2.3 The science of wheat rust

2.3.1 Rusts

Rusts are plant diseases caused by pathogenic fungi of the order **Pucciniales** (previously also known as **Uredinales**). An estimated 168 rust genera and approximately 7000 species, more than half of which belong to the genus *Puccinia*, are currently accepted. [1] Rust fungi are highly specialized parasites with several unique features. A single species may produce up to five morphologically and cytologically distinct spore-producing structures viz., spermagonia, aecia, uredinia, telia, and basidia in successive stages of reproduction. Unlike other plant pathogens, rust usually affects healthy and vigorously growing plants, so the infection is limited to plant parts, such as leaves, petioles, tender shoots, stem, fruits, etc.[1] perennial systemic infection may cause deformities such as growth retardation, witches brooms, stem canker, hypertrophy of the affected tissues or formation of galls. Plants with severe rust infection may appear stunted, yellowed, or otherwise discoloured. Rust sporulates on affected plant parts. [1] Rust is most commonly seen as coloured powder, composed of tiny aeciospores which land on vegetation producing pustules, or uredia, that form on the lower surfaces. During late spring or early summer, yellow orange or brown, hairlike or ligulate structures called telia grow on the leaves or emerge from bark of woody hosts such as *Juniperus* species. These telia produce teliospores which will germinate into aerial basidiospores, spreading and causing further infection.

Taxonomy

The taxonomy of *Pucciniales* is complex and the darker coloured smuts can be mistaken for rust. Rusts are so named after the reddish rusty looking sori and the disease is usually noticed after rain.

The group is considered one of the most harmful pathogens to agriculture, horticulture and forestry. These fungi are major concerns and limiting factors for successful cultivation of agricultural and forestry crops. White pine blister rust, wheatstem rust, and coffee rust are examples of notoriously damaging, economically important rusts. [1]

Spores

Spore of *Puccinia graminis* model from the late 19th century, Botanical Museum Greifswald

Rusts can produce up to five spores types during their life cycle

- 0-Pycniospores (Spermatia)-Haploid gametes in heterothallic rusts.
- I-Aeciospores-non-repeating dikaryotic vegetative spores
- II-Urediniospores-repeating dikaryotic vegetative spores. These spores are referred to as the repeating stage because they can cause auto-infection (re-infect the same host from which the spores were borne). These spores are red/orange and are a characteristic sign of rust fungus infection.
- III-Teliospores-Diploid spores that produce basidia and are the survival stage of life cycle
- IV-Basidiospores-stem from basidia. Haploid spores which infect the alternate host.[2][3] although these are rarely observed outside of the laboratory.

Rust fungi can be categorized by how many types of spores are produced during the life cycle. Fungi that produce all five spores (sometimes excluding pycniospores) are termed **macrocyclic**. Fungi that lack pycniospores and aeciospores in their life cycle are termed **microcyclic** and always have an autoecious life cycle. **Demicyclic** fungi delete the uredial (repeating) stage from the life cycle. Understanding the life cycles of rust fungi allows for proper disease management.[4]

Life cycle

All rusts are obligate parasites, meaning that they require a living host to complete their life cycle. They generally do not kill the host plant but can severely reduce growth and yield. [5] Cereal crops can be devastated in one season and trees that get infected in the main stem within their first five years by the rust *Cronartium quercuum* often die.[6]

Rust fungi can also be categorized by their life cycle. Three basic types of life cycles are recognized based on the number of spore states as macrocyclic, demicyclic, and microcyclic.[1] The macrocyclic life cycle has all spore states, the demicyclic lacks the uredinial state, and the microcyclic cycle lacks both the aecial and uredinial states, thus possess only spermagonia and telia. Spermagonia may be absent from each type but especially the microcyclic life cycle. In macrocyclic and demicyclic life cycles, the rust may be either host alternating (heteroecious), i.e., the aecial state is on one kind of plant but the telial state on a different and unrelated plant, or non-host alternating (autoecious), i.e., the aecial and telial states on the same kind of plant. [1]**Heteroecious** rust fungi require two unrelated hosts to complete their life cycle, with the primary host being infected by aeciospores and the alternate host being infected with basidiospores. This can be contrasted with an **autoecious** fungus which can complete its life cycle on a single host species.[7]

Host plant-rust fungus relationship

There are definite patterns of relationship with host plant groups and the rust fungi that parasitize them. Some genera of rust fungi, especially *Puccinia* and *Uromyces*, comprise species that are capable of parasitizing plants of many families. But many genera appear to be rather definitely restricted to certain plants. Host restriction may, in heteroecious species, apply to both phases of life cycle or to only one phase. [1]

Infection process

The fungi produce asexual spores which disperse by wind, water or by insect vectors[8] spreading the infection.

Rust fungi are biotrophs taking nutrients from living cells. When airborne spores settle on a plant, weak hydrophobic interactions are formed with the cutin on the plant cell surface, securing it. By a process not fully understood, the production of mucous like substances called 'adhesins', initially stick the spore to the plant surface.[9]

Once attached, the spore germinates by growing a germ tube and eventually locates a stoma by a touch responsive process known as thigmotropism. This involves growing towards a ridge between the epidermal cells, followed by a perpendicular growth which end at the stoma.[10]

Rust hypha attacking stoma (1600x magnification).

Inside the stoma, the hyphae tips flatten out to form structures known as appresorium that lock to the cell walls.[11] It is thought that the whole process is mediated by a mechano-sensitive calcium ion channel, located within the germ tube tip, which produces electric currents that stretch the cell membranes, changing gene expression and forming the appresorium.[12]

Then a peg grows into the plant's mesophyll cells. The peg produces specialised hyphal tips, known as haustorium. These spread around the plant cells without invading the membranes. The plant cell membranes invaginate around the main haustorial body forming a space known as the extra-haustorial matrix. An iron and phosphorus rich neck band bridges the plant and fungal membranes in the space between the cells for water flow, known as the apoplast, thus preventing the nutrients reaching the plant's cells. The haustorium contains amino acid and hexose sugar transporters and H^+-ATPases which are used for active transport of nutrients from the plant nourishing the fungus.[13] It continues growing until spore growth occurs. The process repeats every 10 – 14 days, producing numerous spores, carried by wind to new hosts.

2.3.2 Stem wheat rust

The **stem**, **black** or **cereal rusts** are caused by the fungus *Puccinia graminis* and are a significant disease affecting cereal crops. Crop species which are affected by the disease include bread wheat, durum wheat, barley and triticale.[1d] An epidemic of stem rust on wheat caused by race TTKSK (e.g. isolate*Ug99*) is currently spreading across Africa, Asia and the Middle East and is causing major concern due to the large numbers of people dependent on wheat for sustenance. The strain was named after the country where it was identified

(Uganda) and the year of its discovery (1999).[2d] It spread to Kenya, then Ethiopia, Sudan and Yemen, and is becoming more virulent as it spreads.[2d] Scientists are working on breeding strains of wheat that are resistant to *Ug99*. However, wheat is grown in a broad range of environments. This means that breeding programs would have extensive work remaining to get resistance into regionally adapted germplasms even after resistance is identified.[2d]

There is considerable genetic diversity within the species *P. graminis* and several special forms, *forma specialis*, which vary in host range have been identified.

- *Puccinia graminis f. sp. avenae*, oat
- *Puccinia graminis f. sp. dactylis*
- *Puccinia graminis f. sp. hordei*, barley
- *Puccinia graminis f. sp. lolii*
- *Puccinia graminis f. sp. poae*
- *Puccinia graminis f. sp. secalis*, rye, barley
- *Puccinia graminis f. sp. tritici*, wheat, barley

P. graminis is a member of the Phylum Basidiomycota within the Kingdom Fungi. The characteristic rust color on stems and leaves is typical of a general stem rust as well as any variation of this type of fungus. Different from most fungi, the rust variations have five spore stages and alternate between two hosts. Wheat is the primary host and barberry is the alternate host.

There are two pathotypes (QCC and MCC) affecting barley.[3d]

Pathology of stem wheat rust

The stem rust fungus attacks the parts of the plant which are above ground. Spores that land on green wheat plants form a pustule that invades the outer layers of the stalk.[2d] Infected plants produce fewer tillers and set fewer seed, and in cases of severe infection the plant may die. Infection can reduce what is an apparently healthy crop about three weeks before harvest into a black tangle of broken stems and shrivelled grains by harvest.[1d]

Stem rust of cereals causes yield losses in several ways:[4d]

- Fungus absorbs nutrients that would otherwise be used for grain development.[4d]
- Pustules break through epidermis, which disrupt the plant's control of transpiration and can lead to desiccation and infection by other fungi.[4d]
- Interference with plant vascular tissue leads to shrivelled grains.[4d]
- The fungus weakens the stems, which can lead to lodging (falling over). In severe cases lodging can make mechanical harvesting impossible.[4d]

Signs and symptoms of stem rust

On wheat

Stem rust on wheat is characterized by the presence of uredinia on the plant, which are brick-red, elongated, blister-like pustules which are easily shaken off.[1d] They most frequently occur on the leaf sheaths, but are also found on stems, leaves, glumes and awns.[1d] On leaves they develop mostly on the underside but may penetrate to the upperside.[1d] On leaf sheaths and glumes pustules rupture the epidermis, giving a ragged appearance.[1d]

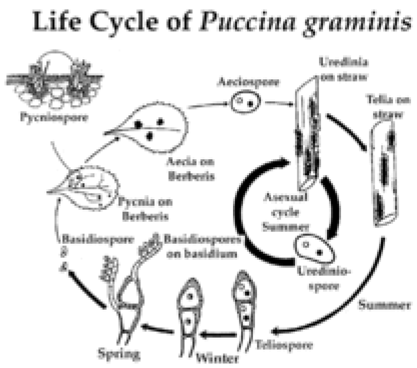
Towards the end of the growing season black telia are produced.[1d] For this reason stem rust is also known as 'black rust'.[1d] The telia are firmly attached to the plant tissue.[1d]

The site of infection is a visible symptom of the disease.

On barberry

Pycnia appear on barberry plants in the spring, usually in the upper leaf surfaces.[4d] They are often in small clusters and exude pycniospores in a sticky honeydew.[4d] Five to 10 days later, cup-shaped structures filled with orange-yellow, powdery aeciospores break through the lower leaf surface.[4d] The aecial cups are yellow and sometimes elongate to extend up to 5 mm from the leaf surface.[4d]

Life cycle of stem rust



Life cycle of *Puccinia graminis*

Like other *Puccinia* species, *P. graminis* is an obligate biotroph and has a complex life cycle featuring alternation of generations. The fungus is heteroecius, requiring two hosts to complete its life cycle - the cereal host and the alternate host.[4d] There are many species in *Berberis* and *Mahonia* that are susceptible to stem rust, but the common barberry is considered to be the most important alternate host.[1d] *P. graminis* is macrocyclic[4d] (exhibits all five of the spore types that are known for rust fungi[5d]).

P. graminis can complete its life cycle either with or without barberry (the alternate host).[4d]

Life cycle of barberry

Due to its cyclical nature, there is no true 'start point' for this process. Here, the production of urediniospores is arbitrarily chosen as a start point.

Urediniospores are formed in structures called uredinia, which are produced by fungal mycelia on the cereal host 1–2 weeks after infection.[4d] The urediniospores are dikaryotic (contain two un-fused, haploid nuclei in one cell) and are formed on individual stalks within the uredinium.[4d] They are spiny and brick-red.[4d] Urediniospores are the only type of spores in the rust fungus life cycle which are capable of infecting the host on which they are produced and this is therefore referred to as the 'repeating stage' of the life cycle.[4d] It is the spread of urediniospores which allows infection to spread from one cereal plant to another.[4d] This phase can rapidly spread the infection over a wide area.

Towards the end of the cereal host's growing season, the mycelia produce structures called telia.[4d] Telia produce a type of spore called teliospores.[4d] These black, thick-walled spores are dikariotic.[4d] They are the only form in which *Puccinia graminis* is able to overwinter independently of a host.[4d]

Each teliospore undergoes karyogamy (fusion of nuclei) and meiosis to form four haploid spores called basidiospores.[4d] This is an important source of genetic recombination in the life cycle.[4d] Basidiospores are thin-walled and coloured.[4d] They cannot infect the cereal host, but can infect the alternative host (Usually barberry).[4d] They are usually carried to the alternative host by wind.

Once basidiospores arrive on a leaf of the alternative host, they germinate to produce a haploid mycelium which directly penetrates the epidermis and colonises the leaf.[4d] Once inside the leaf the mycelium produces specialised infection structures called pycnia.[4d] The pycnia produce two types of haploid gametes, the pcyniospores and the receptive hyphae.[4d] The pycniospores are produced in a sticky honeydew which attracts insects.[4d] The insects carry pcyniospores from one leaf to another.[4d] Splashing raindrops can also spread pcyniospores.[4d] A pycniospore can fertilise a receptive hypha of the opposite mating type, leading to the production of a dikaryotic mycelium.[4d] This is the sexual stage of the life cycle and cross-fertilisation provides an important source of genetic recombination.[4d]

This dikaryotic mycelium then forms structures called aecia, which produced a type of dikaryotic spores called aeciospores.[4d] These have a worty appearance and are formed in chains – unlike the urediniospores which are spiny and are produced on individual stalks.[4d] The chains of aeciospores are surrounded by a bell-like enclosure of fungal cells. The aeciospores are able to germinate on the cereal host but not on the alternative host (the are produced on the alternative host, which is usually barberry).[4d] They are carried by wind to the cereal host where they germinate and the germ tubes penetrate into the plant.[4d] The fungus grows inside the plant as a dikaryotic mycelium.[4d] Within 1–2 weeks the mycelium produces uredinia and the cycle is complete.[4d]

Life cycle without barberry

Since the urediniospores are produced on the cereal host and can infect the cereal host, it is possible for the infection to pass from one year's crop to the next without infecting the alternative host (barberry).[4d] For example, infected volunteer wheat plants can serve as a bridge from one growing season to another.[4d] In other cases the fungus passes between winter wheat and spring wheat, meaning that it has a cereal host all year round.[4d] Since the urediniospores are wind dispersed, this can occur over large distances.[4d] Note that this cycle consists simply of vegetative propagation – urediniospores infect one wheat plant, leading to the production of more urediniospores which then infect other wheat plants.

Spore dispersal of stem rust

Puccinia graminis produces all five of the spore types that are known for rust fungi.[4d]

Spores are typically deposited close to the source, but long-distance dispersal is also well documented.[1d] The following three categories of long-distance dispersal are known to occur:[1d]

Extremely long-distance dispersal
This can occur unassisted (the robust nature of the spores allows them to be carried long distances in the air and then deposited by rain-scrubbing) or assisted (typically on human clothing or infected plant material that is transported between regions).[1d] This type of dispersal is rare and is very difficult to predict.[1d]

Step-wise range expansion
This is probably the most common mode of long-distance dispersal and usually occurs within a country or region.[1d]

Extinction and recolonisation
This occurs in areas that have unsuitable conditions for year-round survival of Puccinia graminis - typically temperate regions where hosts are absent during either the winter or summer.[1d] Spores overwinter or oversummer in another region and then recolonise when conditions are favorable.[1d]

2.3.3 Yellow (stripe) rust

"Yellow rust" takes its name from the appearance of yellow-colored stripes produced parallel along the venations of each leaf blade. These yellow stripes are actually characteristic of uredinia that produce yellow colored urediniospores. Primary hosts of yellow rust are *Triticum aestivum* (bread wheat), *Triticum turgidum* (durum wheat), triticale, and a few *Hordeum vulgare* (barley) cultivars. The alternate host was discovered by accident in 2010.[2g] Species of common barberry plants (the alternate host of wheat stem rust) were found harbouring stripe rust. When transferred to grass hosts, Kentucky Bluegrass was successfully infected and urediniospores were produced. Several species of Berberis were then investigated as alternate hosts of Wheat stripe rust and inoculations were successful.

The disease usually occurs early in the growth season, when temperature ranges between 2 and 15°C; but it may occur to a maximum of 23°C. High humidity and rainfall are favorable conditions for increasing the infection on both leaf blade and leaf sheath, even on spikes when in epidemic form. Symptoms are stunted and weakened plants, shriveled grains, fewer spikes, loss in number of grains per spike and grain weight. Losses can be 50%, but in severe situation 100% is vulnerable. In countries where wheat is grown in winters or at high elevations, yellow rust is a common threat, but not more significant than wheat leaf rust and stem rust, which are continuous threats in all wheat-growing countries.

Worldwide population structure of yellow rust

The evidence of both spatial structuring and invasion has been shown for this disease.[3g] Population genetic analyses indicate a strong regional heterogeneity in levels of recombination, with clear signatures of recombination in the Himalayan and near-Himalayan regions and a predominant clonal population structure in other regions. The existence of a high genotypic diversity, recombinant population structure, high sexual reproduction ability and the abundance of alternate host (Berberis spp.) in the Himalayan and neighboring regions suggest the region as plausible PST center of origin or at least the most closer to its centre of origin. However, further exploration may be useful from Central Asia to East Asian regions.[4g]

2.3.4 Wheat leaf rust

Wheat leaf rust, is fungal disease that affects wheat, barley and rye stems, leaves and grains. In temperate zones it is destructive on winter wheat because the pathogen overwinters. Infections can lead up to 20% yield loss - exacerbated by dying leaves which fertilize the fungus. The pathogen is **Puccinia rust fungus.** *Puccinia triticina* causes 'black rust', *P.recondita* causes 'brown rust' and *P.striiformis* causes 'Yellow rust'. It is the most prevalent of all the wheat rust diseases, occurring in most wheat growing regions. It causes serious epidemics in North America, Mexico and South America and is a devastating seasonal disease in India. All three types of *Puccinia* are heteroecious requiring two distinct and distantly related hosts (alternate hosts). Rust and the similar smut are members of the class Pucciniomycetes but rust is not normally a black powdery mass.

Life cycle of wheat leaf rust

Wheat leaf rust is spreads via airborne spores. Five types of spores are formed in the life cycle. Uredospores, teleutospores, and basidiospores develop on wheat plants and pycnidiospores and aeciospores develop on the alternate hosts.[10] The germination process requires moisture, and works best at 100% humidity. Optimum temperature for germination is between 15-20 C. Before sporulation, wheat plants appear completely asymptomatic. In the Asian Subcontinent, the spores cannot survive the hot dry weather but are re-introduced every year from the Himalayas or surrounding hills, possibly coming from *Berberis spp*, *Thalictrum flavum* and *Muehlenbergia huglet* which is a main reason for bread mouldes or even some grasses. Wheat rust pathogens are biotrophic and require living plant cells to survive.

P. triticina has an asexual and sexual life cycle. In order to complete its sexual life cycle *P. triticina* requires a second host *Thalictrum spp*. which it will overwinter on. In places where *Thalictrum* does not grow, such as Australia, the pathogen will only undergo its asexual life cycle and will overwinter as mycelium or uredinia. The germination process requires moisture and temperatures between 15 – 20 °C. After around 10 – 14 days of infection, the fungi will begin to sporulate and the symptoms will become visible on the wheat leaves.[11]

The pathogen has an asexual and sexual cycle. In North America, South America and Australia the pathogen only undergoes its asexual cycle. However this does not seem to be a disadvantage to it, and wheat leaf rust has many races with different virulence. The sexual life cycle of wheat leaf rust requires a different host species, *Thalictrumn spp*.

Symptoms of wheat leaf rust

Small brown pustules develop on the leaf blades in a random scatter distribution. They may group into patches in serious cases. Infectious spores are transmitted via the soil. Onset of the disease is slow but accelerated in temperatures above 15°C, making it a disease of the mature cereal plant in summer, usually too late to cause significant damage in temperate areas. Losses of between 5 and 20% are normal but may reach 50% in severe cases.

Leaf rust is one of the most common and most important wheat diseases. Leaf rust is caused by a parasitic fungus called Puccinia recondita f. sp. tritici. For example, Kansas annual wheat yield losses due to leaf rust have ranged from trace to 11%, averaging about 4% over the last two decades. Losses due to leaf rust in individual fields can range from trace to over 40%. Conditions are most favourable for leaf rust in the eastern two thirds of Kansas, but significant losses can also occur in western Kansas under irrigation or during years with high precipitation.

Leaf rust causes very small (about 1/32 inch long by 1/64 inch wide), orange-brown pustules

that are raised above the leaf surface. These pustules can be rubbed off the leaf leaving an orange-brown mark on the finger. The pustules scattered across the leaf surface are circular to oval in shape, and confined chiefly to the upper surface of the leaf (Figure 1). Later in the season, black teliospores develop on mature plants, usually on the under surface of the leaf or on the leaf sheath. These black spores are not of concern.