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The Success of the Small Tea Growers of Sittong Evolving Rural Darjeeling into a Model Small-Scale Organic Tea Cultivation Center

Soumya Majumder[1], Diksha Gurung[1], Sahadeb Sarkar[1], Sudeshna Nandi[1], Arindam Ghosh[1], Preeti Subba[1], Sukanya Acharyya[1], Sumedha Saha[1], Sourav Chakraborty[1,2], *Malay Bhattacharya[1]

[1]Molecular Biology and Tissue Culture Laboratory, Department of Tea Science, University of North Bengal, Raja Rammohunpur 734013, India

[2]Postgraduate Department of Botany, Darjeeling Government College, Darjeeling 734101, West Bengal, India

Abstract. The present study was focused on organic small tea plantations of Sittong (a village in Darjeeling) mainly due to their sudden rise in the Indian tea industry with a striking tactic i.e., organic tea cultivation. This cumulative survey and laboratorial experiments-based research focuses on the agricultural conversion in Sittong where farmers started to shift from vegetable and grain cultivation to small tea gardens. Soil physicochemical (pH and electric conductivity; organic carbon; organic matter; and available nitrogen content) and microbiological (determination of microbial cell mass and isolation of consortia; antibiotic and antifungal susceptibility test) characteristics were considered to assess the viability of this shifting agriculture practice and cross-verify the reflections of organic farming practices. The survey revealed that farmers have cultivated and rehabilitated the land in an acceptable manner before planting; they carried out the soil nutrient management practices organically. Survey also revealed economic perspectives including prices of their harvested tea leaves. Further, the moisture content analysis revealed its adequacy in the tea garden soils. Organic matter, organic carbon and available nitrogen were measured that reflected very high results compared to the optimum values suggested by the Tea Board of India. Microbial analysis results, as a cross-verifying tool, affirmed their organic farming practice by revealing microbes' susceptible nature towards antibiotics and antifungals. Overall, the findings of this study suggest that the small tea growers in Sittong-3 are well-positioned to produce a high-quality of organic tea. Sittong was found to have potential to promote the rural areas of Darjeeling into an ideal place for small-scale organic tea cultivation, while also serving as an inspiration for small tea growers across the country.

Key words: small tea grower, soil, organic farming, organic tea, Darjeeling tea.

Introduction

In India, tea is the most economically significant beverage culture. Its cultivation and trade significantly contribute to the country's income from abroad. The quality indicators, such as flavour or aroma, taste, colour of liquor, etc., are greatly influenced by ecophysiological aspects i.e., geographic characteristics and environmental elements including soil quality- its moisture content, pH and nutrient availability; climate- rainfall, temperature, humidity and other weather parameters; seasons; and topography or altitude of tea gardens. More than 55% of India's total tea production comes from the small tea grower (STG) category (Das *et al.,* 2023). Farmers having tea cultivation area of up to 25 acres are considered as STGs, as defined by the Tea Board of India (Das *et al.,* 2023). However, the majority of producers currently possess less than 2 acres of land (Hannan, 2021; Das *et al.,* 2023). STGs of Darjeeling and adjacent hills, and sub-Himalayan plains (Terai and Dooars) are frequently based in extremely remote locations. Due to their dispersed and isolated existence, they lack organization and negotiation skills even as their numbers grow. To sell their harvest or leaves, they are reliant on unfair trade routes. Numerous other problems pose a threat to their existence and way of life. For instance, growers struggle with a lack of funding and poor levels of technical and business

* Corresponding Author's email: malaytsnbu@gmail.com

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understanding. They are also unaware of government assistance.

The uprise of small tea growers has been noticed over the past two decades. Since then, tea industry of India transformed significantly. Das *et al.* (2023) emphasized emergence of BLF (bought-leaf factories), shifting from inorganic to organic farming, foreign market competition, and struggles due to old bushes and depletion of soil nutrients as supplementary issues while describing STGs' scenario in West Bengal. Indian STGs are now holding the major share of tea business producing more than half of the total production (Das *et al.*, 2023). In North Bengal, there are more than fifty thousand STGs as per the recent data (https://www.millenniumpost.in/bengal/small-tea-growers-seek-reduction-in-land-fees-540005). Sinha (2020) reported that, according to Tea Board of India and small tea growers associations and forums, only 15,000 small tea growers among fifty thousand are affiliated to the tea board. Hannan (2021) reported the exact figure of small tea growers in northern West Bengal that was 20,352 in 2009 and their average land area was 3 acres. A study by Rama and Dasgupta (1987) revealed that in 1980's farmers of Terai region of West Bengal started cultivating tea in small pieces of their agricultural land which are previously used for pineapple cultivation. Similarly, in Dooars region, the same practice has been initiated where tea plantations emerged in the cultivation lands of grains like paddy, wheat, maize, etc.; vegetables (mainly potato); and jute. District Uttar Dinajpur stills holds the largest figure of STGs. Small tea growers have also emerged in the hills (Darjeeling district) but little information is available regarding them in the scholarly world. Literature documented by Rai (2017) is one of the few documentations available on small farmers of "Darjeeling Tea" where he reported competence of small farmers from remote locations such as Dabaipani (or Mineral Spring), Yangkhoo and adjacent hills of Darjeeling to produce quality Darjeeling tea.

Soil testing plays an important role in the use of fertilizers and other agricultural inputs. Soil is one of the most important factors that controls the quality and yield of tea. Several soil factors like soil moisture, pH, electrical conductivity, organic matter, organic carbon, total nitrogen, potash, phosphate, sulphur and micronutrients play vital role for the crop. Tea board of India has recommended ranges for soil parameters (Mukherjee *et al.*, 2020). Soil microbial consortia play a pivotal role in determining the success of tea plantations (Saha *et al.*, 2021). These consortia, composed of a diverse group of microorganisms, including bacteria, fungi, and other microbes, collaboratively engages in essential functions within the soil ecosystem. The antibiotic tolerance of these soil microorganisms might present another dimension of inquiry. As antibiotics are used in tea gardens to combat bacterial infections, it was important to explore how the microbial consortia respond to these compounds and whether their tolerance holds implications for sustainable agricultural practices (Cycoń *et al.*, 2019). This study also demonstrates the tolerance pattern of microbial consortia against antifungals commonly employed in tea gardens. By discussing the complexities of such tolerance patterns, the aim was to evaluate the relationship between soil microorganisms and chemical inputs which may help in understanding the effects of organic practices, impact of conversion from inorganic to organic agriculture and resilience and adaptability of these vital soil inhabitants. Therefore, alongside survey, soil physicochemical (moisture, pH, electric conductivity, organic matter, organic carbon and nitrogen content) and microbiological characterization (determination of microbial presence and isolation of consortia; antibiotic and antifungal susceptibility test) were aimed for this research. The organic STGs of Sittong have not been the subject of any investigation to date. However, no literature is available that describes either their plantations, soil physicochemical and microbiological characteristics, morphological and physiological studies or socio-economic status. The aim of this research work was to bring the STGs of Sittong in highlight and to learn about their history of cultivation, their cultural practices, and analysis of different soil physicochemical and microbial parameters. Present study has been designed to further assist in improving tea plantations of this study area.

Materials and Methods

Study area

Sittong or Sitong is a "Khasmahal", a cluster of villages located at the Kurseong division of Darjeeling district in the state of West Bengal, India. Sittong-3 located at 26°56'10.3"N 88°22'02.5"E (Figure 1) is in a village in Sittong Khasmahal and has been our study area. There are a total of thirty small tea growers in Sittong Khasmahal among which twelve STGs reside at village Sittong-3 and practice organic farming in their tea gardens who were selected for this study.

Survey

A survey work was carried out on small tea growers of Sittong-3 who either used virgin lands or converted a major portion of their agricultural lands into organic tea plantations. Total twelve farmers participated in this survey whose details such as land type, clonal variety, cultivation time, seasonal cost on cultivation, pruning cycle, planting pattern, fertilizers and pesticides uses etc. have been collected though questionnaires as given in Table 1.

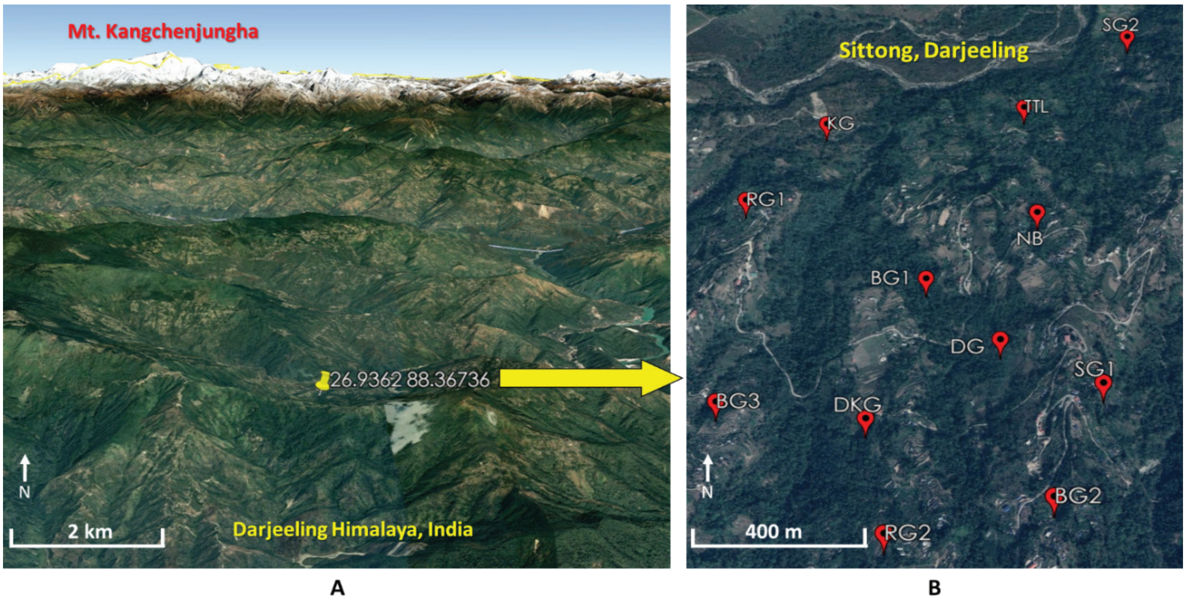

Figure 1. Study area of this research.
(A) Location of Sittong-3 (26°56’10.3”N 88°22’02.5”E) in Darjeeling Himalaya; (B) Sample collection sites and surveyed plantations in Sittong-3 [Map source: Google earth (https://earth.google.com/web/)]

Soil sampling and preparation of soil for analysis

A total of approximately 100 g soil were collected from each small tea gardens of Sittong-3 (Figure 1-B) and the sampling was done in a zigzag pattern at several locations to ensure homogeneity. Only topsoil (15 cm of depth) from the rhizosphere region was earlier marked as ideal by Saha *et al.* (2021) for soil microbiology studies, a part of this research. Soil collection was done in the post-monsoon period (October-December) which was also considered ideal for soil-microbial studies by Saha *et al.* (2021). From the collected soil sample all the unwanted debris like debris like pebbles, stones, roots, twigs etc. was removed manually from collected soil samples. Soil samples were divided into two proportions for physicochemical tests and microbial analysis. The samples collected were coded as: BG1, NB, SG2, DG, TTL, DKG, SG1, BG2, BG3, RG1, KG, RG2 (according to the initials of the farmer’s name and surname) (Table 1). The soil collection and subsequent physicochemical analysis were carried out during post-monsoon period (autumn season) for consecutive three years (2021- 2023) and the results have been expressed as mean ± SD.

Analysis of soil physicochemical properties

For this comparative analysis, selected physiochemical parameters like pH, electrical conductivity or EC, organic matter, organic carbon and available nitrogen were taken into account to characterize the soil samples following the protocols reported by Sarkar *et al.* (2022) and Ghosh *et al.* (2022). These protocols were the ones approved by the Tea Board of India as mentioned by the authors and performed by the Tea Research Association (TRA) as well (Mukherjee *et al.,* 2020).

Soil moisture

Absolute soil moisture was quantified following Mukherjee *et al.* (2020) and Das *et al.* (2023). Each pre-weighed fresh soil sample was taken in a beaker and dried in hot-air oven at 110°C for 48 hours. The final weight (weight of the dried soil) was then determined, and moisture content was estimated using the following formula. Three replicas were used for each sample in this experiment.

$$Soil\ moisture\ (\%) = \frac{W1-W2}{W1} \times 100,$$

where:
W1 - the weight of fresh soil (g);
W2 - the final weight (g) or weight of the same soil after drying.

Soil pH and EC (electrical conductivity)

The pH of the soil sample was determined using a pre-calibrated pH meter. Inside a 100 ml conical flask, 20 g of each soil (after sieving using a 2 mm sieve) were taken and had been air dried. Each flask was then filled with 50 ml of distilled water to maintain a 1 : 2.5 (soil : water) ratio, which was then shaken vigorously for 30 minutes in a mechanical shaker. The pH meter (LMPH-10) was calibrated using standard buffer solutions with pH values of 4.1, 7.0, and 10.01, respectively. After calibration, pH of each sample was measured. A digital EC meter was used to measure electrical conductivity, and all results were recorded in triplicates (Ghosh *et al.,* 2022; Sarkar *et al.,* 2022).

Soil organic carbon (OC), organic matter (OM) and available nitrogen (N)

The titration method for estimation of organic carbon or OC (%) and calculated organic matter or OM (%) were followed as developed by pioneer scientists Walkley and Black (Walkley and Black, 1934; Sarkar *et al.,* 2022; Ghosh *et al.,* 2022). The amount of organic carbon was measured using the chromic acid method. To assess this, 0.5 g of each air-dried and sieved (2 mm) soil was combined with 10 ml of potassium dichromate ($K_2Cr_2O_7$) solution and 20 ml of concentrated sulfuric acid (H_2SO_4), and the mixture was left undisturbed for 30 minutes to complete the digestion process. Then, using distilled water, the volume of each solution was increased to 200 ml. Then 0.2 g of NaF (sodium fluoride) and 1 ml of diphenylamine indicator were added before titration. Sodium fluoride (NaF) enhances gradual colour change and gives the solution a distinctive sharpness. Further titration was performed with 0.5 N ferrous ammonium sulphate ($(NH_4)_2Fe_3(SO_4)_2.6H_20$) until a bright green colour appeared. The amount of titrant i.e., volume of the ferrous ammonium sulphate reagent used for titration was recorded. One conical flask was prepared following the same method but without sample for blank. The soil organic carbon was quantified using the given formula.

$$OC\ (\%\ of\ soil\ organic\ carbon) = \frac{B - S}{B} \times 7.79,$$

where:
B - titration value (volume of the ferrous ammonium sulphate used for titration) for the blank;
S - titration value shown by the soil sample.

The percentage organic matter and available nitrogen were calculated from organic carbon using the formulas given below.

$$OM\ (\%\ of\ soil\ organic\ matter) = OC \times 1.72$$
$$N\ (\%\ of\ available\ nitrogen) = OC \times 0.086$$

Analysis of soil microbial properties

Following the protocols of Saha *et al.* (2021), microbial consortium was isolated from each sample (collected in 2023) and subjected for follow-up experiments to fulfill the objectives. Determination of overall tolerance pattern exhibited by the microbial community isolated from tea garden soil against antibiotics and antifungals was considered as an index to judge the soil health of the land which has either been considered for organic farming from the beginning or converted to tea gardens for organic farming from inorganic agricultural field. Consideration of microbial parameters as index is a novel approach in studies on STGs. Microbe play important role in organic farming, and these experiments will reflect soil composition through the mirror of toxicity potentials of antibiotic and antifungal towards isolated microbes. Moreover, the consequences of organic farming practice in a virgin land or a converted agricultural land (containing inorganically exploited soil) could be studied from such soil-microbiological research. All the microbial experiments were carried out in a sterile environment (laminar air flow).

Isolation of microbial consortium from soil sample

Isolation of the microbial community from the soil sample was done following procedures reported by Saha *et al.* (2021) with slight modifications. In a digital balance, 500 mg of freshly obtained soil samples were measured (excluding pebbles, stones and organic debris) and taken in an autoclaved Eppendorf tube, and 1 ml of sterile distilled water was added. The solution was then vortexed for one minute to ensure optimum soil mixing. The microbial consortium was isolated using Nutrient Broth as the preferred media (Saha *et al.,* 2021). The media was sterilized by autoclaving at 250°F (121°C) for 30 minutes at 15 psi. 10 ml of autoclaved nutrient broth was poured inside each sterilized culture tube. 500 µl sample solution was inoculated in the media using sterilized micropipette. The inoculated media was incubated at 35°C for 48 hours in a shaking incubator. After incubation, the samples were stored at 4°C for further experiments.

Microbial cell dry weight determination assay

This is a preliminary and low-cost but very useful method to determine cell-mass for measurement of microbial population. The protocol was obtained from Bratbak and Dundas (1984) and carried out with slight modification. 500 µl sample solution (vortexed mixture of 500 mg soil and 1 ml sterile distilled water) solution was inoculated in culture tube containing 10 ml nutrient broth and incubated at 35°C for 48 hours in a shaking incubator. Following incubation, 2 ml cultured broth from each tube were centrifuged, supernatant was discarded carefully, pellet was again washed three times with sterile distilled water, the resulting sediment was then dried in oven at 50-55**°C** (https://www.biotechfront.com/2021/09/determination-of-cell-mass-for.html) and dry weight of the sediment was determined which corresponds to the microbial load in the original sample. Three replicas were used for each sample in this experiment.

Antibiotic and antifungal susceptibility assay

Antibiotics have been regularly utilized since their discovery until recently. Because of their widespread use, the targeted bacterial population has developed tolerance to these chemicals as a self-defense mechanism. Protocol reported by Saha *et al.* (2021) was used to perform the *in vitro* antibiotic tolerance assay. Antibiotics i.e., bacitracin, chloramphenicol, penicillin-g, polymyxin-B, gentamicin, neomycin, cefotaxime, augmentin, erythromycin, chloramphenicol, ofloxacin, and co-trimoxazole (HiMedia, India) were used in this experiment. In an autoclaved petri-plate, 200 µl of sample culture was employed as an inoculum, and nutrient agar media (autoclaved) was poured. The plates were securely shaken to ensure consistent mixing of the inoculum. The petri-plates were left undisturbed for a few minutes to allow the agar to solidify. After hardening the agar, antibiotic hexa-discs were placed aseptically on top of the agar and incubated at 35°C for 24 hours. Inhibition zones surrounding the discs were measured.

In vitro antifungal susceptibility test was also performed on microbial consortia isolated from soil samples. The antifungal tolerance by each consortium culture was tested using HiMedia antifungal discs following Saha *et al.* (2021). Six antifungals namely amphotericin-B, clotrimazole, fluconazole, itraconazole, ketoconazole and nystatin were used here. For antifungal susceptibility assay, again pour-plate technique was followed as described in the antibiotic susceptibility assay methodology. Inhibition zones surrounding the discs were measured.

Data analyses

Data Analysis Tools of Microsoft Excel (v2303) was used to get the correlation coefficient. The Pearson correlation matrix from selected experimental data (Soil moisture%, pH, EC, OC and microbial cell dry weight or MCDW) was built using the in Excel Correlation tool (Majumder *et al.*, 2024).

Results and Discussion

Results of the survey

The survey conducted in small tea gardens of Sittong-3 generated a pile of information as described here. A copy of the filled-up questionnaires is attached in Table 1. Small tea growers of Sittong are based in very remote areas and in clusters. The farmers of Sittong started practicing organic tea farming as organic tea production by the set gardens of Darjeeling hills started becoming more accepted due to growing awareness for health concerns among the consumers; fetching good prices; high demand in the domestic and international markets; and the urge to generate an ecofriendly and sustainable industry. Farmers of Sittong started recognizing the potential of organic farming in maintaining soil fertility over longer time without nutritional depletion. Aiming at long-term benefits, some of the STGs of Sittong cultivated virgin land in their properties (deforested areas) for tea plantation while others shifted a portion of their agricultural lands to tea plantations where they were earlier engaged in the organic cultivation of vegetables like squash, potatoes, ginger, carrots etc. 50% of the surveyed participants (six STGs) stated their objective as shifting agriculture to cultivate tea for more profit. Their land was previously used for the cultivation of grain crops like maize and millets, and vegetables like potato, ginger, carrots, squash etc. as revealed by the survey. Thus, shifting agriculture, which was earlier only reported in the plains i.e., Dooars region, has also become common in the hills (Das *et al.,* 2023). Tea is slowly becoming a farmer's crop, providing better economic profits and ushering in new avenues for employment. The STGs of this area are dependent on bought-leaf factories (BLFs) for selling their tea leaves. Currently, there are more than fifty STGs in Sittong-3. A sizeable amount of information, scientifically called "secondary data", has been acquired through this survey work, which has been described below.

The society in this area named "Shelpu Tham Dara Small Tea Growers", was formed in the year 2013. In the beginning, there was a total of eleven members which has grown to thirty-eight members in 2023. Some quantitative comparisons were also made in this survey. According to the report obtained from the data, the areas were mostly hills with steep slopes (Figure 2), varying from 1 acre to 6.5 acres, with a promising rate of tea leaf production ranging between 650–5,500 kg per plantation (approx. 750 kg per acre). The leaf production rate by STGs of the study is less compared to the STGs of sub-Himalayan plains (Terai and Dooars) (Das *et al.,* 2023). Generally, Assam (*Camellia sinensis* var. *assamica*) type of clones (TV varieties) is cultivated in the plains, while in Darjeeling hills, Chinese bushes are usually preferred which although produces a lower harvest, contributes better quality (aroma and flavour) in the cup. Quality of plucking can also be a reason as only two leaves and a bud are plucked in Sittong to maintain the liquor quality while in the plains this is not followed.

Cultivation area of more than 40% of the STGs were recorded to be between 1-2 acres while around 33% of the rest planted in an area of 2-3 acres. Therefore, the survey revealed about 75% of participating STGs of Sittong are presently cultivating tea in a land which is not more than 3 acres. Only three farmers were found in Sittong-3 who use more than 4 acres of their land for tea cultivation and were included in the survey as

Table 1

Results of survey

Owner/ Name of the STG	**Bhupendra Gurung**	**Nar Bahadur Gurung**	**Samir Gurung**	**Dipendra Gurung**	**Ten Tshering Lepcha**	**Dev Kumar Gurung**	**Suresh Gurung**	**Bimal Gurung**	**Binod Gurung**	**Reman Gurung**	**Kishan Gurung**	**Reema Gurung**
Sample code (for soil analysis)	BG1	NB	SG2	DG	TTL	DKG	SG1	BG2	BG3	RG1	KG	RG2
Address	Sittong 3 Tham Dara, Darjeeling, West Bengal, India											
Cultivation Area	4 acres	2 acres	1 acre	2 acres	6.5 acres	1 acre	1.75 acres	5 acres	2 acres	1 acre	2 acres	1 acre
Land Type	Agricultural and virgin land	Virgin land	Agricultural land	Agricultural and virgin land	Virgin land	Agricultural and virgin land	Virgin land	Virgin land	Agricultural land	Agricultural and virgin land	Virgin land	Virgin land
Rehabilitation crops	Guatemala and citronella grass	Napier and citronella grass	Guatemala grass and mimosa	Guatemala grass	Guatemala grass	Guatemala grass	Citronella grass	Citronella grass	Guatemala grass	Guatemala grass	Napier grass	Napier and Citronella grass
Bank Loan	Nil.											
Tea Board of India affiliation	Yes											
Enlisted under STG association	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No	Yes	Yes	No
Age of Bush	12-14 years, 4-5 years	12-14 years	11 years	4-5 years	12-14 years, 8-9 years	11-12 years	5-6 years	13-14 years	3-4 years	12-14 years	12-14 years	3-4 years
Tea Clones	AV2, Phoobsering 312, Nandadevi, Bannockburn 157, Tukdah 135, Goomtee	Nandadevi, Bunnockburn 688, Goomtee, CP1, Badamtam 15/263	Phoobsering 312, Nandadevi, Tukdah 78, Goomtee	Nandadevi, Tukdah 135, Goomtee, AV2, Badamtam 15/263	Nandadevi, Bunnockburn 688, Goomtee, AV2, Goomtee	Tukdah 78, Goomtee, CP1, Bannockburn 688, Badamtam 15/263	Nandadevi, Tukdah 78, Goomtee, AV2, Phoobshering312	AV2, Phoobshering 312, Nandadevi, Tukdah 78, Goomtee	AV2, Bannockburn 157, Nandadevi, Tukdah 135, Tukdah 253, Goomtee	CP1, Bannockburn 688, Tukdah 253, Goomtee	Phoobshering 312, Nandadevi, Tukdah 78, Goomtee	Nandadevi, Tukdah 78, Tukdah 135, Goomtee
Planting Pattern	Single Hedge											
Plucking	Hand Plucking											
Pruning cycle	5 years											
Bush height	24, 26, 28 inches	24 inches	24 inches	25 inches	28 inches	24, 26 inches	24 inches	24 inches	24, 25, 28 inches	25 inches	24, 26, 28 inches	24, 25, 26 inches
Spacing	3/3 feet, 2/3 feet.											
Shade tree	Nil.											
Weed Management	Manually											
Pest Management	Use organic formulations such as extracts of neem and *Artemisia vulgaris*											
Fungicides	Trichoderma											
Foliar Spray	Amrit jal (fermented cow urine-based formulation), Neem oil											

Production (2021-22) (Approx.)	2500 kg	1500-1600 kg	800 kg	1500 kg	5000-5500 kg	800-850 kg	1200 kg	4500-5000 kg	1000 kg	800-850 kg	1600 kg	650 kg
Soil management	Cowdung, Vermicompost											
Price of plucking shoots	80-90 rupees/kg											
Drainage Management	No											
Manufacturing	No	Handmade and Small processing unit	No	No	Handmade and Small processing unit	No	No	No	Handmade	Handmade and Small processing unit	No	No

Figure 2. A small tea plantation of our study area in Sittong-3 based in the hilly terrain of Darjeeling.
The picture shows a single-hedge planting pattern, well maintained tea plants, quality plucking using hands and orange plants distributed in various points of the garden

well. The result has been graphically represented in Figure 3-A.

According to the surveyed farmers, the nursery plants were first bought from Fulbari, Siliguri. In the present scenario, the Small Tea Growers are practicing nursery management with some popular garden series clones (AV2, Phoobsering 312, Bannockburn 157, Bunnockburn 688, Tukdah 78, Tukdah 135, Tukdah 253, Goomtee, CP1, Badamtam 15/263 etc.) by vegetative propagation which includes both high yielding clones (Bunnockburn 688, CP1 and Tukdah 253) as well as high quality clones (AV2, Bannockburn 157, Phoobsering 312, Tukdah 78 etc.) (https://teaboard.gov.in/pdf/LIST_OF_APPROVED_PLANTING_MATERIALS_AND_SOIL_TESTING_LABORATORIES_pdf2583.pdf). Yield results were found clearly corresponding to the type of clones planted by the STGs. STG Dev Kumar Gurung and Reman Gurung, both cultivated high yielding clones- CP1 and Bunnockburn 688 in their gardens. In 2021-2022, they were able to produce around 850 kgs of harvest in just 1 acre of land which was much higher than the average production in this region i.e., 750 kg. Among the participants, Binod Gurung and Reema Gurung, two of the newest in the business (3-4 years old plantation) were not enlisted in the association- "Shelpu Tham Dara Small Tea Growers". However, all the other small tea growers of this area are either listed under the Tea Board of India or have their certification under progress which is an encouraging step by the STGs of the state. The number of small tea growers is gradually increasing year by year in this region.

Age profile assessment revealed that most of the bushes (60%) planted in small tea gardens of Sittong are aged about eleven years old and more. A comparative result has been given in Figure 3-B corresponding to the age of bushes. This assessment also revealed that organic small-scale tea plantations in this region were started around fourteen years ago, four years earlier than the formation of the association. In other terms, the association actually started from the period of tea production (it takes at least 3-4 years to grow pluckable shoots).

Moreover, tea garden management practices were surveyed, which revealed that the removal of earlier existing crops is done manually by hand as is typical of the hilly areas, where slope percentage is high, making it difficult to uproot plants by machinery. Rehabilitation of land is done following the guidelines of Tea Board of India (https://www.teaboard.gov.in/pdf/LIST_OF_APPROVED_PLANTING_MATERIALS_AND_SOIL_TESTING_LABORATORIES_pdf5072.pdf) that included using grasses like *Tripsacum laxum* (commonly known as Guatemala grass), which contribute organic materials to the soil through the loopings from the plant, adding approximately 250 tons of green matter per acre. Leguminous *Mimosa invisa* (thornless mimosa) is used as it has capabilities of fixing atmospheric nitrogen. Some STGs also use citronella grass (*Cymbopogon wintarianus*) and *Pennistetum purpereum* (Pusa Giant Hybrid Napier grass) as their looping also adds humus to the soil through huge amount of green matter (around 500 tons per acre). The farmers prefer single hedge (Figure 2) planting methods. Stalking is done with bamboo sticks to maintain an environment-friendly ecosystem. After the marking is done, each pit is dug manually at a depth not less than 45 cm and width not less than 30 cm. For planting, they select plants which have at least 12-15 leaves and are about 45-60 cm in height, with a stem-thickness of 0.5- 0.8 cm. Planting is done during October to November and April to June. STGs of this region practice cultural methods of mulching accurately as it is the vital step in organic tea farming. They use straw, pruned tea leaves, ferns and leaves from green manure grass as organic mulching materials which help to reduce soil erosion by protecting the soil from the impact of raindrops, help retain soil moisture and add organic

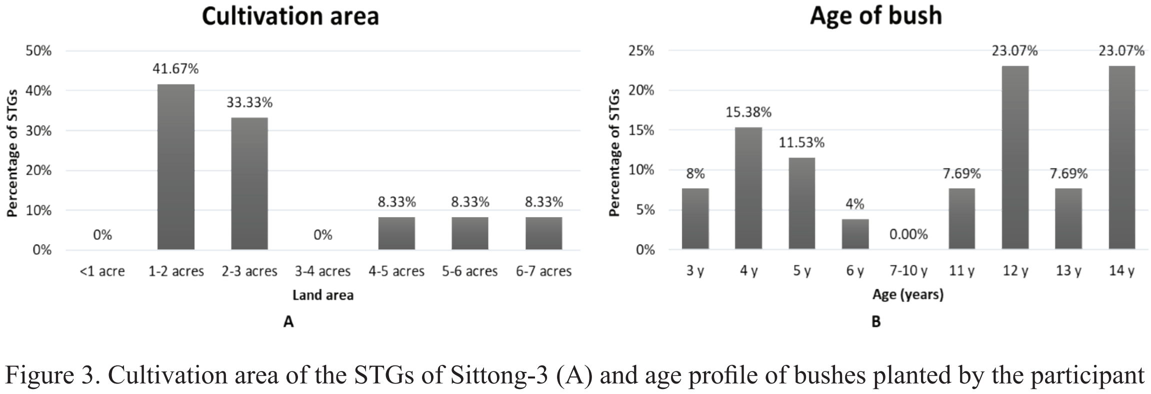

Figure 3. Cultivation area of the STGs of Sittong-3 (A) and age profile of bushes planted by the participant STGs of Sittong-3 (B).

matter to the soil leading to growth of beneficial soil microflora. STGs of Sittong-3 follow the five years' pruning cycle that includes decentering and tipping in the year of planting (0th year); re-center out and skiffing on 1st year; first Frame Forming Pruning (FFP) in the 2nd year of planting; light or medium pruning and later Leveling of Skiff (LoS) in the 3rd year; and "Final Frame Forming Pruning" on 4th year. They also practice knife cleaning out for proper maintenance of their bushes. After the plant reaches 5 years the pruning is done after a gap of 5 years. Shade trees are not planted in these tea gardens, however trees like orange (a common fruit tree of Sittong) and pine can be noticed even inside the tea gardens (Figure 2). No draining system was available in any of the gardens, and they mainly depended on rainwater for irrigation. To fertilize the soil, they mainly use cow dung (3 tons/ha) and FYM or farmyard manure (2 tons/ha) and sometimes vermicompost and green manure from their vegetable fields and households. Weed management is done manually. For pest control, they use organic formulations prepared from neem, artemisia. Tea mosquito bug (*Helopeltis theivora*), green fly, thrips, and looper caterpillars are the major pests in these gardens. Diseases like blister blight and black rot are also present. To prevent and control fungal diseases, they apply Trichoderma powder (2 kg/ha) in a ratio of 800 g/ 200 l of water at the rate of 2–3 times in a year. During this survey we came across a unique foliar spray called "Amrit Jal" in Sittong-3 which is a prepared by fermenting cow dung and cow urine along with some additive herbs, a product of indigenous traditional knowledge. To prepare "Amrit Jal", 20 kgs of raw cow dung, 4 liters of cow urine, 6 kgs of neem-oil cake, 2 kgs of gram flour or besan, 2 kgs of raw mud and Artemisia are added in 200 liters of water and left covered for one week to ferment. Later, they mix 10 liters of the fermented formulation in 100 liters of water to use as a foliar spray. According to the farmers, this formulation not only works as a fertilizer but also has ability to prevent pest attacks. Here, cow dung, cow urine, gram flour and raw mud might serve a role as resources for fermenting microbes as substrates or nutrients for the fermentation process. Surveyed small tea growers also conveyed that they perform soil testing in the Tea Board of India's approved soil analysis laboratory in Siliguri, Darjeeling.

The price of green leaves produced by organic farming is usually very high in this industry compared to others. In the beginning (2013) Organic STGs of Sittong were paid Rs. 25/kg against their green leaf whereas now, in 2023, the price has increased to Rs. 80-90/kg of green leaves. Reportedly, bought leaf factories pay an average Rs. 15 to 17 per kg of green leaves from small tea growers of the sub-Himalayan plains, mostly in Assam and West Bengal (Das *et al.,* 2023; https://www.deccanherald.com/india/low-green-leaf-price-hits-small-tea-growers-in-assam-west-bengal-1013132.html). This comparison is not justified as manufactured organic teas are sold at high end prices in a quality demanding market which is different from the quantity demanding domestic market of CTC and orthodox black tea produced in the plains of West Bengal and Assam. The survey revealed that organic tea farmers of Sittong enjoy high profitability compared to what they were making before shifting to tea. Some of the participant STGs (Nar Bahadur Gurung, Ten Tshering Lepcha, Bimal Gurung and Binod Gurung) have been involved in the manufacturing of handmade tea inside their garden and also installed small-scale processing units for quantity production, which can bring them a huge profit in the near future.

A study by Yao *et al.* (2018) revealed another major tea producing country- China has also started this shifting or agriculture conversion practice to expand its tea cultivation due to the raising demand for tea products in international market. Scientists also criticized this current trend of conversion agriculture in China (Yao *et al.,* 2018). A similar scene of shifting agriculture is also being observed in the sub-Himalayan plains (Jalpaiguri and Darjeeling district) where farmers have already converted a huge portion of their agricultural land to tea plantation aiming for long-term benefits (Das *et al.,* 2023). However, in this rush, a lack of proper maintenance has been reported to be present by Das *et al.* (2023). Poor soil management, ignorance in quality maintenance of bush for yield, excessive use of inorganic fertilizers and chemical pesticides and herbicides as well as unsustainable practices are prevalent. This may lead to the depletion of tea garden ecosystem and cause further economic depression. Encouragingly, the scenario of Sittong was found to be different and can be proposed as an ideal model to be emulated by small tea growers and future entrepreneurs.

Soil physicochemical properties

Moisture content. This test was performed to determine the soil absolute moisture or total water content in soil. A soil moisture content ranging between 20% and 60% is considered optimum for various crop growth and development (https://eos.com/blog/soil-moisture/). According to Mukherjee *et al.* (2020) and Rokhmah *et al.* (2022) tea plant growth is impeded when the soil moisture content falls below 25%, and if the water content decreases to less than 15%, it leads to the death of the plants. In our studied samples moisture content was found in a range between 16.25±1.76% to 40.97±0.8%. The moisture content analysis in the tea garden soils (refer to Figure

4-A) revealed adequacy of moisture in all except sample- BG1 whose moisture content was critically low (16.25±1.76%) because of high proportion of sand in its soil. However, the adequacy of moisture is due to several factors: the diligent land preparation methods employed (even without a drainage system); the effectiveness of the mulching process (which is highly regarded by organic tea farmers in Sittong); and the high organic matter content in the soil. These factors have enabled the tea farmers to overcome the challenges posed by the hilly terrain, inconsistent rainfall, various weather conditions, the sandy loam texture of the soil, and the absence of irrigation.

pH and electrical conductivity (EC) of soil. Tea Research Association (India) demonstrated that a soil pH of 4.5 to 5.5 is suitable for tea plant to grow well which is also recommended by the Tea Board of India (Mukherjee *et al.,* 2020; Sarkar *et al.,* 2022; Das *et al.,* 2023). For microbial function, the soil pH of 5.5is also most favourable. Among the tested soil samples, three samples (BG1, NB and RG2) resulted in a pH that matched the recommended level (pH 4.5-5.5). Eight soil samples collected from STGs i.e., SG2, DG, TTL, DKG, SG1, BG2 (pH 3.7±0.01- the lowest) and BG3 were very acidic, even below the least optimal value (pH <4.5) recommended by the Tea Board of India. A representative graph for the pH values of the tea garden soils is given in Figure 4-B. Limestone application can be suggested to increase the pH value (Bordoloi, 2021) which is applicable in organic farming as well (https://www.growveg.com/guides/soil-ph-for-organic-gardeners/). Various factors (soil moisture, nutrient, and other physicochemical parameters) influence soil pH. The acidity observed in the soil may be attributed to potential factors, such as an excessive application of compost. Organic matter is responsible for an increasing soil acidity where pH actually drops due to release of hydrogen ions which is linked to organic anions or nitrification process in the environment (Ritchie and Dolling, 1985). In a study, Nyawade *et al.* (2019) indicated that organic matter, supplying carbon, nitrogen, and various nutrients to the soil, increases the soil's organic acid content as soil-borne microbes utilize these nutrients, ultimately leading to a decrease in pH. Interestingly, high amount of soil organic carbon as obtained from further experiments correlated with acidic condition of the soil samples analyzed (Figure 5) as revealed in correlation study (Table 4). An *et al.* (2022) reported that soil pH value exhibits a strong negative correlation with and organic carbon that corresponds to this study.

The EC values of the tested soil samples are represented in Figure 4-C. Electrical conductivity (EC) measures the ability of water to carry electrical

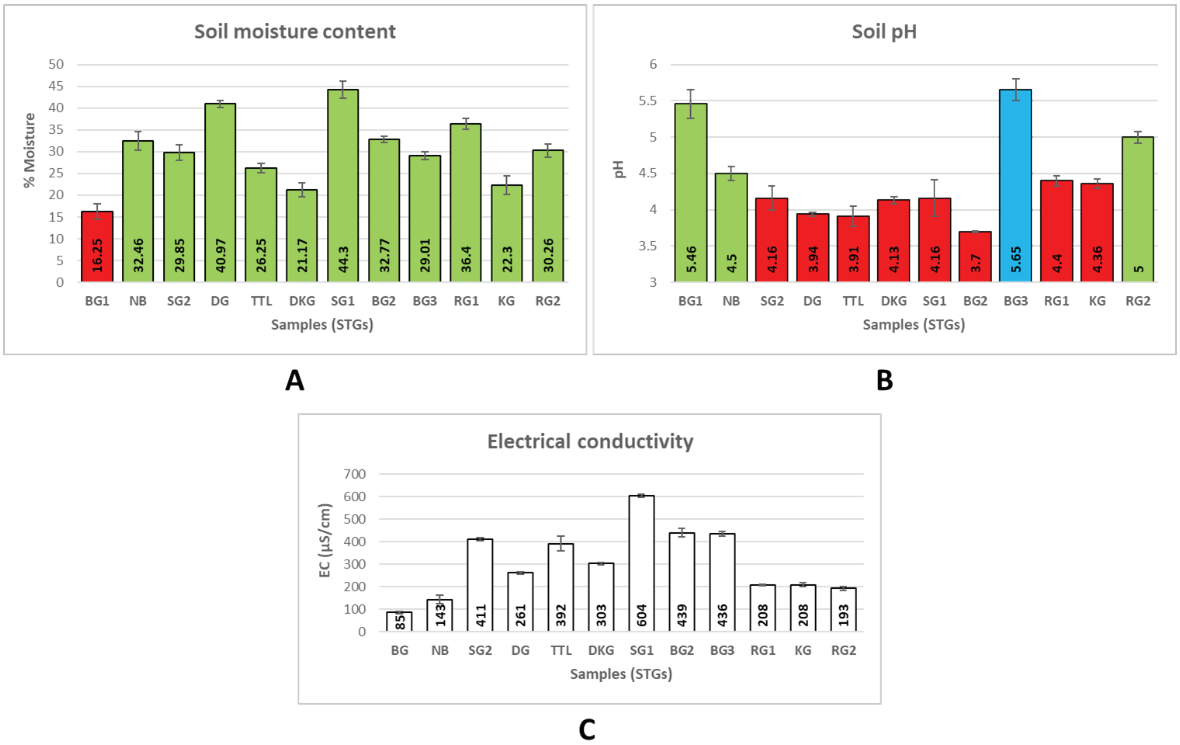

Figure 4. Soil moisture content (A), pH of soil (B) and EC of soil (C) collected from twelve organic small tea gardens of Sittong [Colour codes: Red= Low; Green= Adequate; Blue= Abundant].

current. In agriculture, soil EC values correspond to soil salinity. A low soil EC might be associated with low availability of micronutrients, as a result crop loss might happen (Das *et al.*, 2023). The optimal EC value for plant growth is usually between 0.8-1.8 dS/m (800-1800 μS/cm). However, there is no optimal value or standard for this parameter, specifically for tea plantations in this region. Here, the EC of tested tea garden soils ranged from 85±4 to 604±6 μS/cm (non-saline to slightly-saline) which is quite low if other reported literatures are considered (Mukherjee *et al.* 2020; Das *et al.*, 2023). Recently, in a geostatistical analysis Sahu *et al.* (2023) demonstrated soil EC of tea growing regions of Darjeeling district and represented it through thematic layer diagrams. According to this study the soil EC of high-altitude regions of Darjeeling was comparatively lower than that of less elevated regions which corresponds to the results of this study.

Organic carbon, organic matter and available nitrogen. Among the non-mineral substances of soil, organic carbon is the most vital or the key constituent that maintains quality or fertility of agricultural soil. Organic matter sourced from plant and animal tissue derived products is the source of organic carbon. According to the Tea Board of India, 2-3% of the organic carbon content is considered as suitable in tea garden soil (Mukherjee *et al.,* 2020). And the value below 1% is regarded as unhealthy that requires proper maintenance (Mukherjee *et al.,* 2020; Sarkar *et al.,* 2022; Das *et al.,* 2023). The results of organic carbon and organic matter quantification assay were satisfactory and as represented in Figure 5. The average organic carbon value of tested tea plantation soils was found to be very high (4.37±1.11%) including the highest one recorded for sample SG1 (7.11±0.01%). Even the lowest recorded value, which was 2.87±0.21 for the sample NB, still remained within the optimum range. Deliberately practicing organic farming, itself is the reason behind high organic carbon content in their soil because the fertilizing program entirely depends on application organic manures (cow dung, vermicompost, green manure, FYM etc.). Moreover, results suggest that farmers have cultivated and rehabilitated the land properly by carrying out deep ploughing and mulching before planting and it was also reflected in the survey report. Therefore, the cultural practice has been responsible for this high organic nutrient availability in soil.

According to Chan, 2008, organic matters are very useful elements for maintaining soil fertility and health which actually signifies a cumulative presence of total organic carbon and available nitrogen. Organic matter typically constitutes only 2% to 10% of the total soil mass and our samples also satisfied the optimum range was recorded between 4.94±0.35% (NB) to 12.22±0.02% (SG1) with a mean value of 7.5±1.92%. This result was due to following mulching process regularly and the use of cover crops, green manure, and perennial forage crops.

Nitrogen is the second most important macronutrient in agricultural soil (Ghosh *et al.,* 2022). The results of available nitrogen content has been represented in Figure 5-C. The nitrogen content in the tea gardens of Sittong ranged from 0.25±0.02 to 0.611±0.001% which is suitable if the optimum range (0.1–2.0%) recommended by the Tea Board of India is considered (Mukherjee *et al.,* 2020). In tea gardens of sub-Himalayan plains, leguminous trees are planted as shade trees which add a huge amount of nitrogen in the topsoil by fixing nitrogen with the help of *Rhizobium* bacteria residing in their nodules and by shedding leaves and pods in the winter. But in hilly terrain of Darjeeling shade trees aren't used at all. So, the exposure to organic matter (as described above) solely plays the role here.

Overall, an interrelationship has been observed between the survey results and soil testing results. The top three nutrient rich samples (high organic carbon, organic matter and available nitrogen) were collected from those small tea gardens which were new plantations compared to others i.e., SG1 (5-6 years), DG (4-5 years) and RG2 (3-4 years). Researchers found positive relation between microbial activity and soil organic carbon which might have important consequences. Specifically, soils enriched with organic matter, organic carbon and nitrogen usually improves microbial growth and activity to promote nutrient availability and development of soil structure (Liddle *et al.,* 2020; An *et al.,* 2022). In collected organic small tea garden soil, organic matter, organic carbon and available nitrogen content were very high and pH was substantially low (in accordance with Tea Board of India's recommended standard) which can be ideal for microbial activity. Further microbial biomass analysis (Figure 6) and correlation analysis (Table 4) revealed the same as described below. Moreover, isolation of microbes, biomass determination and their tolerance level against toxic chemicals in collected small tea garden soil will help to produce an overview of soil quality which has not been studied earlier. Therefore, to fulfill these lacunae some microbial experiments have been carried out as described below.

Soil microbial properties

Soil is a vast and major ecosystem that sustains the growth of various life forms and makes complex interactions for holistic growth of all living microorganisms (Curtis and Sloan, 2005). A huge diversity of microbes belongs to soil and many researchers proposed that microbes maintain soil health and increase soil fertility by participating

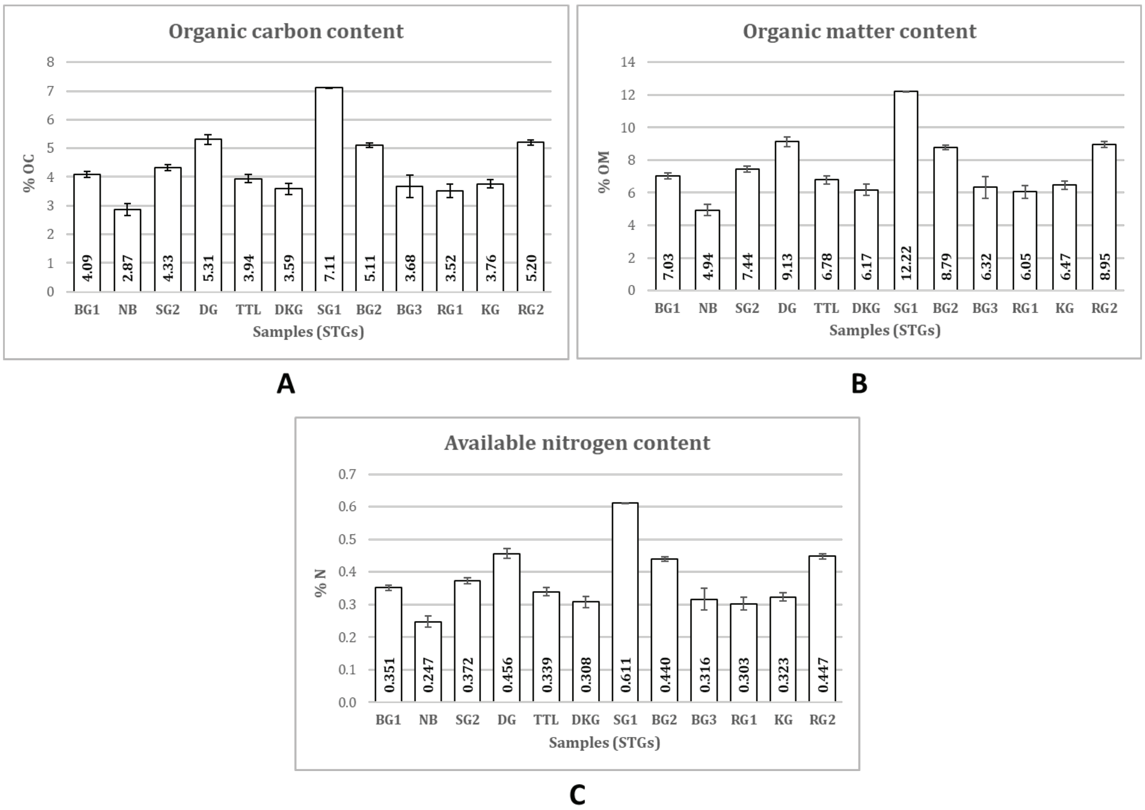

Figure 5. Organic carbon (A), organic matter (B) and available nitrogen (C) content of soil collected from twelve STGs of Sittong.

in the nutrient cycle and decomposing organic materials. The rhizospheric topsoil is upper layer of soil surrounding the plant root which is considered as the most suitable region for microbial growth. Soil health relies significantly on living organisms, their biodiversity, food web structure, ecosystem functions, and the intricate connections of soil microorganisms with plants, animals, and their own interrelationships, whether cooperative or antagonistic. Soil microorganism population or biomass is an important factor in agriculture as it maintains various nutrient cycles and sequestration of soil carbon, specifically in the current scenario of climate change. Soil microorganisms decompose organic matter by degradation of organic particles to available form.

Microbial cell dry weight determination assay. In tea garden soil, microbial consortium biomass refers to the total mass of these diverse microbes present in the soil including bacteria, fungi and other microorganisms that perform critical functions in the soil ecosystem. These microorganisms play important roles in soil nutrient cycling, decomposition of organic matter, and the suppression of plant pathogens (Yadav *et al.*, 2021). Microbial content is high in soil with high organic carbon. Tan *et al.* (2019) during research on effects of organic inputs, reported that organic tea garden soil shows high level of microbial population due to their high organic matter content. Result of microbial cell dry weight determination test (Figure 6) revealed the highest result in sample SG1(92.63±6.85 mg) followed by DG (69.47±1.72) and comparatively less values were recorded for the sample NB followed by RG1, KG and BG3 (Figure 6). High microbial biomass in SG1 and DG corresponded to high organic carbon content in those small tea garden soils. Further, positive correlation was also found with moisture as well as EC, while negative correlation was found with pH (Table 4).

Antibiotic and antifungal susceptibility pattern. Use of antibiotics and antifungals in agricultural fields have also gained momentum to get rid of harmful microbial invasions. But improper knowledge of dosage can often lead to a disastrous end. Exploitation of antibiotics and antifungals may lead to the eradication of the useful bacterial population that inhabits the soil and affect the nutrient uptake processes by the plants. Also there lies the risk of developing antibiotic tolerance by the bacterial population. Nevertheless, in organic farming, the use of antibiotics or antifungals is strictly prohibited to preserve a soil environment that is conducive to beneficial microorganisms. Because, in organic farming microorganisms serve as unseen

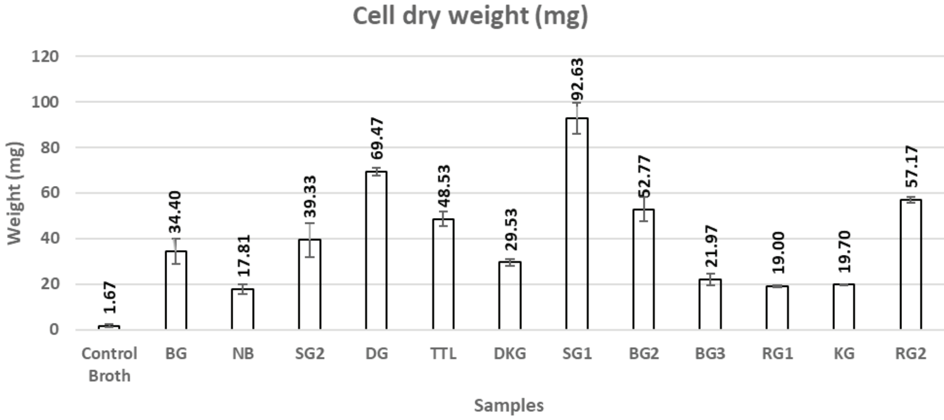

Figure 6. Results of microbial biomass through cell dry weight determination assay.

heroes in nutrient cycling. They transform soil's raw nutrients into available forms, allowing cultivated plants to uptake them, as no inorganic soluble nutrients are used in organic farming. Therefore, this experiment was conducted to assess the performance of the isolated consortia against prevalent antibiotics and antifungals, aiming to cross-verify the practices of organic farming.

Antibiotic susceptibility assay was performed to determine the potentials of consortia against antibiotics. Level of tolerance was determined by measuring the diameter formed around the antibiotics containing discs. According to the data provided in Table 2, all samples tested against twelve antibiotic shows variable zone of inhibition. Where, most susceptible pattern was observed against ofloxacin (average inhibition zone of 27.8 mm) followed by chloramphenicol, co-trimoxazole, chloramphenicol and neomycin and least susceptible nature was recorded against augmentin (average zone of inhibition = 8.4 mm), penicillin-G and cefotaxime (Table 2). Results of the antibiotic susceptibility assay have been expressed in Table 2 using a heatmap and representative photographs of the experimental plates are shown in Figure 7.

Fungal invasions are also a great threat to plants. Thus, needless to say, use of antifungals in agriculture is an inevitable solution to get rid of fungus. With application of antifungals, there comes a possibility of developing antifungal tolerance by the fungal populations. But, in organic farming practice

Table 2

Results of antibiotic susceptibility assay (inhibition zone in millimetres) of different sample tested in different antibiotics (Red-white colour-scale has been used to express high to low results as heatmap)

Antibiotics	Sample (STG)											
	BG	NB	SG2	DG	TTL	DKG	SG1	BG2	BG3	RG1	KG	RG2
Polymyxin B	12	11	11	14	19	12	12	14	15	10	14	19
Gentamicin	18	23	19	22	17	25	9	18	17	12	13	19
Neomycin	14	26	23	20	21	17	22	22	19	26	21	20
Bacitracin	17	9	11	13	16	9	10	18	17	11	11	12
Chloramphenicol	15	21	25	26	24	20	24	24	20	20	20	21
Penicillin-G	9	10	14	12	13	10	9	10	8	9	8	9
Augmentin	7	7	8	10	9	10	8	8	9	9	9	7
Erythromycin	14	18	19	19	12	18	7	7	18	14	11	14
Chloramphenicol	26	29	27	27	21	24	23	25	24	28	28	24
Ofloxacin	26	33	30	26	27	26	27	25	30	31	22	31
Cefotaxime	9	7	9	11	12	9	10	11	11	11	10	9
Co-Trimoxazole	23	25	24	26	26	24	21	23	24	25	25	19

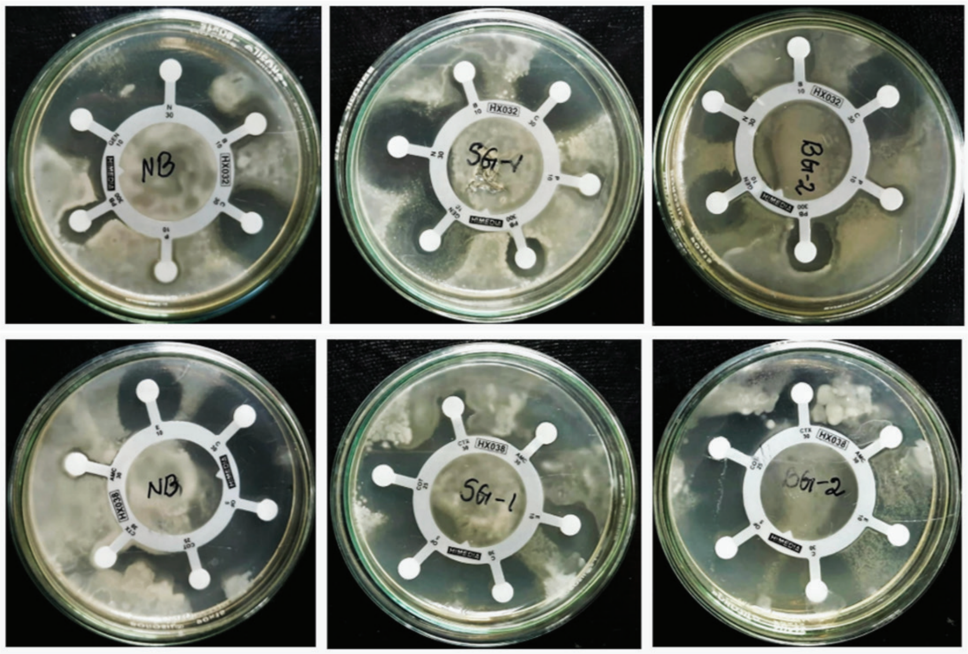

Figure 7. Images of selected sample plates showing inhibition zones in antibiotic susceptibility assay.

application of antifungal is not encouraged. In the result of antifungal susceptibility test, consortia obtained from all samples showed inhibition zone against all the tested antifungals. In comparison amongst the six antifungals itraconazole was most effective showing an average inhibition zone of 30 mm while the susceptibility was recorded against clotrimazole (with mean inhibition zone of 7.5 mm). Results of the antifungal susceptibility assay have been expressed in Table 3 using a heatmap as comparative approach and representative photographs of the experimental plates are shown in Figure 8.

Hence, from the results showing microbes' susceptible nature towards antibiotics and antifungals, it can be assumed that the soil of STGs of Sittong-3 is suitable for organic tea plantation where organic farming has been practiced deliberately and ethically as cross-verified through survey and the cumulative results obtained and analyzed in research.

Correlation study

Six positive correlations were found in the Pearson correlation matrix (Table 4) among ten different combinations. Soil moisture (SM), EC, OC, MCDW were in positive correlation with each other. Among all combinations, the strongest correlation was found between OC (organic carbon) and MCDW (microbial cell dry weight) (95% similarity). Moreover, all negative correlation was found in combinations associated to pH. Earlier, An *et al.* (2022) also stated that soil microbial biomass had a direct positive correlation with the soil organic carbon, total nitrogen content, phosphorus content etc., except for pH.

Table 3

Results of antifungal susceptibility assay (inhibition zone in millimetres) of different sample tested in different antifungals (Red-white colour-scale has been used to express high to low results as heatmap)

Antifungals	**Sample (STG)**											
	BG	**NB**	**SG2**	**DG**	**TTL**	**DKG**	**SG1**	**BG2**	**BG3**	**RG1**	**KG**	**RG2**
Fluconazole	8	7	8	8	7	9	8	7	8	7	8	7
Clotrimazole	8	7	8	7	7	9	7	7	7	8	7	8
Amphotericin	7	7	8	8	8	9	8	7	8	7	9	9
Nystatin	18	12	7	10	14	18	10	16	17	16	15	10
Ketoconazole	8	8	8	7	8	10	7	7	8	8	7	9
Itraconazole	29	34	27	28	34	32	25	35	32	27	26	31

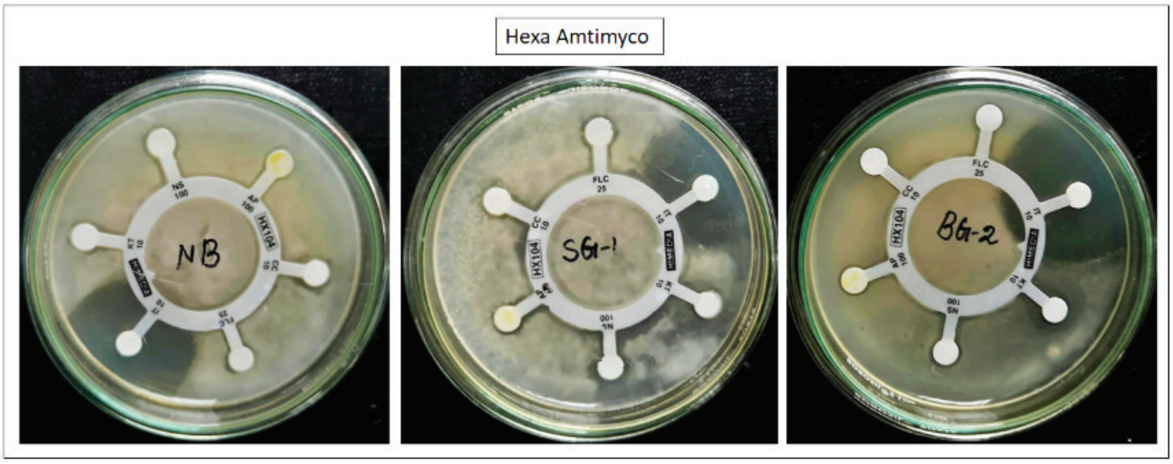

Figure 8. Images of selected sample plates showing inhibition zones in antifungal susceptibility assay.

Table 4

The Pearson correlation matrix from experimental data of six parameters such as Soil moisture% (SM), pH value, electrical conductivity (EC), organic carbon% (OC) and microbial cell dry weight (MCDW) (Red-white colour-scale has been used to express high to low results as heatmap)

	SM	pH	EC	OC	MCDW
SM	1				
pH	-0.39004	1			
EC	0.477857	-0.355	1		
OC	0.594418	-0.256	0.579507	1	
MCDW	0.596795	-0.362	0.552686	0.951751	1

Conclusion

The survey data revealed that the STGs of Sittong-3 own organic tea gardens, and this was also validated by results of different experiments done in this research. The survey of small tea growers revealed that they are often based in remote areas and practice the organic tea planting method. A high level of organic matter, organic carbon, nitrogen content and susceptible nature of soil microflora affirmed that STGs are practicing organic farming to cultivate tea. The soil physicochemical and microbiological characteristics of the small tea growers in Sittong-3 were found to be suitable for tea cultivation. The soil was acidic with high organic carbon content which is suitable for soil microbes to grow and show their activities. The high organic carbon content and diverse microbial community are likely to contribute to the quality of the tea produced in this region. The findings of this study suggest that Sittong-3 can be an ideal model to be followed for the upliftment of small tea growers that too in an ecofriendly way. This information could be used to develop more efficient and sustainable methods for tea cultivation. However, future in-depth research works need to be carried out to gather knowledge regarding consequences of organic farming practice where focus should be on the tea plants; harvested leaf; Sittong's handmade tea; manufactured tea from both bought leaf factories and their own small-scale processing units; and last but not least the economy. This is a promising trend as organic tea is becoming increasingly popular due to its health benefits. Overall, the findings of this study suggest that the small tea growers in Sittong-3 are well-positioned to produce a high-quality of organic tea. This is good news for the tea industry as well as for the local economy.

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