Harnessing Microbial and Agricultural Systems to Transform the Wheat Supply Chain

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Wheat (*Triticum aestivum* L. and related *Triticum* spp.) is one of the most important crops grown worldwide. Wheat and rice (*Oryza* spp.) each provide about 19% of global dietary energy, making these crops two of the most important sources of human nutrition (33). Wheat, however, is unique and complex in many ways. There are several classes of wheat that are adapted to specific geographic areas, have unique value chains, and are used as ingredients in many food products, such as breads, crackers, cookies, cakes, beer, and pasta. The wheat genome is complex as well: durum wheat (*Triticum turgidum* L. subsp. *durum* (Desf.) Husn.) is a tetraploid with A and B genomes, and other classes of wheat are hexaploids with A, B, and D genomes (24). The three genomes originated from natural hybridizations between diploid ancestral species of *Triticum* and *Aegelops* that occurred 6.5 million years ago, evolving into hexaploid wheat species about 400,000 years ago. As agriculture developed over the last 10,000 years, humans selected and cultivated lines that produced high yields, were free-threshing, and had nonbrittle rachis and plump grains.

Despite the critical role of wheat as a staple crop in many regions worldwide, there are concerns that the pace of improvement will diminish, and as the global human population and its affluence increase, it is predicted that wheat production will not meet demands. Ray et al. (33) conducted an analysis of the rates of production gains for four primary global crops: corn (*Zea mays* L.), rice, wheat, and soybean (*Glycine max* (L.) Merr.). From 1961 to 2008, the global average rates of yield increase were 1.6, 1.0, 0.9, and 1.3%/year for corn, rice, wheat, and soybean, respectively. To meet the projected food requirements of 9 billion people by 2050, however, average rates of yield gain must reach 2.4%/year for each of these four crops. Although certain localized areas have been realizing wheat yield gains of 2.4%/year, many more areas are either well below this rate of gain or have even experienced production declines.

The overall economic performance of U.S. agriculture has been slowing as well. From 1948 to 2015, the average annual rate of agricultural output growth was 1.48%/year, resulting in a 2.7-fold increase in output during the period (42). Input use increased by 0.1%/year over this period, so the resulting overall productivity was 1.38%/year. In recent years, however, the economic pace has slowed, with the output rate of gain dropping to 0.72%/year, input use nearly doubling to 0.19%/year, and a net productivity of 0.53%/year, which is 38% of the rate over the entire 1948–2015 period.

Wheat growers have long recognized that they have not benefited from the technological advancements and investments that corn and soybean producers have received, and the rela-

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tively slow yield gains of 0.9%/year for wheat substantiate this concern. In addition to yield, the sustainability of wheat production, which is often studied in terms of land use, soil conservation, and restricted use of irrigation and other practices that contribute to greenhouse gas emissions, has also lagged behind other row crops. From 1980 to 2015, wheat producers globally decreased irrigation water use by 26%, energy use by 22%, and greenhouse gas emissions by 9%; however, for corn and soybeans these metrics were double to triple those for wheat (10). Moreover, as a C_3 species, wheat is expected to be among the most vulnerable crops with regard to rising temperatures resulting from climate change—a 1 degree Celsius temperature increase is predicted to contribute a 4.1–6.4% decline in global wheat production (22). In 2007, the National Association of Wheat Growers and U.S. Wheat Associates passed resolutions to coordinate with growers in Canada and Australia to "research, develop, and commercialize" technologies for wheat that will improve the competitive position of wheat relative to other crops (7).

New technologies need to be developed to drive enhanced productivity and capture value from wheat and other staple crops. Improvements in traditional technologies, such as plant breeding and genetics, will continue to be critical for ongoing gains in agriculture. New technologies must be developed concurrently, however, to better realize the genetic yield potential that already exists in modern cultivars. Lee et al. (21) identified coordination of biological sciences, information and communication technologies, and robotics as necessary to drive improvements in production efficiency (operational excellence), innovative market development (supply chain orchestration), and nearly real-time access to supply chain information (transparency). Implementation of advanced analytical tools throughout the system will play a critical role in achieving the production gains necessary to meet projected demands.

Businesses have begun using advanced analytical tools to enhance agricultural productivity, deliver products that meet consumer needs, protect the environment, and improve economic conditions for growers. Indigo Ag, Inc. is an example of a company using advanced analytical tools to balance the many demands placed on agriculture through the lenses of farmer profitability, environmental sustainability, and consumer well-being. Indigo is using high-throughput genetic sequencing and machine learning to select natural symbiotic endophytes that can improve crop production. It is also using modern technologies, such as blockchains, to segregate and market crops to achieve greater supply chain efficiencies and capture value for growers. Indigo's endophyte technologies are being developed to optimize plant microbiomes that better capture crop yield potential, particularly crops grown under severe abiotic and biotic stresses, while concurrently reducing the use of irrigation, chemical fertilizers, and pesticides. Indigo's on-site crop advisors and postharvest grain management strategies are aimed at more tightly linking crop production to food processors and consumers. Crop quality measures are obtained at the point of field production, so the crop can be delivered to better meet the specifications set by food processors, and crop quality analysis is used to segregate grains for identity preservation. When broadly implemented to support microbiome development, improved crop management, and sophisticated marketing, use of advanced analytical tools will also enhance crop quality for food manufacturers and consumers. Endophytes can be developed to improve the nutritional composition of wheat, such as increased protein concentration and mineral fortification, as well. In addition, routine and rapid analysis of crop quality can provide growers with better supply chain orchestration. Ultimately, use of advanced analytics, the plant microbiome, and postharvest management strategies will generate better economic outcomes for growers and enhanced well-being for consumers. Given the demonstrated lags, compared with other row crops, wheat in particular stands to gain from the transformations that companies like Indigo can inspire.

The urgent needs to increase crop yields and improve the quality, resiliency, and sustainability of wheat go hand-in-hand. The necessary production gains of 2.4%/year must be achieved while concomitantly using less water and fewer chemicals, continuing production on degraded soils, and facing extreme weather shocks, gradual temperature rises, and new pests and diseases (11). Although there is no single solution that can address these obstacles, a systems approach, with the science of the microbiome at its core, can make great strides. This article presents an overview of the technologies that Indigo is using to enable its unique approach to improving agricultural production and rural economic growth.

The Plant Microbiome

Public awareness about the important interrelationships between humans and microorganisms is increasing. Throughout much of the 20th century it was widely believed that human health would benefit if microorganisms were controlled or eliminated on and in the human body. This turned out to be an arduous, risky, and unsustainable approach to human health. As time passed, human health scientists became increasingly aware of how people benefit from microorganism populations on and in our bodies. Savage (38) reported that the ratio of microbial cells to human cells for an individual person was typically 10:1, with the majority of the microbial cells located in the human gastrointestinal tract. More recent research indicates that a balanced microbial population base is an essential aspect of human health (29). In 2001, the term and concept of the "microbiome" was introduced by Lederberg and McCray (20) to "signify the ecological community of commensal, symbiotic, and pathogenic microorganisms that share our body space."

The new emphasis on the human microbiome has led to the development of sensitive tools and approaches to studying and characterizing complex microbial populations. The human microbiome research community developed the approaches of metagenomics, metatranscriptomics, and metaproteomics, as well as the use of 16s rRNA and internal transcribed spacer (ITS) primers, to examine complex microbial populations and data archives with genetic sequence databases that are maintained and shared among scientists. These approaches allow for simultaneous study of the wide array of microbial populations in an ecosystem (41). This rapid development of sophisticated tools also benefits researchers investigating other microbiomes, including soil, animal, and plant microbiomes, as well as ecosystem-wide approaches to studying complex interactions involving microbial populations.

A clearer understanding of the symbiotic relationships between plants and bacteria has been acquired over many decades of research. In the late 19th century, Russian microbiologist Sergei Winogradsky discovered that the anaerobic bacterium *Clostridium pasteurianum* is capable of fixing atmospheric nitrogen. His discovery led to subsequent work on symbiotic relationships between *Rhizobium* spp. and plants in the family *Leguminosae* (6,31). Today, research and development is underway in plants beyond the family *Leguminosae*, and understanding of synergistic plant–microbe interactions is expanding rapidly. The current state of knowledge indicates that all plant tissues are hosts to complex microbial communities, which are dominated by fungi and bacteria (37,41). These complex communities, known as the plant microbiome or phytobiome, are adapted to 1) the rhizosphere, which exists on the surfaces of plant tissues that are underground or in contact with the soil (e.g., roots, rhizomes, and stolons); 2) the phyllosphere, which exists on the surfaces of aerial plant tissues (e.g., leaves, stems, flowers, etc.); and 3) the endosphere, which exists internally within plant tissues.

Rhizospheres are regarded as having microbial species populations that are particularly rich compared with phyllospheric and endospheric populations (2). Consequently, most research on plant–microorganism interactions has focused on the rhizosphere (2). There is considerable overlap of endophytic species with those present in the rhizosphere, indicating that many rhizosphere microorganisms are capable of colonizing internal plant tissues (37,41). Primary access points include root nodules, cracks, wounds, lenticels, and points of emergence of adventitious roots (37). Phyllospheric microorganisms capable of colonizing the endosphere often enter through stomata and floral tissues (28,37). Additionally, plant endophytes may be passed along from one generation to the next by vertical transmission through seeds and vegetative propagation (37). Endophytic bacteria and fungi primarily occupy apoplastic spaces among plant cells or necrotic or senescing tissues (41). Moreover, endophytes can relocate throughout the plant through xylem transport (41).

Endophyte–Host Plant Interactions

Khan et al. (16) provided historic background on early observations of endophytes. Fungal endophytes were first reported in 1898 by Vogl and in 1904 by Freeman, and bacterial endophytes were first reported in 1926 by Perotti (16). Endophytic bacteria and fungi are of particular interest with regard to agronomic crop management. Because of their location inside plants, endophytes are uniquely positioned to influence plant growth and can be managed to help control plant diseases, increase and stabilize crop productivity, reduce chemical inputs, reduce agricultural greenhouse gas emissions, and meet other objectives that support greater sustainability. The interspersed population of endophytes within plant tissues in essence makes these endophytes an extended or second genome of the crop system, collectively functioning as a pan-genome (37,41).

Most of the recent literature defines endophytes as microorganisms that live inside plant tissues without causing symptoms of disease. With regard to bacteria, Turner et al. (41) recently stated "endophytic bacteria are those that reside for at least part of their lives within plant tissues…[t]hey are generally considered to be non-pathogenic, causing no visible symptoms, but they may include latent pathogens that, depending on environmental circumstances and/or host genotype, can cause disease." In contrast, in 1866 De Bary defined endophytes as "any organism occurring within plant tissues" (2). Lata et al. (19) categorized endophytes into three types based on pathogenicity: 1) pathogens of another host that are nonpathogenic in their endophytic relationships; 2) nonpathogenic microorganisms; and 3) pathogens that have been rendered nonpathogenic but are still capable of colonization by selection methods or genetic alterations. Despite these definitions, which are based on degrees of pathogenicity, there is building interest in determining how endophytes enhance plant growth and mechanisms of plant–endophyte symbiosis.

Mechanisms of Growth-Promoting Endophytes

Many bacteria and fungi develop symbiotic communities in the rhizosphere, phyllosphere, and endosphere of host plants. Endophytes are of particular interest for technological development because these microorganisms can be placed to directly interact with and respond to signals from host plants. Khan et al. (16) stated that endophytes have coevolved with plants and developed chemical signals in complex systems, citing Schulz and Boyle (39), who reported that about 51% of biologically active metabolites in plants originate from endophytes.

Santoyo et al. (37) recently posited that endophytes promote host plant growth and health in two ways—either through direct or indirect mechanisms. Direct mechanisms entail facilitating acquisition of nutrients and water, as well as modulation of hormones and plant growth-promoting compounds in the plant (37). Examples of endophyte-produced plant hormones include auxins, abscisic acid (ABA), cytokinin, and gibberellins (32,37). Indirect mechanisms are primarily those that limit damage caused by phytopathogens, insects, nematodes, and herbivores, acting mostly through production of metabolites that confer containment, nutrient starvation, antibiosis, or nonpreference responses.

Accumulating evidence indicates that direct mechanisms of endophytic bacteria and fungi alleviate abiotic stress. One example is bacteria that produce 1-aminocyclopropane-1-carboxylate (ACC) deaminase, such as the endophyte *Burkholderia phytofirmans* PsJN (32,37). These bacteria have the capability to modulate ethylene status in plant cells. Increased ethylene production is a common plant response to chronic abiotic stress, such as osmotic stress from drought or saline conditions, water-saturated soils, heat, or cold. Common morphological and phenological responses to increased ethylene status include inhibited root growth, restricted stem elongation, reduced leaf area, and accelerated senescence. Plants increase production of ACC as a stress response, and ACC oxidase catalyzes conversion of ACC to ethylene. In turn, ethylene signals meristematic tissues to restrict growth. This plant response can be modulated by endophytic bacteria that produce ACC deaminase. The bacteria take up ACC produced by plant cells and metabolize it to ammonia and α-ketobutyrate, thereby preventing ethylene production. As a consequence, plants colonized by these endophytes have greater root growth than those not colonized by ACC deaminase-producing endophytes. Larger root systems provide increased access to water and oxygen in the soil, accumulation of greater carbohydrate reserves, and other factors that later support plant growth.

Endophytic fungi also facilitate host plant health under abiotic stress. Khan et al. (16) and Waqas et al. (44) studied associations between cucumber (*Cucumis sativus* L.) and the endophytes *Phoma glomerata* LWL2, *Penicillium* sp. LWL3, *Exophiala* sp. LHL08, and *Paecilomyces formosus* LHL10. In laboratory-based evaluations, plants colonized by these fungal endophytes had greater plant growth, chlorophyll concentration, and leaf area than noncolonized plants. Notably, the larger root systems of the endophyte-colonized plants were able to retrieve greater amounts of water from sources that noncolonized plants under stress could not access (16). Additionally, ABA synthesis was downregulated, and relatively greater stomatal conductance was likely maintained under stress. Khan et al. (16) postulated that endophyte-mediated modulation of plant ABA levels by fungal production of gibberellins occurred.

Shehata et al. (40) reported on their extensive work regarding vertical transmission of an endophyte for continued delivery of an indirect antibiosis mechanism. Chapalote is a landrace of corn that has been grown continuously by subsistence farmers in southern Mexico for more than 3,000 years because it consistently demonstrates resistance to phytopathogens without use of fungicides. The research team isolated the endophytic bacteria *Burkholderia gladioli* 3A12 from Chapalote and demonstrated that it is vertically conserved in the seed from one generation to the next. The endophyte expresses antifungal properties by first swarming and attaching to fungal phytopathogens and then isolating the fungus with biofilm. Afterward, the endophytic bacteria kill the fungal hyphae. In short, the bacteria are able to recognize the pathogen, contain it, and facilitate control.

Endophytes in Wheat

Several investigators have conducted surveys to characterize endophyte communities in wheat. Marshall et al. (25) assayed wild species of *Triticum* that were collected in Turkey. Their work was done prior to development of metagenomic techniques and instead utilized microscopy to search for fungal hyphae. They found only two fungal endophytes in diploid *Triticum* spp., and although they did not observe expansive endophyte communities, they confirmed vertical transmission of *Neotyphodium* spp. endophytes. Shortly thereafter, Zinniel et al. (45) conducted a survey of 27 prairie plant species and 4 crop species, including wheat, for the presence of endophytic bacteria. They isolated 853 strains, including 28 isolates from wheat. Larran et al. (17) also reported on their survey conducted on wheat leaves. They isolated 130 fungal and 3 bacterial endophytes. They found that leaf endophyte communities became more abundant as the crop developed, but there were no differences among the three cultivars studied. Four of the fungi consistently dominated the endophyte community: *Rhodotorula rubra*, *Alternaria alternata*, *Cladosporium herbarum*, and *Epicoccum nigrum*. Larran et al. (17) concluded that although a large number of endophytic species are often observed, only a few species are present in significant numbers. They categorized the endophytes they observed into three groups: 1) well-known and economically important pathogens of wheat; 2) commonly abundant phylloplane fungi considered to be primarily saprobic and minor pathogens; and 3) species only occasionally present in wheat.

Gdanetz and Trail (12) conducted an extensive field survey of microbiome communities on wheat produced in four production systems: conventional tillage, no-till, low-input, and organic. Although they used sensitive assays (ITS and 16s) and they examined leaf, stem, and root tissues, their study did not distinguish between epiphytes and endophytes. Nevertheless, they isolated 1,634 fungal and 1,112 bacterial species. They found that microbiome communities became more abundant as the crop developed; however, production systems did not have a strong influence on wheat microbiome communities.

Ofek-Lalzar et al. (30) examined fungal endophytes in wheat and two wild ancestors of wheat: wild emmer (*Triticum dicoccoides* L.) and Sharon goatgrass (*Aegilops sharonensis* Eig.). They observed a greater abundance of fungal endophytes in the wild grasses than in wheat, and there was a "narrow core community of *Alternaria* species" in all three plant species examined.

Robinson et al. (35) examined the effects of crop production system on wheat endophyte communities. Surprisingly, bacterial endophytes were most abundant when crop fertilizer inputs were minimized. They estimated that bacterial species abundance was 42% greater for the unfertilized entries than for five other fertilizer treatments, including manure, two nitrogen levels, and a mixture of Mg, P, and K. Manure applications had a negative effect on *Actinobacteria* populations.

Wheat Responses to Endophytes

Research conducted in laboratory, greenhouse, and field settings showed positive responses by wheat plants to endophytes affecting plant growth, grain yield components, and physiology. Reports of field observations conducted by Indigo indicate that the yield of hard red winter wheat produced from certified seed treated with a bacterial endophyte was 8.3% greater than for the crop established from untreated certified seed when averaged across 14 farm locations in Kansas during the 2016–2017 growing season (unpublished data). On average, the yield for endophyte-treated crops was 16% greater than for untreated controls for the seven driest or most heat-stressed farm locations, where yields for untreated controls ranged from 2.3 to 4.3 MT/ha. Initial reports for the 2017–2018 season indicate that the yield for endophyte-treated wheat seed was 13% greater than for untreated control seed on 24 farm fields in Texas, Oklahoma, and Kansas (unpublished data). Indigo endophyte-treated seeds produced 19% greater yields than untreated controls for the 17 driest locations, where yields for untreated controls ranged from 0.7 to 1.5 MT/ha. Indigo's public reports claim improved crop production resulting from inoculation of seed with endophytes, with no difference in the chemical fertilizer or pesticide applications that were used for the untreated controls. Field harvest areas ranged from 0.4 to 63.0 ha. The endophytes used by Indigo are indigenous to U.S. agricultural croplands and are applied to crops as a seed treatment at the time of planting.

Indigo routinely conducts seedling growth studies in the laboratory and under field conditions to screen for endophytes and evaluate processes. Wheat endophytes of interest exhibit enhanced seedling growth (Fig. 1), which influences grain yield components during later stages of phenological development. Enhanced seedling development, especially root growth, is also often observed under field conditions (Fig. 2).

Indigo's commercial-scale observations are consistent with experiments reported in the literature. Colla et al. (5) studied the

Fig. 1. Seedling growth of hard red spring wheat at 7 days postgermination on agar from seeds either treated with a fungal endophyte (right) or not treated with the endophyte (left).

growth responses of durum wheat to a combination of three endophytic fungi applied as a seed treatment: *Glomus intraradices* BEG72, *Glomus mosseae*, and *Trichoderma atroviride* MUCL 45632. In a growth chamber experiment they observed that endophyte-treated entries measured 17 days after planting had a 10.0% higher chlorophyll concentration and 23.1 and 64.2% greater shoot and root dry weights, respectively. Indigo also has often observed noticeably greater chlorophyll concentrations for endophyte-treated wheat than for untreated controls (Fig. 3). Colla et al. (5) conducted field-based experiments during two growing seasons, 2011–2012 and 2012–2013, with 313 and 900 mm of precipitation, respectively. Endophyte-treated entries had 32.1 and 8.3% greater grain yields than untreated entries for the 2011–2012 and 2012–2013 seasons, respectively. Endophyte treatments did not affect mean seed weight, so differences in yield were attributed to greater numbers of seeds per plant for endophyte-treated entries. Grain protein concentration was 6.3% greater for the endophyte-treated entries than for the untreated entries for the dry 2011–2012 season but did not differ significantly between treatments for 2012–2013. Mineral concentrations in leaf tissues were consistently higher for the endophyte-treated entries than for the untreated entries. Liu et al. (23) studied wheat plant responses to the bacterial endophyte *Azorhizobium caulinodans* ORS571. They reported that seedling root and shoot lengths were 17.0 and 8.4% greater, respectively, for the endophyte-treated wheat than for untreated wheat, which was consistent with observations at Indigo and in other reports. Amelioration of water stress can be visualized by multispectral imagery. Multispectral images obtained by Indigo show that endophyte-treated wheat exhibits less stress than untreated controls (Fig. 4). Colla et al. (5) also remarked that their observations were consistent with previous studies that showed endophytic-fungi treatments in wheat ameliorated drought stress and other forms of biotic and abiotic stresses, thereby providing enhanced yield stability and sustainability.

Fig. 2. Seedling growth of hard red winter wheat established from Indigo certified seed treated with a bacterial endophyte (left) or not treated with an endophyte (right). The sample date was January 8, 2018, approximately 14 weeks after planting in a farm field in Gove County, Kansas.

Fig. 3. Adjoining fields of hard red winter wheat established in Kansas for the 2017–2018 growing season. The Indigo endophyte-treated field is on the right, and the untreated control field is on the left.

Hubbard et al. (14) studied the responses of durum wheat to heat and drought stress after treatment with six isolates of an endophytic ascomycetous mitosporic fungus. Three of the six fungal isolates ameliorated heat stress (36°C) better than drought stress. For average seed weight in the heat stress experiments, four of the isolates ameliorated stress, while one of the isolates exacerbated it. Although stem length did not differ among the entries when grown under drought stress, average seed weight was greater for four of the isolates than for the untreated control. Three of the four isolates alleviated both heat stress and drought. Stable carbon isotope discrimination is an integrated measure of stomatal conductance (9). Three of the isolate treatments resulted in increased stable carbon isotope discrimination under drought stress, indicating relatively greater conductance as a result of these endophyte treatments when wheat plants were stressed, which resulted in increased grain yields.

Chen et al. (4) described a novel species of bacteria (proposed as *Pantoea alhagi* sp. nov.) isolated from *Alhagi sparsifolia* Shap., a drought-tolerant legume native to northwestern China. *Pantoea alhagi* LTYR-11Z^T was studied for its effects on growth of water-stressed winter wheat. For well-watered entries, endophyte-treated wheat seedlings had 29.2 and 20.8% greater root length and plant fresh weight, respectively, than the nonstressed control. Water-stressed plants that were inoculated with the endophyte had 17.1, 41.8, and 112% greater shoot length, root length, and plant fresh weight, respectively, than the water-stressed control.

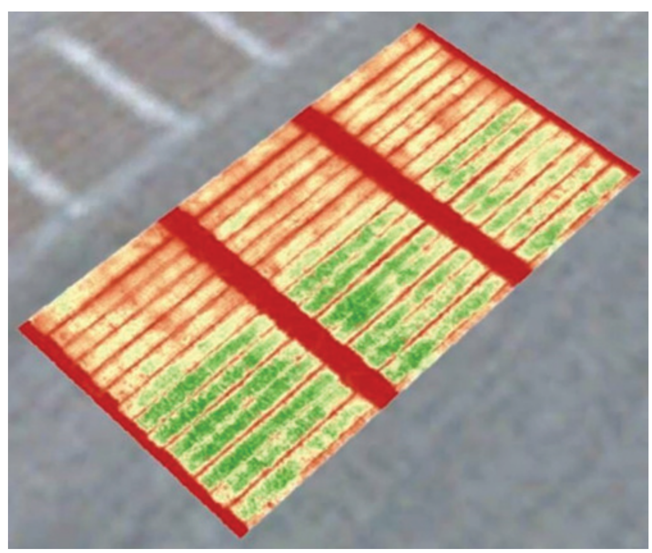

Fig. 4. A multispectral image obtained from an unmanned aerial system platform showing spectral differences between endophyte-treated wheat (lower right) and untreated wheat (upper left). The image indicates the endophyte-treated wheat is under less stress than the untreated wheat.

There are an increasing number of studies reporting on the plant growth-promotion properties of endophytes. In some instances endophytes exhibit no growth promotion when plant growth conditions are nearly optimal, as demonstrated by Sánchez-Rodrígues et al. (36) with bread wheat and durum wheat. Conversely, Chen et al. (4) reported that their novel bacterial endophyte both enhanced growth and ameliorated drought stress. They suggested that endophytes may provide "stress independent," as well as "stress dependent," direct mechanisms.

There have been other reports of direct synergistic mechanisms in wheat. Mitter et al. (28) observed that endophyte-colonized, field-grown wheat plants reached the heading stage of development 5 days earlier than untreated control plants. Vujanovic et al. (43) reported that treatment with a fungal isolate resulted in reductions in seed dormancy, faster germination, and improved seedling vigor in durum wheat. They suggested that the endophyte heightened the gibberellin status, resulting in breakage of dormancy.

Antagonism toward phytopathogens was described earlier as an indirect mechanism of plant growth promotion by endophytes. Recent research has shown potential for use of endophytes as biocontrol agents for tan spot (*Pyrenophora tritici-repentis*) and head blight (*Fusarium graminearum*) in wheat. Herrera et al. (13) examined four bacterial isolates of the genus *Paenibacillus*, one of the genus *Pantoea*, and one isolate that was identified as being from either the genus *Bacillus* or the genus *Fictibacillus*. The *Pantoea* and *Paenibacillus* isolates restricted growth of *Fusarium graminearum* in bioassays, with the *Paenibacillus* isolates releasing antifungal substances into the culture medium. Larran et al. (18) examined nine bacterial and fungal endophytes for control of tan spot in wheat using greenhouse-based, dual-plate bioassays. Endophyte strains of *Bacillus* spp. and *Fusarium* spp. reduced spore germination of *Pyrenophora tritici-repentis* by 82 and 52%, respectively. The endophyte *Trichoderma hamatum* showed the greatest antagonistic effect—the mean percentage of leaf area diseased was 53% less for the endophyte-treated plants than for the controls.

Endophytes and Food

There is a paucity of information on the effects of endophytes on food quality and functionality. Because endophytes occupy internal plant tissues, they have the potential to affect the properties and functionality of foods. Khan et al. (16) listed several bioactive compounds that are produced by endophytes, including flavonoids, peptides, alkaloids, steroids, phenolics, terpenoids, lignans, and volatile organic compounds. These compounds are known to influence the sensory characteristics of foods (e.g., taste and smell), and there is increasing evidence that these groups of compounds may benefit human health (8). Additionally, endophytes in raw food products, such as fresh fruits and vegetables, can influence the human microbiome and potentially improve human health (2). Endophytes may also be used to improve plant access to soil nutrients.

Minervini et al. (26) reported on several endophytic bacteria isolated from durum wheat that produce lactic acid, including *Lactobacillus*, *Streptococcus*, *Enterococcus*, and *Lactococcus* spp. They observed that these bacteria, which were isolated from wheat spikes, were also present in the resulting flour. More recently, Minervini et al. (27) isolated specific lines from durum wheat spikes, including *Lactobacillus plantarum* LA1, LB2, OLB3, OLD1, OLB4, and OLC4; *Lactobacillus rossiae* OLC1; and *Enterococcus faecalis* LA2. They compared sourdough fermentation of these lines with that of *Lactobacillus sanfranciscensis* A4 isolated from sourdough sponge. They reported that *Lactobacillus plantarum* LB2 persisted during dough fermentation and resulted in greater acidification of the dough than did the *Lactobacillus sanfranciscensis* A4 control.

Profitability for Growers

Although pressure is mounting for wheat farmers and technology providers to improve production practices to meet evergrowing consumer demands, the economics make it difficult for them to do so. Commodity wheat prices have historically been volatile but have followed a downward trend over the past decade, partially caused by a surplus global supply. Farmers who rely on wheat as a principal crop may not have sufficient liquidity to invest in more efficient equipment or implement new farming systems. They may also suffer from information asymmetry and not be able to access or afford data that would help them optimize their management decisions.

This cycle points to the need for a systems approach in the wheat sector; however, new technologies and methods for improving sustainability that are brought to market are often inaccessible or unaffordable to wheat farmers. Companies like Indigo are seeing successful adoption of more sustainable practices through sharing of the risks and rewards with growers. For example, Indigo enters into contractual relationships with farmers through which added value is created from 1) endophyte technologies; 2) continuous agronomic insights from crop advisors; 3) on-farm storage capacity; 4) extensive crop quality analysis; and 5) market analysis. Indigo also partners with seed companies to provide certified seed of modern cultivars that are sought by millers and food manufacturers. Farmers that contract their production to Indigo receive on-site consultation by trained agronomists who advise farmers on how to optimize crop yield and end-use quality. A portion of the anticipated added value is promised to the grower at the time of planting, when production contracts are executed. Grain samples are systematically collected during harvest to determine information on grain grade, structure, milling characteristics, and baking characteristics. Indigo also provides financial support for on-farm storage so that harvested crops can be segregated to maintain their inherent value rather than being commingled in community storage systems, where their inherent value is lost. Indigo leads grain marketing efforts before and after harvest to optimize profits for both the growers and Indigo.

When Indigo's endophyte seed treatments are combined with high-yield cultivars, it is reasonable to expect a 20% yield increase in stressful environments. In dryland production systems that typically produce 2.7 MT/ha based on USDA actual production histories (APHs), an increase of 0.6 MT/ha would be expected. The yield increase alone provides for added value to the farmer; however, Indigo also promises a contractual incentive payment for the total contracted crop produced. The incentive for the 2018 harvest was $15.80/MT. In this scenario, the expected increase in gross revenue for the farmer would be $141/ha.

Farmers not only have access to game-changing technologies, like endophyte seed treatments, but are also supported by education, finance, and robust data science systems, including the latest in sensor technologies. With this systems approach, Indigo not only mitigates trade-offs and risks for growers in adopting new technology, but also provides a solid knowledge base for future decision-making. This ultimately leads growers to make optimal decisions concerning planting, irrigation, chemical use, and protecting soils and adjacent watersheds, all while increasing wheat yields and quality to meet and capitalize on growing global demand.

Consumer Benefits

Consumer food selections have historically been driven by taste, price, and convenience—all dubbed "traditional drivers." New factors, including health and wellness, social impact, and transparency, are now influencing the purchasing decisions of roughly half of consumers (34). Consumers are more concerned than ever about food identity preservation and traceability. A growing number of consumers also want to know where their food comes from, what health-promoting attributes it possesses, and how it was produced, processed, and transported. Given the prominence of wheat in human diets, it does not escape consumer scrutiny, and knowing that these consumers are making decisions with their wallets, brands are inclined to be responsive. For example, the Campbell Soup Company recognizes that wheat is a high-priority ingredient and has committed to enrolling 28,000 ha in a fertilizer optimization program (3). The Kellogg Company has also set ambitious goals to reduce energy and water use that extend to its grain suppliers and supply chains, committing to responsible sourcing of wheat and helping more than 500,000 farmers adopt climate-smart agricultural practices (15).

The current commodity-based supply chain does not adequately address consumer desires for improved identity preservation, traceability, and sustainability. To bridge these gaps, companies such as Indigo are implementing several strategies. First, Indigo is more tightly linking growers, processors, and consumers to create a robustly documented and traceable supply chain, which will allow consumers to identify the source and unique features of their chosen wheat products, as well as how they were processed and handled. For example, should there be issues with product recalls, consumers would be reassured that the supply chain could be rapidly retraced to identify the origins and transportation history of the recalled product. Second, the identity of Indigo wheat is preserved by limiting grain comingling in grain elevators and instead supporting greater access to on-farm storage. Third, Indigo is improving agricultural sustainability on two fronts. Endophytes are used to improve plant root structure, enabling crops to access water from deeper within the soil profile and, thereby, reducing irrigation and fertilizer applications. Additionally, longer supply chains with multiple intermediaries negatively affect sustainability because they involve crop transport to nonlocal environments. By reorchestrating the wheat supply chain Indigo will enable greater local production and improve the connection between growers and consumers.

Partnering more directly with systems-minded agricultural technology and service providers (e.g., Indigo) can provide food companies with better access to and more influence in on-farm practices that consumers increasingly care about. These partnerships can also improve data measurement and capture sustainability successes throughout the supply chain, helping food companies to demonstrate accountability against their goals. Finally, more direct linkages to growers enables improved supply chain traceability and transparency, which is a factor increasingly influencing consumer food purchasing choices.

Conclusions

Knowledge about plant microbiomes and mechanisms in wheat growth and production is expanding; however, little is known about how these microorganism communities affect the quality and functionality of foods. Endophytes are known to produce bioactive compounds that are of interest for flavor enhancement and health promotion. It is tempting to speculate that, perhaps, examples already exist where certain food sources are capturing added market value in part because of unique localized endophyte–plant environment combinations. As an example of how understanding these factors influences consumer decisions, the wine industry is built on traceability of production location, variety, vintage, and other factors that link consumers to brands. Use of analytical technologies will develop more of these linkages while also increasing production, improving farmer income, benefiting the environment, and enhancing human health. The global wheat industry is ready for these changes.

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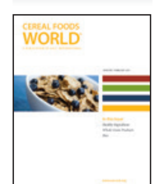

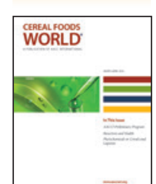

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