Full Length Research Paper

Studying of the Nitrate Contamination in Garden Cress Plant in Dezful, Khouzestan, Iran

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Abstract: In recent years, there has been much attention to the amount of nitrate. Nitrate is not toxic for animals and human. However, if nitrate reduces to nitrite, it can combine with hemoglobin and form met-hemoglobin. Regarding the importance of this subject, they present research was conducted to study the situation of Garden cress in terms of nitrate contamination in vegetable farms in Dezful city. Samples after being shaking, the extract was separated by centrifuge and nitro Sulfosalicylic complex. Then it was read by spectrophotometer in wave length 410 nm in front of standard solution. Results of analyses indicated that the highest amount of nitrate was seen in the north farm samples of Dezful. Results of statistical analysis revealed significant difference between nitrate amounts in areas under study. Moreover, nitrate concentration was reported less than permitted in Garedn cress. As well, nitrate concentration tested in the morning and afternoon samples showed significant difference and the highest difference was related to north of Dezful. Therefore, in order to obtain low levels of nitrate concentration, considering soil Analysis, organic cultivation, consuming organic and biologic fertilizers, micronutrients and eugenics can be regarded.

Key words: Nitrate, Contamination, Garedn cress, spectrophotometer, Vegetable Farms, Dezful City

1. INTRODUCTION

Nitrogen comprises 79% of atmosphere. In addition it is considered the most important element in organic cells in soil after carbon and is a vital element for plants whose utilization by human is controllable and can be used in different forms in agriculture. Also, it is considered as a restrictive factor for most of the agricultural products. One of its functions is to participate in protein and chlorophyll formation, so its shortage leads old eaves to turn yellow. Finally, it results in stopping plant’s growth and decreasing product. On the other hand, excessive nitrogen leads to overgrowth of plant growing dark green leaves and plant lodging (Malakuti and Homaei, 2004).

Vegetables and drinking water contain much more amount of nitrate than before because of overusing (organic and artificial) nitrate fertilizers in agriculture (CECSCF, 1992). The nutritional value of vegetables in providing necessary materials for human health, in order to provide a part of required energy for human body’s metabolism and also supplying a number of essential compounds such as vitamins, minerals, some amino acids and raw fiber. Therefore, harmful compounds in them like nitrate enter the body and have adverse effects on human health. Enter of Nitrate to body and have adverse effects on human health. Nitrate has been placed in the toxicant category (chemicals resulted from agricultural action) in classification of chemical salts inserted in the latest guide line of world Health Organization (WHO) in 2006. Nitrate stability is high in the environment, so that it can remain for 50 years (Peyvast, 2006).

World Health Organization has suggested consuming 400 grams of vegetables and fruits a day (Hord et al., 2009) and nearly 80% of nitrate enters human’s body through consuming vegetables and fruits (Dich, 1996). Nitrate concentration in vegetables depends on season, light intensity, temperature, and growth conditions, fertilizing amount and storage conditions (Malakuti et al., 2005). In various plant’s nitrate is stored is different Organs. Factors such as nitrate supply, plant, and species and plant age influence it. The ratio of nitrate reduction in root rises by increasing the environment temperature (Kennedy, 2003).

Increasing the plant age, nitrate concentration in plant also increases. However, excessive fertilizing in each phase leads to increased nitrate concentration in the plant. Results showed in decreased amount of nitrate. Therefore, vegetables had better be harvested in the afternoon (Alexander, 2008). There are various standard for maximum concentration of nitrate in the vegetables. For instance, European Union has suggested the maximum amount of nitrate concentration for lettuce in hydroponic about 3500 mgkg-1 in spring farming and about 4500 mgkg-1 in fall farming. This amount is about 2500 mgkg-1 for land framings (Hord et al., 2009).
The permitted amount of nitrate for various vegetables has not yet been determined in Iran. Generally speaking, the maximum amount of nitrate entering human body should not exceed 3.65 mg kg\(^{-1}\) of human body a day (Santamaria, 1999). However, a 70 kg individual should not consume more than 255 mg of nitrate a day. Therefore, we should critically try to decrease nitrate concentration to the minimum amount particularly for people who consume a lot of vegetables in their diet.

### 2. MATERIALS AND METHODS

Sampling was conducted from vegetable farms located in northern, southern, eastern and western areas of Dezful City in Khouzestan province in Iran. The samples were collected twice a day, in the morning and afternoon, randomly from first, middle and last rows of the farms. After carrying the samples to the lab, they were washed with distilled water and after being ground they were air dried for 48 hours. Their fresh weights were determined (first the sample dish was weighed, than the dish with the sample was weighed). The samples were dried in 70° C using an oven. After measuring their dry weight, they were ground by an electric mill.

Preparation of Nitrosulfosalicylic complex and reading it using a spectrophotometer:

- Of the important properties of this method are:
  - Simplicity, needing a few equipment, absence of interference by other ions present in plant tissue, high speed and its extensive measurement range.
  - In the vicinity of nitrate, salicylic acid forms Nitrosalicylic which is light yellow.

  The intensity of this color corresponds to the amount of nitrate in plant tissue. This color has the maximum absorption in wavelength 410 nm. Generally, this method is used more to measure nitrate in samples with high nitrate concentration (about 3%).

  First, 0.1 gram oven-dried and ground sample is weighed and 10 ml distilled water is added to it. After that the samples are stored in 45 for an hour. Then the sample mixtures are centrifuged in 1000 Rpm (One can use activated charcoal and Wittman filter paper to filtrate the extracts). Of the resultant extract 0.2 ml is separated and 0.8 ml Sulfosalicylic acid (5%) is added to it. After cooling it, 19 ml NaOH 2 normal is added to it, and then the intensity of the resultant color is read in the wavelength 410 nm using a spectrophotometer.

  Obviously, to read the extracted samples we need to determine a standard from 5 through 300 mg/l for nitrate nitrogen concentration that is produced using dry potassium nitrate (KNO3), than reading unknown samples according to it (Malakuti and Bybourdi, 2004). Results of analyzing the samples were compared using spss software version 18. The differences among the areas and the comparison of measured values with related standard value were conducted using ANOVA and t-test respectively. Using excel software, the charts were drawn.

### 3. RESULTS AND DISCUSSIONS

The Garden cress average nitrate concentrations in vegetable farms located in four areas of Dezful are shown in table 1. According to the findings, there is a significant difference among those areas in terms of nitrate concentration with probability level of 0.05.

The results of variance analysis showed that there is a significant different between amounts of nitrate in Garden cress among four studied areas in two harvest times. Nitrate concentration in north of Dezful was more than other areas (south, east and west). In addition, the comparison of the average nitrate concentration in Garden cress in the morning and afternoon showed statistical signification difference (0.01%). One of the reasons for nitrate changes when exposed to light is nitrate reductase enzyme dependence to Photospectrum changes.

Through shedding light during day, the activity of nitrate reductase enzyme increases as well photosynthesis and protein synthesis speed up, in both cases it results in using nitrate concentrated in plant organs and consequently its amount decreases (Malakuti and Bybourdi, 2004). Therefore, harvesting Garden cress in the afternoon can help in reducing nitrate concentration (Rahmani, 2006).

| Table 1: comparison of nitrate concentration to the values reported in resources and its permissible limit in Garden cress (Santamaria, 2006) |
|-----------------------------|------------------|-----------------|------------------|
| Vegetable                   | Average nitrate  | Concentration  | Permissible Limit |
|                             | concentration    | reported in     | of nitrate       |
|                             | in dry matter    | resources       | concentration    |
|                             | (mg.kg\(^{-1}\)) | (mg.kg\(^{-1}\)) |                  |
| Garden cress                | 1474             | 965.5           | 1500             |
Table 2: comparing the measured nitrate concentration in vegetables with Garden cress nitrate limit.

<table>
<thead>
<tr>
<th>The area Under study</th>
<th>Number of samples Garden cress</th>
<th>Range of measured values In (mg.kg⁻¹)</th>
<th>Average nitrate concentration in dry Garden cress In (mg.kg⁻¹)</th>
<th>Average nitrate concentration in the fresh Garden cress (mg.kg⁻¹)</th>
<th>Limit average comparison with permissible limit</th>
</tr>
</thead>
<tbody>
<tr>
<td>North of Dezful</td>
<td>6</td>
<td>1700-2000</td>
<td>1833.3</td>
<td>516</td>
<td>ns</td>
</tr>
<tr>
<td>South of Dezful</td>
<td>6</td>
<td>1200-1400</td>
<td>1300</td>
<td>196.6</td>
<td>ns</td>
</tr>
<tr>
<td>East of Dezful</td>
<td>6</td>
<td>1100-1400</td>
<td>1233.3</td>
<td>187</td>
<td>ns</td>
</tr>
<tr>
<td>West of Dezful</td>
<td>6</td>
<td>1400-1700</td>
<td>1533.3</td>
<td>384</td>
<td>ns</td>
</tr>
</tbody>
</table>

** and ns indicate the presence and absence of significant differences in Duncan's test.

Table 3: comparison of average nitrate concentrations measured in Garden cress in the morning and afternoon

<table>
<thead>
<tr>
<th>The area Under study</th>
<th>The average nitrate concentration in the fresh in the morning (mg.kg⁻¹)</th>
<th>The average nitrate concentration in the fresh in the afternoon (mg.kg⁻¹)</th>
<th>Nitrate reduction rate in the afternoon</th>
<th>statistical average comparison</th>
</tr>
</thead>
<tbody>
<tr>
<td>North of Dezful</td>
<td>516</td>
<td>399</td>
<td>117</td>
<td>**</td>
</tr>
<tr>
<td>South of Dezful</td>
<td>196</td>
<td>147</td>
<td>49</td>
<td>**</td>
</tr>
<tr>
<td>East of Dezful</td>
<td>187</td>
<td>119</td>
<td>68</td>
<td>**</td>
</tr>
<tr>
<td>West of Dezful</td>
<td>384</td>
<td>301</td>
<td>83</td>
<td>**</td>
</tr>
</tbody>
</table>

**Indicates statistically significant difference in probability level of 1% in Duncan test.

Fig. 1: Nitrate concentration in the Garden cress in the morning and afternoon in the north of Dezful in the fresh (mg.kg⁻¹) (N1, N2 and N3 indicate the amount of nitrate in the first, second and third rows of the farm, respectively)

Fig. 2: Nitrate concentration in Garden cress in the morning and afternoon in the South of Dezful in the fresh (mg.kg⁻¹) (S1, S2 and S3 indicate the amount of nitrate in the first, second and third rows of the farm respectively)
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**Fig. 3:** nitrate concentration in Garden cress in the morning and afternoon in the fresh (mg kg\(^{-1}\)) (E1, E2 and E3 indicate the amount of nitrate in the first, second and third rows of the farm respectively.)

**Fig. 4:** nitrate concentration in Garden cress in the morning and afternoon in the west of Dezful in the fresh (mg kg\(^{-1}\)) (W1, W2 and W3 indicate the amount of nitrate in the first, second and third rows of the farm respectively)

**Fig. 5:** comparison of the average nitrate concentration Garden cress in the four areas North, South, East and West of the city of Dezful in the morning
4. CONCLUSION

The average nitrate concentration in Garden cress in vegetable farms located in north of Dezful Lower than the permitted amount, so it is suggested not to use Garden cress in diets as much as possible specially for children in order to prevent causing met-hemoglobin. However, because of high consumption of it in stews and soup, it is impossible to put it aside. Therefore, it is necessary to control cultivation and harvest of coriander in Dezful. A research done (1998 fall up to 1999 summer) showed that average nitrate in leafy green vegetables is 278.9 mgkg⁻¹ (Rahmani, 2006), and it is much more and about 627.2 mgkg⁻¹ in Kermanshah. While, the average nitrate in Garden cress which is one of the leafy green vegetables is 1474 mgkg⁻¹ for Dezful (Pirsaheb et al., 2012). Regarding the fact that the most harvested water crops yearly with 16.5% belong to Khuzestan, most of farms in Dezful are devoted to vegetables. Therefore, in order to obtain low levels of nitrate concentration, considering soil test, organic cultivation, consuming organic and biologic Fertilizers, micronutrients and eugenics can be regarded. Because of its high consumption and great changed in nitrate concentration in Garden cress, further investigation and continuous control of nitrate concentration is necessary.

REFERENCES


Dr. Ali Gholami is an assistant professor in soil science. He is the editor and reviewer of some international journals. He has studied 15 university research design, and 50 printed papers in national and international conferences and journals. Mr. Ali Gholami has graduated with first grade in M.Sc. degree and PhD coarse book. He was selected as head of department of soil science and manager of research office in Islamic Azad University, Khuzestan Science and Research Branch in 2009 and it now. Also he was selected as superior researcher in 2011 and 2012 in Khuzestan province, Iran.

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