscript{d}N: Negative.

\textsuperscript{e}Cheese gratings 75% Gouda and 25% Parmesan.

\textsuperscript{1}Brine concentration = \frac{Salt \text{ content}}{Salt + moisture} \times 100
mg of NO\textsubscript{3}\textsuperscript{-} per kg, which probably originated from its bacon component.

All cheeses without nitrates were negative for nitrite, whereas cheeses with nitrates were either negative or contained less than 1 mg of NO\textsubscript{2} per kg, irrespective of the nitrate content of the cheese. Similar results have been reported by other investigators (3,11,20,23,24) for ripened cheese. This is attributed to the unstable nature of nitrates which appear transiently as they are further transformed to other reduction products (27) or may be bound to other compounds (32). The disappearance of nitrates in cheese is discussed by some authors (11,12), and it has been observed that nitrite formation is greatest during the most intense nitrate degradation and the maximum content in nitrites may correspond with only about 5% (on mole basis) of the nitrate which has been lost (11).

According to the Food and Agriculture Organization, the unconditional\textsuperscript{2} acceptable daily intake for added nitrates in foods for adults has been set at a maximum of 5 mg/kg body weight and the conditional\textsuperscript{3} at 10 mg/kg body weight. The corresponding values for nitrites are 0.4 and 0.8 mg/kg body weight, respectively. For an average 65-kg man the above figures for unconditional maxima are 325 mg of nitrates and 26 mg of nitrites. On the other hand, investigations for dietary sources of nitrates and nitrites made in USA showed that the total average daily intake of nitrates and nitrites was 99.8 mg and 11.22 mg, respectively, per resident (31). Of this amount only 0.2% came from milk and dairy products while four-fifths came from vegetables. Similar results have been reported for Norway (14). Therefore, the contribution of cheese to the daily ingestion of nitrates and nitrites is not significant. This is held to be true for a resident of Greece as well, despite the fact that his annual cheese consumption in 15 kg as compared to that of 8.6 kg for a United States resident (7). However, one should also consider the findings from a study in which several foods were incubated under simulated gastric conditions and it was revealed that with concentrations of nitrite similar to those used as food preservatives, only cheese yielded detectable amounts of volatile N-nitrosamines (29). On the other hand, nitrates and nitrites are also formed by endogenous synthesis in the human intestine (26). Therefore, the cheese regulations of some countries that require restricted amounts of nitrates and nitrites in cheese (50 mg of NO\textsubscript{3}\textsuperscript{-} and 2 mg of NO\textsubscript{2} per kg) (2,24) are rather arbitrary, whereas those of other countries that do not permit their presence in cheese are not realistic because nitrates and nitrites may be present in the cheese even if no nitrates have been added (3,11,12,24). In any event, some of the examined samples of the domestically processed cheese and part of the imported Teleme and bacon cheese did not comply with the cheese regulations of Greece.

A number of investigators consider inadvisable use of nitrates in cheese manufacture (7,5,29) while others recommend low levels of them in the cheese or cheese milk (3,6,25). Studies are still inadequate on formation of N-nitroso compounds in cheese as well as their in vivo formation in man. There is also a discrepancy in results due to the application of non-standardized analytical methods (30). All these should make one hesitant about using nitrates in cheese-making.

REFERENCES


\textsuperscript{2}Unconditional = The acceptable daily intake which is allocated to those substances for which the biological data available include either the results of adequate short term and long term toxicological investigation or information on the biochemistry and metabolic fate of the compound or both.

\textsuperscript{3}Conditional = The acceptable daily intake which falls short of the unconditional or refers to specific purpose requirements. (FAO nutrition report No. 44.1968. Rome).

Notermans, Dufrenne and Keijbets, con't. from p. 574