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# The impact of seed gall nematode on grain yield, quality and marketing prices on durum wheat in Anatolia, Turkey

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The objective of this study was to draw the attention for this under estimated pest in the South-east of Anatolia. Field studies were carried out in historically known infested farmer fields to assess the effects of seed gall nematode (*Anguina tritici* [Steinbuch]) on grain yield, quality and marketing price of durum wheat (*Triticum durum* Desf.) in the wheat production region of Viranşehir, Anatolia, Turkey during 2008/2009 growing season. Grain yield losses due to A. tritici were reported up to 32%. Seed gall severity corresponded with a decrease in number of grain spike<sup>-1</sup>, grain weight spike<sup>-1</sup>, test weight (kg hl<sup>-1</sup>) and 1000 kernel weight (g). However, the SDS protein content (%) was not affected by increase in seed galls. The price of durum wheat per tonne was reduced from 335 to 297 USD\$ with increasing *A. tritici* contamination (from 5 to 20%). This could be explained by a significant negative regression with grain price against *A. tritici* (P ≤ 0.01; R<sup>2</sup> = 80.2%). *A. tritici* has a potential to be one of the major biotic constraints in the irrigated wheat production regions of Anatolia as both yield and grain price were affected. The use of clean seeds should assist in effective control and careful monitoring is a must.

Key words: Grain yield, quality, marketing price, Anguina tritici, durum wheat, seed gall nematodes.

# INTRODUCTION

Although many nematodes have been found associated with small grain cereals, only a few of them are considered to be economically important (Nicol and Rivoal, 2008). Seed gall nematode *Anguina tritici* ranks fourth for its economical importance after cereal cyst (*Heterodera spp.*), root lesion (*Pratylenchus* spp.) and root knot (*Meloidogyne* spp.) nematodes, respectively (Nicols and Rivoal, 2008). Ten percent of world crop production is estimated to be lost as a result of plant parasitic nematode damage (Whitehead, 1998). The potential populations of seed gall or 'ear cockle' nematode are determined by the amount of seed contaminated with galls caused by *A. tritici* with the contaminated galled seeds. Once the wheat seed is sown

the second stage juveniles emerge in the moist soil, invade the wheat seedling and feed ectoparasitically on the tissues of young leaves near the growing point. The infected seedlings appear more or less severely stunted with characteristic rolling, twisting and crinkling of the leaves. The resulting infested ears are undersized, shorter and thicker than healthy ones, with some or all grains being replaced by the galls (Southey, 1972; Evans et al., 1993). Ear cockle has been reported throughout West Asia and North Africa (WANA) (Sikora, 1988), the Indian sub continent, China, parts of Eastern Europe (Tesic, 1969; Swarup and Sosa-Moss, 1990), Iraq (Stephan, 1988), Pakistan (Maqboll, 1988) and Iran (Bonjar et al., 2004).

In Turkey, the first documented studies on ear cockle (Karamuk in Turkish) were from surveys conducted in the mid eighties. The infestation rate estimates were 0.2% in the Eastern Anatolia (Yüksel et al., 1980), 1.6 to 55.2% in the Western part (Elmali, 2002), 25.4% in the Marmara

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region (Ağdaci and Efe, 1986), 3.6% in the South East Anatolia (İmren, 2007), and none at Aegean (Misirlioğlu and Pehlivan, 2007) and Eastern Mediterranean region (Elekçioğlu, 1996; Gözel, 2001). The yield loss estimates were 0.2% in the Eastern Anatolia (Yüksel et al., 1980) and up to 60.0% in Central Anatolia (Öztürk and Elmali, 1993). However a study approximately ten years later checking in the seed lots of the same region in Central Anatolia showed the losses of only 0.01% (Elmali, 2002). In Irag infection rates ranged from 0.03 to 22.9%, with corresponding yield losses up to 30% (Stephan, 1988). Yield losses caused by A. tritici in Pakistan and China were reported in various studies to be 2.0 to 3.0 and 10.0 to 30.0% (Magbool, 1988; Chu, 1945). In India, the effects of contamination of seed galls on grain and flour marketing prices resulted in reductions between 22.8 and 48.1% in marketing prices (Paruthi and Bhatti, 1988). Despite all listed losses, there are very few studies about the effects of seed galls caused by A. tritici on both yield and grain quality of kernels on both infested as well as healthy spikes with several infected kernels. This study attempted to address this issue drawing attention of many stake holders namely, farmers, seed producers, commodity purchasers, macaroni industry and policy makers both in Turkey and neighboring countries.

#### MATERIALS AND METHODS

Study was carried out in farmer fields in the Sakalar village of Viranşehir, in Şanliurfa province during 2008/2009 wheat growing season wherein A. tritici infestation was first identified in 2006/2007 season when the Durum wheat variety Fuatbey 2000 was grown. Cotton was planted in 2007/2008 followed by the same durum wheat variety in 2008/2009 cropping season. A total area of 18 ha was sown by drill at a rate of 450 seeds m<sup>-2</sup>. Fertilizers namely, di ammonium phosphate (DAP: 18 and 46% of N and P), ammonium nitrate (AN: 33%) and ammonium nitrate (AN: 26%) at the rates of 200, 300 and 150 kg ha<sup>-1</sup> were applied at sowing, early joining and stem elongation stages of crop growth, respectively. Chemical broad- leaf- weeds management was practiced at four leaves stage of weeds. Four flood irrigations one each at sowing and boot stage and twice at milky dough ripening stage was given. All other agronomical practices for a healthy crop production were taken up (Pala and Akulke; 1968; Uzun, 1982, 1983, 1984). Crop was harvested on 21<sup>st</sup> of June, 2009 by combine harvester.

Twin plots  $(1+1 = 2 m^2 \text{ each})$  located 80 m apart were randomly selected at four different places in the field at heading stage. Total number of spikes, number of distorted and totally dried spikes with empty spikelets (nematode damaged) in each plot were counted.

Selected twin plots were harvested by hand and threshed for measuring grain yield. After dockage cleaning and weighing, grains of four twin plots were combined. Healthy grains and grains with galls were separated by hand and five groups (pure healthy, 5, 10, 15 and 20% gall contaminated lots [total 100 g each]) were built up in the laboratory for analysis of market pricing. Employing a randomized complete block design with 5 replications (grain buyers), above seed lots were presented to five randomly selected grain buyers for price estimations in the local commodity market on 10<sup>th</sup> of September, 2009.

A fifty healthy looking and distorted spikes in the experimental field were tagged randomly and harvested by hand at maturity for further analysis of yield and quality components. Student t- test was

performed for comparisons of yield components such as number of grain spike<sup>-1</sup> and grain weight spike<sup>-1</sup> Grain samples obtained from tagged spikes were subjected to quality analysis such as 1000 kernel weights (g) (Williams et al., 1986) ,test weight (kg hl<sup>-1</sup>), kernel vitreousness (%) (Anonymous, 2002a), Sodium Dodecyl Sulphate (SDS) (Anonymous, 1992) and protein content (%) (Anonymous, 2002b) in the laboratory of Faculty of Agriculture. Yield loss (kg ha<sup>-1</sup>) was calculated based on the number of infected spikes (m<sup>2</sup>) X(average grain weight of healthy spike [g]- average grain yield of infected spike [g])  $\times$  10000 (Bora and Karaca, 1970). TARIST (Açikgöz et al., 1994) statistical software was used for variance and regression analysis.

# RESULTS

Total number of spikes m<sup>-2</sup>, the number of seed gall nematode damaged spikes m<sup>-2</sup>, actual yield, yield loss due to *A. tritici* and expected actual yield (kg ha<sup>-1</sup>) based on means of twin plots are given in Table 1. The numbers of spikes m<sup>-2</sup> were almost homogeneous for the four plots. The infestation ratio excluding the rates of healthy looking spikes, was 16.21% = ([(38.75+16.75)/342.25]x 100) in durum wheat. Expected actual grain yield was 6.63 tons ha<sup>-1</sup>. Yield losses due to *A. tritici* were estimated to be 2.13 tons ha<sup>-1</sup> totaling an average yield loss of 32%.

Only 29 (6 healthy+ 14 healthy looking+ 9 distorted) of the 50 tagged spikes were evaluated for losses before harvest. Student t-test results employing only tagged spikes were given in Table 2. These indicated that the highest spike weight of 2.87±0.15 g was obtained from healthy spikes. Average spike weight of healthy looking spikes was 2.77±0.21 g and that of distorted spikes was 0.55±0.12 g. Statistical significance for spike weight of healthy spikes vs. healthy looking was nil (t =  $0.79^{ns}$ ) whereas these turned out to be significant with distorted vs. healthy looking and distorted vs. healthy spikes (t = 11.37<sup>\*\*</sup> and  $t = 6.89^{**}$ ). Table 2 also shows that the highest number of grain spike<sup>-1</sup> was obtained from healthy spikes with 50.00±2.38. That of healthy looking spikes was 43.64±3.72 and this was 10.22±1.94 for distorted spikes. Healthy looking spikes also contained an average 6.28±2.45 grains with galls. T-test for number of grains of healthy spikes vs. healthy looking spikes was non significant (t =  $1.066^{ns}$ ) whereas differences for the number of grains of distorted spikes vs. healthy and that of distorted vs. healthy looking spikes were significant (t=  $12.92^{**}$  and t =  $6.78^{**}$ ).

Data for marketing price estimates ( $kg^{-1}$ ) were subjected to a variance analysis and the results are given in Table 3. Statistical significance for differences among the seed lots with increasing ratios of contaminations (= 130.31\*\*) was noted. Differences among grain buyers (replications) was also significant (F= 21.84\*\*). This indicated that buyer justification is an effective factor in marketing price determination. *A. tritici* damaged kernels with increasing ratios were offered less marketing price than pure healthy grains of same cultivar. Pure healthy

| No. of twin plots | NOS1 <sup>1</sup> | NOS2 <sup>2</sup> | NOS3 <sup>3</sup> | NOS4 <sup>4</sup> | AY⁵(kg ha⁻¹) | YL <sup>6</sup> (kg ha⁻¹) | PY <sup>7</sup> (kg ha <sup>-1</sup> ) |
|-------------------|-------------------|-------------------|-------------------|-------------------|--------------|---------------------------|--|
| 1                 | 330               | 40                | 17                | 273               | 4604.0       | 2181.3                    | 6785.0                                 |
| 2                 | 352               | 25                | 13                | 314               | 5351.0       | 1862.0                    | 7213.0                                 |
| 3                 | 350               | 58                | 21                | 271               | 4508.0       | 2684.0                    | 7192.0                                 |
| 4                 | 337               | 32                | 16                | 209               | 3553.0       | 1783.0                    | 5336.0                                 |
| Mean              | 342.25            | 38.75             | 16.75             | 266.75            | 4504.0       | 2127.5                    | 6631.0                                 |
| ±SD               | ±39.57            | ±3.55             | ±0.82             | ±10.82            | ±184.3       | ±102.23                   | ±221.46                                |
|                   |                   | SW2 <sup>8</sup>  | SW3 <sup>9</sup>  | SW4 <sup>10</sup> |              |                           | SW5 <sup>11</sup>                      |
| Mean              |                   | 0.555             | 0.0               | 2.468             |              |                           | 2.77                                   |
| ±SD               |                   | 0.03              |                   | 0.05              |              |                           | 0.06                                   |

Table 1. Expected and actual yields, yield loss due to A.tritici obtained from 4 randomly sampled unit area in the infected field.

<sup>1</sup>NOS1: Average no. of spike  $(m^2)$ , <sup>2</sup>NOS2: average no. of distorted spikes  $(m^2)$ , <sup>3</sup>NOS3: ave. no. of dried spikes  $(m^2)$ , <sup>4</sup>NOS4: ave. no. of healthy looking spikes  $(m^2)$ , <sup>5</sup>AY: yield obtained from combine harvester, <sup>6</sup>YL: yield loss=(NOS2×SW2)+(NOS3×SW3)+(NOS4×SW4), <sup>7</sup>PY=Potential yield= NOS1 × SW5, <sup>8</sup>SW2: distorted spike weight, <sup>9</sup>SW3: dried spike weight, <sup>10</sup>healthy looking spike weight, <sup>11</sup>healthy spike weight.

Table 2. Student t-tests for number of grain spike<sup>-1</sup> and grain weight spike<sup>-1</sup>

| Number of grain spike <sup>-1</sup> |   |         |                     | Spike weight |   |        |                     |
|-------------------------------------|---|---------|---------------------|--------------|---|--------|---------------------|
|                                     | 1 | 2       | 3                   |              | 1 | 2      | 3                   |
| 1                                   |   | 6.783** | 12.92**             | 1            |   | 6.89** | 11.36**             |
| 2                                   |   |         | 1.066 <sup>ns</sup> | 2            |   |        | 0.793 <sup>ns</sup> |
| 3                                   |   |         |                     | 3            |   |        |                     |

ns: p≥0.05 statistical significance.\*\*: p≤0.01 Statistical significance, 1.distorted spike, 2.healthy looking spike, 3.healthy spike.

**Table 3.** LSD groups for marketing prices (\$ ton<sup>-1</sup>) for increasing ratios of *A.tritici* contaminated grains of durum wheat.

| Contamination ratios | Means (\$ ton <sup>-1</sup> ) |
|----------------------|-------------------------------|
| Pure grains          | 335.09 <sup>a</sup>           |
| 5 % contamination    | 319.20 <sup>b</sup>           |
| 10 % contamination   | 314.56 <sup>°</sup>           |
| 15 % contamination   | 307.94 <sup>d</sup>           |
| 20 % contamination   | 297.35 <sup>e</sup>           |

CV% = 0.87 LSD = 5.549.

grains received the highest marketing price offer ( $335.09 \text{ ton}^{-1}$ ) and decreased for the grain lots having 5, 10, 15 and 20% contaminations giving 319.20, 314.56, 307.94 and  $297.35 \text{ ton}^{-1}$  respectively. The relation between marketing price offers and contamination ratios with seed galls was significant (y =  $514.7 - 13.10x^{**}$ ; F =  $93.28^{**}$ ; R<sup>2</sup> = 80.2%) (Figure 1).

Quality analysis results for distorted, healthy looking and healthy spikes and the seed lots with various ratios of contamination with seed galls are given in Table 4. Vitreousness (%) was the same for different categories of tagged spikes a, b, c and d. (98%). 1000 kernel weight was highest for d and the lowest for a as observed for 1000 kernel weights. Crude protein (%) was the highest for category b and increased slightly in the seed lots from f to i with increasing amount of seed galls as observed for protein content (%), and SDS value. Hectoliter weight of grains obtained from healthy looking spikes was 80 kg, however, there was no adequate amount of grain to weight hectoliter weight for the grains of infected spikes.

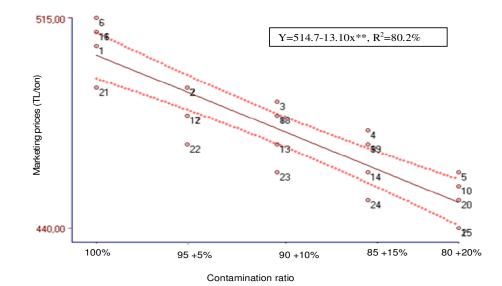
### **DISCUSSION AND CONCLUSION**

The infestation rate of *A. tritici* excluding healthy looking spikes in durum wheat (cv. Fuat bey 2000) was 16.21%.

Table 4. Quality analysis of tagged spikes and grain lots with increasing ratios of A.tritici.

| Samples  | 1000 K.W <sup>a</sup><br>(g) | 1000 K.W (g)(At dry<br>matter) | Protein <sup>b</sup><br>(%) | Protein (%)<br>(At dry matter) | SDS <sup>c</sup><br>(ml) | Vitreousness <sup>d</sup><br>(%) |
|--|------------------------------|--------------------------------|-----------------------------|--------------------------------|--------------------------|----------------------------------|
| °Tagged spikes:  |                              |                                |                             |                                |                          |                                  |
| a. 100 %Pure grains  | 48.72                        | 44.92                          | 14.56                       | 15.80                          | 19.00                    | 98                               |
| b. Healthy grains of distorted spikes                          | 53.38                        | 49.48                          | 15.68                       | 16.95                          | 20.00                    | 98                               |
| c. Healthy grains of healthy looking spikes                    | 56.39                        | 52.38                          | 15.52                       | 16.71                          | 20.00                    | 98                               |
| d. Healthy grains of healthy spikes                            | 56.90                        | 52.56                          | 14.85                       | 16.08                          | 22.00                    | 98                               |
| <sup>f</sup> Seed lots prepared for marketing price estimation | ons                          |                                |                             |                                |                          |                                  |
| e. 100% Pure grains(= a)                                       |                              |                                | 14.60                       | 15.77                          | 19.00                    |                                  |
| f. 95%+5% contam.  |                              |                                | 15.39                       | 16.61                          | 20.50                    |                                  |
| g. 90%+ 10%contam.   |                              |                                | 15.43                       | 16.63                          | 20.50                    |                                  |
| h. 85%+ 15% contam.  |                              |                                | 15.46                       | 16.62                          | 21.00                    |                                  |
| i. 80%+20% contam.   |                              |                                | 15.52                       | 16.69                          | 21.50                    |                                  |

<sup>a</sup>William et al.,1986; <sup>b</sup> Anonymous, 2002a; <sup>c</sup> anonymous 1992; <sup>d</sup> anonymous 2002b<sup>e</sup> 100% pure grains were obtained from thresher output for unit area spikes , dockage cleaned and other grains were separated by hand, others (b, c and d) were obtained spike base; <sup>f</sup> Grains of a(= e) were added increasing amount of grains with galls and f, g, h and 'i' were established.



**Figure 1.** Regression line marketing price versus contamination ratios of *A.tritici* and 95% confidence interval. 1USD\$ = 1.51TL.

Similar or even more infestation ratios were found by Ağdaci and Efe (1986) and Elmali (2002) in Marmara and Central Anatolia in Turkey. However the findings of Yüksel et al. (1980) and İmren (2007) were significantly 14.39% contamination was observed under natural infection immediate to threshing. But actual infection ratio accounting harvesting operation losses was much higher than that of grain lot score. Yield loss (32%) obtained from this study was higher than those of Yüksel et al. (1980), Sasser (1987), Magbool (1988), Stephan (1988) and Elmali (2002) but lower than that of Öztürk and Enneli (1993). Some similarities were observed in terms of yield losses with the research findings of Akhiyani (2001). This could be both due to the methods employed and seasonal effects. 1000 kernel weight of pure grains, healthy grains of infected, healthy looking and healthy spikes increased as the infection of spikes decreased. Healthy grains of infected spikes possessed the highest protein content (%) due to shriveled grains. Increase in both crude protein content (%) and SDS values of seed lots prepared for marketing price estimations (100% pure grains; 95% pure +5% contamination with galls; 90% pure + 10% contamination, 85% pure + 15% contamination; 80% pure + 20% contamination) can be attributed possibly to the presence of A. tritici and associated galls as additional sources of protein. A.tritici infection resulted in some reductions on 1000 kernel weight however other quality parameters under study were not affected. Şanliurfa commodity market is a quite guality conscious (Özberk et al., 2005 a, b) with grain purchase being affected by some measurable characters such as 1000 kernel and hectoliter weights. Damaged kernels can be easily identified by visual observation; however the cause is not always clear. Many grain purchasers call such grains as 'hard bunt' or 'iron bunt' grains, and those having similar density with healthy grains can not be easily separated by floating in water tanks before boiling in bulgur industry. Furthermore while processing cracked wheat grains with galls after boiling and drying processes in bulgur production appear as inert materials thus reducing the bulgur marketing price. As a result farmers are offered less marketing price for the grains with seed galls in market place. The higher coefficient of determination for the relation between the marketing prices with various degrees of ear cockle infestation provides supporting evidence.

South–East Anatolia is known as the durum wheat belt of the country. Southern part of region including Şanliurfa is the center of durum wheat production and commerce with one million tons of annual production. Due to the tough seed hygiene measures taken by government *A. tritici* has been excluded from internal quarantine list in Turkey recently (İmren, 2007). Farmers in the region are encouraged by government giving extra subsidy to use certified seeds in commercial crop production certified seed use increased sharply in the region. Despite the efforts, A *.tritici* first occurred in the south part of Harran

Plain (Akçakale) almost 7 to 8 years ago owing to seed imported from the west without having any internal quarantine measure. Viranşehir region got A .tritici 3 to 4 years ago through non controlled seed transfer. Plant pathologists in the region are very much aware of the presence of nematodes such as H. avenae, Pratylenchus spp., Angiuna tritici and Merlinus brevidens. .A.tritici contamination was about 3.6% of target study area (Anonymous, 1995) in the region. Irrigation has boosted the acreage contaminated by such nematodes. There are several control measures such as two-year crop rotation (Aytan and Ediz, 1978) with non host crops, one year full fallow land implementation (Limber, 1976; Paruthi and Gupta, 1987; Öztürk and Enneli, 1993), spring sowing (Yüksel et al., 1980), chemical control by Sulphur dioxide (SO<sub>2</sub>) (Kausar et al., 2005), growing of resistant varieties (Saleh and Fattah, 1990; Shahina et al., 1989; Fattah, 1990; Swarup and Sosa-Moss, 1990) and seed hygiene (Sing and Agrawal, 1987: Swarup and Sosa Moss, 1990).

The negative effect of *A.tritici* on grain yield, marketing price and 1000 kernel weight cannot be neglected, and the farmers in the region must be encouraged to follow crop rotation with non host crops and use certified seeds. It also becomes highly essential that the nematode must be monitored carefully, and the appropriate measures such as reintroduction of quarantine measures must be taken rapidly. Quarantine measures across WANA region must be taken by the governments to prevent large scale damage due to *A.tritici*.

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