



Research article

## Microbial treatments enhance nutritional quality and *in-vitro* dry matter digestibility of maize (*Zea mays* L.) stover

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### Abstract

Low nutritional value, poor voluntary intake and digestibility limit the utilization of maize stover in livestock feeding. Different microbial treatments (*Trichoderma harzianum*, *Trichoderma viride*, *Saccharomyces cerevisiae* and *Pseudomonas fluorescens*) were evaluated for improving the quality and digestibility of 24 genotypes of maize stover. Treatment of maize stover with *P. fluorescens* and *S. cerevisiae* resulted in increase of CP content (11.82% and 10.56%) compared to control (8.54%). Out of four microbial strains *T. viride* has a positive effect on enhancing the total carbohydrate content, where maize stover (Vivek) exhibited significantly highest increase in total carbohydrate content (21.81%). Treatment of maize stover with *T. harzianum* reduced the NDF content to 65% in African Tall while treatment with *P. fluorescens* reduced it to 65.8% in Palam Shankar Makka in comparison to control (69 and 67.2%, respectively). Reduction in lignin content of maize stover (3.00% in PMG-52-1) was observed after treatment with *T. viride* in comparison to control (7.20%). Treatment of maize stover with *T. viride* and *P. fluorescens* was most effective in reducing the total phenolic content of PMG-97 and PMG-95 and African Tall genotypes of maize stovers, respectively. Treatment with *T. harzianum* and *P. fluorescens* reduced total tannins contents most effectively in Palam Shankar Makka and PMG-93-1, respectively. *P. fluorescens* followed by *S. cerevisiae* treatment resulted in a decrease in silica content of maize stover to the extent of 0.87% and 0.6% in maize stover of PMG-95 genotype, compared to control (1.8%). Digestibility of maize stover improved with all biological treatments, the highest significant increase (69.2%) was observed with *T. harzianum* treatment compared to control (52.1%). The results of the study revealed that *T. viride* is the most promising fungi for improving the quality and digestibility of maize stover of different genotypes, followed by *T. harzianum*. The bacteria *P. fluorescens* was found to be equally efficient in improving the quality and digestibility of maize stover. The genotype x treatment interaction was found to be significant for all the traits indicating that the microbes could efficiently enhance different quality traits in maize stover.

**Keywords:** Biological treatment, Crude protein, Digestibility, Lignin, Maize stover, Silica.

### Introduction

The increasing expansion of the agro-industry has resulted large accumulation of crop residues in the world. Alone in India, about 500 MT of crop residues are produced every year (Bhuvaneshwari *et al.*, 2019). It is a concerning fact that the drastic increase in the human population has intensified the rate of depletion of natural feed resources. Moreover, the increasing livestock population and their growing feeding requirements have further complicated the situation (Katoch *et al.*, 2018a). Different crop residues not only provide a relatively cheap feed source to livestock but also ensure feed security during the unavailability of green

fodder. Though crop residues hold vast energy potential, they are not utilized to their full potential in livestock feeding due to low voluntary intake, poor digestibility, low protein and mineral contents, which classify them as a non-maintenance type of feeds. Therefore, it is of utmost importance to develop different technologies for improving the quality and digestibility of crop residues to meet the feeding as well as nutritional requirements of livestock.

India stands fourth in global maize (*Zea mays* L.) production and has vast potential for utilization of remaining crop residue (stover) in livestock feeding. In the past, maize was mainly grown as a food crop in

India, but now has been recognized as an ideal food-feed crop (Katoch *et al.*, 2009; Katoch and Kumar, 2014). Every year large amount of maize stover is produced but its limited amount is utilized for livestock feeding. A major portion of maize stover is either removed from the field, burned on site, piled, spread over fields or used for mulching. Despite some limitations in livestock feeding, it still has comparatively high crude protein content and dry matter digestibility than other crop residues. Maize stover contains 3.50 to 8.70%, crude protein, 31.30 to 41.00% cellulose, 20.00 to 34.40% hemicellulose, 3.10 to 5.0% ADL and 4.20 to 7.50 %, ash content (Apoorva *et al.*, 2019). The utilization of maize stover in livestock feeding on a large scale is limited by its low voluntary intake and poor digestibility because of high lignin, and silica, and relatively low protein and energy content (Katoch *et al.*, 2017).

Several physical and chemical treatments have been investigated for improving quality and digestibility of crop residues. These treatments primarily target lignocellulosic complex in the cell wall and release bound cellulosic polysaccharides for enzymatic hydrolysis into simpler carbohydrate moieties in the gastrointestinal tract of animals. Though these treatments have been reported to improve the quality and degradability of crop residues but some disadvantages, such as high operational cost, are associated with their practical use (Chen *et al.*, 1995). Biological treatments offer several advantages over physical and chemical processes which include mild reaction conditions, low capital cost, avoidance of toxic chemicals and high product yield (Katoch *et al.*, 2017; Ragunathan *et al.*, 1996). The potential of biological treatments for improving digestibility of crop residues has been explained by ability of microbes to release oxidative ligninolytic enzymes such as lignin peroxidase, manganese peroxidase and laccase, hydrolytic enzymes such as cellulase and xylanases and tannase (Tannin acyl hydrolase) that target lingo-cellulose and tannin-protein complex in crop residues (Ramirez-Bribiesca *et al.*, 2010; Mahesh and Mohini, 2013).

The easy and cost-effective treatment of maize stover could improve its feeding quality and provide an option of feed resource during the periods of fodder scarcity. Keeping in view the availability of maize stover in the Indian farming system, the present study was undertaken to investigate different biological treatments for improving the quality and digestibility of maize stover for providing better feed resources during the lean season.

## Materials and Methods

**Study site and materials:** Different maize genotypes were sown in the experimental farm of Chaudhary Sarwan Kumar Himachal Pradesh Krishi Vishvavidyalaya

(CSKHPKV), Palampur, Himachal Pradesh. The samples of stover of 24 maize genotypes (Palam Shankar Makka; PMG-36; PMG-9; African Tall; PMG-93-1; PMG-52; SIRI-5455; PMG-49; PMG-41; PMG-95; PMG-93-2; PMG-33; PMG-97; PMG-3; PMG-40; PMG-93; PMG-96; PMG-18; Vivek; PMG-62; PMG-14; PMG-52-1; PMZ-4; Local Chadhiar) was oven dried to the constant weight at  $60 \pm 2^\circ\text{C}$ . The samples were ground in a Willey mill (1mm sieve) and preserved in air tight polythene bags for further treatment.

**Samples treatment:** The stover from different maize genotypes was inoculated with four different microorganisms, three fungi, *Trichoderma harzianum*, *Trichoderma viride*, *Saccharomyces cerevisiae* and a bacteria, *Pseudomonas fluorescens* associated with dissociation of lignocellulosic complexes. The mother culture of these microorganisms was obtained from the Department of Plant Pathology, CSKHPKV, Palampur and subsequently sub-cultured. *T. harzianum*, *T. viride* and *S. cerevisiae* were sub-cultured on potato dextrose broth, while *P. fluorescens* was sub-cultured using King's B broth. Prior to treatment with microbes the maize stovers were autoclaved and cooled down to remove any microbial load from unknown sources. The concentrations of inoculum were 0.6 OD for all microbes, moisture of stover was maintained around 60%, temperature was maintained at  $28 \pm 2^\circ\text{C}$  for fungus while it was  $37 \pm 2^\circ\text{C}$  for bacterial cultures, the pH was near neutral 6.8 to 7.2 and the particle size of stover were 1-2 cm. The pasteurized samples were inoculated and incubated for 7 days at  $28 \pm 2^\circ\text{C}$  temp and approximately 70-75% relative humidity. After 7 days, inoculated samples were oven dried, stored in a polythene bags for analysis.

**Nutritional analysis:** Crude protein (CP) and ash contents were estimated by the Micro-Kjeldahl method (AOAC, 2000). Fibre content [Neutral detergent fibre (NDF), Acid detergent fibre (ADF)] and cell wall constituents [Acid detergent lignin (ADL), Hemicellulose (HC) and Cellulose (CL)] were determined according to the method of Goering and Van Soest (1975). The *in vitro* dry matter digestibility (IVDMD) was estimated by two stage digestion method of Tilley and Terry (1963). The rumen liquor was collected from cattle. Total phenols, simple phenols and total tannins were estimated by following the method of Makkar (2003). Total carbohydrate content in stover samples was estimated according to the method of Hedge and Hofreiter (1962). Silica content was determined by following the method of Van Soest and Wine (1968). The significant differences in results were detected by analysis of variance (One-Way ANOVA) (Panse and Sukhatme, 1985). A probability of  $\leq 0.05$  was selected as a criterion for a statistically significant difference.

**Data analysis:** The data obtained were analyzed by statistical software. The PAST software was used for cluster analysis and MEV 4.0 was used for generating the heatmap for clear visualization of data sets.

## Results and Discussion

**Crude protein (CP) and total carbohydrate contents:** Crude protein content in untreated stover from different maize genotypes ranged from 7.00% (PMG-96) to 9.80% (PMG-93-2). Treatment of stover with *P. fluorescens* revealed highest increase (15.75%) in CP content of stover from genotype PMG-14 in comparison to control (7.87%). There was 13.13%, 12.08% and 12.25% increase

in CP content of stover from genotypes PMG-49 (control 9.8%), PMG-96 (control 7%) and PMG-49 (control 7.35) which were incubated with *T. harzianum*, *T. viride* and *S. cerevisiae*, respectively (Table-1). Islamiyati *et al.* (2013) also reported 27.58% increase in CP content of maize stover treated with *Trichoderma sp.* RI-7. Treatment with oyster mushroom (*Pleurotus ostreatus*) has also been reported effective in increasing the CP content of stover to the extent of 171.89% (Ramirez-Bribiesca *et al.*, 2010; Adenipekun and Okunlade, 2012). Chen *et al.* (2025) showed that microorganisms and enzymes pre-treatment play synergistic role in improving nutritive values of maize stover where increase in CP content was significantly high (8.48%) as compared to control (7.73%).

**Table 1.** Crude protein and total carbohydrate contents (%) of raw and treated maize stover from different genotypes

| Genotypes           | UT            | Th                 | Tv                 | Sv                 | Ps                 | SE  | CD   | UT                   | Th                 | Tv                 | Sv                 | Ps                 | SE  | CD   |
|---------------------|---------------|--------------------|--------------------|--------------------|--------------------|-----|------|----------------------|--------------------|--------------------|--------------------|--------------------|-----|------|
|                     | Crude protein |                    |                    |                    |                    |     |      | Carbohydrate content |                    |                    |                    |                    |     |      |
| PMG-3               | 7.7           | 9.28*              | 10.85 <sup>a</sup> | 9.10*              | 13.13 <sup>a</sup> | 0.7 | 1.63 | 14.1                 | 5.37 <sup>b</sup>  | 14.97*             | 10.20 <sup>b</sup> | 8.09 <sup>b</sup>  | 1   | 2.11 |
| SIRI-5455           | 8.2           | 8.25*              | 9.28 <sup>a</sup>  | 11.03 <sup>a</sup> | 10.33 <sup>a</sup> | 0.2 | 0.52 | 9.09                 | 7.82*              | 16.69 <sup>a</sup> | 9.26*              | 8.30*              | 0.8 | 1.73 |
| PMG-9               | 8.1           | 11.38 <sup>a</sup> | 11.88 <sup>a</sup> | 9.10 <sup>a</sup>  | 12.08 <sup>a</sup> | 0.3 | 0.68 | 9.64                 | 6.92 <sup>b</sup>  | 15.44 <sup>a</sup> | 15.19 <sup>a</sup> | 8.88*              | 0.8 | 1.69 |
| PMG-14              | 7.9           | 12.08 <sup>a</sup> | 12.05 <sup>a</sup> | 10.15 <sup>a</sup> | 15.75 <sup>a</sup> | 0.5 | 1.03 | 14.6                 | 6.50 <sup>b</sup>  | 18.31 <sup>a</sup> | 11.56 <sup>b</sup> | 10.94 <sup>b</sup> | 0.4 | 0.94 |
| PMG-18              | 8.4           | 9.98 <sup>a</sup>  | 13.83 <sup>a</sup> | 9.63 <sup>a</sup>  | 8.58*              | 0.3 | 0.7  | 11                   | 6.28 <sup>b</sup>  | 16.72 <sup>a</sup> | 10.77*             | 8.73 <sup>b</sup>  | 0.6 | 1.42 |
| Vivek               | 9.1           | 8.75*              | 10.15*             | 11.73 <sup>a</sup> | 11.90 <sup>a</sup> | 0.7 | 1.62 | 12.7                 | 14.28 <sup>a</sup> | 21.81 <sup>a</sup> | 14.59 <sup>a</sup> | 9.34 <sup>b</sup>  | 0.4 | 0.94 |
| PMG-33              | 9.1           | 10.83 <sup>a</sup> | 9.10*              | 8.23*              | 10.33 <sup>a</sup> | 0.6 | 1.22 | 12.5                 | 9.82 <sup>b</sup>  | 18.29 <sup>a</sup> | 15.23 <sup>a</sup> | 10.34*             | 1.2 | 2.58 |
| PMG-36              | 8.9           | 9.28*              | 9.80*              | 11.38 <sup>a</sup> | 11.55 <sup>a</sup> | 0.7 | 1.47 | 15.5                 | 9.71 <sup>b</sup>  | 15.38*             | 12.62 <sup>b</sup> | 14.25*             | 1   | 2.19 |
| PMG-40              | 7.7           | 8.25*              | 8.93*              | 11.03 <sup>a</sup> | 10.85 <sup>a</sup> | 0.9 | 2.09 | 11.3                 | 8.01 <sup>b</sup>  | 15.70 <sup>a</sup> | 12.77*             | 10.26*             | 0.8 | 1.83 |
| PMG-41              | 9.1           | 10.85 <sup>a</sup> | 11.20 <sup>a</sup> | 9.63*              | 11.38 <sup>a</sup> | 0.5 | 1.12 | 10.8                 | 12.55*             | 16.97 <sup>a</sup> | 11.37*             | 10.55*             | 1   | 2.27 |
| PMG-49              | 7.4           | 13.13 <sup>a</sup> | 11.20 <sup>a</sup> | 12.25 <sup>a</sup> | 11.55 <sup>a</sup> | 0.5 | 1    | 13.9                 | 7.59 <sup>b</sup>  | 14.44*             | 15.98 <sup>a</sup> | 8.69 <sup>b</sup>  | 0.3 | 0.62 |
| PMZ-4               | 9.3           | 11.90 <sup>a</sup> | 13.13 <sup>a</sup> | 11.20 <sup>a</sup> | 11.20 <sup>a</sup> | 0.7 | 1.47 | 11.9                 | 10.47 <sup>b</sup> | 14.09 <sup>a</sup> | 8.16 <sup>b</sup>  | 9.92 <sup>b</sup>  | 0.4 | 0.9  |
| PMG-52              | 7.7           | 11.18 <sup>a</sup> | 9.28 <sup>a</sup>  | 8.75 <sup>a</sup>  | 11.20 <sup>a</sup> | 0.3 | 0.64 | 13.5                 | 8.96 <sup>b</sup>  | 14.36*             | 13.04*             | 12.71*             | 0.8 | 1.85 |
| PMG-52-1            | 8.9           | 10.85 <sup>a</sup> | 8.40*              | 9.28*              | 13.65 <sup>a</sup> | 0.7 | 1.57 | 12.9                 | 10.70 <sup>b</sup> | 17.10 <sup>a</sup> | 13.15*             | 11.80 <sup>b</sup> | 0.4 | 0.99 |
| PMG-62              | 8.6           | 8.75*              | 9.28 <sup>a</sup>  | 11.73 <sup>a</sup> | 13.48 <sup>a</sup> | 0.2 | 0.4  | 14.1                 | 7.82 <sup>b</sup>  | 16.84*             | 9.71 <sup>b</sup>  | 11.57*             | 1.3 | 2.91 |
| PMG-93              | 9.1           | 12.78 <sup>a</sup> | 10.83 <sup>a</sup> | 8.93*              | 13.83 <sup>a</sup> | 0.3 | 0.6  | 12.9                 | 6.27 <sup>b</sup>  | 15.61 <sup>a</sup> | 11.11 <sup>b</sup> | 11.94 <sup>b</sup> | 0.4 | 0.88 |
| PMG-93-1            | 8.6           | 11.18 <sup>a</sup> | 9.28 <sup>a</sup>  | 13.30 <sup>a</sup> | 9.98 <sup>a</sup>  | 0.3 | 0.68 | 13.3                 | 8.47 <sup>b</sup>  | 17.42 <sup>a</sup> | 11.94 <sup>b</sup> | 8.05 <sup>b</sup>  | 0.4 | 0.79 |
| PMG-93-2            | 9.8           | 13.13 <sup>a</sup> | 11.55*             | 11.20*             | 15.05 <sup>a</sup> | 1   | 2.36 | 13                   | 8.14 <sup>b</sup>  | 13.95*             | 9.87 <sup>b</sup>  | 13.13*             | 0.9 | 1.96 |
| PMG-95              | 7.7           | 7.53*              | 8.93 <sup>a</sup>  | 10.50 <sup>a</sup> | 9.63 <sup>a</sup>  | 0.3 | 0.68 | 11                   | 11.41*             | 17.44 <sup>a</sup> | 11.79*             | 10.60*             | 0.5 | 1.11 |
| PMG-96              | 7             | 8.75 <sup>a</sup>  | 12.08 <sup>a</sup> | 11.38 <sup>a</sup> | 12.25 <sup>a</sup> | 0.3 | 0.67 | 18                   | 6.43 <sup>b</sup>  | 19.58 <sup>a</sup> | 13.79 <sup>b</sup> | 9.38 <sup>b</sup>  | 0.5 | 1.16 |
| Palam Shankar Makka | 9.5           | 9.28*              | 8.75 <sup>b</sup>  | 12.40 <sup>a</sup> | 11.55 <sup>a</sup> | 0.2 | 0.36 | 14.4                 | 9.37 <sup>b</sup>  | 17.55 <sup>a</sup> | 14.77*             | 13.58 <sup>b</sup> | 0.3 | 0.56 |
| PMG-97              | 8.9           | 8.75*              | 9.80 <sup>a</sup>  | 9.63 <sup>a</sup>  | 12.08 <sup>a</sup> | 0.4 | 0.79 | 13                   | 13.64*             | 15.49 <sup>a</sup> | 8.43 <sup>b</sup>  | 8.30 <sup>b</sup>  | 0.7 | 1.47 |
| Local Chadhiar      | 9.1           | 11.90 <sup>a</sup> | 10.33*             | 9.98*              | 11.03 <sup>a</sup> | 0.6 | 1.3  | 13.9                 | 7.56 <sup>b</sup>  | 12.57 <sup>b</sup> | 8.73 <sup>b</sup>  | 9.16 <sup>b</sup>  | 0.3 | 0.69 |
| African Tall        | 8.6           | 12.25 <sup>a</sup> | 9.98 <sup>a</sup>  | 10.50 <sup>a</sup> | 12.78 <sup>a</sup> | 0.4 | 0.93 | 13                   | 8.69 <sup>b</sup>  | 16.65 <sup>a</sup> | 8.58 <sup>b</sup>  | 7.92 <sup>b</sup>  | 0.4 | 0.89 |

In table(s) 1-7, superscripts 'a', 'b' and '\*\*' represent significant increase, significant decrease and statistically at par values as compared to untreated, respectively at  $P \leq 0.05$ ;

NS: Non-significant; CD: Critical difference; SE: Standard error;

UT: Untreated; Th: *T. harzianum*; Tv: *T. viride*; Sv: *S. cerevisiae*; Pf: *P. fluorescens*

**Table 2.** Neutral Detergent Fiber (NDF) and Acid Detergent Fiber (ADF) contents (%) of raw and treated maize stover from different genotypes

| Genotypes           | UT  | Th                 | Tv                 | Sv                 | Ps                 | SE  | CD   | UT   | Th                 | Tv                 | Sv                 | Ps                 | SE  | CD   |
|---------------------|-----|--------------------|--------------------|--------------------|--------------------|-----|------|------|--------------------|--------------------|--------------------|--------------------|-----|------|
|                     | NDF |                    |                    |                    |                    |     |      | ADF  |                    |                    |                    |                    |     |      |
| PMG-3               | 72  | 67.5               | 68                 | 69                 | 69.5               | 2.2 | NS   | 40.5 | 39.80 <sup>b</sup> | 39.80 <sup>b</sup> | 39.73 <sup>b</sup> | 38.00 <sup>b</sup> | 0.3 | 0.55 |
| SIRI-5455           | 70  | 65.70 <sup>b</sup> | 67.10 <sup>b</sup> | 67.60 <sup>b</sup> | 68.70 <sup>b</sup> | 0.5 | 1.16 | 46.4 | 45.40 <sup>b</sup> | 44.80 <sup>b</sup> | 45.20 <sup>b</sup> | 43.20 <sup>b</sup> | 0.3 | 0.59 |
| PMG-9               | 68  | 65.60 <sup>b</sup> | 65.40 <sup>b</sup> | 65.80 <sup>b</sup> | 66.90 <sup>b</sup> | 0.3 | 0.7  | 48.6 | 47.13 <sup>b</sup> | 47.67 <sup>b</sup> | 46.20 <sup>b</sup> | 44.20 <sup>b</sup> | 0.3 | 0.64 |
| PMG-14              | 72  | 69.20 <sup>b</sup> | 69.60 <sup>b</sup> | 69.90 <sup>b</sup> | 70.40 <sup>b</sup> | 0.4 | 0.79 | 43.4 | 40.07 <sup>b</sup> | 42.73 <sup>*</sup> | 40.00 <sup>b</sup> | 35.40 <sup>b</sup> | 1.2 | 2.62 |
| PMG-18              | 72  | 67.8               | 67.6               | 68.3               | 70.2               | 1.8 | NS   | 46.7 | 40.80 <sup>b</sup> | 38.80 <sup>b</sup> | 39.27 <sup>b</sup> | 33.93 <sup>b</sup> | 0.9 | 1.91 |
| Vivek               | 72  | 68.90 <sup>b</sup> | 68.30 <sup>b</sup> | 69.70 <sup>b</sup> | 70.30 <sup>b</sup> | 0.8 | 1.74 | 46.9 | 46.13 <sup>*</sup> | 44.00 <sup>b</sup> | 42.13 <sup>b</sup> | 35.20 <sup>b</sup> | 0.4 | 0.88 |
| PMG-33              | 72  | 67.9               | 67.6               | 68.7               | 69.3               | 1.5 | NS   | 48.2 | 40.20 <sup>b</sup> | 38.93 <sup>b</sup> | 42.40 <sup>b</sup> | 36.40 <sup>b</sup> | 0.2 | 0.38 |
| PMG-36              | 69  | 66.30 <sup>b</sup> | 65.80 <sup>b</sup> | 66.30 <sup>b</sup> | 66.70 <sup>b</sup> | 0.5 | 1.12 | 44.4 | 43.33 <sup>*</sup> | 41.67 <sup>b</sup> | 40.07 <sup>b</sup> | 34.47 <sup>b</sup> | 0.6 | 1.3  |
| PMG-40              | 72  | 68.1               | 68.8               | 69                 | 69.6               | 2.3 | NS   | 44.9 | 44.27 <sup>*</sup> | 42.53 <sup>b</sup> | 44.20 <sup>*</sup> | 41.67 <sup>b</sup> | 1   | 2.11 |
| PMG-41              | 72  | 68.9               | 68                 | 69.4               | 68.7               | 2.7 | NS   | 46   | 45.33 <sup>*</sup> | 44.20 <sup>*</sup> | 44.47 <sup>*</sup> | 41.53 <sup>b</sup> | 1   | 2.33 |
| PMG-49              | 71  | 67.9               | 67.9               | 68.7               | 68.7               | 1.5 | NS   | 47.5 | 45.87 <sup>b</sup> | 40.07 <sup>b</sup> | 45.80 <sup>b</sup> | 42.27 <sup>b</sup> | 0.4 | 0.94 |
| PMZ-4               | 75  | 70.30 <sup>b</sup> | 70.90 <sup>b</sup> | 71.90 <sup>b</sup> | 72.40 <sup>b</sup> | 0.3 | 0.72 | 44.2 | 41.80 <sup>b</sup> | 42.93 <sup>b</sup> | 41.47 <sup>b</sup> | 38.93 <sup>b</sup> | 0.5 | 1.03 |
| PMG-52              | 71  | 66.9               | 67.7               | 69                 | 68.4               | 3.3 | NS   | 48   | 46.33 <sup>b</sup> | 43.53 <sup>b</sup> | 45.60 <sup>b</sup> | 44.73 <sup>b</sup> | 0.3 | 0.65 |
| PMG-52-1            | 73  | 68.2               | 68.6               | 70.2               | 70.6               | 2   | NS   | 44.4 | 42.8               | 41.87              | 41.6               | 40.6               | 3.6 | NS   |
| PMG-62              | 73  | 68.80 <sup>b</sup> | 68.90 <sup>b</sup> | 69.80 <sup>b</sup> | 70.40 <sup>b</sup> | 0.4 | 0.95 | 49   | 41.70 <sup>b</sup> | 44.80 <sup>b</sup> | 46.60 <sup>b</sup> | 43.47 <sup>b</sup> | 0.4 | 0.87 |
| PMG-93              | 73  | 67.6               | 68.9               | 69.8               | 69.7               | 2.5 | NS   | 46.4 | 42.93 <sup>b</sup> | 45.60 <sup>*</sup> | 44.73 <sup>*</sup> | 40.73 <sup>b</sup> | 1.1 | 2.54 |
| PMG-93-1            | 71  | 66.8               | 68.2               | 68.4               | 68.2               | 1.8 | NS   | 47.6 | 43.07 <sup>b</sup> | 45.40 <sup>b</sup> | 47.00 <sup>*</sup> | 41.60 <sup>b</sup> | 0.6 | 1.31 |
| PMG-93-2            | 71  | 66.60 <sup>b</sup> | 67.90 <sup>b</sup> | 68.70 <sup>b</sup> | 69.20 <sup>b</sup> | 0.7 | 1.52 | 45.6 | 44.87 <sup>*</sup> | 43.80 <sup>*</sup> | 44.60 <sup>*</sup> | 41.50 <sup>b</sup> | 1.2 | 2.61 |
| PMG-95              | 72  | 66.1               | 68                 | 69.5               | 68.9               | 2.1 | NS   | 46.2 | 45.4               | 45.27              | 44.87              | 44.73              | 0.9 | NS   |
| PMG-96              | 72  | 66.8               | 68.3               | 69.7               | 69.8               | 3.2 | NS   | 43   | 43.33 <sup>*</sup> | 41.40 <sup>*</sup> | 43.67 <sup>*</sup> | 39.87 <sup>b</sup> | 0.9 | 2.05 |
| Palam Shankar Makka | 67  | 66.3               | 64.8               | 65.3               | 65.8               | 1   | NS   | 47.5 | 45.20 <sup>b</sup> | 44.47 <sup>b</sup> | 45.33 <sup>b</sup> | 44.10 <sup>b</sup> | 0.4 | 0.89 |
| PMG-97              | 72  | 68.4               | 68.9               | 69.2               | 69.4               | 1.4 | NS   | 45.6 | 45.67 <sup>*</sup> | 45.13 <sup>*</sup> | 41.13 <sup>b</sup> | 40.47 <sup>b</sup> | 0.7 | 1.45 |
| Local Chadhiar      | 75  | 69                 | 70.7               | 72.2               | 72.7               | 2.7 | NS   | 47.8 | 46.93 <sup>*</sup> | 42.33 <sup>b</sup> | 41.47 <sup>b</sup> | 39.73 <sup>b</sup> | 0.7 | 1.44 |
| African Tall        | 69  | 65                 | 67                 | 67.1               | 67                 | 1.1 | NS   | 46.2 | 45.97              | 44.03              | 46                 | 43.7               | 2.2 | NS   |

The increase in CP content after the biological treatment may be due to the addition of protein rich fungal biomass and degradation of tannin-protein complex by tannase enzyme (Raghuwanshi *et al.*, 2014). Shrivastava *et al.* (2011) also recorded 5.08% and 4.77% CP content of wheat straw inoculated with *Pleurotus ostreatus* and *Trametes versicolor* as compared to control straw (3.37%) for 30 days, respectively. The biological treatments revealed variable effects on total carbohydrate content of stover. Variation in total carbohydrate content of untreated stover ranged from 9.09% to 18.02% from different maize genotypes. Treatment with *T. viride*, resulted increase in carbohydrate content of stover from genotype Vivek to 21.81% while *T. harzianum* showed upaccumulation of carbohydrate to 14.28% in comparison to control 12.73% in the same genotype, implying *T. viride* is better in increasing carbohydrate. The bacterial inoculation of

stover with *P. fluorescens* was ineffective in improving carbohydrate content of maize stover (Table-1). The increase in carbohydrate content of stover may be due to the addition of cellular contents of microorganisms and degradation of cell wall phenolics associated with carbohydrate moieties (Li *et al.*, 2021; Katoch 2022).

**Fiber and lignin content:** Fibre content (NDF and ADF) in forages has an inverse relation to voluntary feed intake and digestible energy intake to livestock (Katoch *et al.*, 2018b). In this study, variation in NDF content of stover from different 24 maize genotypes were recorded from 67.20% (Palam Shankar Makka) to 74.80% (PMZ-4). The highest decrease in NDF content (was observed after treatment with *T. harzianum* treatment in stover from genotype PMG-95 (66.10%) followed by from *T. viride* (68.00%) in comparison to the control (71.60%), (Table-2).

**Table 3.** Acid detergent lignin content (%) of raw and treated maize stover from different genotypes

| Genotypes           | Untreated | <i>T. harzianum</i> | <i>T. viride</i>  | <i>S. cerevisiae</i> | <i>P. fluorescens</i> | SE   | CD   |
|---------------------|-----------|---------------------|-------------------|----------------------|-----------------------|------|------|
| PMG-3               | 5.30      | 4.10 <sup>b</sup>   | 3.20 <sup>b</sup> | 3.47 <sup>b</sup>    | 4.40 <sup>b</sup>     | 0.19 | 0.43 |
| SIRI-5455           | 5.10      | 4.70 <sup>b</sup>   | 3.60 <sup>b</sup> | 3.93 <sup>b</sup>    | 4.30 <sup>b</sup>     | 0.18 | 0.40 |
| PMG-9               | 4.80      | 4.33 <sup>*</sup>   | 3.80 <sup>b</sup> | 4.47 <sup>*</sup>    | 4.00 <sup>b</sup>     | 0.23 | 0.51 |
| PMG-14              | 6.30      | 5.50 <sup>b</sup>   | 5.57 <sup>b</sup> | 4.47 <sup>b</sup>    | 5.00 <sup>b</sup>     | 0.26 | 0.59 |
| PMG-18              | 5.20      | 2.70 <sup>b</sup>   | 4.47 <sup>b</sup> | 3.53 <sup>b</sup>    | 4.40 <sup>b</sup>     | 0.30 | 0.67 |
| Vivek               | 5.40      | 5.13                | 5.00              | 4.73                 | 3.80                  | 0.95 | NS   |
| PMG-33              | 4.80      | 4.40 <sup>b</sup>   | 3.70 <sup>b</sup> | 3.30 <sup>b</sup>    | 3.60 <sup>b</sup>     | 0.15 | 0.35 |
| PMG-36              | 5.70      | 4.13                | 4.00              | 3.53                 | 4.90                  | 0.73 | NS   |
| PMG-40              | 6.50      | 5.47 <sup>b</sup>   | 4.20 <sup>b</sup> | 4.53 <sup>b</sup>    | 5.10 <sup>b</sup>     | 0.17 | 0.37 |
| PMG-41              | 3.70      | 3.20                | 3.00              | 2.93                 | 3.00                  | 0.42 | NS   |
| PMG-49              | 5.50      | 4.20 <sup>b</sup>   | 5.07 <sup>b</sup> | 4.70 <sup>b</sup>    | 4.30 <sup>b</sup>     | 0.15 | 0.33 |
| PMZ-4               | 5.90      | 5.53 <sup>b</sup>   | 4.53 <sup>b</sup> | 3.73 <sup>b</sup>    | 4.10 <sup>b</sup>     | 0.14 | 0.30 |
| PMG-52              | 5.70      | 4.27 <sup>b</sup>   | 4.67 <sup>b</sup> | 4.20 <sup>b</sup>    | 4.90 <sup>b</sup>     | 0.13 | 0.29 |
| PMG-52-1            | 7.20      | 4.87 <sup>b</sup>   | 3.00 <sup>b</sup> | 4.60 <sup>b</sup>    | 5.70 <sup>b</sup>     | 0.52 | 1.17 |
| PMG-62              | 6.10      | 5.60 <sup>*</sup>   | 3.40 <sup>b</sup> | 3.27 <sup>b</sup>    | 5.00 <sup>*</sup>     | 0.58 | 1.29 |
| PMG-93              | 4.40      | 3.60 <sup>b</sup>   | 3.13 <sup>b</sup> | 2.00 <sup>b</sup>    | 3.40 <sup>b</sup>     | 0.30 | 0.68 |
| PMG-93-1            | 4.70      | 3.30 <sup>b</sup>   | 4.00 <sup>*</sup> | 2.47 <sup>b</sup>    | 3.90 <sup>*</sup>     | 0.47 | 1.04 |
| PMG-93-2            | 5.50      | 3.60 <sup>b</sup>   | 3.40 <sup>b</sup> | 2.60 <sup>b</sup>    | 4.40 <sup>b</sup>     | 0.17 | 0.37 |
| PMG-95              | 6.30      | 5.27 <sup>*</sup>   | 4.80 <sup>b</sup> | 2.80 <sup>b</sup>    | 5.10 <sup>b</sup>     | 0.54 | 1.19 |
| PMG-96              | 5.60      | 4.73 <sup>*</sup>   | 4.00 <sup>b</sup> | 2.47 <sup>b</sup>    | 4.30 <sup>*</sup>     | 0.61 | 1.35 |
| Palam Shankar Makka | 4.80      | 2.87 <sup>b</sup>   | 2.47 <sup>b</sup> | 3.00 <sup>b</sup>    | 3.60 <sup>b</sup>     | 0.24 | 0.53 |
| PMG-97              | 6.20      | 4.73 <sup>b</sup>   | 4.33 <sup>b</sup> | 3.47 <sup>b</sup>    | 5.00 <sup>b</sup>     | 0.28 | 0.63 |
| Local Chadhiar      | 4.40      | 3.93 <sup>*</sup>   | 3.67 <sup>b</sup> | 3.00 <sup>b</sup>    | 2.70 <sup>b</sup>     | 0.26 | 0.59 |
| African Tall        | 4.70      | 3.60 <sup>b</sup>   | 2.00 <sup>b</sup> | 4.20 <sup>b</sup>    | 2.90 <sup>b</sup>     | 0.15 | 0.35 |

Similar to above results, the inoculation of moist rice straw with microbes like *Azospirillum brasilense* and *Saccharomyces cerevisiae* significantly reduced the NDF (55.39%) and ADF (42.46%), (Zayed, 2018).

The variations in ADF content of maize stover ranged from 40.50% (PMG-3) to 49.00% (PMG-62). The highest reduction in ADF content as compared with control was observed in stover from genotype PMG-18 (33.93%) with *P. fluorescens* treatment. It was followed by 38.80% in PMG-18 with *T. viride*, 40.20% in PMG-33 with *T. harzianum* and 39.27% in PMG-18 with *S. cerevisiae* (Table-4). El-Galil *et al.* (2011) also observed reduction in NDF content (from 59.79 to 48.65%) of bean straw treated with *Bacillus sp.* Abdel-Azim *et al.* (2011) also observed decrease in NDF and ADF content (from 66.40 to 58.39% and 46.27 to 39.43%, respectively) of corn stalks inoculated with *T. viride*.

A decrease of NDF content from 73.20 to 60.20% have also been reported in sugarcane bagasse inoculated with *T.*

*viride* (Mahrous *et al.*, 2011). Yalchi and Hajighrari (2010) also revealed significant reduction in NDF and ADF content in wheat straw inoculated with *T. harzianum*. It has also been reported that fungi belonging to *Trichoderma* genus liberate several enzymes which hydrolyze complex polysaccharides in the cell wall to their monosaccharide constituents (Safari Sinegani *et al.*, 2005), thus resulting in decreased fibre content in stover.

Lignification of cell wall is one of the major factors for low nutritive value and digestibility of maize stover (Katoch *et al.*, 2017). ADL content in untreated stover of different maize genotypes varied from 3.70 (PMG-41) to 7.20% (PMG-52-1). Treatment with *P. fluorescens* and *T. harzianum* revealed significant reduction in ADL content of stover from genotypes Local Chadhiar (2.70%) and PMG-18 (3.93%), respectively. Among all treatments, *T. viride* followed by *S. cerevisiae* treatment revealed highest reduction in ADL content of stover from genotypes PMG-52-1 (3.00%) and PMG-96 (4.00%), respectively (Table-3).

**Table 4.** Total phenolic and total tannin contents (mg/g) of raw and treated maize stover from different genotypes

| Genotypes                 | UT                     | Th                 | Tv                 | Sv                 | Ps                 | SE  | CD   | UT                   | Th                | Tv                | Sv                | Ps                | SE  | CD   |
|---------------------------|------------------------|--------------------|--------------------|--------------------|--------------------|-----|------|----------------------|-------------------|-------------------|-------------------|-------------------|-----|------|
|                           | Total phenolic content |                    |                    |                    |                    |     |      | Total tannin content |                   |                   |                   |                   |     |      |
| PMG-3                     | 9.3                    | 8.65 <sup>b</sup>  | 7.30 <sup>b</sup>  | 8.87 <sup>b</sup>  | 8.88 <sup>b</sup>  | 0.1 | 18   | 3.96                 | 3.61 <sup>*</sup> | 2.93 <sup>b</sup> | 3.88 <sup>*</sup> | 3.23 <sup>b</sup> | 0.3 | 0.7  |
| SIRI-5455                 | 9.1                    | 8.78 <sup>*</sup>  | 8.41 <sup>*</sup>  | 6.06 <sup>b</sup>  | 7.68 <sup>b</sup>  | 0.4 | 0.83 | 3.04                 | 2.79              | 2.69              | 2.1               | 2.06              | 0.5 | NS   |
| PMG-9                     | 10                     | 9.75 <sup>*</sup>  | 7.40 <sup>b</sup>  | 8.46 <sup>b</sup>  | 9.43 <sup>*</sup>  | 0.4 | 0.81 | 4.86                 | 4.14 <sup>b</sup> | 3.36 <sup>b</sup> | 3.33 <sup>b</sup> | 3.72 <sup>b</sup> | 0.3 | 0.63 |
| PMG-14                    | 12                     | 8.51 <sup>b</sup>  | 10.16 <sup>b</sup> | 10.57 <sup>b</sup> | 10.40 <sup>b</sup> | 0.3 | 0.68 | 6.45                 | 5.27 <sup>b</sup> | 4.50 <sup>b</sup> | 5.86 <sup>*</sup> | 5.53 <sup>*</sup> | 0.5 | 1.06 |
| PMG-18                    | 9.9                    | 8.51               | 8.98               | 8.64               | 7.23               | 0.8 | NS   | 4.78                 | 3.76              | 4.34              | 4.39              | 3.79              | 0.4 | NS   |
| Vivek                     | 10                     | 8.59 <sup>b</sup>  | 7.02 <sup>b</sup>  | 9.53 <sup>b</sup>  | 7.54 <sup>b</sup>  | 0.2 | 0.51 | 3.97                 | 3.27 <sup>b</sup> | 3.13 <sup>b</sup> | 2.98 <sup>b</sup> | 3.52 <sup>*</sup> | 0.3 | 0.65 |
| PMG-33                    | 9.8                    | 9.13               | 7.92               | 8.56               | 8.54               | 0.8 | NS   | 4.9                  | 4.4               | 3.35              | 4.49              | 4.08              | 0.5 | NS   |
| PMG-36                    | 10                     | 9.23 <sup>*</sup>  | 9.31 <sup>*</sup>  | 8.61 <sup>b</sup>  | 7.25 <sup>b</sup>  | 0.6 | 1.37 | 5.34                 | 4.58 <sup>*</sup> | 4.88 <sup>*</sup> | 4.02 <sup>b</sup> | 3.20 <sup>b</sup> | 0.4 | 0.84 |
| PMG-40                    | 11                     | 9.42               | 10.27              | 9.92               | 8.87               | 1.2 | NS   | 5.13                 | 4.89              | 4.23              | 4.93              | 3.93              | 0.7 | NS   |
| PMG-41                    | 11                     | 10.08              | 8.31               | 9.4                | 9.68               | 0.9 | NS   | 5.99                 | 5.61 <sup>*</sup> | 5.54 <sup>*</sup> | 4.42 <sup>b</sup> | 4.94 <sup>b</sup> | 0.4 | 0.99 |
| PMG-49                    | 11                     | 10.16              | 9.98               | 10.57              | 10.7               | 0.3 | NS   | 5.26                 | 4.82 <sup>*</sup> | 3.52 <sup>b</sup> | 4.76 <sup>*</sup> | 4.85 <sup>*</sup> | 0.5 | 1.09 |
| PMZ-4                     | 11                     | 11.28 <sup>*</sup> | 8.88 <sup>b</sup>  | 10.06 <sup>*</sup> | 9.22 <sup>b</sup>  | 0.7 | 1.61 | 6.15                 | 6.03 <sup>*</sup> | 5.37 <sup>b</sup> | 5.21 <sup>b</sup> | 4.99 <sup>b</sup> | 0.3 | 0.77 |
| PMG-52                    | 11                     | 9.23               | 10.3               | 8.72               | 10.53              | 1.6 | NS   | 6.67                 | 5.7               | 6.33              | 5.65              | 6                 | 1.7 | NS   |
| PMG-52-1                  | 12                     | 11.22 <sup>*</sup> | 8.21 <sup>b</sup>  | 10.91 <sup>*</sup> | 9.49 <sup>b</sup>  | 0.7 | 1.6  | 7.06                 | 6.74              | 6.1               | 6.87              | 5.25              | 0.9 | NS   |
| PMG-62                    | 12                     | 10.47              | 8.65               | 9.74               | 8.78               | 1.1 | NS   | 6.94                 | 6.09 <sup>*</sup> | 4.74 <sup>b</sup> | 5.60 <sup>b</sup> | 5.87 <sup>*</sup> | 0.5 | 1.2  |
| PMG-93                    | 12                     | 11.35 <sup>*</sup> | 9.58 <sup>b</sup>  | 9.89 <sup>b</sup>  | 9.09 <sup>b</sup>  | 0.8 | 1.84 | 4                    | 3.59              | 3.79              | 3.47              | 3.48              | 0.6 | NS   |
| PMG-93-1                  | 11                     | 10.48 <sup>*</sup> | 9.83 <sup>b</sup>  | 8.27 <sup>b</sup>  | 9.37 <sup>b</sup>  | 0.4 | 0.82 | 4.87                 | 3.84 <sup>b</sup> | 4.67 <sup>*</sup> | 4.22 <sup>*</sup> | 2.83 <sup>b</sup> | 0.3 | 0.67 |
| PMG-93-2                  | 12                     | 9.98               | 10.11              | 10.84              | 10.71              | 1.6 | NS   | 6.16                 | 5.08              | 5.81              | 5.38              | 5.41              | 0.8 | NS   |
| PMG-95                    | 10                     | 8.93               | 7.05               | 9.42               | 9.96               | 1.7 | NS   | 3.48                 | 3.17              | 3.11              | 2.81              | 2.91              | 0.4 | NS   |
| PMG-96                    | 10                     | 9.93 <sup>*</sup>  | 8.26 <sup>b</sup>  | 9.17 <sup>b</sup>  | 8.13 <sup>b</sup>  | 0.4 | 0.97 | 3.41                 | 3.17              | 2.48              | 2.75              | 2.92              | 0.3 | NS   |
| Palam<br>Shankar<br>Makka | 11                     | 10.37 <sup>*</sup> | 9.46 <sup>b</sup>  | 9.42 <sup>b</sup>  | 7.19 <sup>b</sup>  | 0.3 | 0.56 | 2.48                 | 1.45              | 1.68              | 1.92              | 2.43              | 0.4 | NS   |
| PMG-97                    | 11                     | 8.02 <sup>b</sup>  | 7.49 <sup>b</sup>  | 10.50 <sup>b</sup> | 7.57 <sup>b</sup>  | 0.1 | 0.29 | 4.43                 | 3.95              | 3.55              | 3.92              | 3.7               | 0.4 | NS   |
| Local<br>Chadhiar         | 9.2                    | 7.29 <sup>b</sup>  | 8.54 <sup>*</sup>  | 8.56 <sup>*</sup>  | 7.50 <sup>b</sup>  | 0.4 | 0.9  | 3.18                 | 2.87              | 3.06              | 2.44              | 2.35              | 0.4 | NS   |
| African<br>Tall           | 13                     | 8.25 <sup>b</sup>  | 8.69 <sup>b</sup>  | 9.96 <sup>b</sup>  | 8.26 <sup>b</sup>  | 0.4 | 0.87 | 6.17                 | 5.00 <sup>b</sup> | 5.05 <sup>b</sup> | 5.86 <sup>*</sup> | 5.90 <sup>*</sup> | 0.4 | 0.89 |

Abdel-Azim *et al.* (2011) also reported decrease in ADL content (from 11.19 to 10.24%) of maize stalks inoculated with *T. viride*. The decrease in ADL content of maize stalks inoculated with *P. ostreatus* and *S. cerevisiae* has also been reported by Darwish *et al.* (2012). The delignification in maize stover after treatment with microorganisms may be due to secretion of oxidative ligninolytic enzymes which aid the enzymatic digestion of lignin (Zhou *et al.*, 2024).

**Phenolic, tannin and silica content:** High phenolic and tannin content inhibit the growth and activity of ruminal microbiota in livestock thus affects digestibility and absorption of nutrients from forages. The total phenolic content in untreated maize stover was observed

in the range of 9.12 (SIRI- 5455) to 11.78 mg/g (PMG-93). The highest reduction in total phenolic content was observed in stover from genotype African Tall (8.25% and 8.26%, respectively) after treatment with *T. harzianum* and *P. fluorescens*. The decrease in phenolic content after the treatment may be due to their utilization as the sole source of carbon and energy by *P. fluorescens* (Mahiudddin *et al.*, 2012). Treatment of maize stover with *T. viride* also resulted in a significant reduction in total phenolic content. Maize stover inoculated with *S. cerevisiae* revealed variation in total phenolic content from 6.06 (SIRI-5455) to 10.91 mg/g (PMG-52-1) where highest reduction in total phenolic content was observed in stover from genotype SIRI-5455 (Table 4).

**Table 5.** Silica content and *in vitro* dry matter digestibility (IVDMD) of raw and treated maize stover from different genotypes

| Genotypes           | UT                 | Th                | Tv                | Sv                | Ps                | SE  | CD   | UT        | Th                 | Tv                 | Sv                 | Ps                 | SE  | CD   |
|---------------------|--------------------|-------------------|-------------------|-------------------|-------------------|-----|------|-----------|--------------------|--------------------|--------------------|--------------------|-----|------|
|                     | Silica content (%) |                   |                   |                   |                   |     |      | IVDMD (%) |                    |                    |                    |                    |     |      |
| PMG-3               | 2.1                | 1.60 <sup>b</sup> | 1.80 <sup>b</sup> | 1.73 <sup>b</sup> | 1.33 <sup>b</sup> | 0.1 | 0.23 | 52.1      | 69.20 <sup>a</sup> | 65.20 <sup>a</sup> | 68.30 <sup>a</sup> | 62.40 <sup>a</sup> | 0.2 | 0.42 |
| SIRI-5455           | 1.9                | 1.40 <sup>b</sup> | 1.27 <sup>b</sup> | 1.20 <sup>b</sup> | 1.20 <sup>b</sup> | 0.2 | 0.35 | 55.4      | 67.40 <sup>a</sup> | 63.20 <sup>a</sup> | 65.40 <sup>a</sup> | 63.20 <sup>a</sup> | 0.2 | 0.43 |
| PMG-9               | 1.6                | 1.33 <sup>*</sup> | 1.13 <sup>b</sup> | 0.93 <sup>b</sup> | 0.87 <sup>b</sup> | 0.2 | 0.41 | 56.2      | 65.30 <sup>a</sup> | 69.00 <sup>a</sup> | 62.40 <sup>a</sup> | 60.40 <sup>a</sup> | 0.3 | 0.62 |
| PMG-14              | 3.4                | 2.00 <sup>b</sup> | 2.47 <sup>b</sup> | 2.67 <sup>b</sup> | 2.30 <sup>b</sup> | 0.3 | 0.56 | 55.2      | 69.20 <sup>a</sup> | 67.00 <sup>a</sup> | 68.10 <sup>a</sup> | 65.40 <sup>a</sup> | 0.2 | 0.41 |
| PMG-18              | 2.6                | 2.13 <sup>b</sup> | 1.93 <sup>b</sup> | 2.00 <sup>b</sup> | 1.80 <sup>b</sup> | 0.1 | 0.21 | 53.2      | 65.40 <sup>a</sup> | 67.00 <sup>a</sup> | 67.90 <sup>a</sup> | 63.20 <sup>a</sup> | 0.4 | 0.91 |
| Vivek               | 2.5                | 2.07 <sup>b</sup> | 1.67 <sup>b</sup> | 1.60 <sup>b</sup> | 1.20 <sup>b</sup> | 0.1 | 0.28 | 56.2      | 67.20 <sup>a</sup> | 68.20 <sup>a</sup> | 67.20 <sup>a</sup> | 68.40 <sup>a</sup> | 0.2 | 0.4  |
| PMG-33              | 1.9                | 1.60 <sup>b</sup> | 1.20 <sup>b</sup> | 1.00 <sup>b</sup> | 1.10 <sup>b</sup> | 0.1 | 0.26 | 57.6      | 69.00 <sup>a</sup> | 67.20 <sup>a</sup> | 68.00 <sup>a</sup> | 62.20 <sup>a</sup> | 0.4 | 0.97 |
| PMG-36              | 2.5                | 2.13 <sup>b</sup> | 1.87 <sup>b</sup> | 1.73 <sup>b</sup> | 2.10 <sup>b</sup> | 0.1 | 0.3  | 57.1      | 68.20 <sup>a</sup> | 65.40 <sup>a</sup> | 67.20 <sup>a</sup> | 69.40 <sup>a</sup> | 0.4 | 0.9  |
| PMG-40              | 2.5                | 1.90 <sup>b</sup> | 1.80 <sup>b</sup> | 2.07 <sup>b</sup> | 1.47 <sup>b</sup> | 0.1 | 0.29 | 58.6      | 69.40 <sup>a</sup> | 67.20 <sup>a</sup> | 65.40 <sup>a</sup> | 68.00 <sup>a</sup> | 0.3 | 0.69 |
| PMG-41              | 2.1                | 1.73 <sup>b</sup> | 1.40 <sup>b</sup> | 1.80 <sup>*</sup> | 1.70 <sup>b</sup> | 0.1 | 0.32 | 59.2      | 70.00 <sup>a</sup> | 69.40 <sup>a</sup> | 66.20 <sup>a</sup> | 65.40 <sup>a</sup> | 0.2 | 0.36 |
| PMG-49              | 1.3                | 1                 | 0.73              | 1.07              | 0.73              | 0.2 | NS   | 58.3      | 72.20 <sup>a</sup> | 70.00 <sup>a</sup> | 65.30 <sup>a</sup> | 67.30 <sup>a</sup> | 0.1 | 0.22 |
| PMZ-4               | 2.7                | 2.20 <sup>b</sup> | 1.73 <sup>b</sup> | 1.87 <sup>b</sup> | 1.70 <sup>b</sup> | 0.2 | 0.4  | 56.4      | 69.20 <sup>a</sup> | 70.20 <sup>a</sup> | 67.40 <sup>a</sup> | 69.40 <sup>a</sup> | 0.2 | 0.4  |
| PMG-52              | 2.9                | 2.47 <sup>b</sup> | 1.93 <sup>b</sup> | 2.00 <sup>b</sup> | 1.53 <sup>b</sup> | 0.2 | 0.37 | 57.4      | 68.40 <sup>a</sup> | 69.80 <sup>a</sup> | 69.00 <sup>a</sup> | 62.40 <sup>a</sup> | 0.2 | 0.36 |
| PMG-52-1            | 2.7                | 2.33 <sup>*</sup> | 2.33 <sup>*</sup> | 1.47 <sup>b</sup> | 1.73 <sup>b</sup> | 0.3 | 0.55 | 52.8      | 65.90 <sup>a</sup> | 68.20 <sup>a</sup> | 66.20 <sup>a</sup> | 65.30 <sup>a</sup> | 0.1 | 0.26 |
| PMG-62              | 3.1                | 2.27 <sup>b</sup> | 2.53 <sup>b</sup> | 2.73 <sup>b</sup> | 2.10 <sup>b</sup> | 0.1 | 0.26 | 56.9      | 67.80 <sup>a</sup> | 65.00 <sup>a</sup> | 66.00 <sup>a</sup> | 61.40 <sup>a</sup> | 0.2 | 0.37 |
| PMG-93              | 2.1                | 1.73 <sup>*</sup> | 1.40 <sup>b</sup> | 1.20 <sup>b</sup> | 1.00 <sup>b</sup> | 0.2 | 0.42 | 57.8      | 68.00 <sup>a</sup> | 69.20 <sup>a</sup> | 67.20 <sup>a</sup> | 65.90 <sup>a</sup> | 0.2 | 0.41 |
| PMG-93-1            | 2.3                | 1.60 <sup>b</sup> | 2.00 <sup>b</sup> | 2.00 <sup>b</sup> | 1.40 <sup>b</sup> | 0.1 | 0.29 | 56.4      | 69.20 <sup>a</sup> | 70.00 <sup>a</sup> | 68.40 <sup>a</sup> | 66.30 <sup>a</sup> | 0.2 | 0.41 |
| PMG-93-2            | 1.6                | 1.33 <sup>*</sup> | 1.40 <sup>*</sup> | 1.00 <sup>b</sup> | 0.73 <sup>b</sup> | 0.1 | 0.31 | 56.3      | 67.00 <sup>a</sup> | 65.40 <sup>a</sup> | 69.40 <sup>a</sup> | 69.80 <sup>a</sup> | 0.3 | 0.55 |
| PMG-95              | 1.8                | 1.20 <sup>b</sup> | 1.13 <sup>b</sup> | 0.87 <sup>b</sup> | 0.60 <sup>b</sup> | 0.2 | 0.39 | 53.4      | 66.00 <sup>a</sup> | 65.00 <sup>a</sup> | 66.00 <sup>a</sup> | 67.90 <sup>a</sup> | 0.6 | 1.3  |
| PMG-96              | 2.5                | 2.00 <sup>b</sup> | 2.13 <sup>b</sup> | 1.67 <sup>b</sup> | 1.60 <sup>b</sup> | 0.2 | 0.36 | 54.8      | 65.20 <sup>a</sup> | 66.20 <sup>a</sup> | 67.60 <sup>a</sup> | 66.80 <sup>a</sup> | 0.3 | 0.63 |
| Palam Shankar Makka | 2.6                | 1.93 <sup>b</sup> | 1.40 <sup>b</sup> | 1.20 <sup>b</sup> | 1.70 <sup>b</sup> | 0.2 | 0.38 | 59.4      | 69.20 <sup>a</sup> | 69.20 <sup>a</sup> | 68.20 <sup>a</sup> | 67.40 <sup>a</sup> | 0.2 | 0.43 |
| PMG-97              | 2.6                | 2.20 <sup>*</sup> | 2.33 <sup>*</sup> | 1.80 <sup>b</sup> | 1.30 <sup>b</sup> | 0.2 | 0.42 | 57.6      | 67.40 <sup>a</sup> | 68.00 <sup>a</sup> | 64.30 <sup>a</sup> | 67.80 <sup>a</sup> | 0.2 | 0.34 |
| Local Chadhiar      | 1.5                | 1.40 <sup>*</sup> | 1.47 <sup>*</sup> | 1.07 <sup>b</sup> | 1.00 <sup>b</sup> | 0.1 | 0.28 | 58.6      | 65.40 <sup>a</sup> | 67.20 <sup>a</sup> | 64.90 <sup>a</sup> | 69.40 <sup>a</sup> | 0.2 | 0.52 |
| African Tall        | 1.9                | 1.60 <sup>b</sup> | 1.20 <sup>b</sup> | 1.00 <sup>b</sup> | 1.30 <sup>b</sup> | 0.1 | 0.2  | 59.4      | 70.00 <sup>a</sup> | 69.40 <sup>a</sup> | 65.40 <sup>a</sup> | 66.60 <sup>a</sup> | 0.4 | 0.91 |

**Table 6.** Categorization of genotypes based on microbial responsiveness

| Response group                   | Genotypes                                                                                                                          | CP mean range (%) | IVDMD mean range (%) | Key characteristic                             |
|----------------------------------|------------------------------------------------------------------------------------------------------------------------------------|-------------------|----------------------|------------------------------------------------|
| Group I (Highly responsive)      | PMG-93-2, PMG-14, PMZ-4, PMG-49, PMG-93                                                                                            | 11.09-12.15       | 66.52-66.68          | Lowest fibre; highest digestibility gains      |
| Group II (Moderately responsive) | PMG-3, PMG-9, PMG-18, Vivek, PMG-36, PMG-41, PMG-52-1, PMG-62, PMG-93-1, PMG-96, Palam Shankar Makka, Local Chadhiar, African Tall | 10.01-10.50       | 63.34-66.68          | Consistent but intermediate response           |
| Group III (Least responsive)     | SIRI-5455, PMG-33, PMG-40, PMG-52, PMG-95, PMG-97                                                                                  | 8.86-9.84         | 62.66-65.02          | Narrow response range; cell-wall recalcitrance |

Classification based on Duncan's homogeneous subset analysis ( $\alpha = 0.05$ ) for crude protein (CP) and *in vitro* dry matter digestibility (IVDMD) as primary discriminators

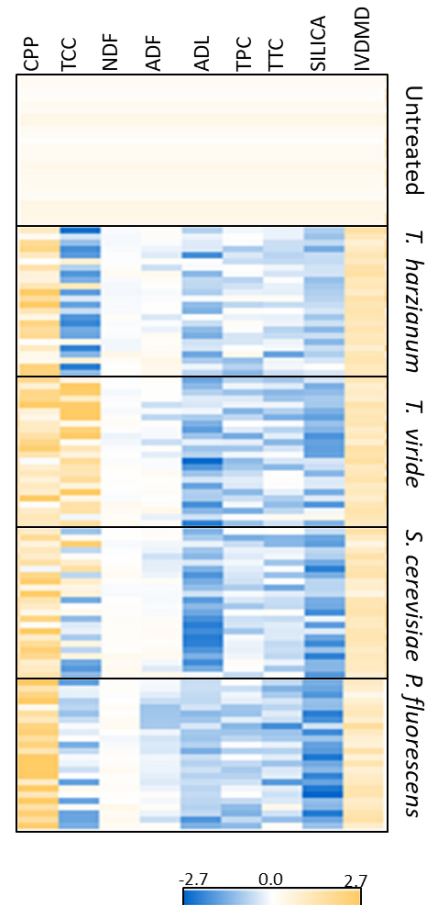
**Table 7.** Summary of Genotype × Treatment interaction effects

| Traits                 | F (Genotype) | F (Treatment) | F (G×T) | p (G×T) | R <sup>2</sup> |
|------------------------|--------------|---------------|---------|---------|----------------|
| Crude protein          | 781.31       | 9642.11       | 505.41  | < .001  | 0.998          |
| Carbohydrate           | 1646.25      | 48174.32      | 947.97  | < .001  | 0.998          |
| NDF                    | 347.41       | 1743.79       | 9.27    | < .001  | 0.992          |
| ADF                    | 326.70       | 1687.93       | 8.97    | < .001  | 0.997          |
| Lignin                 | 518.65       | 1232.45       | 11.13   | < .001  | 0.987          |
| Total phenolic content | 1381.35      | 4679.49       | 141.97  | < .001  | 0.996          |
| Tannin                 | 4593.97      | 1998.63       | 108.73  | < .001  | 0.998          |
| Silica                 | 1673.08      | 4651.53       | 79.86   | < .001  | 0.996          |
| IVDMD                  | 211.24       | 15251.61      | 101.42  | < .001  | 0.997          |

F-values for Genotype (df = 23, 238), Treatment (df = 4, 238), and Genotype × Treatment interaction (df = 92, 238); R<sup>2</sup> = Model R<sup>2</sup> from full two-way ANOVA including replicate; All interaction F-values significant at P<.001, confirming that microbial treatment effects were genotype-dependent across all measured traits  
NDF: Neutral detergent fiber; ADF: Acid detergent fiber; IVDMD: *in vitro* dry matter digestibility.

Total tannins content revealed variation from 2.48 (Palam Shankar Makka) to 7.06 mg/g (PMG-52-1) in untreated maize stover. Treatment of stover with *T. viride* revealed a significant decrease (3.52%) in tannin content of stover from genotype PMG-49. Treatment with *S. cerevisiae* exhibited a significant decrease in total tannin content ranging from 1.92 to 6.87 mg/g of stover from different maize genotypes, where highest significant decrease was observed for the stover from genotype PMG-9 (Table 4). The presence of silica in cell wall is also considered as one of the important factors affecting the digestibility of crop residues. The stover from genotypes PMG-14 (3.40%) and PMG- 49 (1.30%) revealed significantly higher and lower silica content, respectively. Treatment of stover with *T. harzianum* and *T. viride* resulted in a significant decrease in silica content to the extent of 1.0% and 0.73%, respectively in PGM-49 genotype. After treatment with *S. cerevisiae* the significant reduction in silica content (0.87%) was observed for stover from genotype PGM-95 (Table-5), while the reduction in same genotype by *P. fluorescens* was 0.6% in comparison to control (1.8%). Among all treatments, inoculation of stover with *P. fluorescens* was highly effective in reducing silica content. The decrease in silica content in stover may be the result of increased solubilization of insoluble silicates by micro-organisms (Umamaheswari *et al.*, 2016).

***In vitro* dry matter digestibility (IVDMD):** The *in-vitro* dry matter digestibility of stover from different maize genotypes varied from 52.10% (PMG-3) to 59.40% (Palam Shankar Makka). *T. harzianum* treatment followed by *S. cerevisiae* treatment resulted in a significant increase (69.20% and 68.30 %, respectively) in the digestibility of stover from genotype PMG-3. Inoculation of stover with *T. viride* and *P. fluorescens* also improved the digestibility



**Fig 1.** Heatmap of maize genotypes response to different microbial treatments on biochemical composition. The untreated samples were considered as 100% of particular biochemical and the respective proportional enhancement or reduction has been represented as color code to provide a holistic view of treatment effect.

to the extent of 59.80% (PMG-52-1) and 67.90% (PMG-95), respectively (Table-5). The increase in digestibility of wheat straw (from 25.89 to 30.72%) has also been observed after treatment with *Trichoderma* (Yalchi and Hajieghrari, 2010). Chen *et al.* (1995) also reported an increase in the digestibility of maize stover with the inoculation of different white-rot fungi. The increase in digestibility might be due to solubilization of structural polymers in the cell wall (Fazaeli *et al.*, 2004; Yalchi and Hajieghrari, 2010).

## Conclusion

In conclusion, the treatment of maize stovers of different genotypes with three fungal and one bacterial strain revealed an effective method to improve the nutritive value as assessed by several biochemical analyses in comparison to control. The overall study concluded that *T. viride* is the most promising strain for improving the feed quality of maize stovers followed by *T. harzianum* and *P. fluorescens* (Fig 1). Different genotypes have been grouped as highly responsive, moderately responsive and least responsive groups based on crude protein and in vitro digestibility traits in response to microbial treatment, resulting in 5, 13 and 6 genotypes, respectively (Table 6). All the genotypes responded well to different microbial treatments as revealed by genotype X treatment interaction study (Table 7).

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