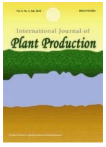

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Farmer's seed sources and seed quality: 2. seed health

Z. Bishaw[a,*], P.C. Struik[b], A.J.G. Van Gastel[c]

[a]*Seed Section, ICARDA, P.O. Box 5466, Aleppo, Syria.*
[b]*Center for Crop Systems Analysis, Wageningen University, Wageningen, The Netherlands.*
[c]*Harspit 10, 8493KB, Terherne, the Netherlands.*
*Corresponding author. E-mail: z.bishaw@cgiar.org

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Abstract

The study assessed the health quality of wheat (*Triticum aestivum* L.) and barley (*Hordeum vulgare* L.) seed samples collected from formal and informal sector in Ethiopia and Syria. In Ethiopia, several seed-borne fungi were found on wheat samples: *Cochliobolus sativum*, *Fusarium avenaceum*, *F. graminearum*, *F. nivale*, *F. poae* and *Septoria nodorum*. *C. sativum* was predominant with 84% of samples infected (frequency) and 1.85% mean infection level (rate) followed by *F. graminearum* with 74% and 1.54%, respectively. Certified seed consistently showed less infection for most seed-borne pathogens. In Syria, 68% and 14%, respectively, of wheat samples were infected with common bunt (*Tilletia* spp) and loose smut (*Ustilago tritici*). Mean loose smut infection rate was 0.79%. In barley, 85% of samples were infected with covered smut (*Ustilago hordei*) and 83% with loose smut (*Ustilago nuda*). Mean loose smut infection rate was 18%. Wheat seed health was better than of barley in terms of frequency and rate of infection. In Ethiopia, significant difference (P<0.001) in infection levels was detected for most pathogens from different seed sources, but not in Syria. There were significant differences (P<0.001) in mean infection levels across regions and districts for both crops in Ethiopia and Syria. All seed samples infected with loose smut of wheat or barley were in excess of minimum standards for seed certification across West Asia and North Africa, showing fundamental weaknesses in seed health from both formal and informal sources. National seed programs should set realistic standards and introduce routine testing to produce healthy seed.

Keywords: Wheat; Barley; Seed system; Seed health; Ethiopia; Syria.

Introduction

Seed health is one of the most important attributes of seed quality. Seeds can serve as a vehicle for dissemination of plant pathogens, causing serious disease outbreaks. Infected seeds may fail to germinate, produce abnormal seedlings and have low germination or low seedling vigor affecting grain yield and quality. Rennie et al. (1983) reported that glume blotch (*Septoria nodorum*) reduce both laboratory germination and seedling emergence of wheat, particularly at low temperatures. Wheat seed infected by Karnal bunt (*Neovossia indica*) either fail to germinate or produce abnormal seedlings (Singh, 1980; Singh and Krishna, 1982). Loose smut-infected wheat plants produce fewer tillers and reduced tiller height (Agarwal and Gupta, 1989). About 5-7% yield loss was reported for common bunt in West Asia and North Africa (Mamulk, 1991) and in Ethiopia (Niemann et al., 1980). Gorfu et al. (2012) cited yield losses from 1-25% for wheat seed-borne diseases in Ethiopia. Infection with certain pathogens causes discoloration and shriveling of seed, reducing grain quality (Agarwal, 1986; Wiese, 1987; Mathur and Jørgensen, 1992). *F. graminearum* causes head scab, a devastating disease leading to reduced seed quality and vigor, discolored and shriveled kernels, reduced seed weights and yields; and infected grains produce mycotoxins (deoxynivalenol (DON) with greater health risk for humans and livestock (Windels, 2000; Rosewich et al., 2002).

Seed certification has become valuable among other means in prevention of seed-borne diseases and produce healthy seed. Consequently, seed health standards are prescribed for certification (Besri, 1983; Diekmann, 1993; Kashyap and Duhan, 1994). However, almost all countries have standards for purity and germination, while only a few countries have standards for seed health certification (ICARDA, 2002). Moreover, even in countries where such standards do exist, it remains incomplete lacking either field or seed health standards (ICARDA, 2002; Bishaw, 2004).

Several seed-borne diseases were reported on wheat and barley from Syria (Azmeh and Kousaji, 1982; Mamluk et al., 1990; Mamluk, 1991; Mamluk et al., 1992; El-Ahmed, 1999; Kyali et al., 2010) and on wheat in Ethiopia (Neimann et al., 1980; Abate, 1985; Gebremariam et al., 1991; Gorfu et al., 2012). However, there is limited information on wheat and barley seed quality in general and seed health in particular from the formal and informal sectors (Bishaw, 2004; Bishaw et al., 2010). Therefore, the main objectives of this study were to assess the status of health quality of seeds obtained from formal and informal sources and the distribution of seed-borne diseases in different regions.

Materials and Methods

A total of 304 wheat farmers in Ethiopia and 206 wheat and 200 barley farmers in Syria were interviewed as part of seed system studies (Figures 1 a,b). A stratified sampling procedure followed by random sampling of farmers, as described by Bishaw et al. (2011), was employed. In Ethiopia, four regions were included covering nine districts and 81 villages. In Syria, three provinces were included, covering six districts and 59 villages. Farmers were interviewed about seed sources, perception of seed health and seed management practices. Finally one kg sample was collected from each farmer from the seed lot planted or intended for planting, for laboratory analysis.

a b

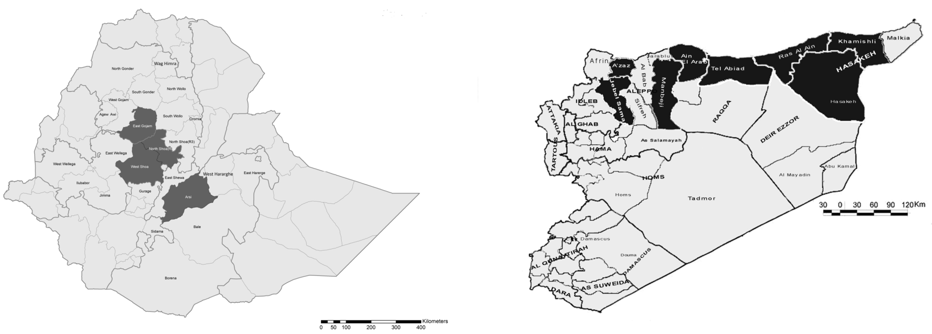

Figure 1. Wheat seed system study areas (shaded) in Ethiopia (a) and Syria (b).

Laboratory Tests

All wheat and barley seed samples collected during the survey were analyzed for seed health (ISTA, 1984). Seed health tests were conducted at Holetta Agricultural Research Center for seed samples from Ethiopia, and at ICARDA for seed samples from Syria.

Freezing blotter test

A deep-freezing blotter test was used to assess infection with *Fusarium* spp. and *Cochliobolus sativum* (ISTA, 1984). Four replicates of 100 seeds from each sample were planted in germination boxes using a blotter/pleated paper and incubated in a germinator for 24 hours at 20 °C; and transferred to a deep freezer set at -18 °C overnight. The seeds were further incubated at 20 °C for seven days under near UV light in alternating cycles of 12 hours light and darkness to stimulate sporulation. After seven days, the infected seedlings were counted and the pathogens were identified on the basis of their spore morphology.

Agar plate test

The agar plate method was used to assess incidence of glume blotch (*Septoria nodorum*) (ISTA, 1984). Four hundred seeds from each sample were pretreated with 1% aqueous solution of NaCl for 10 min and then rinsed with sterilized water. Seeds were plated on Petri dishes and incubated at 22 °C for 7 days with 12 hours of alternating cycles of daylight and darkness. After seven days, the seeds were further incubated for four days in 12-hour cycles of darkness and near ultra violet light. Using pycnidia count and colony characters, the seeds were assessed for glume blotch infection.

Centrifuge wash test

The centrifuge wash test was conducted to assess common bunt (*Tilletia* spp.) for wheat and covered smut (*Ustilago hordei*) for barley (ISTA, 1984). From each sample, 50 g seed of four replicates was sampled. Each sample was mixed with 50 ml sterilized water in a conical flask and 0.15% liquid soap was added to suspend the spores. The mixture was shaken for 5 min using a rotary shaker and centrifuged at 2000 rpm for 10 min. The water was decanted and 0.5 ml of water added to the remaining suspension. A few drops of the suspension were transferred to a counting chamber. The spore load was counted under a compound microscope and number of spores per g of seed was determined. In Syria, however, 4 replicates of 100 seeds each were used following the procedures of the ICARDA's Seed Health Laboratory to detect low level contamination of spores (Siham Assad, personal communication, 2000).

Embryo count test

An embryo count method was used both for detection of loose smut of wheat (*U. tritici*) and barley (*U. nuda*) (ISTA, 1984). A working sample of

2000 seeds from each sample was soaked in 1 liter 5% fresh solution of NaOH containing 200 mg/liter tryphan blue at 20 °C for 24 hours. After soaking, the sample was washed in warm water and agitated to facilitate the separation of embryos and passed through 2.5 mm and 1mm mesh sieves to collect the endosperm and the embryo, respectively. The sample was transferred to a funnel closed with a rubber tube and stop-cork, and covered with a mixture of lactophenol and water. Finally, the embryos were transferred into fresh lactophenol and boiled for 30 sec. Embryos were immersed in fresh glycerol, arranged in rows in grooved perspex plates and examined using stereomicroscope with sub-stage illumination. Embryos with loose smut mycelium were recorded as infected (ISTA, 1984) and the percent infection was calculated based on the number of embryos examined.

Wheat gall nematode

Two replicates of 50 g each from each sample were taken and examined under a stereomicroscope and seeds with nematode galls were separated and counted (ISTA, 1984). To confirm the presence of the nematodes, suspected seeds were soaked in water for about 1h and then cut open. The materials were transferred to clean water and were observed under microscope to record mobile and infective juvenile nematodes. The frequency and rate of infection were determined by the following formulae:

$$\text{Frequency of infection} = \frac{\text{Number of samples with infection}}{\text{Number of samples collected}} \times 100$$

$$\text{Rate of infection} = \frac{\text{Number of seeds infected}}{\text{Number of seeds tested}} \times 100$$

Field Experiments

In Ethiopia, wheat was planted at 30 g per plot (6 rows × 2.5 m length × 0.2 m between rows) on 3 and 4 July 1999 at Gonde Basic Seed Farm (8° 02' N and 39° 10' E) where the soils are classified as an intergrade (between an eutric Nitosol and a luvic Phaeozem). Fifty samples of 14 bread [6 modern (*ET13, HAR1685, HAR1709, HAR710, K6295, Pavon76*), 3 obsolete (*Batu, Dashen, Kenya*) and 5 landraces (*Goli, Israel, Menze, Rash* *and Zombolel*) and 11 durum (1 modern (*Boohai*) and 10 landraces (*Baunde, Baherseded, Enat-sende, Gojam-gura, Gotoro, Local, Key-sende, Legedadi, Nech-shemet, Shemet*)] wheat varieties/landraces were planted and emergence recorded.

In Syria, wheat was planted at 60 g for bread and 70 g for durum on plot of 5 m^2 on 4 and 6 January 2000 at Tel Hadya (36° 01′ N and 36° 56′ E) on soil with fine clay (montmorillonitic) of pH from 7.9 to 8.2. Sixty seed samples representing 6 bread [4 modern (*Bohouth4, Bohouth6, Cham2, Cham4*), 1 obsolete (*Mexipak*) and 1 landrace (*Hamari*)] and 12 durum [7 modern (*Acsad65, Bohouth5, Cham1, Cham3, Cham5, Gezira17 and Lahan*) and 4 landraces (*Bayadi, Hamari, Hourani, Swadi*)] wheat varieties/ landraces were planted and emergence was recorded. Barley was planted at 50 g on plot of 5 m^2 in December 1998. In all experiments, seedling emergence was measured on 1m^2 twice, first when emergence was stabilized and two weeks after the first count. A randomized complete-block design with three replications was used for field experiments.

Data Analyses

The statistical analysis of the data from the laboratory tests was based on a completely randomized design using Genstat 6.1 statistical package and the LSD was used to find significance between tests and to separate the means among different treatments. Simple Pearson correlation coefficients were calculated to quantify the association among different seed-borne diseases with laboratory tests and field emergence.

Results and Discussion

Wheat Seed Health in Ethiopia

Cochliobolus, Fusarium and Septoria spp

Different seed-borne fungi (*Cochliobolus, Fusarium* and *Septoria* spp.) were identified from seed samples collected from major wheat production regions, which revealed their widespread distribution (Table 1). There was a significant difference in health quality of samples from different regions and districts. Wheat seed samples from Arsi consistently showed higher levels of infection than other regions.

Table 1. Wheat seed health quality (% mean infection rates) of samples collected from different districts in Ethiopia.

Districts	*Cochliobolus sativum*	*Fusarium avenaceum*	*Fusarium graminearum*	*Fusarium nivale*	*Fusarium poae*	*Septoria nodorum*
Gedeb	2.90	0.91	2.58	0.37	1.26	0.47
Munesa	2.18	1.07	1.96	0.23	1.57	0.61
Hetosa	2.47	0.75	2.48	0.04	0.98	1.14
Dodota	1.88	1.18	1.85	0.19	1.10	0.73
Dendie	1.62	0.19	1.28	0.07	0.84	0.70
Chelia	1.88	0.32	1.37	0.15	0.78	0.46
Ensaro Wayu	0.91	0.08	0.71	0.52	0.51	0.00
Hulet Eju	1.32	0.39	0.79	0.13	0.63	0.05
Machakal	0.45	0.11	0.36	0.2	0.43	0.00
Mean	1.85	0.56	1.58	0.21	0.90	0.50
Significance	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

From 304 wheat samples tested for seed health, infection was as follows: 84% of seed samples (*C. sativum*), 31% (*F. avenaceum*), 74% (*F. graminearum*), 13% (*F. nivale*), 52% (*F. poae*) and 31% (*S. nodorum*). Infection with *C. sativum* was the highest across the regions and districts where 84% of the seed samples were infected with a mean infection rate of 1.85%. The highest infection rate was observed on seed samples from Gedeb, Munesa and Hetosa districts in Arsi (southeastern) followed by samples from Hulet Eju and Machakal districts in Eastern Gojam (northwestern) of Ethiopia. Tanner (1994) also recorded the highest infection of *C. sativum* from samples collected from northwestern Ethiopia.

Five *Fusarium* spp. were isolated from wheat seed samples and occasionally with more than one species from each sample. *F. graminearum* was predominant both in frequency (74%) and rate (1.58%) of infection, followed by *F. poae, F. avenaceum* and *F. nivale* (Table 1). The results indicated relatively higher infection from samples in southeastern Ethiopia, which is consistent with earlier reports (Tanner et al., 1990; Gebremariam et al., 1991). Bekele and Kar (1997) also found high levels of *Fusarium* infection of wheat seeds from different regions of Ethiopia. Mean infection rates with *F. avenaceum* was relatively low, though earlier studies reported up to 20% infection in wheat (Abate, 1985). The prevalence of *F. graminearum* was also reported from wheat seed samples elsewhere (Hajihasani et al., 2012; Mobasser et al., 2012).

The frequency (31%) and mean infection rate (0.5%) was the least for glume blotch (*S. nodorum*) compared to other seed-borne pathogens except *F. nivale*. Wheat samples from different districts showed infection with glume blotch except from Ensaro-Wayu and Machakal districts. Tanner (1994) also identified no glume blotch except on two wheat seed samples collected from north-western Ethiopia. In Morocco, 20% of non-certified seed was infected by glume blotch, with an infection level of up to 1% (Besri, 1983).

The health of wheat seed samples obtained from different sources is presented in Table 2. Infection by seed-borne diseases was observed across different seed sources and appeared to be significant except for *F. avenaceum*. The infection rate for seed lots from the formal sector was consistently lower than from other sources except for *F. nivale* and *F. poae*. Abate (1985) and Bekele and Kar (1997), however, reported that infection with certain *Fusarium* spp. from farmer's fields were lower than that from research stations or state farms where commercial seed is produced. Mean infection with *S. nodorum* was low for certified seed compared with seed obtained from informal sector and significant. In summary, the health of certified seed was consistently better than own-saved seed which was followed by seed obtained from other farmers and then from local markets for *C. sativum*, *F. avenaceum*, *F. graminearum*, *F. poae* and *S. nodorum*.

Table 2. Wheat seed health quality (% mean infection rates) of samples collected from different sources in Ethiopia.

Seed sources	*Cochliobolus sativum*	*Fusarium avenaceum*	*Fusarium graminearum*	*Fusarium nivale*	*Fusarium poae*	*Septoria nodorum*
Formal sector	1.63	0.47	1.45	0.32	0.95	0.10
Farmer/neighbors	2.47	0.52	2.03	0.09	1.34	0.56
Markets/traders	2.32	0.63	2.0	0.03	1.05	1.10
Own-saved seed	1.8	0.57	1.55	0.22	0.86	0.49
Mean	1.85	0.56	1.58	0.21	0.90	0.50
Significance	<0.001	0.83	<0.008	<0.051	<0.007	<0.001

Loose smut (Ustilago tritici)

Although, the frequency and rate of infection with loose smut of wheat was low (Table 3), it was found across regions and districts of the country. About 34 samples (11%) were infected with loose smut. Loose smut infection was reported previously in Arsi, Gojam, and North Shoa (Gebremariam et al., 1991). No infection was reported from certified seed, but one sample each for seed obtained from neighbors and traders, and the remainder from own saved seed. All infected samples (11%) had loose smut infection in excess of the standard for any category of certified seed; and 4% of samples had an infection level ≥1%. Infection was observed on modern bread wheat varieties (*Batu, Dashen, ET13, HAR710, Pavon76*) and durum wheat landraces (*Baherseded, Shemet*) showing their susceptibility to loose smut. Abdelfattah (1994) also reported loose smut infection with significant difference between certified and non-certified wheat seed in Jordan.

Table 3. Wheat seed samples with loose smut, common bunt and gall nematode infections (%) in Ethiopia.

Districts	Loose smut		Common bunt		Gall nematode	
	No. of samples infected (%)	Mean infection (%)	No. of samples infected (%)	Spores per g of seed	No. of samples infected (%)	Mean infection (%)
Gedeb	12	0.08	5	5.79	17	1.02
Munesa	23	0.25	0	0	18	0.93
Hetosa	4	0.05	2	0.52	23	1.48
Dodota	7	0.07	0	0	3	0.17
Dendi	13	0.10	7	54.6	7	0.29
Chelia	11	0.09	0	0	0	0
Ensaro-Wayu	13	0.10	5	5.46	3	0.16
Hulet-Eju	12	0.19	0	0	0	0
Machakal	21	0.09	0	0	0	0
Total (% infection)	11		2		9	
Mean		0.10		7.1		0.50
Minimum		0		0		0
Maximum (no of teliospores)		2.1		(1400)		13.5

Common bunt (Tilletia spp)

The number and rate of infection was extremely low for common bunt (Table 3) compared to previous surveys. No infection was observed on certified seed, but seven seed samples (2.3%) from informal sources were contaminated with bunt. Three wheat seed samples from Arsi, two each from West Shoa and North Shoa had bunt contamination, with no infection from East Gojam. Tanner (1994), however, found that most of the wheat seed samples from northwestern Ethiopia contained bunted kernels.

Mean bunt contamination from 45 to 1400 spores per 1 g of seed was observed on obsolete bread varieties in Arsi and on durum landraces in West Shoa and North Shoa. The highest mean spore load was observed on a landrace (*Baunde*) with 1400 spores per 1 g of seed. Abate (1985), however, reported 49, 1623 and 1554 spores per seed from samples collected in Arsi, West Shoa and Gojam, respectively. In Iran, Hajihasani et al. (2012) reported 7.1% average infection rate for *T. laevis* and *T. tritici* with 1-48 spores per seed while Hamidi et al. (2010) found 1.2 spores per seed for *T. laevis.*

Neimann et al. (1980) concluded that under favorable environments, a natural bunt contamination of 100 spores per sample (approx. 10 mg spores/ 100 g seeds) could cause 70% or more infection, whereas under unfavorable conditions or in moderately susceptible wheat varieties up to 1000 spores/ sample might be required. Besri (1983) reported that bunt contamination with 500 and 1500 spores/seed resulted in 0.5% and 21% infection in durum wheat depending on environmental conditions and varietal susceptibility.

Gall nematode (Anguina tritici)

Although, the frequency and rate of infection with gall nematode was low infection was reported across different regions and districts of the country (Table 3). Twenty six (9%) wheat seed samples were infected, with a mean gall nematode count of 0.5 per 50 g of seed. All samples infected were from the informal sources, except one sample of certified seed; and 23 samples were from the Arsi region. Gall nematode infection was observed on modern bread wheat varieties (*Batu, ET13, HAR710, Pavon76*) and durum landrace (*Gotoro*). Wheat gall nematode appears a major seed-borne disease and earlier surveys reported high infection rates in the same region or elsewhere in the country (Gebremariam et al., 1991).

Association between seed health and quality attributes

A significant negative correlation ($P<0.01$) was observed between germination and infection with seed-borne pathogens, such as *C. sativum, F. graminearium* and *S. nodorum* (Table 4). It is evident that wheat seed samples infected with these seed-borne pathogens had lower germination and increased abnormal seedlings and dead seeds which were significant.

The association of *C. sativum, F. graminearium* and *S. nodorum* with seedling abnormalities, dead seeds and reduced germination indicates their effects on seed quality. Mobasser et al. (2012) found negative and significant correlation between loose smut and germination, but not with common bunt and *F. graminearum*.

Table 4. Simple correlation coefficients between germination, vigor and health quality and field emergence of wheat seed in Ethiopia.

Laboratory tests and field emergence	CS	FA	FG	FN	FP	SN	LS	AT
Standard germination	-0.25**	-0.04	-0.20**	0.09	-0.04	-0.17**	0.03	-0.08
Abnormal seedlings	0.17**	0.01	0.18**	-0.07	0.01	0.18**	-0.03	0.08
Dead seeds	0.28**	0.06	0.19**	-0.09	0.06	0.13**	-0.02	0.08
Speed of germination	-0.18**	-0.02	-0.12**	0.12**	-0.06	-0.17**	0,03	-0.03
Seedling shoot length	0.06	-0.02	0.09	-0.04	0.11	-0.09	-0.02	-0.16
Seed ling root length	-0.19	-0.03	-0.15	-0.03	-0.11	-0.12	-0.01	-0.04
Seedling dry weight	-0.13	-0.15	0.05	-0.13	-0.09	0.02	-0.01	-0.01*
Field emergence	-0.38**	-0.09	-0.32**	0.01	-0.35**	-0.27**	0.12	-0.01

Note: *, ** Significant at $P<0.05$ level and $P<0.01$, respectively; CS=*C. sativum; FA= F. avenaceum; FG= F. graminearum; FN= F.nivale; FP= F. poae; SN= S. nodorum.*

A significant negative correlation was observed between speed of germination and field emergence and infection with *C. sativum*, *F. graminearum* and *S. nodorum.* The result showed a negative effect of seed-borne diseases on seed vigor including speed of germination and field emergence similar to what is reported for *S. nodorum* (Rennie et al., 1983) and *F. graminearum* (Windels, 2000). In our findings none of the pathogens investigated showed significant correlation with seedling vigor contrary to what were reported earlier for other fungal pathogens (Tanner and Mwangi, 1992; Tanner, 1994).

Farmers' perception of seed health quality

Farmers identified plant diseases as major constraints for wheat production; and 77% indicated rusts compared to only 7.4% for smuts. Farmers experienced periodic outbreaks of rust epidemics with devastating consequences, particularly in Arsi region where modern bread wheat varieties are widely grown. In contrast, a few farmers perceived seed-borne diseases, such as smuts, common bunts and gall nematodes, as important, which could be attributed to less disease incidence, the difficulty to recognize the diseases or minor losses incurred in crop production. Our results also revealed that only 11.2, 8.6 and 2.3% of the samples were infected with smut, gall nematode and bunt respectively, though they occurred across regions which were consistent with previous surveys. Moreover, infection rates with *C. sativum, S. nodorum* and *Fusarium* spp. were also to too low for farmers to detect their impact on crop production.

In Ethiopia, the study on wheat seed health can be summarized as follows. First, important seed-borne diseases were found across major wheat production areas with higher infection levels in southeastern as compared with central and northeastern regions with significant difference in mean infection rates among districts and regions. Second, certified seed consistently has lower number and rates of infection compared with non-certified seed and were significant for some seed-borne diseases. Third, the low level of occurrence of common bunt, loose smut and gall nematode in wheat seed samples could be attributed to the environmental conditions under which the crops were grown prior to the survey year which might have influenced disease infection. Fourth, infection with wheat seed-borne pathogens was observed both on modern varieties and landraces showing their susceptibility. Fifth, although chemical seed treatments have been identified against some seed-borne diseases, neither certified seed destined for small-scale farmers is treated nor do farmers practice on-farm seed treatment. Sixth, apart from purity and germination, no regular health tests were conducted for certified seed. Incidentally wheat seed production is concentrated in southeastern Ethiopia where higher-level infection was reported in this and earlier studies. Cognizant of the threats of seed-borne diseases, appropriate measures need to be undertaken to produce healthy seed through integrated seed health management practices.

Wheat and Barley Seed Health in Syria

Loose smuts (U. tritici and U. nuda)

Wheat and barley seed samples collected from different agro-climatic zones, regions and districts showed significant differences ($P<0.001$) in seed health, except for loose smut of wheat from different zones (data not shown). In wheat, only 13.6% of samples were infected with loose smut with mean infection rate of 0.79%. Wheat samples from zone 1 (more rainfall) had a relatively higher loose smut infection rate (0.85%) compared with samples from zone 2 (0.76%), although not statistically significant, showing the association between rainfall and disease infection. Infection with loose smut appeared to be localized and was found on samples from Hasakeh (5.3%) and Khamishli (8.3%) districts. In both cases, the infection rate was more than most standards for certified seed2 (ICARDA, 2002).

In barley, 83% of seed samples were infected with loose smut with mean infection rate of 17.9% which was significant ($P<0.001$) among different agro-climatic zones and regions. Average infection levels were 24.2, 14 and 8.5% in zone2, zone3 and zone4, respectively showing decreasing trend with declining rainfall patterns. At the district level, samples from Ain El-Arab had the highest infection level, followed by samples from Al-Bab and Manbeji (Figure 2).

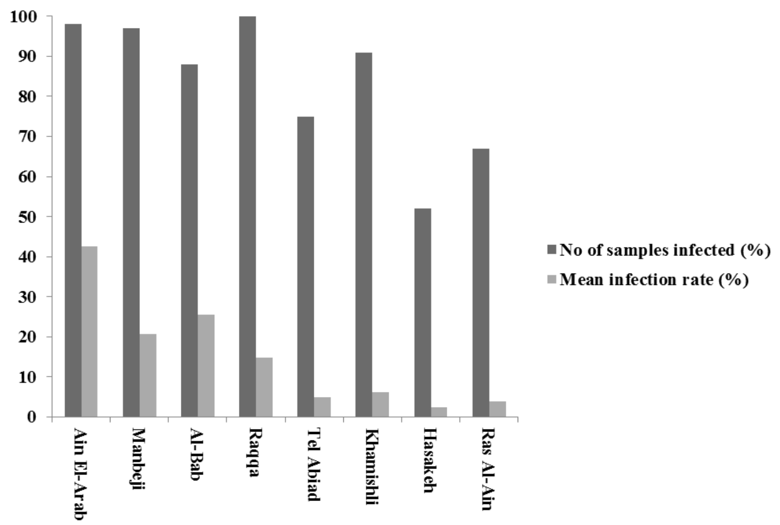

Figure 2. Barley seed health quality of samples collected from different districts in Syria.

The frequency and rate of wheat loose smut infection was not significantly different among different seed sources. About 10-15% of samples from different sources (formal and informal) were infected by loose smut. The lowest average infection level was found on own saved (0.14%), followed by seed from the formal sector (0.22%), farmers (0.52%) and traders (1.08%). We found that all wheat certified seed and non-certified seed were treated with chemicals before planting in Syria. The widespread use of chemical treatment could possibly have contributed to the relatively low infection with loose smut.

In barley, all seed samples were non-certified and there was no significant difference in loose smut infection among different sources. All infected samples from different seed sources had more than 0.5% infection level. In low-input agriculture where barley is the main crop, most farmers did not use certified seed and/or chemical treatment (Bishaw, 2004; Bishaw et al., 2011) which might have contributed to high level loose smut infection.

Although most countries ins WANA region had no seed health certification standards (ICARDA, 2002), a few countries have established standards as strict as allowing a maximum of 0.05% seed infection, whereas others still allow 0.2% infection for certified seed 2. If seed health standards for both wheat and barley are relaxed to 0.2%, none of the loose smut-infected samples could meet this standard. It is worth noting that wheat loose smut infection of certified seed found in this study and reported elsewhere (Besri, 1983; Abdelfattah, 1994; Bishaw, 2004) showed the ineffectiveness of certification schemes in producing healthy seed.

Common bunt (Tilletia spp)

About 68% wheat seed samples were contaminated with common bunt spores (*T. caries* or *T. foetida*) across different regions, districts and sources. The mean count per sample was 12 spores removing samples with extremely high spore count (12.6%). Wheat seed samples with 1-5 spores, 6-10 spores, 11-50 spores or 51-100 spores per sample accounted for 14, 13, 23 and 5% of all samples, respectively. Azaz and Jebel-Saman districts had low number of wheat seed samples with bunt contamination, whereas 70-80% of samples from Ras Al-Ain were contaminated. Kyali et al. (2010) reported mean frequencies of 87.7 and 12.3%, respectively for *T. caries* and *T. foetida* in durum wheat and 19.1 and 80.9%, respectively bread wheat. Mobasser et al. (2012) found that *T. caries* and *T. leavis* as dominant species.

There was a significant difference in the number of spores per sample from different sources. The proportion of samples with common bunt contamination was 58% for wheat seed samples from the formal sector compared with 70-75% of samples from the informal sector. With the tolerance level of 5 spores per sample about 46% of samples would maintain the requirement and if this tolerance level were increased to 10 spores per sample, 59% of the wheat seed samples were within the tolerance level. However, it should be noted that both certified and non-certified seeds had spores in excess of these tolerance levels. With increased use of mechanical harvesting which spreads bunt contamination from infected fields, it would be necessary to prescribe adequate isolation distances combined with use of effective seed treatment to produce healthy seed.

Covered smut (U. hordei)

The results revealed the presence and widespread distribution of covered smut where 85% of barley seed samples had smut contamination. All samples from Aleppo and Raqqa provinces had the contamination irrespective of the agro-climatic zones. A limited number of samples were free of covered smut (15%), mainly from zones 3 and 4 in Hasakeh (10.5%) and Ras Al-Ain (4.5%) districts, from Hasakeh province. However, from samples that were not contaminated, 13% were own-saved seed. The frequency of contamination showed a widespread threat of covered smut in barley production.

Other seed-borne diseases

Flag smut (*U. agropyri)* was identified on 11 barley seed samples (5.5%) from Raqqa and Hasakeh provinces. *Fusarium* spp. and *Helminthosporium* spp. were isolated from six (3%) barley seed samples in Aleppo, Al-Raqqa and Hasakeh provinces with low infection levels (<0.75%). The number and level of infection were not high, given a few farmers using irrigation for barley production where the disease is most prevalent.

Association between health and seed quality attributes

In wheat, there was no significant correlation of loose smut infection with germination, abnormal seedlings and dead seeds. Similarly, loose smut infection did not show any significant relationship with vigor tests and field emergence, contrary to reports from Iran (Mobasser et al., 2012). In barley, loose smut infection showed a negative correlation with germination but a positive correlation with abnormal seedlings and dead seeds; both correlations were significant ($P<0.01$). In barley as in wheat, loose smut infection did not correlate with seed vigor (except for root length) and seedling emergence.

Farmers' perception of seed health quality

All barley and most wheat seed samples were obtained from informal sources. Wheat growers indicated smuts (57%), black point (9%) and nematodes (3%) as important seed-borne diseases. In barley, farmers mentioned smuts (17%) and *Abu-Elawi* (26%) as important seed-borne diseases. Among barley farmers, who reported *Abu-Elawi* (gall nematode), 22% were from Ain Al-Arab, Manbeji and Al-Bab districts, although the problem was also reported from Tel Abiad and Hasakeh districts. Earlier field surveys confirmed the widespread problem of barley head sterility and its association with gall nematode (Khatib et al., 2000).

The study revealed remarkable difference in the frequency and rate of infection of wheat and barley seed samples First, the majority of barley samples were contaminated with covered smut (85%) and infected with loose smut (83%). On the other hand, 68% of wheat samples had bunt contamination and only 13.6% had loose smut infection showing low frequency and rates of infection compared to barley. Second, in wheat, loose smut infection was observed from the Hasakeh province, whereas barley infection was observed throughout the three regions, the highest being in Aleppo province. Third, in both wheat and barley, seed samples from wetter zones had relatively higher infection rates compared with samples from drier areas and this was significant for barley loose smut infection. Fourth, wheat farmers perceived that the problem of smut less serious compared to barley growers, which confirmed their perception as revealed with health tests. Fifth, wheat farmers applied better seed management practices where almost all own-saved seed received chemical treatment compared to 7% in barley. Sixth, although chemical treatment was widely used for wheat, there was threat from loose smut infection attributed to lack of appropriate chemicals. The widespread use of mechanical harvesting exacerbated the problem of common bunt contamination. The majority of farmers surveyed grew both wheat and barley crops. However, seed health of wheat is better than of barley similar to another study on physical and physiological seed quality (Bishaw et al., 2011). Therefore, health quality of both certified seed and non-certified seed was a cause for concern in crop production.

Farmers' Seed Management and Seed Health

Farmers employed different seed management practices to ensure the quality of their seed for planting purposes (Tables 5 and 6). Seed management practices with profound effect on seed health were harvesting methods, cleaning, treatment and storage. In Ethiopia, only harvesting method had significant effect on infection with seed-borne pathogens and germination. Harvesting methods may predispose the seed to mechanical damage, chemical treatment injury, fungal invasion or storage insect infestation. Machine-harvested seed consistently gave the highest infection across different seed-borne diseases, indicating the contribution of mechanical damage to disease infection. In Syria, the widespread use of mechanical harvesting may have contributed to high level contamination with covered smut and common bunt. Harvesting methods was also significant ($P<0.001$) for loose smut infection of barley samples.

Table 5. Farmers' seed management and seed health quality (% mean infection rates) of wheat seed in Ethiopia.

Farmer's seed management	Number of samples	CS	FA	FG	FN	FP	SN	LS	AT
Hand harvesting	204	1.4	0.38	1.14	0.17	0.63	0.3	0.11	0.21
Machine harvesting	59	2.47	0.92	2.1	0.19	1.05	0.86	0.09	1.66
No seed cleaning	30	1.13	0.3	0.93	0.27	0.8	0.27	0.08	0.37
Seed cleaning	233	1.68	0.53	1.41	0.16	0.72	0.45	0.11	0.56
No seed treatment	253	1.62	0.49	1.37	0.18	0.73	0.44	0.09	0.56
Seed treatment	10	1.5	0.7	0.9	0.10	0.7	0.1	0.29	0

Note: CS=*C. sativum; FA= F. avenaceum; FG= F. graminearum; FN= F. nivale; FP= F. poae; SN= S. nodorum; AN=Anguina tritici*.

Table 6. Farmers' seed management and health quality (% mean infection rates) of wheat and barley seeds in Syria.

Farmer's seed management	Loose smut of wheat		Loose smut of barley	
	Number of farmers	%	Number of farmers	%
Hand harvesting	5	0.2	99	0.53
Machine harvesting	122	1.1	66	1.41
No seed cleaning	-	-	8	0.72
Seed cleaning	127	1.1	157	0.89
No seed treatment	-	-	154	0.89
Seed treatment	127	1.1	11	0.72

In Ethiopia, cleaned and treated seed had a lower number of samples infected or contaminated compared with non-cleaned or non-treated samples, but the difference was not significant. Cleaning improved health quality by removing shriveled seeds heavily infected with seed-borne fungi, particularly if infection occurred during crop maturity. Cleaning wheat seed lots infected with *Fusarium* spp. reported to improve germination and decrease the proportion of infected and shriveled seeds (Gutormson et al., 1993). However, the results from Ethiopia and Syria appeared to be inconsistent for different pathogens, showing the inefficiency of traditional cleaning practices.

Wheat seed samples treated by farmers showed increased germination, speed of germination and seedling length but reduced field emergence. Treated seed had low bunt contamination but slightly higher loose smut infection compared with non-treated seed because a few farmers (2.6%) used Vitavax, which is effective against loose smut infection. Abu-Yahya (1997) reported effects of seed treatment on yield and yield components and reduction of spores of common bunt-infected wheat seed lots.

In Syria, there was no significant difference in mean infection or contamination between seed stored separately or together with grain for both crops except for loose smut ($P<0.05\%$). The storage environment had an effect on physiological and health quality, particularly if seed were stored under high relative humidity, moisture content or temperature, which encouraged infection with storage fungi or infestation with storage insects reducing seed quality. Kashyap and Duhan (1994) found that insect damage on wheat seed embryo substantially reduced germination and seedling vigor, with a subsequent increase in abnormal or dead seeds.

The results on seed management practices did agree well with previous studies on physical and physiological quality of wheat and barley seeds from different sources (Bishaw et al., 2011). In principle introducing on-farm seed cleaning, seed treatment and better storage would benefit the majority of farmers using non-certified and non-treated seed from the informal sector.

Conclusion

The present study demonstrated the occurrence and the geographic distribution of major seed-borne fungi of wheat and barley in Ethiopia and Syria. The seed health quality of wheat and barley seed samples showed significant inter-regional differences (agro-climatic zones, regions, districts) in terms of the frequency and rate of infection with seed-borne pathogens. The health tests have revealed the extent of the problem of seed-borne diseases from both the formal and informal sectors. For example, all seed samples infected with loose smut of wheat or barley were in excess of the minimum standards prescribed for seed certification across the WANA region, showing fundamental weaknesses of seed health from both formal and informal sectors. It is recommended that national seed programs: establish realistic seed health standards for certified seed production; introduce integrated seed health management combining field inspection and seed quality testing; introduce need-based seed treatment for certified seed production; identify regions with less disease pressure to produce healthy seed; introduce better on-farm seed health management and create awareness among farmers; strengthen national capacity of seed quality assurance agencies through training and provision of facilities; and initiate breeding new varieties with resistance to major seed-borne diseases. It is imperative that systematic and well-coordinated seed pathology research be initiated to conduct comprehensive surveys to identify major seed-borne diseases, understand their epidemiology and rate of transmission and economic importance to set priorities on control strategies.

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