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## REVIEW ON BLACK STEM RUST OF WHEAT

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### ABSTRACT

Throughout history, *Puccinia graminis tritici*, Ug99 has been regarded as a significant danger to the production of wheat because it causes the stem or "black rust" disease. The main biotic factors limiting wheat yield are the three rust diseases that affect wheat: stem, leaf, and stripe rust. Stem rust, which can result in losses of up to 70%, is a significant threat to global food security. In cultivars that are susceptible, stem rust mostly parasitizes the surface of the leaf and stem, but it can also attack the leaf sheaths, spikes, glumes, awns, and grains. Heavy dews, high humidity, and warm temperatures (24–30 °C) are all favorable for the development of stem rust. Urediniospore production occurs in these circumstances 7–14 days following infection. Its expansion to other nations in Africa, Asia, and beyond is evident, whether caused by the wind or human intervention. The issue caused by rust can be solved by understanding the illness, locating resistance lines, and subsequently creating resistant types with the aim of shortening the disease. Cultural traditions, the use of pesticides, and the use of resistant varieties like Vijaya, Tilottama, Banganga, Gaura, Dhaulagiri, Danphe, Swargadwari, etc. are all part of management techniques.

### KEY WORDS

Wheat, Ug99, *Puccinia graminis tritici*, virulence, resistance, rust control.

Wheat (*Triticum aestivum* L.) is cultivated globally and its demand continues to increase at an annual rate of 1.6% some estimates indicate that 60% more wheat will be needed by 2050 (CGIAR, 2013). In Nepal, wheat is the third most important crop that is cultivated in the area of 703,992 hectares (MOAD, 2020). It is the world's most preferred staple food and is also a good source of B-group of vitamins, protein, minerals, and dietary fiber (Kandel et al., 2018; Shewry, 2007). In the year 2018/19, the national production of wheat is 2,005,665 metric tons and the productivity was 2.85 t/ha (MOAD, 2020). Through the intensification in the field of agriculture, the incidence of pest and pathogen are increasing day by day. New biotic threats like Ug99 of stem rust, a new risk of the wheat blast, and virulence for resistance gene Yr9 in the stripe rust pathogen have appeared. To withstand and increase wheat production, the impact of biotic and abiotic stresses on production must be reduced. Besides these stresses, the wheat rust diseases are best known for their devastating and widespread nature. Under favorable conditions, stem rust may cause yield losses up to 100 % to the susceptible varieties (Leonard & Szabo, 2005; Roelfs & Budhnell, 1985).

The obligate biotrophic pathogen *Puccinia graminis f.sp. tritici* is responsible for wheat stem rust, which may appear anywhere wheat is grown (Roelfs et al., 1992). The leaf blade, leaf sheath, stem, and spike of the plant are all destroyed by stem rust, which is arguably the most harmful of the three rusts (Eversmeyer & Kramer, 2000). After development of semi-dwarf resistant varieties in the 1960s and 1970s, there hasn't been a serious loss of it anywhere in the world (Iqbal et al., 2010). Stem rust can be avoided by resistance breeding and the removal of the alternative host barberry (*Berberis vulgaris*) close to wheat-growing regions (Kolmer et al., 1991). Stem rust is common where the temperature consistently approaches 25°C (Ravi P. Singh et al., 2015). Infected plants has leaf sheaths, stems,



glumes, and an awn all covered by elongated blister pustules (Ravi P. Singh et al., 2008). The National Wheat Research Program (NWRP) in Nepal has created improved wheat varieties that are high yielding and disease resistant, including the popular varieties such as Dhaulagiri, Tilottama, Aditya, WK1204, Vijay, Bhrikuti, and NL 971. (Subedi et al., 2019).

## METHODS OF RESEARCH

The review has been made by consulting various available journal articles, proceedings papers, annual technical reports, statistical papers, and survey papers of various research and academic institution of the government organization (GOs), Non-government organizations (NGOs), and International Organizations (INGOs). Various information published by the Ministry of Agriculture and Livestock Development was also collected in the review work. Various pathological findings were collected and categorized into different sub-contexts such as history, symptoms and epidemiology, biology and ecology, alternate host and loss assessment, management options, etc.

## RESULTS AND DISCUSSION

*Stem rust (Ug99)*. Stem or black rust is an important sporadic disease of wheat caused by the fungus *Puccinia graminis f. sp. tritici* Ericks and Henn. An obligate biotroph, *puccinia* is heterothallic and heteroecious (Alexopoulos et al., 1996). It was first observed in Uganda in 1998 (Pretorius et al., 2000) and is considered to be the main risk to global wheat production (Ravi P. Singh et al., 2011, 2015). Under the North American nomenclature system, Ug99 was designated as TTKS (R. Wanyera and M. G. Kinyua, 2006). These obligate parasites are highly specialized and have a significant variation in their population for virulence to specific resistance genes (Ydoğdu & Boyraz, 2012). Their evolution and selection occur frequently by relocation, mutation, and recombination of existing virulence genes (Ravi P. Singh et al., 2008). At present, eight Ug99 on the eastern rim of the African continent, as well as from Yemen and Iran, have been described within this race group (Sharma et al., 2013).

Table 1 – Detection of Ug99 lineage races of stem rust of wheat

| Race  | First Detected country |
|-------|------------------------|
| TTKSK | Uganda (1998/99)       |
| PTKSK | Uganda (1998/99)       |
| TTKST | Kenya (2006)           |
| TTTSK | Kenya (2007)           |
| TTKSP | South Africa (2007)    |
| PTKST | Ethiopia (2007)        |
| TTKSF | South Africa (2010)    |
| TRTTF | South Africa (2010)    |
| TRTTF | Kenya (2014)           |
| TTKTT | Ethiopia (2018)        |

Note: Adopted from Sharma et al., 2013 and Nazari et al., 2021.

*Virulence of Ug99*. Incidence of virulence of stem rust of new races in a geographic area can be linked to various phenomena such as migration and gene flow, mutation, and selection for a specific virulence and recombination (Ydoğdu & Boyraz, 2012). The emergence and spread of these new races pose a great threat as it made breeding for resistance difficult. Gene flow and migration is the basic source of variance which can occur from one place to another where a pathogen was previously absent; new genes can be established and again spread to different regions along with evolution forming new pathotypes (S. Singh, 2016). Similarly, sexual reproduction assembles unique virulence combinations through sexual recombination generating new pathotypes (Yue Jin et al., 2010). In the mid-1930s, Race 56 in North America was accountable for the major stem rust epidemic that originated from barberry (Yue Jin, 2011). Likewise, in somatic hybridization exchange of nuclei between anastomosing fungal hyphae occur (S. Singh, 2016). Gene flow



via hybridization is a quick source of the development of new genetic material and rapid evolutionary change (Brasier, 2000). The spontaneous mutation is also the main cause of new variation and development of new pathotypes that are able to overcome resistance; and these mutations may have occurred due to deletion, insertion, transpositions, and chromosome polymorphism along with chromosomal instability (S. Singh, 2016).

Infections in cereals or grasses occur mainly as orange-red pustules on leaves, leaf sheaths, and stems, but occasionally occur on leaf blades and glumes as well (Schumann & Leonard, 2000). Typically the first macroscopic symptom occurs around 8–10 days after infection; a pustule emerges by rupturing the host epidermis from the pressure of a mass of urediniospores (Zhu et al., 2014). Uredinal pustules are distinctly eruptive, with torn epidermal tissues at their margins, and are usually linear or diamond-shaped and up to 10 mm long (Komen, 2007; Leonard & Szabo, 2005). As the infection continues, later production of urediniospores stops, and then a layer of black teliospores appears, which makes the infected stem appear blackened in the later phase (Leonard & Szabo, 2005).



Figure 1 – Orange-red pustules of *Puccinia graminis* f. sp. tritici on the stem of a susceptible plant (image courtesy of Z.A. Pretorius)



Figure 2 –Ug99 infested plant. Oblong reddish brown pustules (raised blisters) are visible on stem, also on leaves. (<http://www.topnews.in/files/wheat-virus-ug99.jpg>)

**Epidemiology.** The life cycle of stem rust is macro-cyclic and heteroecious, there are 5 spore stages basidiospores, pycniospore, aeciospore, urediospore, teliospore and includes a sexual stage that occurs on barberry and mahonia species & asexual stages that occurs on wheat and related grasses (S. Singh, 2016). Its lifecycle consists of recurrent uredinal generations in most areas of the world. In a gramineous host, *P. graminis* typically produce a thick-walled, dikaryotic resting spore stage called teliospore in temperate climates; each of which remains dormant in the infected straw until spring (Zhu et al., 2014). During this period, karyogamy occurs, and then it becomes diploid; then after the spring rains and favorable environment teliospore germinates, undergoes meiosis, and produces a four-celled basidium (Roelfs et al., 1992). Then a single haploid basidiospore is produced via each cell which is windborne to the barberry plant which germinates and penetrates directly resulting in the production of flask-shaped pycnia (Roelfs et al., 1992).

For maximum infection, the barberry leaf tissue should be less than 2 weeks old (Roelfs et al., 1992). Single-celled pycniospores and receptive hyphae (n) are produced by pycnia that acts as gametes whereby fertilization occurs between them forming aecium which ultimately produces dikaryotic aeciospores in chains in the aecium causing an infection on hosts (Roelfs, 1985; S. Singh, 2016). Aeciospores can spread via water and air over distances of meters to perhaps a few kilometers (Leonard & Szabo, 2005). After successful infection, a dense mat of hyphae is produced beneath the host epidermis; where sporophores grow and ultimately produces masses of single-celled dikaryotic urediniospores which rupture the host epidermis generating uredinium (Zhu et al., 2014). Then repeating 14 days asexual cycle begins which involves the production of uredinia (Roelfs et al., 1992). Teliospores are produced once the host matures whereas uredinia cease production of urediniospores (Zhu et al., 2014).

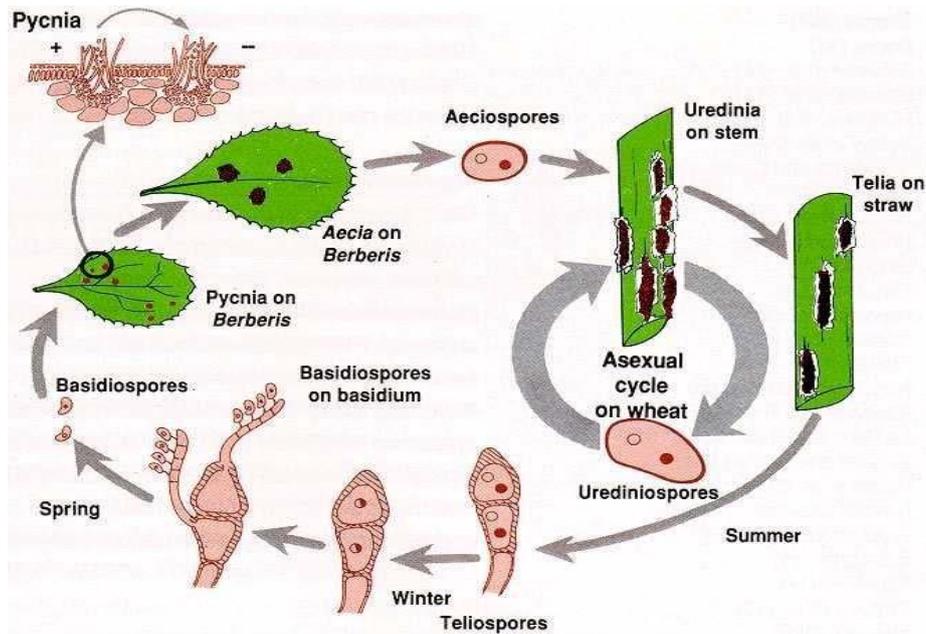


Figure 3 – The life cycle of *Puccinia graminis* f. sp. tritici (Roelfs et al., 1992).

Many species of *Berberis*, *Mahonia*, and their hybrid (*X Mahoberberis*) are susceptible to *Puccinia graminis* (Ro, 1985). The most important of the susceptible species is *Berberis vulgaris* L., although other susceptible species exist throughout the world; it also plays a major role in creating virulence and aggressiveness in the pathogen and thus made resistance breeding difficult (Roelfs et al., 1992). Barberry acts as the source of aeciospores early in the season and about  $64 \times 10^9$  aeciospores can be produced by a single barberry bush in about a few weeks which is equivalent to the daily output of 20 million uredinia, in an area of 400 m<sup>2</sup> (Roelfs et al., 1992). *Berberis* spp. plays a major role in the sexual reproduction of *P. graminis* f. sp. Tritici under natural conditions as more than 100 isolates of *P. graminis* f. sp. tritici were found from the three *Berberis* spp, as found in China (Zhao et al., 2015). Thus, studies have shown that *Berberis* spp. can create new races under natural conditions and also develop virulence variation in the pathogen.



Figure 4 – Barberry leaf infected with wheat stem rust fungus producing aecia which will produce aeciospores ([https://www.ars.usda.gov/images/docs/10755\\_10949/aecia.jpg](https://www.ars.usda.gov/images/docs/10755_10949/aecia.jpg))

Environmental factor strongly plays an important role in the development, extent, and severity of rust pathogen. For the development of spore, a susceptible host along with suitable temperature, moisture condition, and the direction velocity of winds are necessary for their dissemination (Komen, 2007; Staples & Macko, 1984). Various factors affect the development of rust. Such as:

- Availability of abundant viable inoculums;
- Dissemination and distribution of such inoculums;



- Favorable climatic conditions;
- Availability population of susceptible hosts;
- Suitable growth stage for infection.

The urediniospores are most likely to be found in high numbers above the seed canopy during favorable conditions (Roelfs et al., 1992). Excessive nitrogen fertilizer application along with other soil conditions affects rust severity as it causes a delay in maturity of the host crop (Komen, 2007). There is a prominent effect of relative humidity on the viability of urediniospores and is closely linked to temperature i.e. at the medium relative humidity germination percent is high and spore viable period is long; but germination percent and viable period decrease in high temperature while at low temperatures opposite occurs (Peltier, 2015).

Table 2 – Environmental conditions required for the stem rusts of wheat

| Stage        | Optimum Temperature (°C) | Light | Water     |
|--------------|--------------------------|-------|-----------|
| Germination  | 15-24                    | Low   | Essential |
| Germling     | 20                       | Low   | Essential |
| Appressorium | 16-27                    | None  | Essential |
| Penetration  | 29                       | High  | Essential |
| Growth       | 30                       | High  | None      |
| Sporulation  | 30                       | High  | None      |

Note: Adopted from Roelfs et al., 1992.

Rust infects or colonizes new crops via migration and dispersion (Nagarajan & Singh, 1990). Wind and rain splash plays a major role in the long-distance dispersion of rust spore (S. Singh, 2016). The distribution of urediospores over long distances is affected by wind pattern and latitude (Roelfs et al., 1992). For a short distance, steady step-wise range expansion occurs for Ug99 within a region (Ravi P. Singh et al., 2006). Urediniospores of stem rust are relatively resistant to light and temperatures at humidity up to 30 % and can disperse up to 100 km in viable environmental conditions and sometimes up to 2000 km by wind frequency (Roelfs et al., 1992; S. Singh, 2016). In 90 days or less, *P. graminis* travels 2000 km from the southern winter wheat to the northernmost spring wheat in North America (Roelfs et al., 1992). Aerodynamically rust spores are modified to get removed and transported via wind i.e. anemochory (Fig. 1); however, spores were not easily caught and carried by rain drops i.e. ombrochory (Fig. 2) was supposed (Sache, 2000).

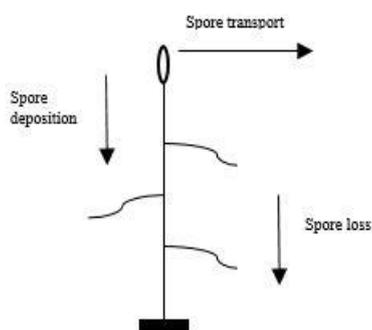


Figure 5 – Impact of wind on spore distribution for cereal rust (Sache, 2000)

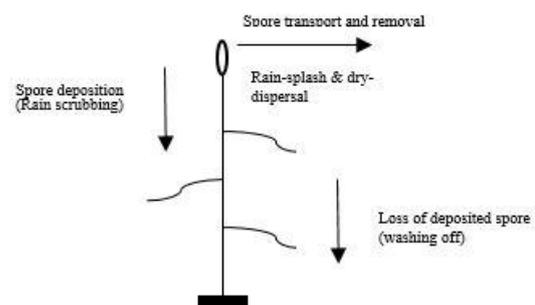


Figure 6 – Impact of rain on spore distribution for cereal rust (Sache, 2000)

*Stem rust resistance genes and their efficiency against the Ug99 race.* Stem rust pathogen has more than 50 wheat genes that give resistance among which many are developed from alien wheat varieties but only a few are effective in providing resistance against Ug99 (S. Singh, 2016). CIMMYT Expert panel, 2005 also determine some of the effective genes as Sr-2, 13, 14, 22, 24, 25, 26, 27, 28, 32, 33, 35, 36, 37, 39, 49, 44, Tmp, Tt-3, 1A, 1R etc. (S. Singh, 2016). New races of Ug99 overcame Sr genes and made them susceptible like Sr5, 6, 7a, 7b, 8a, 8b, 9a, 9b, 9d, 9g, 10, 11, 12, 15, 16, 17, 18, 19, 20, 23,



30, 31, 34, 38, and Wld-1 (Y. Jin et al., 2007). Among all of these genes, Sr2 was only non-race specific (R. P. Singh, Huerta-Espino, et al., 2007; Ravi P. Singh et al., 2006; Spielmeyer et al., 2003). Two genes Sr9b and Sr6 found in the US wheat variety were also found to be susceptible to Ug99 (Kolmer et al., 2007; Volkova et al., 2020). Similarly, in trials conducted in Ethiopia Sr genes like Sr13, Sr36 and SrTmp were found effective while Sr31 and Sr38 were found ineffective against Ug99 (Admassu et al., 2009). In 2017-18, northwest Ethiopia host carrying Sr24 and Sr31 genes were found to be effectively followed by Sr11, Sr9b, and Sr30 (Azmeraw et al., 2020). All other Sr genes were ineffective to all races while some of the effective genes were widely deployed in all wheat varieties by CYMMYT among which gene Sr2 is found in more than half of current spring wheat germplasm as this gene was found to be recessive (McIntosh, 1988; Rajaram et al., 1988; R. P. Singh, Kinyua, et al., 2007; Ravi P. Singh et al., 2006). This Sr2 non-race-specific gene effect can be enhanced by combining it with other race-specific Sr genes (S. Singh, 2016). The narrow genetic base for resistance and the continuously evolving pathogen is the key cause of the ineffectiveness of wheat cultivars against Ug99 (Assefa & Fehrmann, 2004; Ydoğdu & Boyraz, 2012). Combination techniques where a major resistant gene combines with a minor resistant gene would be effective against Ug99 (Babiker et al., 2009). Gene pyramiding can also be an effective strategy to use resistance genes to increase resistance against stem rust but it is a difficult and time-consuming method (Leonard & Szabo, 2005; Ydoğdu & Boyraz, 2012). So, an alternative quick method to develop resistant variety; marker-assisted selection (MAS) can be utilized for gene deployment and gene pyramiding (Wu et al., 2009).

*Control options.* Cultural practices emphasize the use of early maturing and early planting varieties which induce a chance of rust incidence; so delaying planting along with the removal of green bridges a few weeks before planting may prevent infestation (Ro, 1985; S. Singh, 2016). The use of excess nitrogen fertilizers must be avoided and frequent light irrigation must be done (Roelfs, 1985). Proper spacing and cultivar diversity in the field can give significant results (Ro, 1985). It has been founded that reduced stand densities may promote the development of stem rust in barley (Dill-Macky & Roelfs, 2000). Avoidance can also be done by growing early cultivars as it can escape damage avoiding the growth period of the fungus (S. Singh, 2016).

An alternate host of wheat stem rust is barberry (*Berberis vulgaris* L.) which has the potential to produce new races of pathogen Ug99 (Yue Jin et al., 2010). So, barberry must be destroyed to delay disease onset, reduce initial inoculum level, decrease the number of pathogen phenotypes, and stabilize these pathogen phenotypes (Roelfs, 1985). Alternate host eradication was started in Rouen, France in 1660 (Roelfs & Budhnell, 1985) Similarly in 1918, an eradication campaign was initiated in the US and the elimination of these barberries from most of the areas was completed in the 1930s (Peterson, 2018).

The use of chemical has a very minor role in controlling stem rust because of the effective host resistance, very high rate of disease intensification under ideal conditions, and high cost of chemical applications (Roelfs, 1985; S. Singh, 2016). Nine fungicides were evaluated between 2005 and 2006 in Kenya among which only four were found effective against stem rust namely AmistarXtra 280 SC, Folicur 250 EC, Silvarcur 375 EC, and Orius 25 EW (Wanyera et al., 2009). But if used on large scale other races of Ug99 may develop resistance creating a potential threat (S. Singh, 2016). Fungicide application is only effective for slightly infected plans, but not for heavily infected and is more effective if applied immediately (Loughman et al., 2005).

Based on efficiency and safety, the application of arbuscular mycorrhiza (AM) fungi and *Trichoderma* spp. is found to be effective in reducing disease measures along with increasing the peroxidase and polyphenol oxidase enzymes, phenol content and improving other growth parameters (El-Sharkawy et al., 2018). To be precise the synergistic effect of the combination of AM Fungi, *T. harzianum* HL1, and *T. viride* HL5 was found to be superior against stem rust (El-Sharkawy et al., 2018). Similarly, *Bacillus subtilis*, *B. polymyxa* and *B. megaterium*, Eugenol, and leaf extract of *Artemisia cina* were also found to be effective in controlling wheat stem rust disease (Omara et al., 2020).



The most effective, environmentally friendly, and economical way to control Ug99 is genetic resistance. Scientifically identified race-specific effective resistance genes against Ug99 can be used for the development and release of new resistant varieties (S. Singh, 2016). There are several types of resistance to stem rust in wheat such as seedling resistance, adult-plant resistance, durable resistance, all-stage resistance (ASR), slowly rusting and gene pyramiding (Komen, 2007; S. Singh, 2016). The main aim of resistance breeding is to learn about the expression and inheritance of resistance along with the range of available genetic diversity (McIntosh, 1988). However, resistance is overcome by pathogens via genetic adaptation (Komen, 2007).

## CONCLUSION

In developing countries, Ug99 is a serious threat to wheat production as wheat is a major crop and causes a significant impact on their economy. These rust pathogens of wheat evolve continuously, and their urediospores can spread over long distances by air. Due to this, there is a failure of crop resistance and severe losses to wheat production. As the incidence of a new virulent strain of stem rust spread the pathogen to larger areas and cultivar resistance is lost. So, necessary precautions must be undertaken to overcome this situation. Research and development for new varieties; their rapid seed multiplication and dissemination are important. Globally monitoring must be done for the incidence of new pathotypes. Awareness programs related to the potential impact of Ug99 must be conducted among farmers along with the promotion of new resistance cultivars. A regular survey of disease, development of information centers, increase research capacity, resistant breeding, and proper use of chemicals might be led to effective management of rust disease in developing countries.

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