IOSR Journal of Biotechnology and Biochemistry (IOSR-JBB)
ISSN: 2455-264X, Volume 5, Issue 6 (Nov. – Dec. 2019), PP 01-09
www.iosrjournals.org

Biochemical and Molecular Biological Alterations in Angiospermic Plants upon Infection by Angiosperm Parasite, *Cuscuta spp*.: An Overview

Danish Inam, Indra Vikram Singh, Nabeel Ahmad and Sanjay Mishra*
School of Biotechnology, IFTM University, Delhi Road (NH-24), Moradabad -244102, UP, India
[*]Corresponding Author: Dr. Sanjay Mishra
Professor, School of Biotechnology, IFTM University, Delhi Road (NH 24), Moradabad 244 102, UP, India

***Abstract**: Angiospermic plants have been observed to be adversely affected by certain plant parasites like Cuscuta spp. Consequent to their invasion into host plants, a variety of physiological, biochemical and molecular biological alterations occur, and therefore affect the quality as well as quantity of yield from angiospermic plants. The present article embodies an overview of angiosperm plants in reference to host-parasite interaction with Cuscuta spp, and collectively may provide new insights into developing biochemical as well as molecular biological strategies for expressing solutions for certain problems concerning with agricultural perturbation of crop yield.*

***Key words:** Angiosperm plants; Cuscuta spp; host- parasite interaction*

Date of Submission: 04-11-2019 Date of Acceptance: 20-11-2019

I. Introduction

Cuscuta, generally considered to as dodders, reflect little resemblance to their photoautotrophic relatives. The genus has been observed to develop gradually within the Convolvulaceae in the order Solanales and includes nearly two hundred species of obligate shoot holoparasitic plants (Garcia et al., 2014). *Cuscuta* is divided into the three subgenera, namely, *Monogynella*, *Cuscuta* and *Grammica*. Whilst species of *Monogynella* characteristically have thick stems, the other two subgenera include more delicate species (Dawson et al., 1994).

Though of higher abundance in tropical and subtropical areas, stumbles are known to have a global distribution and can be found on every continent excluding Antarctica. Species of *Cuscuta* adversely affect important crops (e.g. alfalfa and tomato) and express severe problems to agriculture as they result in downgrading crop yields (Aly, 2007) .Upon developing bud, *Cuscuta* seedlings appear without cotyledons or leaves and with a reduced root-like structure, which rapidly degenerates (Sherman et al., 2008). A few species species of *Cuscuta* are obviously green e.g. *C. reflexa*, reflecting photosynthetic activity. Nevertheless, even in these relatively green species the rate of carbohydrate synthesis is rather low to sustain the parasite (van der Kooij et al., 2000). Thus, for enabling to complete its life cycle, freshly germinated seedlings must quickly locate, adhere and infect a vulnerable host plant. In the later stages of contamination, tip-growing cells called searching hyphae stick out from the main haustorium and grow in search of the vascular tissues of the susceptible host. When they approach contact with xylem or phloem of the host, they differentiate into the respective cell types to direct the uptake of water and sugar from the host plant (Vaughan, 2006; Lee, 2009). In a fully developed feeding haustorium of *C. reflexa* on its vulnerable host *P. zonale*, the xylem via duct between the two species is evidently visible. Furthermore, plasmodesmata have been noticed between host cells and developing parasite hyphae (Lee, 2009; Birschwilks et al., 2006; Birschwilks et al., 2007; Vaughan, 2003). This happening reveals the evidence of interspecific symplastic connections being established. When it reaches to the range of host species parasitized, certain *Cuscuta* species are very common that can successfully infect a large number of different host plants, whilst others are confined to only a single host species (Dawson et al., 1994). The parasite seems to approach dicotyledonous plants, as monocotyledonous plants are barely infected. Grasses, especially, are shown to be in but very few cases, immune to *Cuscuta* (Vaughan, 2003). Notwithstanding, there are also dicotyledonous species, which display resistant mechanisms against *Cuscuta* attacks (Kaiser et al., 2015).

Certain physiological and biochemical alterations in angiospermic plants as a consequence of *Cuscuta* infection have been overviewed under following headings:

1. Plant Responses to Invasion

Considering that plants possess systems to identify and act against attacks by other plant pathogens (Boller and Felix, 2009), it appears improbable that the intruding risk of the haustorium should go on unnoticed. As such, the zonale, display no clear defence reactions when infected by *C. reflexa* is quite remarkable. The haustoria of *C. reflexa* were noticed to be able to invade the tropical liana *Ancistrocladus heyneanus,* but eventually deteriorated as a response to the production of pathogen repellents by the host. Poinsettia *(Euphorbia pulcherrima)* responds to haustoria of *C. reflexa* and *C. japonica* by producing localized bark projections, which force out the infection organs (Christensen et al., 2009). Anyhow, the host has been demarcated as only partially incompatible as the parasite could bear haustorium rejections by developing new infection sites. The cultivated tomato *Solanum lycopersicum* has been shown to be resistant to infection by *C. reflexa,* whilst the wild tomato *Solanum pennellii* is susceptible (Johnsen, 2014; Hegenauer et al., 2016). These closely concerned species provide an excellent system for working on host-parasite interactions.

Expression of genes encoding a cell wall enzyme and an aquaporin have been associated with the hypersensitive- type response of this tomato against *C. reflexa* (Werner et al., 2001; Albert et al., 2004; Boller and Felix, 2009). Though proposed to be involved in the detail of host cells at the site of adherence with the parasite, the exact roles of these genes in the defense response remain obscure (Vaughan, 2003). The obstacle of *S. lycopersicum* appears to be specific against *C. reflexa* as many other species inclusive of *Cuscuta pentagona* are able to successfully infect this plant species. However, *S. lycopersicum* responds with strong inductions of the defense-concerned plant hormones, namely, jasmonic acid and salicylic acid as a consequence of being contagious by *C. pentagon* (Runyon et al., 2010). Both hormones are recognized to play pivotal roles in plant immunity (Pieterse et al., 2012; Shigenaga and Argueso, 2016). Infection by *C. reflexa* also induces release of Ca^{2+}, a major player in signal transduction pathways, in the resistant tomato (Albert et al., 2010). It is remainder to be investigated whether the hormones and the second messenger act through the same or different pathways and where the threshold for complete resistance lies (Vaughan, 2003; Kaiser et al., 2015).

Recently, it was reported that the pattern recognition receptor, namely, *Cuscuta* Receptor 1 of *S. lyopersicum find out* a small peptide factor from *C. reflexa* and commences the above mentioned defense responses (Hegenauer et al., 2016). This reveals that plants can sense for *Cuscuta* much in the same way as they do accept other plant pathogens (Malinovsky et al., 2014; Mitsumasu et al., 2015).

2. Invasion of Host Tissues

The unilineal inflamentation of the parasite's stem facing the host surface reflects the initiation of haustorium development. *Cuscuta* epidermal cells then secrete a cementing substance, which secures the attachment of parasite to host (Vaughan, 2002; Lee, 2008). Proper adherence to the host surface seems to be essential for intrusion as the parasite also induces the host to produce substances that further augment the adhesion force (Albert et al., 2006); the activity of a cysteine protease from *C. reflexa* was shown to be needed for flourishing host infection, proposed by degrading host proteins (Bleischwitz et al., 2010). Interestingly, the prohibit of a similar cysteine protease in the obligate root parasite *Phelipanche aegyptiaca* also reduced infection rates (Rehker et al., 2012). A Shoot meristemless-like protein is essential to host infection by *C. pentagona* as gene silencing by RNA interference distorts the growth and development of the parasite haustorium (Alakonya et al., 2012). Presumably involved in directing growth of searching hyphae, the exact function of this protein remains obscure. Recently, transcriptome analyses have correponded the high expression of genes encoding products concerning with transport activity and cell wall with haustoria development in both *Orobancheae* (Orobanchaceae) and *Cuscuta* (Convolvulaceae) (Ranjan et al., 2014; Ikeue et al., 2015; Yang et al., 2015). The enhanced production of transporters is consistent with the parasite preparing to feed on its host.

3. Plant Cell Wall

Plant cells are encircled by walls and, given the pivotal role of this barrier in maintaining plant coherence, the structural components and dynamics of plant walls will be briefly presented in this chapter. One may identify three layers in a plant cell wall (Popper, 2008): (i) The middle lamella forms the fixative boundary between cell walls of adjoining cells. (ii) The primary wall is a strong but extensible layer continuously synthesized and transformed by growing cells. Some cell types (e.g. xylem cells) deposit a (iii) secondary wall between their primary wall and the plasma membrane to give further strength and rigidity after growth cessation. Cellulose is the principle structural component of plant cell walls. This polysaccharide is synthesized by large enzyme complexes at the plasma membrane and consists of long linear chains of β-(1→4)-linked glucose packed tightly into microfibrils (Doblin et al., 2002). Cellulose microfibrils are stiff rods of enormous mechanical strength that contribute structural stability to the cell wall when cross-linked by other wall polysaccharides.

The term hemicellulose encompasses non-cellulosic polysaccharides with β-(1→4)-linked backbones that are often extensively branched. This includes xyloglucan, xylan, mannan and mixed-linkage glucan (which

have β-(1→3)-linkages interspersed at regular intervals within their β-(1→4)-linked backbone) (Scheller and Ulvskov, 2010). Hemicelluloses are synthesized in the Golgi and secreted into the apoplast where their main role is to strengthen the cell wall by interacting with cellulose. Pectin is a heterogeneous group of complex cell wall polysaccharides that are rich in galacturonic acid. The three main types The three main types The three main types The three main types of pectin are homogalacturonan, rhamnogalacturonan and rhamnogalacturonan-I (Willats et al., 2001). Pectic polysaccharides form a gel-like matrix between cellulose microfibrils that is associated with several different functions. In the middle lamellae of dicotyledonous plants, pectins are proposed to facilitate adhesion between adjacent cells (Jarvis et al., 2003). As with hemicelluloses, pectins are synthesized in the Golgi and delivered to the wall through vesicles.

The type of part that make up the cell wall networks can vary between various plant groups and species. Notably, the cell wall configuration of grasses differs from that of dicotyledonous plants. Whereas xyloglucan is the most large hemicellulose in the primary walls of dicotyledonous plants, cell walls of grasses contain extra arabinoxylan (typically decorated with ferulic acid) and mixed-linkage glucan, but little xyloglucan (Vogel, 2008).

Classically, the primary cell wall of dicotyledonous plants is related as a load-bearing network of xyloglucan-coated cellulose microfibrils embedded in a pectin matrix. However, increasing verification argues that linkages exist between pectins and other cell wall components including cellulose and xyloglucan (Thompson and Fry, 2000; Popper and Fry, 2008; Chebli and Geitmann, 2017). These networks of polysaccharides make up a complex matrix that allows plant cells to keep high turgor pressures and thus enable plants to stand tall to harvest sunlight efficiently. Although, as plant growth and development require cell division, expansion and differentiation, plant cells must be able to control and modify the mechanical properties of their walls.

4. **Cell Wall Modifiers**

In fill out cells, new wall polymers are secreted into the cell wall to avert thinning as the cell surface increases. However, the structure, strength and flexibility of the primary cell wall are not only decided by what components are synthesized and deposited into the apoplast. Various proteins regulate plant growth and development by acting on the polysaccharides, which build up the cell wall. Some of these are glycoside hydrolases that develop cell wall loosening by splitting more or less specific polysaccharides. Others split one polysaccharide chain and graft it onto another (transglycosylation) and can thus facilitate either loosening or the reversal strengthening of the cell wall (Frankova and Fry, 2013). The xyloglucan endotransglucosylases/hydrolases (XTHs) modify wall strength by transglycosylating or hydrolytically cleaving the hemicellulose xyloglucan, respectively termed xyloglucan endotransglucosylation (XET) and xyloglucan endohydrolysis (Rose et al., 2002). Pectins are uploaded into the cell wall in a highly methyl-esterified state. The de-esterification of pectins by pectin methyl esterases makes the polysaccharide extra prone to degradation by pectate lyases and polygalacturonases (Wakabayashi et al., 2003). Though, as de-esterification of pectins also facilitates cross-linking and gel formation, pectin methyl esterases can be promoters of both cell wall loosening and strengthening (Willats et al., 2001; Chebli and Geitmann, 2017). Expansions are proteins that induce wall loosening without exhibiting enzymatic activity, probably by disrupting hydrogen bonds between wall polysaccharides (Cosgrove, 2005). Whereas plants modify their own walls to regulate growth and development, plant tissue invaders have been shown to employ cell wall modifiers to achieve access across the cell wall.

5. Genetic Transformation of *Cuscuta*

The ability to knock out specific genes in an organism is a potential tool for investigating gene function. We have established a protocol for the transient transformation of *C. reflexa* using a biolistic particle delivery system. This system takes helium pressure to shoot DNA-coated microcarriers (e.g. gold) into cells where, if reaching any of the DNA-containing nucleus, mitochondria and plastids, the transgene may be incorporated into the respective genome or expressed transiently from a plasmid vector. Evidently, gold particles coated with a declaration plasmid encoding a *Discosoma* sp. red fluorescent protein (dsRed) was bombarded onto the surface of *C. reflexa* stem pieces. The presence of polyphenol oxidases in *Cuscuta* results in the production of brown pigments in tissues damage by the microcarriers (Cosgrove, 2005). However, to be able to address the biological function of a gene product, stable genius of the transgene into the plant genome is necessary in order to pass on the trait to subsequent generations (Anami et al., 2017). Protocols for *Agrobacterium*-mediated transformation of *Cuscuta* calli have been reported earlier (Borsics et al., 2002; Svubova and Blehova, 2013), but none of these were able to revive genetically modified *Cuscuta* plants. An important facet of stable transformation is the use o proper selective agents to only allow the proliferation of resistance-acquired transformed cells. Callus cultures of *C. reflexa* were enlarged on Murashige and Skoog medium supplemented with sucrose (50 g/l), the cytokinin kinetin (3 mg/l), the auxin 1-Naphthaleneacetic acid (3 mg/l) and gibberellic acid (0.1 mg/l). A 5 µg/ml concentration of the protein synthesis-fend hygromycin B killed

C. reflexa calli within a week. Bombarded tissues of *Cuscuta* would require more time for treatment recovery and genomic unification to establish a stable expression of transgenes, probably enabling survival and proliferation on the selective medium. Therefore, in order to find a suitable concentration, which might allow transformed cells this recuperation time, the growth of callus on medium containing 0.2 and 1 µg/ml hygromycin B was tested. Both 0.2 and 1 µg/ml hygromycin B diminished the growth of *C. reflexa* calli. After 3 weeks, the callus on growth medium with 1 µg/ml hygromycin B point signs of cell death, which were even more pronounced after 8 weeks. As reported earlier by Das et al. (2011), equal concentrations of auxin and cytokinin start regeneration of *C. reflexa* shoots from callus. . This demonstrates the possibility to selectively regenerate transgenic *Cuscuta* if transgenes encoding hygromycin-resistance can be stably expressed in the parasitic plant. Genomic integration and retention of transgenes are challenges left to tackle in the continuing efforts to establish a protocol for the stable transformation of the parasitic plant *Cuscuta*.

6. Biochemical Defense Mechanism in Rapeseed-Mustard Genotypes against Alternaria blight Disease

Biochemical defense response in rapeseed-mustard genotypes has been observed upon infection with *Alternaria brassicae* (Berk.) Sacc. causing blight at different growth stage (Mathpal et al, 2011). Three genotypes viz. *Brassica juncea* cv. Varuna (susceptible), *B. juncea* cv. PAB-9534 (moderately resistant) and *B. alba* (tolerant) were selected. The results revealed that all the genotypes showed variable disease severity. It was observed that in all stages of pathogen infection, disease severity and characteristic symptoms were more prominent insusceptible genotype than the other two. The biochemical analysis of leaves of different varieties of mustardrevealed that total phenol, o-dihydroxy phenol, total sugar, reducing sugar, chlorophyll content and flavonol contents were observed to be more in resistant genotype (*B. alba*) than others. With progress of infection, total phenol, o-dihydroxy phenol and protein content increased in all three genotypes while the chlorophyll, total sugar, reducing sugar and flavonol content decreased. The results indicated that factors conditioning the host response to *A. brassicae* might be the outcome of complex biochemical changes operated in hostgenotypes (Mathpal et al, 2011).

7. Yield and Yield Attributes of Rapeseed-Mustard (*Brassica*) Genotypes Grown under Late Sown Condition

A field experiment was conducted at the Central Research Station of BARI, Gazipur, India for two consecutive years 2010-11 and 2011-12 with 30 varieties/ genotypes of rapeseed-mustard under three dates of sowing viz., 25 November, 5 December, and 15 December to determine changes in crop phenology, growth and yield of mustard genotypes under late sown condition when the crop faced high temperature (Alam et al., 2014). Days to flowering and maturity were different at different planting times. Date of sowing significantly influenced plant height, siliquae/plant, seeds/siliqua, seed yield,and oil content of seed in both the years. The highest seed yield (1310 and 1535 kg/ha) wasobtained from the first planting (25 November) in both the years, which was significantly different from two other dates of sowing. Yield and yield attributes of different varieties varied significantly. Among the varieties, BARI Sarisha-16 of *Brassica juncea* gave significantly the highest seed yield (1495 and 1415 kg/ha), which was statistically identical to BJDH-11, BJDH-12, BJDH-05, BJDH-20, and BARI Sarisha-6 and significantly different from all other varieties (Alam et al., 2014). Interaction effect of variety and sowing date significantly influenced plant height, number of siliquaeper plant, number of seeds per siliqua, seed yield,and strover yield. The highest seed yield (1758 and 1825 kg/ha) were recorded from BJDH-11 and BARI Sarisha-16 of *Brassica juncea* at 25 November planting and BJDH-11 produced the highest yield at 15 December in both the years. The maximum strover yield (3758 and 3825 kg/ha) were obtained from BJDH-11 and BARI Sarisha-16 of Brassica junceaat 25 November planting during 2010-11 and 2011-12. The highest oil content of seeds (44.4 % and 45.9%) was obtained from the seed of BARI Sarisha-6 and BARI Sarisha-14 at 25 November planting in both the years (Alam et al., 2014).

8. Advances in Agronomic Management of Indian Mustard (*Brassica juncea* (L.) Czernj. Cosson

India is the fourth largest oilseed economy in the world. Among the seven edible oilseeds cultivated in India, rapeseed-mustard contributes 28.6% in the total oilseeds production and ranks second after groundnut sharing 27.8% in the India's oilseed economy. The mustard growing areas in India are experiencing the vast diversity in the agro climatic conditions and different species of rapeseed-mustard are grown in some or other part of the country (Shekhawat et al., 2012). Under marginal resource situation, cultivation of rapeseed-mustard becomes less remunerative to the farmers. This results in a big gap between requirement and production of mustard in India. Therefore site-specific nutrient management through soil-test recommendation based should be adopted to improve upon the existing yield levels obtained at farmers field. Effective management of natural resources, integrated approach to plant-water, nutrient and pest management and extension of rapeseed-mustard cultivation to newer areas under different cropping systems will play a key role in further increasing and stabilizing the productivity and production of rapeseed-mustard. The paper reviews the advances in proper land

and seedbed preparation, optimum seed and sowing, planting technique, crop geometry, plant canopy, appropriate cropping system, integrated nutrient management and so forth to meet the ever growing demand of oil in the country and to realize the goal of production of 24 million tonnes of oilseed by 2020 AD through these advanced management techniques (Shekhawat et al., 2012).

9. **Parasitic Plants of the Genus *Cuscuta* and their Interaction with Susceptible and Resistant Host Plants**

By comparison with plant-microbe interaction, little is known about the interaction of parasitic plants with their hosts. Plants of the genus Cuscuta belong to the family of Cuscutaceae and comprise about 200 species, all of which live as stem holoparasites on other plants (Das et al., 2011; Mathpal et al, 2011). *Cuscuta spp.* possesses nor roots nor fully expanded leaves and the vegetative portion appears to be a stem only. The parasite winds around plants and penetrates the host stems via haustoria, forming direct connections to the vascular bundles of their hosts to withdraw water, carbohydrates, and other solutes. Besides susceptible hosts, a few plants exist that exhibit an active resistance against infestation by *Cuscuta spp.* For example, cultivated tomato (*Solanum lycopersicum*) fends off *Cuscuta reflexa* by means of a hypersensitive-type response occurring in the early penetration phase. This report on the plant-plant dialog between *Cuscuta spp.* and its host plants focuses on the incompatible interaction of *C. reflexa* with tomato (Mathpal et al, 2011).

10. Cell Wall Composition of Parasitic Dodder (*Cuscuta reflexa*) and its Hosts: A Priori Differences and Induced Alterations

Host plant penetration is the gateway to survival for holoparasitic *Cuscuta* and requires host cell wall degradation. Compositional differences of cell walls may explain why some hosts are amenable to such degradation while others can resist infection (Borsics et al., 2002). Antibody-based techniques for comprehensive profiling of cell wall epitopes and cell wall-modifying enzymes were applied to several susceptible hosts and a resistant host of *Cuscuta reflexa* and to the parasite itself. Infected tissue of Pelargonium zonale contained high concentrations of de-esterified homogalacturonans in the cell walls, particularly adjacent to the parasite's haustoria. High pectinolytic activity in haustorial extracts and high expression levels of pectate lyase genes suggest that the parasite contributes directly to wall remodeling. Mannan and xylan concentrations were low in P.Â zonale and in five susceptible tomato introgression lines, but high in the resistant Solanum lycopersicum cv M82, and in C. reflexa itself. Knowledge of the composition of resistant host cell walls and the parasite's own cell walls is useful in developing strategies to prevent infection by parasitic plants (Borsics et al., 2002).

11. Detection of the Angiosperm Parasite, *Cuscuta reflexa* by a Tomato Cell Surface Receptor

Parasitic plants are a constraint on agriculture worldwide. *Cuscuta reflexa* is a stem holoparasite that infests most dicotyledonous plants. One exception is tomato, which is resistant to *C. reflexa* (Hegenauer et al., 2016). Researchers discovered that tomato responds to a small peptide factor occurring in *Cuscuta spp.* with immune responses typically activated after perception of microbe-associated molecular patterns. Cell surface receptor-like protein CUSCUTA RECEPTOR 1 (CuRe1) is essential for the perception of this parasite-associated molecular pattern. CuRe1 is sufficient to confer responsiveness to the *Cuscuta* factor and increased resistance to parasitic *C. reflexa* when heterologously expressed in otherwise susceptible host plants. These findings reveal that plants recognize parasitic plants in a manner similar to perception of microbial pathogens (Hegenauer et al., 2016).

12. Natural Products from *Cuscuta reflexa* Roxb. with Antiproliferation Abilities in HCT116 Colorectal Cell Lines

Parasitic *Cuscuta reflexa* Roxb. possesses many medicinal properties and is a rich source of a variety of biologically relevant natural products (Riaz et al., 2017). Natural products are the prime source of leads, drugs, and drug templates, and many of the anticancer and antiviral drugs are either based on natural product or derived from them. This search for anticancer natural products from *C. reflexa* has yielded four natural products: Scoparone (1), p-coumaric acid (2), stigmasta-3,5-diene (3) and 1-O-p-hydroxycinnamoylglucose (4) and among them 1-O-p-hydroxycinnamoyldlucose (4) showed promising antiproliferative activities in HCT116 colorectal cell lines, whereas compounds 1-3 showed moderate activities (Riaz et al., 2017).

13. Identification of Tomato Initiation Lines with Increased Susceptibility or Resistance to Infection by Angiosperm Parasitic Extremely Large Dodder (*Cuscuta reflexa*)

The parasitic flowering plant genus *Cuscuta* (dodder) is a parasitic weed, which infects many economically crucialt crops. Once it winds around the shoots of potential host plants and initiates the development of penetration organs, called haustoria, only a few plant species have been shown to deploy effective defense mechanisms to ward off *Cuscuta* parasitization (Kaiser et al., 2015). However, a notable

exception is *Solanum lycopersicum* (tomato), which exhibits a local hypersensitive reaction when attacked by giant dodder (*Cuscuta reflexa*). Interestingly, the closely related wild desert tomato, *Solanum pennellii*, is unable to stop the penetration of its tissue by the *C. reflexa* haustoria. In this study, researchers observed that grafting a *S. pennellii* scion onto the rootstock of the resistant *S. lycopersicum* did not change the susceptibility phenotype of *S. pennellii*. This indicates that hormones, or other mobile substances, produced by *S. lycopersicum* do not induce a defense reaction in the susceptible tissue. Screening of a population of introgression lines harboring chromosome fragments from *S. pennellii* in the genome of the recurrent parent *S. lycopersicum*, revealed that most lines exhibit the same defense reaction as revealed by the *S. lycopersicum* parental line. Though, several lines showed different responses and exhibited either susceptibility, or cell death, which extended considerably beyond the infection site. These lines will be valuable for the future identification of key loci involved in the perception of, and resistance to, *C. reflexa* and for creating strategies to enhance resistance to infection in crop species (Kaiser et al., 2015).

14. Impact of Infection by *Cuscuta reflexa* Roxb on Androgen-Induced Alopecia

Alopecia is a psychologically distressing condition. Androgenetic alopecia, which affects millions of men and women, is an androgen-driven disorder. *Cuscuta reflexa* Roxb is evaluated for hair growth activity in androgen-induced alopecia (Pundir et al., 2008). Petroleum ether extract of *C. reflexa* was successfully studied for its hair growth-promoting activity. Alopecia was induced in albino mice through testosterone administration for the period of twenty days. Its inhibition by simultaneous administration of extract was monitored using follicular density, anagen/telogen ratio, and microscopic observation of skin sections. For investigating the mechanism of observed activity, *in vitro* experiments were conducted to study the effect of extract and its major component on activity of 5-alpha-reductase enzyme. Petroleum ether extract of *C. reflexa* exhibited promising hair growth-promoting activity as reflected from follicular density, anagen/telogen ratio, and skin sections. Inhibition of 5-alpha-reductase activity by extract and isolate indicate that the extract reversed androgen-induced alopecia by inhibiting conversion of testosterone to dihydrotestosterone. The petroleum ether extract of C. *reflexa* and its isolate is useful in treatment of androgen-induced alopecia by inhibiting the enzyme 5-alpha-reductase (Pundir et al., 2008).

15. Evidence for Abscisic Acid Biosynthesis in *Cuscuta reflexa*, A Parasitic Plant Lacking Neoxanthin

Abscisic acid (ABA) is a plant hormone observed in all higher plants; it plays a pivotal role in seed dormancy, embryo development, and adaptation to environmental stresses, most remarkably drought. The regulatory step in ABA synthesis is the cleavage reaction of a 9-cis-epoxy-carotenoid catalyzed by the 9-cis-epoxy-carotenoid dioxygenases (NCEDs). The parasitic angiosperm *Cuscuta reflexa* lacks neoxanthin, one of the common precursors of ABA in all higher plants (Qin et al., 2008). Therefore still a brainstorming concern of *C. reflexa* of being capable of synthesizing ABA, or does it acquire ABA from its host plants? Stem tips of *C. reflexa* were cultured in vitro and found to accumulate ABA in the absence of host plants (Qin et al., 2008); this demonstrating that this parasitic plant is capable of synthesizing ABA. Dehydration of detached stem tips caused a big rise in ABA content. Two NCED genes, CrNCED1 and CrNCED2, were cloned from *C. reflexa*. Expression of CrNCEDs was up-regulated significantly by dehydration. *In vitro* enzyme assays with recombinant CrNCED1 protein showed that the protein is able to cleave both 9-cis-violaxanthin and 9'-cis-neoxanthin to give xanthoxin. Therefore, despite the absence of neoxanthin in *C. reflexa*, the biochemical activity of CrNCED1 is similar to that of NCEDs from other higher plants (Qin et al., 2008). These data provide evidence for conservation of the ABA biosynthesis pathway among members of the plant kingdom (Qin et al., 2008).

16. *Cuscuta reflexa* Invasion Induces Calcium Release in Its Host.

Cuscuta reflexa has been reported to induce a variety of reaction in its hosts. Some of these are visual reactions, and it is clear that these morphological changes are preceded by events at the molecular level, where signal transduction is one of the early processes (Albert et al., 2010). Calcium (Ca^{2+}) release is the principal second messenger during signal transduction, and it has therefore been studied for monitoring Ca^{2+} spiking in tomato during infection with *C. reflexa*. Bioluminescence in aequorin-expressing tomato was successfully noticed for 48 h after the onset of *Cuscuta* infestation. Signals at the attachment sites were observed from 30 to 48 h. Treatment of aequorin-expressing tomato leaf disks with *Cuscuta* plant extracts suggested that the substance that induced Ca^{2+} release from the host was closely linked to parasitic haustoria (Albert et al., 2010).

17. Plastidic Genome Conformation and Degradation of Photosynthetic Activity in *Cuscuta*

The genus *Cuscuta* (dodder) is composed of parasitic plants, some species of that appear to be losing the ability to photosynthesize. A molecular phylogeny was constructed using 15 species of *Cuscuta* in order to assess whether changes in photosynthetic ability and alterations in structure of the plastid genome relate to

phylogenetic position within the genus (Revill et al., 2005). The molecular phylogeny provides evidence for four major clades within *Cuscuta*. Though DNA blot analysis revealed that *Cuscuta* species have smaller plastid genomes than tobacco, and that plastome size varied remarkably even within one *Cuscuta* clade, dot blot analysis reflected that the dodders possess homologous sequence to 101 genes from the tobacco plastome (Revill et al., 2005). Evidence is provided for significant rates of DNA transfer from plastid to nucleus in *Cuscuta*. Size and structure of *Cuscuta* plastid genomes, as well as photosynthetic ability, appear to vary on its own of position within the phylogeny, probably supporting the hypothesis that within *Cuscuta* photosynthetic capability and organization of the plastid genome are altering in a clumsy fashion (Revill et al., 2005).

18. Assessment of Total DNA Sequences of Plastidic Genomes of Two Parasitic Plant Species, Namely, *Cuscuta reflexa* and *Cuscuta gronovii*

The holoparasitic plant genus *Cuscuta* comprises species with photosynthetic capacity and functional chloroplasts as well as achlorophyllous and intermediate forms with restricted photosynthetic activity and degenerated chloroplasts (Funk et al., 2007). Previous data reflected significant differences with respect to the plastid genome coding capacity in different *Cuscuta* species, which could correlate with their photosynthetic activity. In order to getting new insights into molecular alterations accompanying the parasitic lifestyle, researchers sequenced the plastid chromosomes of the two species viz. *Cuscuta reflexa* and *Cuscuta* gronovii (Funk et al., 2007). Both species are having the potential of performing photosynthesis, though with varying efficiencies. Together with the plastid genome of *Epifagus virginiana*, an achlorophyllous parasitic plant whose plastid genome has been sequenced, these species represent a series of progression towards complete dependency on the host plant, ranging from reduced levels of photosynthesis in *C. reflexa* to a restricted photosynthetic activity and degenerated chloroplasts in *C. gronovii* to an achlorophyllous state in *E. virginiana* (Funk et al., 2007).. The newly sequenced plastid genomes of *C. reflexa* and *C. gronovii* reveal that the chromosome structures are basically very similar to that of non-parasitic plants, though a number of species-specific insertions, deletions and sequence inversions were recognized (Funk et al., 2007). Overall, the comparative genomic analysis of plastid DNA from parasitic plants reflects a partiality towards a simplification of the plastidic gene expression (Funk et al., 2007).

II. Conclusion

Conclusively, studies covered by this review article together entail that there are a number of complications, which are developed as a consequence of angiosperm host- parasite interaction, ultimately badly affecting crop yield (Mishra and Sanwal, 1992, 1994, 1995, 1997; Gibot-Leclerc et al., 2012; Fernández-Aparicio et al., 2016). Never-the-less, certain strategies may be developed based on background gathered herein, and probably be assisting to agricultural field in view of overcoming the concern of crop yield in near future.

Acknowledgement

Authors are grateful to Prof. M.P. Pandey (Vice Chancellor, IFTM University, Moradabad, India) for constant encouragement during accomplishment of this piece of work of author Danish Inam for his Ph.D. program.

References

[1]. Alakonya A, Kumar R, Koenig D, Kimura S, Townsley B, Runo S, Garces HM, Kang J, Yanez A, David-Schwartz R, Machuka J, Sinha N. 2012. Interspecific RNA interference of shoot meristemless-like disrupts Cuscuta pentagona plant parasitism. The Plant Cell 24: 3153-3166.

[2]. Alam MM, Begum F, Roy P. 2014. Yield and yield attributes of rapeseed-mustard (Brassica). Bangladesh Journal of Agricultural Research 39 (2): 311-336.

[3]. Albert M, Werner M, Proksch P, Fry SC, Kaldenhoff R. 2004. The cell wall-modifying xyloglucan endotransglycosylase/hydrolase LeXTH1 is expressed during the defence reaction of tomato against the plant parasite Cuscuta reflexa. Plant Biology 6: 402-407.

[4]. Albert M, Belastegui-Macadam X, Kaldenhoff R. 2006. An attack of the plant parasite Cuscuta reflexa induces the expression of attAGP, an attachment protein of the host tomato. The Plant Journal 48: 548-556

[5]. Albert M, van der Krol S, Kaldenhoff R. 2010. Cuscuta reflexa invasion induces Ca release in its host. Plant Biol (Stuttg) 12 (3): 554-557.

[6]. Aly R. 2007. Conventional and biotechnological approaches for control of parasitic weeds. In Vitro Cellular and Developmental Biology-Plant 43: 304-317.

[7]. Anami S, Njuguna E, Coussens G, Aesaert S, Van Lijsebettens M. 2013. Higher plant transformation: principles and molecular tools. The International Journal of Developmental Biology 57: 483-494.

[8]. Birschwilks M, Haupt S, Hofius D, Neumann S. 2006. Transfer of phloem-mobile substances from the host plants to the holoplastic Cuscuta Spp. Journal of Experimental Botany 57: 911-921.

[9]. Birschwilks M, Sauer N, Scheel D, Neumann S. 2007. Arabidopsis thaliana is a susceptible host plant for the holoparasite Cuscuta spec. Planta 226: 1231-1241.

[10]. Bleischwitz M, Albert M, Fuchsbauer HL, Kaldenhoff R. 2010. Significance of Cuscutain, a cysteine protease from Cuscuta reflexa, in host-parasite interactions. BMC Plant Biology 10: 227.

[11]. Boller T, Felix G. 2009. A renaissance of elicitors: perception of microbe-associated molecular patterns and danger signals by pattern-recognition receptors. Annual Review of Plant Biology 60: 379-406.

[12]. Borsics T, Mihalka V, Oreifig AS, Barany I, Lados M, Nagy I, Jenes B, Toldi O. 2002. Methods for genetic transformation of the parasitic weed dodder (Cuscuta trifolii Bab. et Gibs) and for PCR-based detection of early transformation events. Plant Science 162: 193-199.

[13]. Chebli Y, Geitmann A. 2017. Cellular growth in plants requires regulation of cell wall biochemistry. Current Opinion in Cell Biology 44: 28-35.

[14]. Christensen NM, Dorr I, Hansen M, van der Kooij TA, Schulz A. 2003. Development of Cuscuta species on a partially incompatible host: induction of xylem transfer cells. Protoplasma 220: 131-142.

[15]. Cosgrove DJ. 2005. Growth of the plant cell wall. Nature Reviews Molecular Cell Biology 6: 850-861.

[16]. Das P, Kar M, Sahoo S. 2011. In vitro hormone-regulated growth and floral induction of Cuscuta reflexa: a parasitic angiosperm. Acta Physiologiae Plantarum 33: 1031-1035

[17]. Dawson JH, Musselman LJ, Wolswinkel P, Dorr I. 1994. Biology and control of Cuscuta. Reviews of Weed Science 6: 265-317.

[18]. Doblin MS, Kurek I, Jacob-Wilk D, Delmer DP. 2002. Cellulose biosynthesis in plants: from genes to rosettes. Plant and Cell Physiology 43: 1407-1420.

[19]. Fernández-Aparicio M, Flores F, and Rubiales D. 2016. The effect of Orobanche crenata infection severity in faba bean, field pea, and grass pea productivity. Front Plant Sci. 7: 1409.

[20]. Frankova L, Fry SC. 2013. Biochemistry and physiological roles of enzymes that 'cut and paste' plant cell-wall polysaccharides. Journal of Experimental Botany 64: 3519-3550.

[21]. Funk HT, Berg S, Krupinska K, Maier UG, Krause K. 2007. Complete DNA sequences of the plastid genomes of two parasitic flowering plant species, Cuscuta reflexa and Cuscuta gronovii. BMC Plant Biol. 7: 45.

[22]. Garcia MA, Costea M, Kuzmina M, Stefanovic S. 2014. Phylogeny, character evolution, and biogeography of Cuscuta (dodders; Convolvulaceae) inferred from coding plastid and nuclear sequences. American Journal of Botany 101: 670-690.

[23]. Gibot-Leclerc S, Sallé G, Reboud X, Moreau D. 2012. What are the traits of Phelipanche ramosa (L.) Pomel that contribute to the success of its biological cycle on its host Brassica napus L? Flora 207: 512–521.

[24]. Hegenauer V, Fürst U, Kaiser B, Smoker M, Zipfel C, Felix G, Stahl M, Albert M. 2016.

[25]. Detection of the plant parasite Cuscuta reflexa by a tomato cell surface receptor. Science 353 (6298): 478-481.

[26]. Ikeue D, Schudoma C, Zhang W, Ogata Y, Sakamoto T, Kurata T, Furuhashi T, Kragler F, Aoki K. 2015. A bioinformatics approach to distinguish plantparasite and host transcriptomes in interface tissue by classifying RNA-Seq reads. Plant Methods 11: 34.

[27]. Jarvis MC, Briggs SPH, Knox JP. 2003. Intercellular adhesion and cell separation in plants. Plant, Cell and Environment 26: 977-989.

[28]. Johnsen HR. 2014. Doctoral thesis: Analysis of processes at the haustorial interfaces between Cuscuta reflexa and its hosts. UiT The Arctic University of Norway. Available at: http://hdl.handle.net/10037/9124.

[29]. Kaiser B, Vogg G, Furst UB, Albert M. 2015. Parasitic plants of the genus Cuscuta and their interaction with susceptible and resistant host plants. Frontiers in Plant Science 6: 45.

[30]. Lee KB. 2008. Anatomy and ultrastructure of epidermal cells in the haustorium of a parasitic flowering plant, Cuscuta japonica, during attachment to the host. Journal of Plant Biology 51: 366-372.

[31]. Lee KB. 2009. Structure and development of the endophyte in the parasitic angiosperm Cuscuta japonica. Journal of Plant Biology 52: 355-363.

[32]. Malinovsky FG, Fangel JU, Willats WG. 2014. The role of the cell wall in plant immunity. Frontiers in Plant Science 5: 178.

[33]. Mathpal P, Punetha H, Tewari AK, and S. Agrawal S. 2011. Investigation on defensive enzymes'activity of Brassica juncea genotypes. Journal of Oilseed Brassica 2(2): 87-94.

[34]. Mitsumasu K, Seto Y, Yoshida S. 2015. Apoplastic interactions between plants and plant root intruders. Frontiers in Plant Science 6: 617.

[35]. Mishra S, Sanwal GG. 1992. Alterations in lipid composition of seed oil from Brassica juncea upon infection by Cuscuta reflexa. Journal of Agricultural & Food Chemistry 40: 52-55.

[36]. Mishra S, Sanwal GG. 1994. Effect of Cuscuta infection on chloroplast lipid composition of Brassica leaves. European Journal of Plant Pathology 100: 61-70.

[37]. Mishra S, Sanwal GG. 1995. Changes in lipid composition of Brassica siliquae upon infection by Cuscuta. Journal of Plant Physiology 146: 303-306.

[38]. Mishra S, Sanwal GG. 1997. Effect of Cuscuta infection on chloroplast lipid composition of developing Brassica siliquae. Indian Journal of Agricultural Biochemistry 10: 6-10.

[39]. Pieterse CM, Van der Does D, Zamioudis C, Leon-Reyes A, Van Wees SC. 2012. Hormonal modulation of plant immunity. Annual Review of Cell and Developmental Biology 28: 489-521.

[40]. Popper ZA. 2008. Evolution and diversity of green plant cell walls. Current Opinion in Plant Biology 11: 286-292.

[41]. Popper ZA, Fry SC. 2008. Xyloglucan-pectin linkages are formed intra-protoplasmically, contribute to wall-assembly, and remain stable in the cell wall. Planta 227: 781-794.

[42]. Pandir S, Chauhan NS, Dixit VK. 2008. Effect of Cuscuta reflexa Roxb on androgen-induced alopecia. J Cosmet Dermatol. 7(3): 199-204.

[43]. Qin XQ, Yang SH, Kepsel AC, Schwartz SH, Zeevaart JAD. 2008. Evidence for abscisic acid biosynthesis in Cuscuta reflexa, a parasitic plant lacking neoxanthin. Plant Physiology 147 (2): 816-822.

[44]. Ranjan A, Ichihashi Y, Farhi M, Zumstein K, Townsley B, David-Schwartz R, Sinha NR. 2014. De novo assembly and characterization of the transcriptome of the parasitic weed Cuscuta pentagona identifies genes associated with plant parasitism. Plant Physiology 166: 1186– 1199.

[45]. Rehker J, Lachnit M, Kaldenhoff R. 2012. Molecular convergence of the parasitic plant species Cuscuta reflexa and Phelipanche aegyptiaca. Planta 236, 557-566.

[46]. Revill MJW, Stanley S, Hibberd JM. 2005. Plastid genome structure and loss of photosynthetic ability in the parasitic genus Cuscuta. Journal of Experimental Botany 56 (419): 2477- 2486.

[47]. Riaz M, Bilal A, Ali MS, Fatima I, Faisal A, Sherkheli MA, Asghar A. 2017. Natural products from Cuscuta reflexa Roxb. with antiproliferation activities in HCT116 colorectal cell lines. Natural Product Research 31 (5): 583-587.

[48]. Rose JKC, Braam J, Fry SC, Nishitani K. 2002. The XTH family of enzymes involved in xyloglucan endotransglucosylation and endohydrolysis: current perspectives and a new unifying nomenclature. Plant and Cell Physiology 43: 1421-1435.

[49]. Runyon JB, Mescher MC, Felton GW, De Moraes CM. 2010. Parasitism by Cuscuta pentagona sequentially induces JA and SA defence pathways in tomato. Plant, Cell and Environment 33: 290-303.

[50]. Shekhawat K, Rathore SS, Premi OP, Kandpal BK, and Chauhan JS. 2012. Advances in Agronomic Management of Indian Mustard

(Brassica juncea (L.) Czernj. Cosson): An
[51]. Overview. International Journal of Agronomy 2012: Article ID 408284:14 pages; doi:10.1155/2012/408284.
[52]. Sherman TD, Bowling AJ, Barger TW, Vaughn KC. 2008. The vestigial root of dodder (Cuscuta Pentagona) seedlings. International Journal of Plant Sciences 169: 998-1012.
[53]. Scheller HV, Ulvskov P. 2010. Hemicelluloses. Annual Review of Plant Biology 61: 263-289.
[54]. Shigenaga AM, Argueso CT. 2016. No hormone to rule them all: Interactions of plant hormones during the responses of plants to pathogens. Seminars in Cell Developmental Biology 56: 174-189.
[55]. Svubova R, Blehova A. 2013. Stable transformation and actin visualization in callus cultures of dodder (Cuscuta europaea). Biologia 68: 633-640.
[56]. Thompson JE, Fry SC. 2000. Evidence for covalent linkage between xyloglucan and acidic pectins in suspension-cultured rose cells. Planta 211: 275-286.
[57]. van der Kooij TA, Krause K, Dorr I, Krupinska K. 2000. Molecular, functional and ultrastructural characterisation of plastids from six species of the parasitic flowering plant genus Cuscuta. Planta 210: 701-707.
[58]. Vaughan KC. 2002. Attachment of the parasitic weed dodder to the host. Protoplasma 219: 227- 237.
[59]. Vaughan KC. 2003. Dodder hyphae invade the host: a structural and immunocytochemical characterization. Protoplasma 220: 189-200.
[60]. Vaughan KC. 2006. Conversion of the searching hyphae of dodder into xylic and phloic hyphae: a cytochemical and immunocytochemical investigation. International Journal of Plant Sciences 167: 1099-1114.
[61]. Wakabayashi K, Hoson T, Huber DJ. 2003. Methyl de-esterification as a major factor regulating the extent of pectin depolymerization during fruit ripening: a comparison of the action of avocado (Persea americana) and tomato (Lycopersicon esculentum) polygalacturonases. Journal of Plant Physiology 160: 667-673.
[62]. Werner M, Uehlein N, Proksch P, Kaldenhoff R. 2001. Characterization of two tomato aquaporins and expression during the incompatible interaction of tomato with the plant parasite Cuscuta reflexa. Planta 213: 550-555.
[63]. Willats WG, McCartney L, Mackie W, Knox JP. 2001. Pectin: cell biology and prospects for functional analysis. Plant Molecular Biology 47: 9-27.
[64]. Yang Z, Wafula EK, Honaas LA, Zhang H, Das M, Fernandez-Aparicio M, Huang K, Bandaranayake PC, Wu B, Der JP, Clarke CR, Ralph PE, Landherr L, Altman NS, Timko MP, Yoder JI, Westwood JH, dePamphilis CW. 2015. Comparative transcriptome analyses reveal core parasitism genes and suggest gene duplication and repurposing as sources of structural novelty. Molecular Biology and Evolution 32: 767-790.

IOSR Journal of Biotechnology and Biochemistry (IOSR-JBB) is UGC approved Journal with Sl. No. 4033, Journal no. 44202.

Dr. Sanjay Mishra "Biochemical and Molecular Biological Alterations in Angiospermic Plants upon Infection by Angiosperm Parasite, Cuscuta spp.: An Overview." IOSR Journal of Biotechnology and Biochemistry (IOSR-JBB) 5.6 (2019): 01-09.