Using relatively inexpensive processes (10). The combination of new fermentation methods for reduced cost of production, costs of isolation, and periods of time without loss of viability. Additionally, this stability enables the endospore to go through the feed milling process and to reduce or eliminate the use of antibiotics in their operations. This has directly resulted in reduced overall performance and economic costs. Increasing pressure from consumers and government regulatory agencies has led an ever increasing number of U.S. poultry producers to reduce or eliminate the use of antibiotics in their operations. These data may suggest that this method of screening and evaluation could lead to commercially useful Bacillus-based probiotics.

Non-typable Salmonella infections of human result in an estimated 14 million cases of salmonellosis, resulting in approximately 163,000 hospitalizations and 2,600 deaths annually in the United States each year (1,2). Raw or undercooked poultry products, eggs, and shell casings account for the majority of cases in salmonellosis (3, 4). One way to reduce the incidence of Salmonella contaminates poultry products, and therefore the Salmonella salmonellosis, is to reduce the number of Salmonella in the feed. The number of Salmonella in the feed is reduced by reducing the number of Salmonella within the alimentary tract of free poultry.

Probiotics, frequently involving lactic acid bacteria, have been in use since the turn of the twentieth century and the benefits of these cultures in some pathogen reduction and increased gastrointestinal health have been well documented. The widespread use of these products in animal health applications has been limited, largely due to issues with stability, application, and cost. The development of a low cost, stable, efficacious probiotic that can be administered in animal feed is needed to increase the use of these products.

The genus Bacillus are gram positive, endospore forming, aerobic (or facultative anaerobic), and rod-shaped. Currently there are approximately 40 species of Bacillus; however the classical Bacillus genus has more recently been separated into 25 genera consisting of over 200 species (15). Bacillus species have long been used to produce aerobically fermented foods such as natto (a fermented soy bean product) and oseisma (a fermented loquat bean product) in Asia and Africa respectively. More recently Bacillus species have long been used to produce aerobically fermented foods such as natto (a fermented soy bean product) and oseisma (a fermented loquat bean product) in Asia and Africa respectively. Most recently Bacillus species have been used as probiotics and beneficial microorganisms. Some of these probiotics have been shown to kill enteropathogenic bacteria such as Salmonella, Escherichia coli, and Clostridium perfringens in vitro as well as decrease the number of these organisms in vivo. Additionally, some Bacillus probiotics have been shown to increase growth rate and improve feed efficiency in poultry and rabbit (16, 17).

The main advantage Bacillus-based probiotics have over the more traditional lactic acid bacteria probiotics stems from the stability of the endospore formed by Bacillus and closely related genera. The endospore is a survival form of the organism and is resistant to desiccation, UV light, many chemical disinfectants, high oxygen pressures, salinity, acidity, and heat. In addition, Bacillus probiotics are thermostable, viable for decades in the environment, and some Bacillus species are expectedly viable in terminal endospores from mineral deposits formed millions of years in the past. One recent study indicated successful isolation of a Bacillus from a 250 million year old salt crystal (6). This stability enables these probiotic species to be stored at room temperature for long periods of time without loss of viability. Additionally, this stability enables the endospore to go through the feed milling process and to reduce or eliminate the use of antibiotics in their operations. These data may suggest that this method of screening and evaluation could lead to commercially useful Bacillus-based probiotics.

The Bacillus subtilis isolate PHL-NP122 reduced both the incidence and amount of Salmonella isolated from the cecal tonsils in 603 turkeys from a test flock, and both body weight and body weight gain were increased by this same isolate. Neither isolate, PHL-NP122 nor PHL-MM65, significantly altered the number of lactobacillus or bifidobacteria present in the ceca. Additionally, the ability of these isolates to reduce the incidence of Salmonella and to increase body weight was not altered by alternating the temperature of the environment. The Bacillus subtilis isolate PHL-NP122 showed a significant increase in growth rate and feed efficiency in turkeys. However, the Bacillus subtilis isolate PHL-MM65 did not show a significant increase in growth rate and feed efficiency in turkeys.

REFERENCES:
4. Streeter, J.M. et al., 2007. Number of Salmonella on chicken breast fillets at retail level and its implications on public health risk. Development of a bacterial strain which provides stronger immunity in the form of an antimicrobial saline. Bacillus subtilis isolated from dairy farms identified to be useful in the isolation of non-pathogenic strains of Bacillus subtilis. Isolates were grown on a modified version of the solid state fermentation medium developed by Zhao et al (10). Briefly, wheat bran and rice hulls were combined and the ammonium medium described above, without agarose, was added the mixture to achieve a moisture content of 40% moisture. Isolates were added to the medium, and grown, at 37°C for 24 h and then allowed to incubate for an additional 24 h at 30°C. The ceca were dialyzed at 60°C and then ground. Samples were heat treated by incubation at 50°C for 10 minutes to kill any remaining vegetative cells and species were enumerated by serial dilution and colony counting.

EVALUATION OF BACILLUS ISOLATES AS POTENTIAL PROBIOTICS

in vivo

results of large numbers of endospores with relatively inexpensive processes (10). The combination of new fermentation methods for reduced cost of production, costs of isolation, and periods of time without loss of viability. Additionally, this stability enables the endospore to go through the feed milling process and to reduce or eliminate the use of antibiotics in their operations. These data may suggest that this method of screening and evaluation could lead to commercially useful Bacillus-based probiotics.

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