STATISTICS ANALYSIS REGARDING NITRATE AND NITRITE CONTENT IN LETTUCE FROM THE WEST SIDE OF ROMANIA

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Abstract: In this paper we studied the statistics interpretation of nitrate and nitrite content in lettuce. During three years, between 2005 - 2007 have been made determinations regarding nitrate and nitrite content in garden lettuce. The samples have been taken from agro-food markets in Timisoara, proceed from a few places in Timis County, in the west part of Romania. Nitrate and nitrite content determination have been realized colorimetrically in the Laboratory for the Measurement of Residues of the Department of Agro-techniques of the U.S.A-V.M.B in Timisoara. The analysis were done with the help of rapid tests AQUA MERCK, with the Spectrophotometer SQ 118 at a wavelength of 515 and 525 nm for nitrate, nitrites. The statistics interpretation of the results was done by using the analysis of some statistics parameters like: mean, range, kurtosis, skewness.

Keywords: lettuce, nitrate, nitrite, statistical parameter

INTRODUCTION

Nitrates and nitrites are natural compounds of soil, resulted from mineralization of animal and vegetal substances. A part of nitrate is absorbed by the roots of the plant and is used as a raw material for protein synthesis, and other is taken by the surface waters or of those which cross soil and we find them in rivers, lakes, and underground waters [1].

Between nitrate and nitrite in soil, water and plants establish a balance, which can be broken by the intensive use of the organic fertilizers, natural or synthetic in agriculture or horticulture.

Nitrate and nitrite degradation compounds richness the soil and can be accumulated in crops through injuring levels for consumers.

Nitrate and nitrite analysis supply as a food diet, indicate a predominant level of nitrate ingested daily at once with vegetables and in a smaller doses at once with animal food.

According with literature studies, in balanced food diet, 79% of ingested nitrates proceed from vegetables, 20% from drinking water, 6% from meat, 1% from cereal products and 1.3% from fruits and milk products [3].

Nitrate radical (NO$_3^-$) can be absorbed by a vegetable and converted into amino acid, and finally gone through the biosynthesis to compose the amino acid into the usable proteins stored in the stems and leaves of the vegetable. In the nature, only some of the root modules of the plants can live with a group of nitrogen fixation bacteria and absorb the ammonia (NH$_3$) element in the nature directly or the nitrogen (N$_2$) element in air. The ammonia (NH$_3$) element is transformed into chemicals absorbable by plants, and such process is called nitrification, and its chemical formula is given as follows: (NO$_3^-$) $\rightarrow$ (nitrite bacteria decomposition) $\rightarrow$ (NO$_2^-$) $\rightarrow$ (nitrobacteria decomposition) $\rightarrow$ (NO$_3^-$) [2].

The level of nitrites in vegetables is smaller than the level of nitrates because in the transformation process of the nitrates in plants, the nitrites stage is provisional, so the nitrites result under the influence of nitrate-bacteria, it turn further on ammonia form under the influence of nitrite-bacteria. The maximum limit allowed for nitrate in lettuce is 2000 ppm according to Order No. 293/640/2001-1/2002 regarding condition of security and quality for vegetables and fresh fruits for human consumption [9]. The nitrite content in vegetable should not exceed 1-2 ppm according with the other authors [1].

MATERIALS AND METHODS

The nitrate and nitrite content in lettuce was done with the help of rapid tests AQUA MERCK, with the Spectrophotometer SQ 118 at a wavelength of 515, 525 nm for nitrate and nitrites. Minimum detection limits according to work method are: 1 mg/l for nitrates, 0.02 mg/l for nitrites.

This vegetable has been selected, because is well-known that have a high nitrate content, who is generate by the genetic potential of plant, by the small content of nitrate-bacteria and the absence of minor elements, very important factors in the activity of reduction bacterial. Samples of lettuce were washed and than we cut finely and put in distillation water. After an hour we filtrated the sample and we took 1.5 ml filtrated lettuce sample and put it over the Aqua Merk nitrate solution. After 10 minutes we read the value of nitrate to Spectrophotometer SQ 118. Regarding the nitrite we took 10 ml of filtrated lettuce sample and put it over the nitrite-Aqua Merk solution, and after 10 minutes we read the value of nitrite to the same Spectrophotometer SQ 118, but to another wavelength (525).

Analysis concerning nitrate content in lettuce was done colorimetrically in the Laboratory for the Measurement of Residues of the Department of Agrotechniques of the U.S.A-V.M.B in Timisoara. Samples have been taken from agro-food markets in Timisoara, proceed from a few places in Timis County, in the west part of Romania.

The conclusions will be express after the interpretation of some statistics parameters. Beside the classic parameters of position and variation ($mean - \bar{X}, amplitude - A$), we will analyze in this paper a few
aspects concerning the symmetry or obliquity (skewness) and flattening or vaulting (kurtosis). "Normality" of static distribution is given by the approximating of normal curve form (Gauss). In the case of normal distribution, a perfect symmetric distribution, arithmetic average, median line and modulus coincide [10].

Asymmetric coefficient (skewness) returns the skewness of a distribution. Skewness characterizes the degree of asymmetry of a distribution around its mean. Positive skewness indicates a distribution with an asymmetric tail extending toward more positive values. Negative skewness indicates a distribution with an asymmetric tail extending toward more negative values [4]. The equation for skewness is defined as:

$$s = \sqrt{\frac{1}{n(n-1)(n-2)}} \sum_{i=1}^{n} \left( \frac{x_i - \bar{x}}{s} \right)^3$$

for 1 to 30 arguments for which we want to calculate skewness, where $s$ is the sample standard deviation.

Excess or kurtosis characterizes as the relative peakedness or flatness of a distribution compared with the normal distribution. Positive kurtosis indicates a relatively peaked distribution (leptokurtic). Negative kurtosis indicates a relatively flat distribution (platikurtic) [5]. Kurtosis is defined as:

$$k = \frac{n(n-1)(n-2)}{(n-3)} \sum_{i=1}^{n} \left( \frac{x_i - \bar{x}}{s} \right)^4 - \frac{3(n-1)^2}{(n-2)(n-3)}$$

for 1 to 30 arguments for which we want to calculate kurtosis, where $s$ is the sample standard deviation [4].

### RESULTS

Table 1. NO$_3$ content (ppm), in lettuce, in agro-food markets; average for 2005-2007

<table>
<thead>
<tr>
<th>Period</th>
<th>Fratelii</th>
<th>Săcălaz</th>
<th>Dinaș</th>
<th>Sândre</th>
<th>Utin</th>
<th>Cenad</th>
<th>Dudești</th>
<th>Jimboli</th>
<th>Gelu</th>
</tr>
</thead>
<tbody>
<tr>
<td>Spring</td>
<td>177.33</td>
<td>180.33</td>
<td>185</td>
<td>178.33</td>
<td>207</td>
<td>219</td>
<td>199</td>
<td>206.66</td>
<td>167</td>
</tr>
<tr>
<td>Autumn</td>
<td>173</td>
<td>145.33</td>
<td>139.33</td>
<td>155</td>
<td>180.33</td>
<td>168.66</td>
<td>169</td>
<td>182.33</td>
<td>152</td>
</tr>
</tbody>
</table>

Regarding to the all nitrate content values obtained, it observe that the variation of parameters values (amplitude and standard deviation) have values inferior to mean, so the obtained data are platikurtic and with asymmetry to right. For the autumn values, the repartition is platikurtic and with asymmetry to left.

Table 2. NO$_3$ content (ppm), in lettuce, statistics interpretation

<table>
<thead>
<tr>
<th>Period</th>
<th>Media</th>
<th>Min</th>
<th>Max</th>
<th>Relative amplitude</th>
<th>Kurtosis</th>
<th>Skewness</th>
<th>Standard deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Spring</td>
<td>191.0722</td>
<td>167</td>
<td>219</td>
<td>27.21%</td>
<td>-1.21724</td>
<td>0.305995</td>
<td>17.40128</td>
</tr>
<tr>
<td>Autumn</td>
<td>162.7756</td>
<td>139.33</td>
<td>182.33</td>
<td>26.41%</td>
<td>-1.40218</td>
<td>-0.24248</td>
<td>15.40721</td>
</tr>
</tbody>
</table>

Regarding to the all nitrite content values obtained, it observe that the variation of parameters values (amplitude and standard deviation) have values inferior to mean, that thing leads to a low degree of dispersion. For the spring values, the repartition is platikurtic and with asymmetry to right. For the autumn values, the repartition is platikurtic and with asymmetry to right.

Table 3. NO$_2$ content (ppm), in lettuce from agro-food markets; average for 2005-2007

<table>
<thead>
<tr>
<th>Period</th>
<th>Fratelii</th>
<th>Săcălaz</th>
<th>Dinaș</th>
<th>Sândre</th>
<th>Utin</th>
<th>Cenad</th>
<th>Dudești</th>
<th>Jimboli</th>
<th>Gelu</th>
</tr>
</thead>
<tbody>
<tr>
<td>Spring</td>
<td>1.06</td>
<td>1.21</td>
<td>1.14</td>
<td>1.17</td>
<td>1.11</td>
<td>1.49</td>
<td>1.23</td>
<td>1.1</td>
<td>1.22</td>
</tr>
<tr>
<td>Autumn</td>
<td>0.44</td>
<td>0.715</td>
<td>0.59</td>
<td>0.5</td>
<td>0.52</td>
<td>0.54</td>
<td>0.71</td>
<td>0.64</td>
<td>0.56</td>
</tr>
</tbody>
</table>

Regarding to the all nitrate content values obtained in this 3 years of study, it observe that the variation parameters values (amplitude, standard deviation) have values inferior to mean, that thing leads to a low degree of dispersion. For the spring values the repartition is leptokurtic and with asymmetry to right. For autumn values the repartition is platikurtic and with asymmetry to right.

### DISCUSSIONS

In Table 1 it shows the nitrate content in lettuce from agro-food markets of Timisoara, average for 2005-2007. The all nitrate content values registered between 2005 - 2007 did not exceed maximum limit allowed for this culture, which is 2000 ppm according with the Order No. 293/640/2001-1/2002 [9]. Through the studies made by other foreign researches [6] we can see that exist some correlation between the nitrates values obtained in lettuce leaves in Italy, where have been done similar experimental researches, with the values obtained by us in the west part of Romania. In Italy the mean value for nitrates content was 185.07 for spring lettuce and 174.21 for autumn lettuce. In the study made by us, in the west part of Romania the mean values regarding nitrates content for spring culture we obtained 191.0722 and for autumn culture 162.7756, which means that the researchers from Italy used almost the same culture technology like us.

Regarding nitrate content statistics parameters (Table 2) relative amplitude and standard deviation have values inferior to mean, so the obtained data are...
not dispersal which means that the vegetable producers use approximately the same technology to lettuce culture in all the localities from which we took samples.

Regarding the symmetry or obliquity (skewness) of the nitrate content values obtained spring, the skewness coefficient is 0.305995, is positive, so positive skewness indicates a distribution with an asymmetric tail extending toward more positive values, results asymmetry to right. The values of nitrate content in autumn have a negative skewness coefficient -0.24248, is negative, so negative skewness indicates a distribution with an asymmetric tail extending toward more negative values, results asymmetry to left. So, in spring the most data obtained are under the mean which means that the most producers use small quantities of fertilisers so the nitrate content is lower in lettuce in spring in the most localities from which we took samples. In autumn the data obtained are over the mean so the most producers used higher synthetic fertilisers but the nitrate content did not accumulated and the values did not exceed maximum limit allowed for this culture.

Regarding the excess or kurtosis coefficient of nitrate content values, in spring was -1.21724 and in autumn was -1.40218 had negative values, so negative kurtosis indicates a relatively flat distribution (platikurtic), so we didn’t registered the same values which to appear in high number, either in spring and in autumn.

Regarding the nitrite content in lettuce between 2005 – 2007 (Table 3), the maximum limit allowed according with another authors [1] is 1 – 2 ppm, in all the 3 years of research the obtained values did not exceed this limit, in all the localities from which we took lettuce sample. These results indicate that the values of parameters variation (amplitude, standard deviation) have values inferior to mean, so we had a low dispersion degree of data.

In the case of nitrite content regarding the symmetry, (Table 4) the skewness coefficient had positive values either spring (1.819002) and autumn (0.28172), results asymmetry to right, so in spring the most data obtained are under the mean which means that the most producers use small quantities of fertilisers so the nitrate content is lower in lettuce in spring in the most samples.

Nitrite content values had a kurtosis coefficient different in spring and autumn, in spring kurtosis was 4.225287, had positive value indicating a relatively peaked distribution (leptokurtic) and in autumn was negative, -0.85678, negative kurtosis indicates a relatively flat distribution (platikurtic). So, in spring the nitrite content values were in high number the same, around 1 ppm, this means that climatic condition influenced the nitrite accumulation in lettuce in all the localities in the west part of Romania. In spring of 2005 and 2006, in the west part of Romania was inundations and in spring of 2007 was a large drought. Both excess and deficiency of water in lettuce culture lead to accumulation of nitrite in plants, by inhibiting action of micro-organisms which concur to mineralization process of nitrogen fertilisers for development of plant.

Between nitrate and nitrite content in lettuce the correlation coefficient is 0.71, which means that between nitrates and nitrates is a dependence because the level of nitrates in vegetables is smaller than the level of nitrates because in the transformation process of the nitrates in plants, the nitrates stage is provisional, so the nitrates result under the influence of nitrate-bacteria, it turn further on ammonia form under the influence of nitrate-bacteria [7].

Ten years ago, to Timisoara Agricultural Station, have been made studies by the Banat’s University researchers [8] with the purpose of establish some correlations between the increasing nitrogen level in term of administrated nitrogen dose. This correlation were very significant in the case of manure fertilisation, to dose who exceed 25 t/ha. The increase of nitrogen content in spring lettuce, proportionally with the laystall administrated dose. The correlations are significant for the variants with 40 – 50 t/ha, becoming distinct significant for the maximum dose. Comparison between the two types of organic fertilisers (manure and laystall) allow to accentuate a high increasing of nitrates content when it fertilize with manure (241% to 30 t/ha), unlike the laystall fertilisation (179% to 60 t/ha). In the both types of organic fertilisation, the nitrates content increase in lettuce culture was low comparing with the one determinate by the ammonium nitrogen synthetic fertiliser (402% to 300 kg N/ha) [8].

Explanation of these dynamics, in the three types of fertilisation exposed earlier depend by the form of the nitrogen in the applied fertilisers assortments. Higher values, determinate in ammonium nitrogen variants, due to the fact that here the nitrogen appear as synthetic form, accessible directly to plants of 50% N-NH₄ and 50% N-NO₃.

As a conclusion we can say that the results data distribution is not a normal one because of the use synthetic fertilisers and organic fertilisers to lettuce culture, by the vegetable producers from west part of Romania. Another cause can be the climatic condition which was chaotic in these 3 years of research, with inundations and drought.

REFERENCES
