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Preface

We would like to present, with great pleasure, the inaugural volume-9, Issue-4, April 2023, of a scholarly journal, *International Journal of Environmental & Agriculture Research*. This journal is part of the AD Publications series *in the field of Environmental & Agriculture Research Development*, and is devoted to the gamut of Environmental & Agriculture issues, from theoretical aspects to application-dependent studies and the validation of emerging technologies.

This journal was envisioned and founded to represent the growing needs of Environmental & Agriculture as an emerging and increasingly vital field, now widely recognized as an integral part of scientific and technical investigations. Its mission is to become a voice of the Environmental & Agriculture community, addressing researchers and practitioners in below areas.

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Environmental science and regulation, Ecotoxicology, Environmental health issues, Atmosphere and climate, Terrestrial ecosystems, Aquatic ecosystems, Energy and environment, Marine research, Biodiversity, Pharmaceuticals in the environment, Genetically modified organisms, Biotechnology, Risk assessment, Environment society, Agricultural engineering, Animal science, Agronomy, including plant science, theoretical production ecology, horticulture, plant, breeding, plant fertilization, soil science and all field related to Environmental Research.

Agriculture Research:

Agriculture, Biological engineering, including genetic engineering, microbiology, Environmental impacts of agriculture, forestry, Food science, Husbandry, Irrigation and water management, Land use, Waste management and all fields related to Agriculture.

Each article in this issue provides an example of a concrete industrial application or a case study of the presented methodology to amplify the impact of the contribution. We are very thankful to everybody within that community who supported the idea of creating a new Research with *IJOEAR*. We are certain that this issue will be followed by many others, reporting new developments in the Environment and Agriculture Research Science field. This issue would not have been possible without the great support of the Reviewer, Editorial Board members and also with our Advisory Board Members, and we would like to express our sincere thanks to all of them. We would also like to express our gratitude to the editorial staff of AD Publications, who supported us at every stage of the project. It is our hope that this fine collection of articles will be a valuable resource for *IJOEAR* readers and will stimulate further research into the vibrant area of Environmental & Agriculture Research.



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Dr. Salvinder received MacKnight Foundation Fellowship for pre-doc training at WSU, USA – January 2000- March 2002 and DBT overseas Associateship for Post-Doc at WSU, USA – April, 2012 to October, 2012.

Dr. V K Joshi

Professor V.K.Joshi is M.Sc., Ph.D. (Microbiology) from Punjab Agricultural University, Ludhiana and Guru Nanak Dev University, Amritsar, respectively with more than 35 years experience in Fruit Fermentation Technology, Indigenous fermented foods, patulin ,biocolour ,Quality Control and Waste Utilization. Presently, heading the dept. of Food Science and Technology in University of Horticulture and Forestry, Nauni-Solan (HP), India.

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Dr. Chiti Agarwal

Dr. Chiti Agarwal works as a postdoctoral associate at the University of Maryland in College Park, Maryland, USA. Her research focuses on fungicide resistance to fungal diseases that affect small fruits such as strawberries. She graduated from North Dakota State University in Fargo, North Dakota, with a B.S. in biotechnology and an M.S. in plant sciences. Dr. Agarwal completed her doctorate in Plant Pathology while working as a research and teaching assistant. During her time as a graduate research assistant, she learned about plant breeding, molecular genetics, quantitative trait locus mapping, genome-wide association analysis, and marker-assisted selection. She wants to engage with researchers from many fields and have a beneficial impact on a larger audience.

DR. Owais Yousuf

Presently working as Assistant professor in the Department of Bioengineering, Integral University-Lucknow, Uttar Pradesh, India.

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Presently working as Assistant Professor in Dept. of Plant Pathology, College of Agriculture polytechnic, NAU, Waghai.

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Mr. Jiban Shrestha

Scientist (Plant Breeding & Genetics)

Presently working as Scientist (Plant Breeding and Genetics) at National Maize Research Programme (NMRP), Rampur, Chitwan under Nepal Agricultural Research Council (NARC), Singhdarbar Plaza, Kathmandu, Nepal.

Mr. Aklilu Bajigo Madalcho

Working at Jigjiga University, Ethiopia, as lecturer and researcher at the College of Dry land Agriculture, department of Natural Resources Management.

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MPhil. in Entomology, from University of Ghana.











He has extensive knowledge in tree fruit orchard pest management to evaluate insecticides and other control strategies such as use of pheromone traps and biological control to manage insect pests of horticultural crops. He has knowledge in agronomy, plant pathology and other areas in Agriculture which I can use to support any research from production to marketing.







Mr. Bimal Bahadur Kunwar

He received his Master Degree in Botany from Central Department of Botany, T.U., Kirtipur, Nepal. Currently working as consultant to prepare CCA-DRR Plan for Hariyo Ban Program/CARE in Nepal/GONESA.

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Ultrastructural Changes of Secondary Phloem Cells in the Cambium Annual Activities of *Taxodium Ascendens*

Youming Xu^{1*}, Huiling Wang², Jieyun Tao³, Han Lin⁴

College of Horticulture and Forestry Science, Huazhong Agricultural University, Wuhan 430070

*Corresponding Author

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Abstract— Tree bark is an important part of tree trunk and it derives from the activities of cambium. More studies were focused on the xylem cells development of trees at the ultrastructural level and not on phloem cells. To explore the ultrastructural changes of all kinds of secondary phloem cells in the cambium annual activities of *Taxodium ascendens*, and to study the mechanism of its wood formation. The samples of *Taxodium ascendens* were collected in different growth periods. The ultrastructural changes of all kinds of cells in the secondary phloem of *Taxodium ascendens* and the process of wood formation were observed by transmission electron microscope. The Results show as follows:

The sieve cells began to differentiate in late March and their cell walls thickened continuously during the periods of April to June. At the end of August, part of the sieve cells entered programmed death, with nucleus pyknosis, chromatin agglomerated, nucleoli disappeared, and the nucleoplasm decreased. At the same time, the size of the nucleus became smaller, and when the sieve cells stopped functioning, the protoplasts died. The whole cell was squeezed and deformed. Phloem parenchyma cells began to differentiate in mid-April. In the early stage of cambium activity (from mid-April to early May), the resin in the parenchyma cells was distributed in the form of large droplets. At the peak of cambium activity (from the end of May to the end of June), the resin was dispersed in the cells as fog, and the resin droplets were evenly distributed along the cell wall during the dormant period. In mid-April, newly differentiated young ray parenchyma cells were observed in the phloem near the cambium. From mid-late April to early June, the young phloem ray cells developed to mature and remained in this state until the middle of November. After that, the cells went into programmed death, and the cells contained only a large number of oil droplets and empty vesicles. The development and maturation of phloem fibers occur in all periods. Phloem fibers can be watched in any period of phloem development. The proportion of mature and developing phloem fibers differs slightly in each period.

The active period of cambium cells lasted from the beginning of March to the end of November. Newly differentiated sieve cells could be observed in March, but newly differentiated phloem parenchyma cells and ray parenchyma cells were not observed until the middle of April. Therefore, the sieve cells differentiated earliest, while the phloem parenchyma cells and ray parenchyma cells were about half a month later than the sieve cells. During the period from the beginning of phloem differentiation to the end of cambium activity, the division and differentiation of secondary phloem cells can be observed continuously.

Keywords— *Taxodium ascendens*, Secondary phloem, Sieve cell, Parenchyma cell, Phloem fiber, Ultrastructure.

I. INTRODUCTION

The secondary phloem derived from the vascular cambium of trees is responsible for the synthesis and storage of organic matter and the transport and distribution of photosynthates (Esau K, 1969). At present, there have been a large number of studies on the phloem development of hardwood phloem, with a comprehensive summary of all kinds of cells in the phloem of hardwood (Derry *et al.* 1967; Zhao 2012), while most of the studies on softwood are focused on xylem (Rossi *et al.* 2016; Pattathil *et al.* 2016; Zhang *et al.* 2018; Zheng *et al.* 2022) and cambium (Rossi *et al.* 2006; Prislán *et al.* 2016; Myśkow *et al.* 2019), there are few systematic reports on the structure and development of all kinds of cells in the phloem of softwood. The phloem of softwood is usually composed of axial sieve cells, phloem fibers, phloem parenchyma cells, and radial phloem ray parenchyma cells (Abbe *et al.* 1939). In different developmental stages, the morphology, contents, and composition of all kinds

of cells are different (Gričar *et al.* 2017). For more than half a century, scholars have carried out a large number of phloem development studies on various tree species and combined them with climatic factors to explore (Gričar *et al.* 2007; Luis *et al.* 2007; 2011; Dié *et al.* 2012; Salmon *et al.* 2019). However, most studies focus on the seasonal growth pattern and growth of phloem (Cardoso *et al.* 2019; Kopanina *et al.* 2022; Ohse *et al.* 2022). The studies on the seasonal changes of various cell morphology of softwood phloem are mainly focused on the microscopic layer (Mullendore 2010; Prislán *et al.* 2013; Baba *et al.* 2019). At present, the ultrastructural layer studies on phloem are mainly focused on sieve cells (Knoblauch *et al.* 2018; Prislán *et al.* 2018), the study of other phloem cells is not reported comprehensively. The resolution of modern electron microscopy has gone deep into the most basic structural units of wood cells (Singh *et al.* 2001; Fromm *et al.* 2003; Chukhchin *et al.* 2020). Using electronic microscope to explore the process of wood formation will be the research trend in the future.

Taxodium ascendens is a successful fast-growing timber tree species introduced and cultivated in the Yangtze River in China. In this paper, the ultrastructural changes of sieve cells, phloem parenchyma cells, phloem ray parenchyma cells, and phloem fibers were observed by transmission electron microscope, to obtain more information about the seasonal activity of cambium and secondary phloem changes in gymnosperms. The purpose of this paper is to enrich the biological knowledge of the growth and development of coniferous wood, combine the formation of phloem and xylem, and connect with the overall growth of trees. This not only helps to consolidate the theory of cambium development but also helps to provide a basis for practical problems such as forest management measures and regulation of wood and partial bark growth. It also provides a scientific basis for the cultivation of *Taxodium ascendens* plantations, wood material improvement, and forest resource utilization, and contributes to the efficient utilization of bark resources.

II. MATERIAL AND METHODS

In this study, five normally grown trees were meticulously selected from the forest of *Taxodium ascendens* near South Lake in Wuhan, Hubei Province of China. Samples of 2-3-year-old branches were cut from the middle part of the crown of *Taxodium ascendens* trees, which were twenty years old and their heights are about 20-meter in tall.

Samples are taken every 3 days from March to June 2019, every 7 days from July to October 2019, and every two weeks from November 2019 to March 2020. When sampling, select the 2-3-year-old branches in the middle and upper part of the crown, which are about 3-4 m above the ground and grow well, and then cut the middle of them into several small sections of 0.5-1cm. Afterwards, fix them with glutaraldehyde and vacuum treatment to make them completely fixed, and pay attention to avoid the branches of reaction wood. Detailed records were maintained for each sampling site, including the longitude and latitude, altitude, sampling date, and weather conditions, to facilitate further analysis.

Samples were softened with 40% hydrofluoric acid (HF) to allow free cutting with double-sided blades. Then fixed at 4°C for 5-6 hours, rinsed with phosphate buffer solution (PBS) 3 times for 20 minutes each time, and then fixed for 4 hours. Then rinse with pH7.2 PBS 3 times, followed by gradient alcohol dehydration, propylene oxide replacement, Epon812 resin embedding, slicing with LKB-V ultramicrotome, lead citrate-uranium acetate staining, transmission electron microscope observation. The images obtained were collated and analyzed by Photoshop software, and the phloem ray morphological characteristics of sieve cells, phloem fibers, phloem parenchyma cells, and radial system of the axial system were described in detail, and the changes in the characteristics of secondary phloem during the branch activity were summarized.

III. RESULTS

3.1 Ultrastructural changes of phloem sieve cells in the annual activities of cambium

The sieve element in the phloem of gymnosperms is sieve cells, and they are of the same shape, a single narrow spindle-shaped cell, usually rectangular in cross-section and tapering at the end. Granular or crystalline inclusions dispersed in the cytoplasm were observed in the sieve element of *Taxodium ascendens*. In this experiment, the changes in ultrastructure and wall structure of sieve cells were observed by making ultrathin sections and transmission electron microscope.

During the period of cambium recovery activity on March 18, a small number of functional sieve cells that survived the previous year could be seen in the phloem; most of them were nonfunctional sieve cells, and high-density crystalline P plastids could be seen in the sieve cells (Figs. 1A, 1B). The newly differentiated young sieve cells were observed in the materials collected on March 26, indicating that the vascular cambium started activity and differentiated into the phloem. The nucleus of the young sieve cells was not obvious, and a large number of dispersed cytoplasm could be seen in the cells, the sieve cells showed vacuolization, and the nucleus began to be visible, but the size was small. Like the primordial cells that produced them, the protoplasts of immature functional sieve cells have visible nuclei, vacuoles, and other normal organelles (Fig. 1C). On

April 11, cambium entered the active phase and cytoplasmic content significantly increased. A small number of mitochondria can be seen along the cell wall (Fig. 1D). The obvious nucleus can be seen in the material on April 22 (Fig. 1E). At the end of May, S plastids were observed for the first time near the nucleus of the sieve cells (Fig. 1F). On June 5, the diameter of the sieve cell began to expand, the developing plastids could be observed in the cytoplasm, the number of plastids also increased, and the nucleus was darker. Long oval S plastids were observed, S plastids contained spherical osmiophilic bodies, and P plastids began to condense to a small extent. At this time, *Taxodium ascendens* was in the active stage, and the transfer and transport of assimilation products on the sieve cells had reached an active peak. The cell wall began to thicken (Figs. 1G, 1H). On July 10, it was observed that the black osmiophilic substance condensed into a sphere in some sieve elements (Fig. 1I).

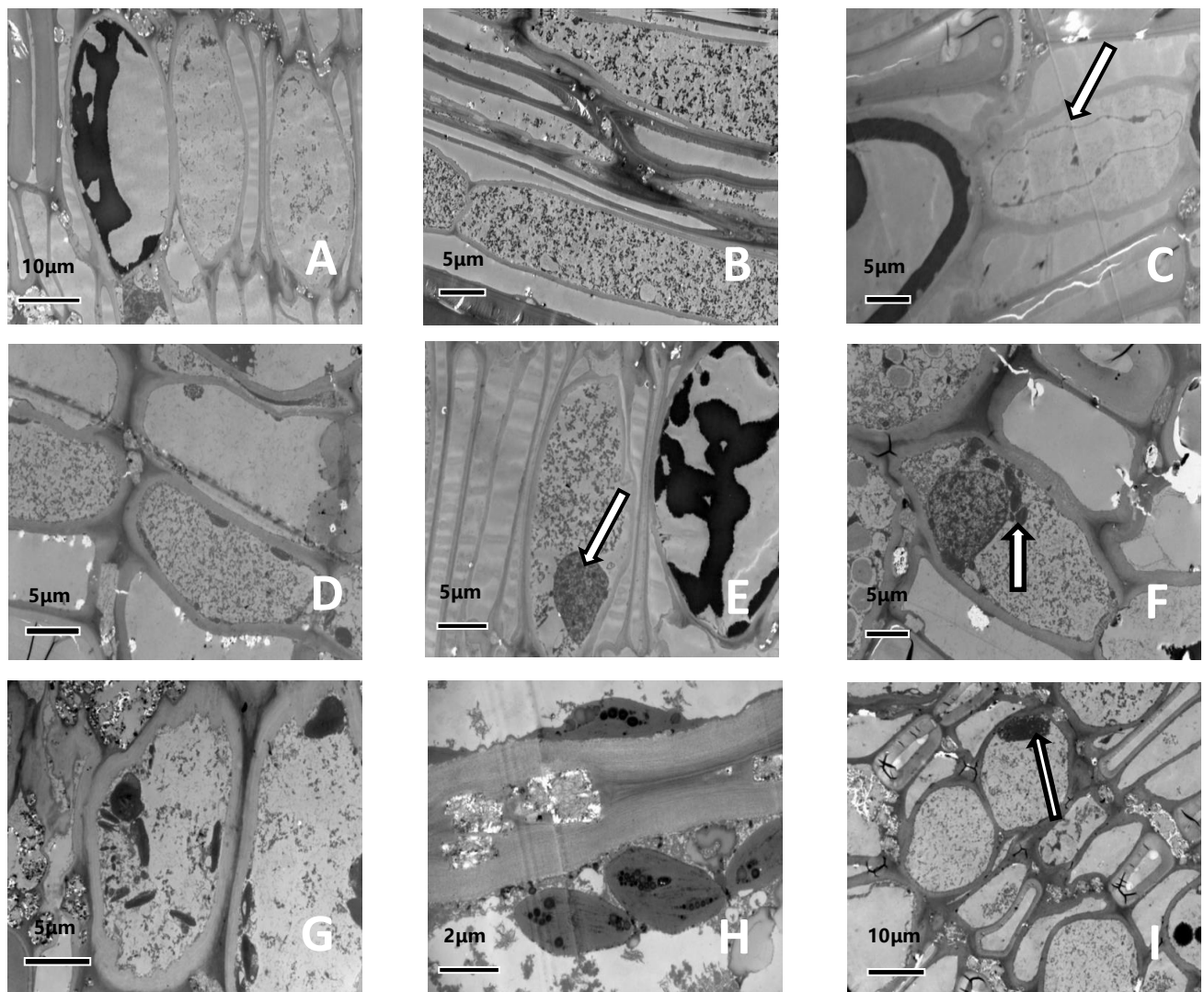


FIGURE 1: Ultrastructural changes of sieve cells during the recovery active and active period of cambium

A: Cross section of the material on March 18, a small number of functional sieve cells survived in the previous year, most of them are nonfunctional sieve cells. **B:** March 18 material diameter section. **C:** March 26 material, new division and differentiation of sieve cells. **D:** April 11 cross section, increased cytoplasmic concentration. **E:** April 22 material, mature nucleus developed in the sieve cell (shown by the arrow). **F:** S plastids appeared near the mature nucleus on May 31. **G:** Cross section of the material on June 5, a large number of S plastids can be seen. **H:** On the radial section of the material, S plastids are enlarged, and osmiophilic bodies can be seen in the cytoplasm. **I:** On the cross section of July 10, black osmiophilic substances are observed to condense into spheres in some sieve molecules (shown by the arrow).

In the late active phase (August 13), the chromatin and nucleoplasm disappeared to almost invisibility, and some of the sieve plastids collapsed and released osmiophilic substances (Fig. 2A). On August 27, it was observed that ribosomes, microtubules,

microfilaments, Golgi apparatus, and tonoplast disintegrated to disappear, with only one small pyknotic nucleus, obvious thickening of the cell wall, and obvious layered structure of the cell wall, it can be distinguished from the inner layer (S3), middle layer (S2), and outer layer (S3). A small amount of cytoplasm is distributed on the edge of the cell, which is mainly composed of the plasma membrane, a small number of mitochondria and sieve plastids (Fig. 2B). On October 22, *Taxodium ascendens* began to enter the dormant period, only a small amount of functional phloem could be seen in the phloem, and most of the sieve cells developed into dead cells with only transport and support function, that is, the nonfunctional sieve cells were obviously delaminated and the cell wall was thicker (Fig. 2C). On November 19, few functional sieve cells were seen. During this period, most of the nonfunctional sieve cells were compressed by phloem parenchyma cells and swollen stone cells, and the cambium was in a dormant phase (Fig. 2D). The secondary phloem still has a very small number of functional sieve cells to maintain the transport function until the following spring, ensuring the transport of assimilates in the overwintering stage.

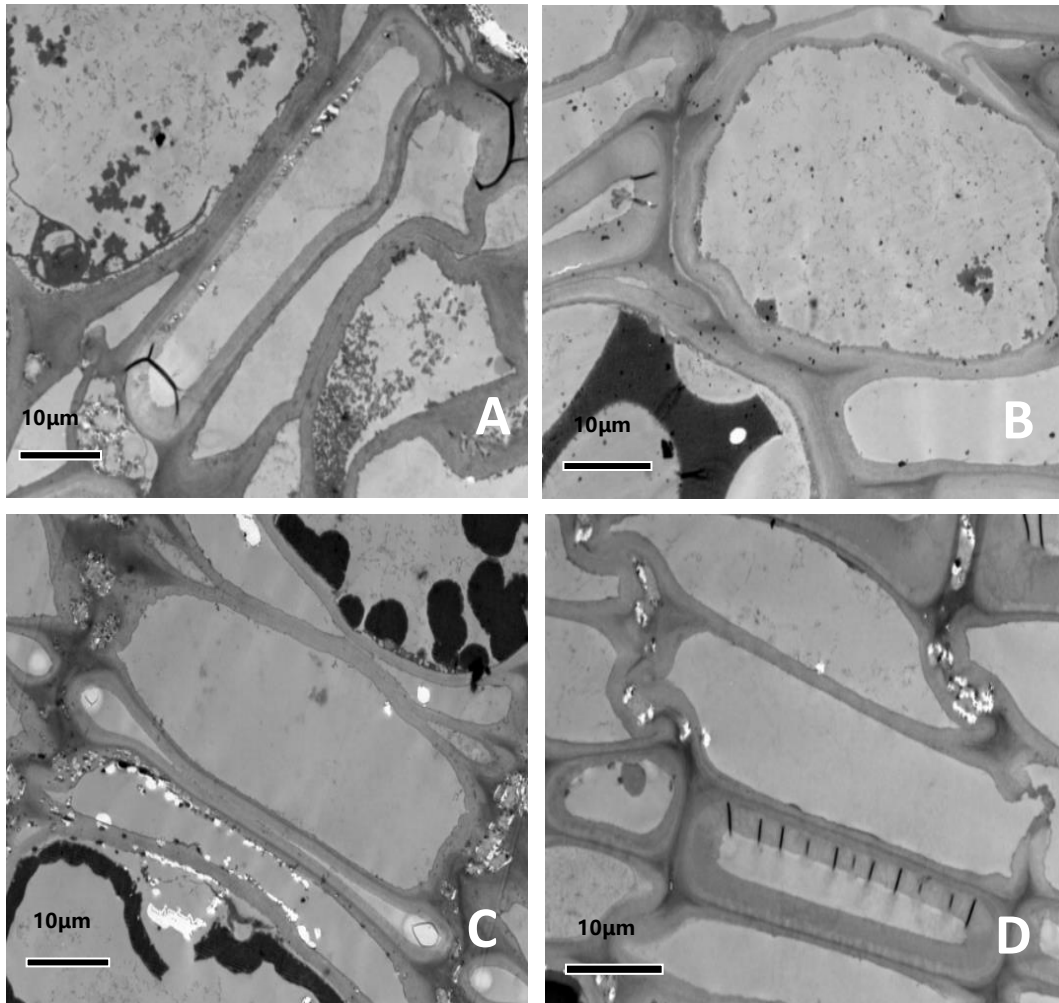


FIGURE 2: Ultrastructural changes of sieve cells during the late active and dormant period of cambium

A: In the cross section of the material on August 13, the chromatin and nucleoplasm disappeared to almost invisible, and some of the sieve intracellular plastids collapsed and release osmiophilic substances. **B:** On August 27, the ribosome, microtubule, microfilament, Golgi apparatus and tonoplast basically disintegrated to disappear, with only one small suspected pyknotic nucleus, obvious thickening of the cell wall and obvious layered structure of cell wall. **C:** In the cross section of the material on October 22, only a small amount of functional phloem can be seen in the phloem, and most of the sieve cells develop into dead cells with only dredging and supporting functions, that is nonfunctional sieve cells, with obvious delamination of nonfunctional cell wall and thicker cell wall. **D:** On November 19, most of the materials are nonfunctional sieve cells

3.2 Ultrastructural changes of phloem parenchyma cells in the annual activities of cambium

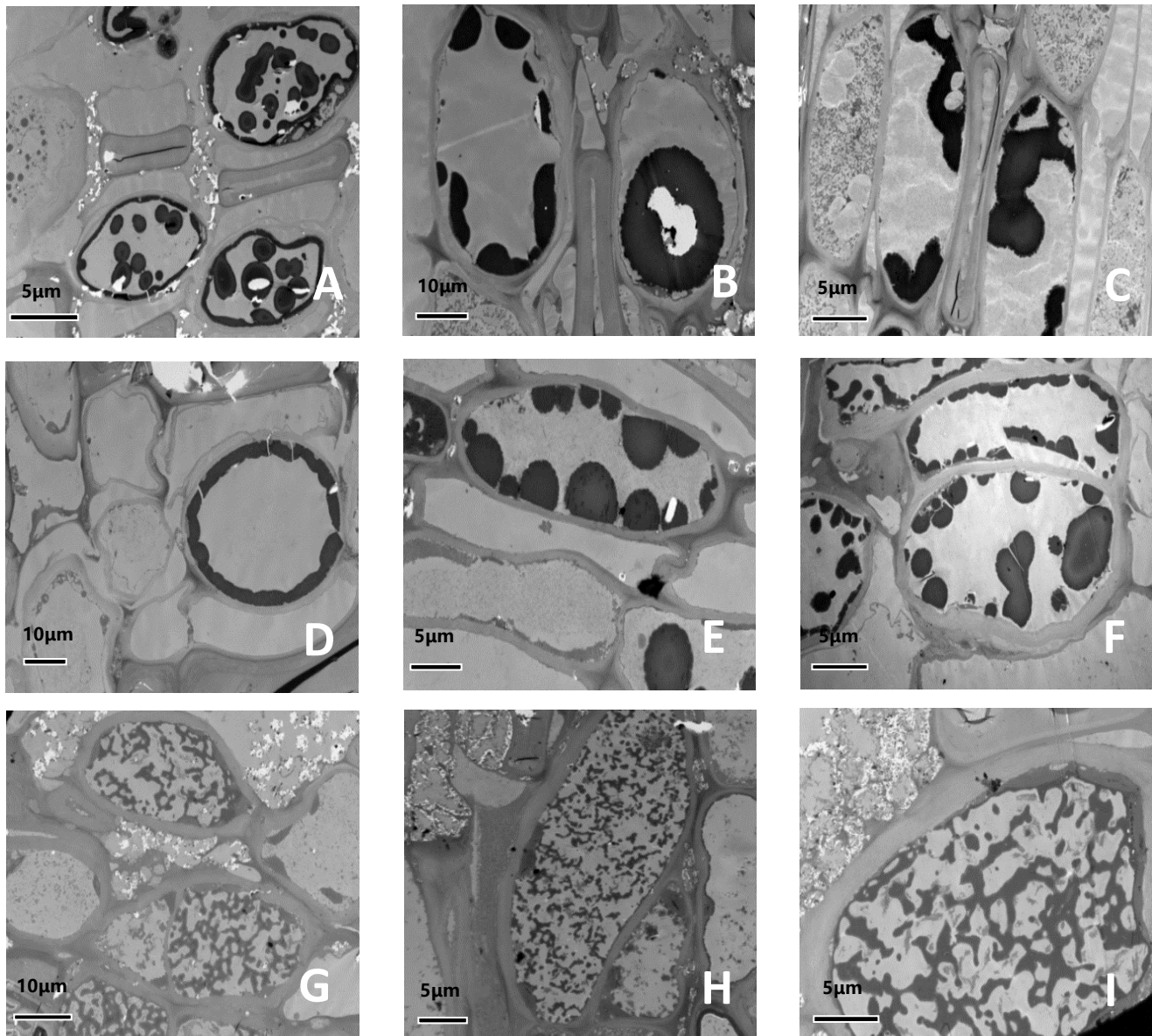


FIGURE 3: Ultrastructural changes of parenchyma cells during the recovery active and active period of cambium

A: On March 13 the material cross section, phloem parenchyma cells are in dormancy, there are small droplets inside the cells, and some resin droplets are homogeneous along the wall. **B:** Dark resin in phloem parenchyma cells in the cross section of material on 18 March appears droplet like. **C:** On March 26 the material was largely maintained. **D:** Cross section of the material on April 11, newly differentiated phloem parenchyma cells were observed, and the intracellular resin was evenly distributed along the cell wall in a candidate shape. **E:** Small droplets of resin on the inner wall of the cell reaggregate on April 22. **F:** 17 May material observed droplets dispersing intracellularly. **G:** Cross section of material May 31, showing dispersion in small droplets. **H:** Material cross section on June 1 with droplets almost misty highly dispersed. **I:** 26 June resin droplets condensed to a small extent

On March 13, the cambium of *Taxodium ascendens* was in the dormant period, and part of the resin distribution along the inner wall of the cell could be observed (Fig. 3A). On March 18, *Taxodium ascendens* entered the cambium recovery period, and the dark resin in the phloem parenchyma cells showed a droplet shape (Fig. 3B), and gradually dispersed to the cell wall in late March (Fig. 3C). On April 11, the newly differentiated phloem parenchyma cells were observed, the cell wall was very thin, and the intracellular resin was uniformly distributed along the cell wall (Fig. 3D). On April 22, the resin droplets on the inner wall of the cell gathered again and then gradually dispersed into the cell (Fig. 3E). The resin in the phloem parenchyma of *Taxodium ascendens* was highly dispersed from May to June, and the division and differentiation were more active (Figs. 3F,

3G). Almost foggy and uniformly distributed in the cell fluid was observed on June 5, but no resin droplets were observed (Fig. 3H). At this time, the cambium activity was in the active period, and the cambium differentiation rate reached the peak, and then the cambium differentiation rate decreased gradually. A small condensation of resin droplets was observed on the material on June 26 (Fig. 3I). On July 10, the resin droplets condensed into clusters (Figs. 4A, 4B, 4C). By September 11, the resin was in the shape of large droplets, accounting for most of the intracellular volume, and a small number of starch granules were observed (Fig. 4D), and the cambium activity entered the end of the active period. In early January, the resin was redispersed, and in mid-November, the resin was again dispersed as a droplet to the inner wall of the cell, and starch granules could be seen along the cell wall (Figs. 4E, 4F). After that, the resin in the phloem parenchyma cells remained in this state through the winter, until the following spring, when the cambium resumed activity and the resin droplets gathered again in the cell (Fig. 3A).

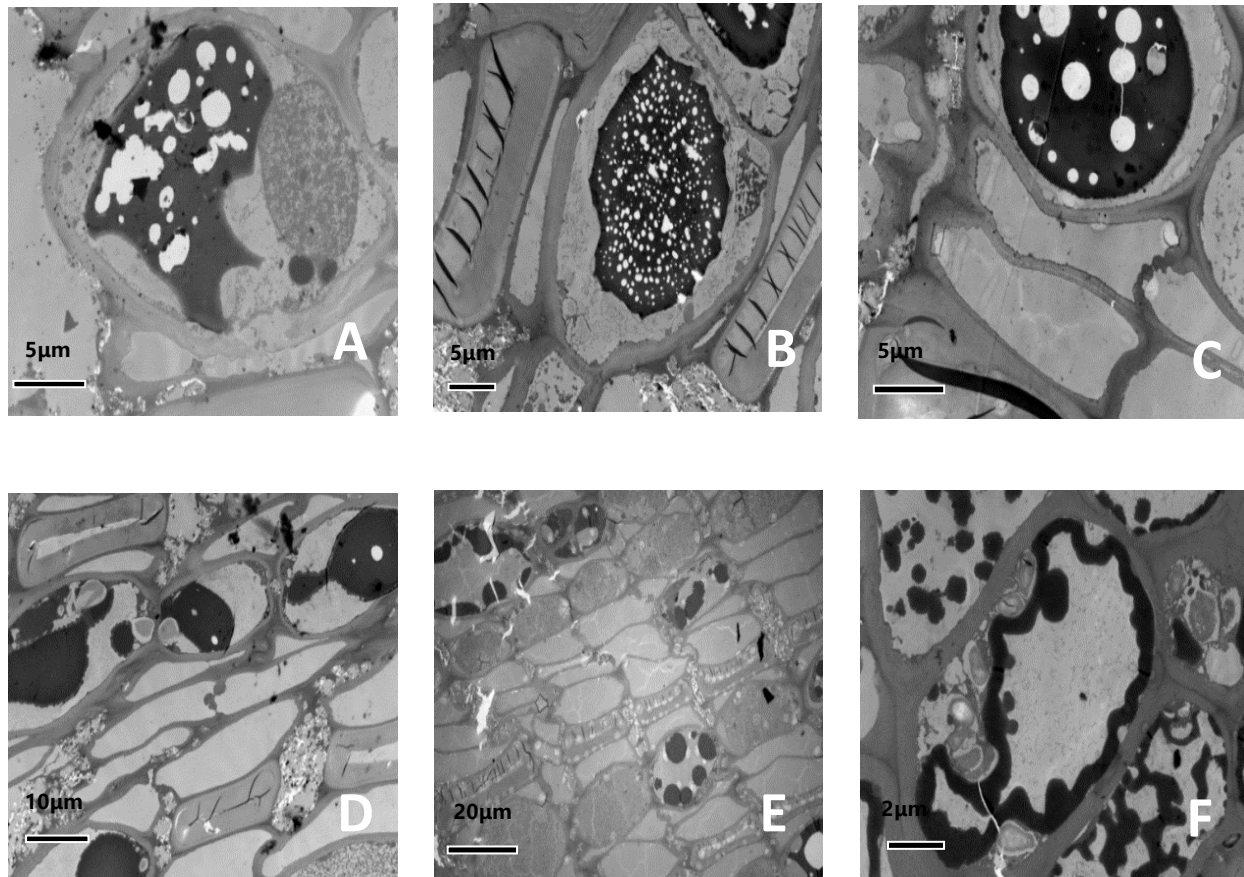


FIGURE 4: Ultrastructural changes of parenchyma cells during the late active and dormant period of cambium

A-C: In July and August materials, resin droplets condensed into agglomerates. D: September 11 materials, resin droplets are large droplets, and a small number of starch particles can be seen. D-F: From October 22 to November 19, the resin was dispersed again as a small drop to the inner wall of the cell, and the number of starch granules along the cell wall was significantly more than that of the material in mid-September

3.3 Ultrastructural changes of phloem ray parenchyma cells in the annual activities of cambium

On March 18, the cambium had not yet begun to differentiate into the phloem, and mature phloem ray parenchyma cells were observed in the phloem, with mature nuclei, a small number of multivesicular bodies and less cytoplasm, most of which were distributed along the inner wall of the cells, the cell wall is thicker (Fig. 5A). The young ray parenchyma cells were still not observed on March 26, the nucleus of mature phloem ray parenchyma cells was smaller than that on March 18, and the number of starch granules decreased. It is speculated that the cambium becomes active during this period, and nutrients are needed to meet the needs of cell division and differentiation (Fig. 5B). On April 11, newly differentiated young ray parenchyma cells were observed in the phloem near the cambium; the tangential cell wall was very thin, the cells showed high vacuolization, and vesicles could be seen near the inner wall of the cells. At the same time, the deposition of lignin in the cell wall was observed (Figs. 5C, 5D). On April 18, large nuclei were seen in the cells, nucleoli were clearly visible, multivesicular bodies appeared near the nuclei, and there were no inclusions in the vesicles (Fig. 5E). From early April to mid-May, most of the cell

cavity was occupied by vacuoles (Fig. 5F). On May 31, the number of cell inclusions increased, the large vacuoles had been degraded, and abundant small vacuoles could still be observed (Fig. 5G). By June 5, multivesicular bodies had become smaller, dark substances were deposited inside, and the cambium was active (Fig. 5H). Since then, ray parenchyma cells have maintained the state of larger nuclei and more multivesicular bodies (Fig. 5I). The situation lasted until mid-September.

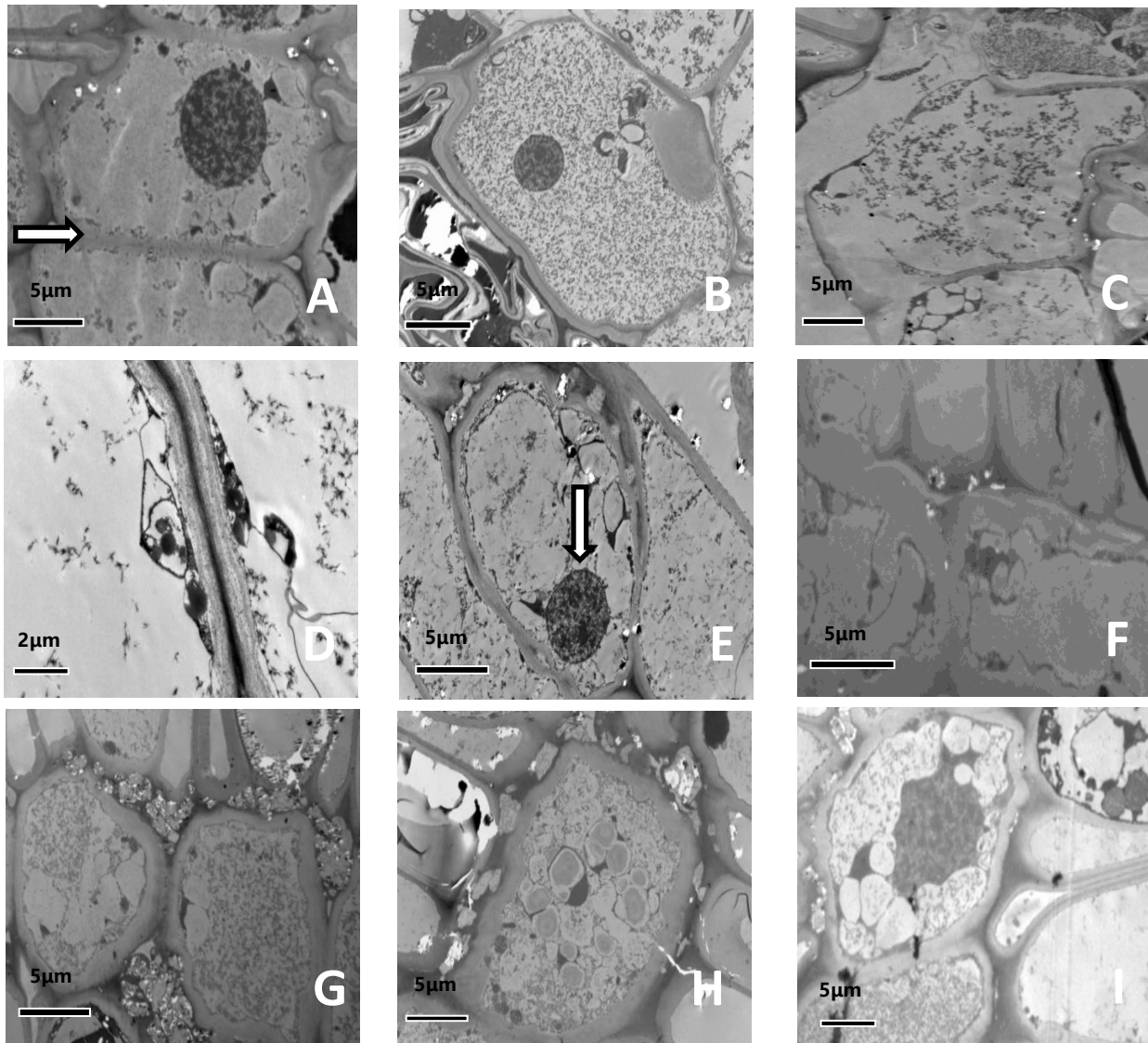


FIGURE 5: Ultrastructural changes of ray parenchyma cells during the recovery active and active period of cambium

A-I: Cross section. A: Mature phloem ray parenchyma cells were observed in the phloem on March 18, with mature nuclei, a small amount of multivesicular bodies, less cytoplasm, mostly distributed along the inner wall and thicker cell wall (shown by the arrow). **B:** The nuclear volume of mature phloem ray parenchyma cells on March 26 was smaller than that on March 18, and the number of starch granules decreased. **C:** Young ray parenchyma cells were observed on April 11, indicating that ray parenchyma cells began to differentiate, during which the cell wall was extremely thin and contained large vacuoles. **D:** On April 11, Golgi apparatus and vesicles were observed near the cell wall. **E:** On April 18, the nucleus of developing phloem ray parenchyma cells became significantly larger (shown by the arrow). **F:** On May 17, the cells were almost occupied by vacuoles. **G:** On May 31, the number of cell inclusions increased, the large vacuoles had been degraded, and abundant small vacuoles could still be observed, and the small vacuoles remained free. **H:** On June 5, the multivesicular bodies became smaller and dark substances were deposited inside. **I:** On July 10, the nucleus was clearly visible, occupying most of the cells

On September 11, it was observed that the fusiform ray primitive cells still maintained the state of division and differentiation, indicating that the vascular cambium was still active. At the same time, secondary lysosomes and residual undigested bodies after digestion were observed, representing the transformation of ray parenchyma cells from a mature state to an aging state (Figs. 6A, 6B), but cell division and differentiation continued. The division and differentiation of ray parenchyma cells could still be observed in mid-October (Fig. 6C). On November 19, it was observed that some of the chromatin condensed in the nucleus, and the nuclear DNA was degraded and broken, finally forming a dense apoptotic body surrounded by the membrane. The apoptotic cells maintained the integrity of the cell membrane; there was no cell inclusion overflow; and the cells were undergoing programmed death (Fig. 6D). On December 6, it was observed that the tonoplast ruptured and the apoptotic bodies in the nucleus overflowed and dispersed into the cytoplasm (Fig. 6E). At the beginning of January, most of the ray parenchyma cells in the dormant phase of cambium activity had become dead cells, and there were only a large number of oil droplets and empty vesicles in the cells (Fig. 6F).

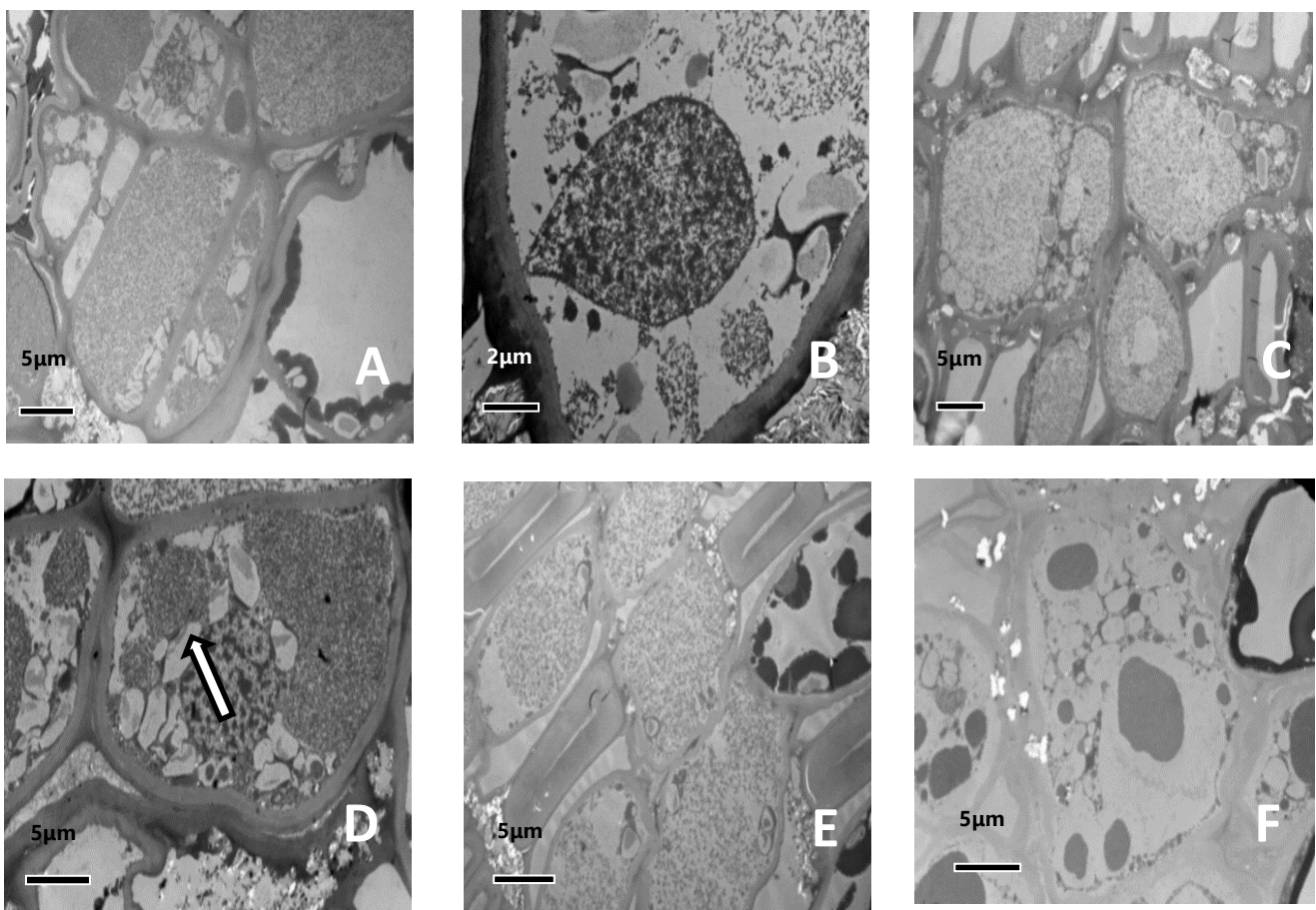


FIGURE 6: Ultrastructural changes of ray parenchyma cells during the late active and dormant period of cambium

A-F: Cross section. A: The fusiform ray primitive cells were still in the state of division and differentiation, indicating that the vascular cambium was still active. B: The enlarged version of the phloem ray cells of the material on September 11, during this period, the organelles were obviously visible, and lysosomes and residual corpuscles were observed. C: The division of phloem ray cells could still be seen on October 22. D: On November 19, the chromatin concentration in part of the ray nucleus (shown by the arrow) was observed, and the nuclear DNA was degraded and broken and finally formed dense apoptotic bodies surrounded by the membrane. E: On December 6, the tonoplast ruptured, the apoptotic bodies in the nucleus spilled and dispersed into the cytoplasm. F: The dead ray parenchyma cells were observed on January 6, which contained only a large number of oil droplets and empty vesicles

3.4 Ultrastructural changes of phloem fibers in the annual activities of cambium

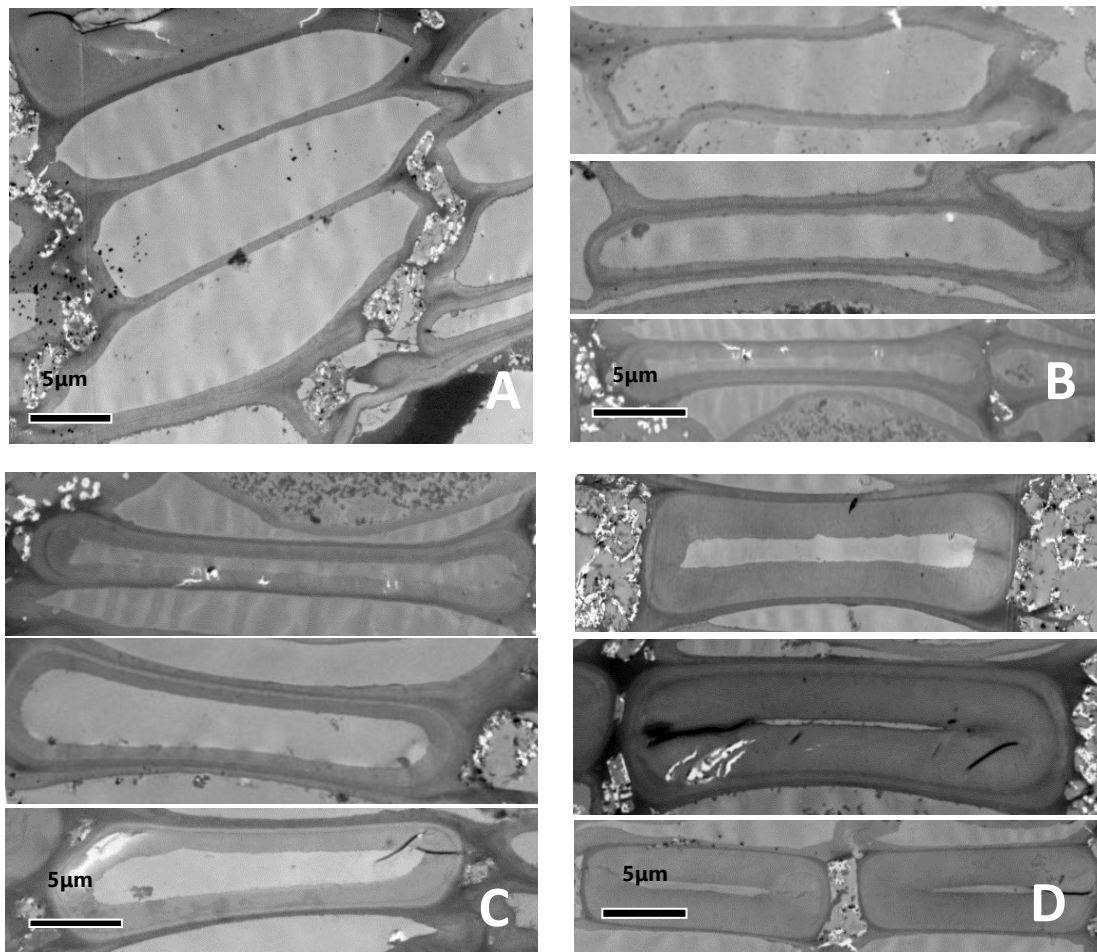


FIGURE 7: Ultrastructural changes of phloem fibers during different periods of cambium activity

A: Nonfunctional sieve cell with just programmed death, the cell has no inclusion and is spindle-shaped. B: Nonfunctional sieve cell-phloem fiber transition period, during which the primary wall thickens. C: Phloem fiber development stage, the secondary wall thickens greatly. D: Phloem fiber mature stage, phloem fiber is basically mature, with different shapes, the common characteristic is that the cell cavity is very small

During the active period of the cambium, that is, from March to November, some of the nonfunctional sieve cells were thickened and transformed into immature phloem fiber cells. At the same time, the dense arrangement of mature phloem fiber cells was observed near the periderm. The development and maturation of phloem fibers occur in all stages. Phloem fibers can be seen in any stage of phloem development. The proportion of mature and developing phloem fibers differs slightly in each stage. The sign of the transition from sieve cells to phloem fiber is that the radial wall begins to thicken, which is much larger than the thickness of the tangential wall.

In this paper, according to the cell wall development stage, the development of phloem cells is divided into three stages: (1) the nonfunctional sieve cell-phloem fiber transition period; (2) the phloem fiber development period; (3) the phloem fiber maturity period. The nonfunctional sieve cells with just programmed death are still spindle-shaped and have no inclusions (Fig. 7A). In the nonfunctional sieve cell-phloem fiber transition period, the cells gradually flattened and the cell wall thickened, which was mainly the intercellular layer thickening; the secondary wall thickening was not obvious in this period (Fig. 7B). During the development of phloem fibers, the morphology of fiber cells was slender, and the delamination of the intercellular layer and secondary wall was gradually obvious. The cells extended radially and contracted in the tangential direction, and the radial width and tangential wall width were about 1:6. Most of them were extruded and deformed by functional sieve cells and phloem parenchyma cells; the secondary wall continued to thicken, the layering of the secondary wall and intercellular layer became obvious gradually, and the cell lumen became smaller until it developed into the mature fiber. During this period, the ratio of the intercellular layer thickness to the secondary wall thickness was about 1:4 (Fig. 7C). At the mature period of phloem

fiber, the cell wall thickness is up to 2 μ m. The ratio of intercellular layer thickness to secondary wall thickness was about 1:6, the cell lumen was very small, and the morphological difference was great (Fig. 7D).

IV. DISCUSSION

The phloem of *Taxodium ascendens* consists of sieve cells, phloem fibers, phloem ray parenchyma cells, and phloem parenchyma cells. From the cross section, it can be seen that there is an obvious "stratification" phenomenon in the secondary phloem. Sieve cell molecules, phloem fibers, and parenchyma cells are arranged alternately in tangential bands, and the ultrastructure and composition of various molecules are different in different developmental stages. In the Wuhan area, the active phase of cambium cells lasted from the beginning of March to the end of November, and the newly differentiated sieve cells were observed in the materials on March 26. However, the newly differentiated phloem parenchyma cells and ray parenchyma cells, that is, the earliest differentiation of sieve cells, were observed on April 11, that is, in the middle of April. The appearance of S plastids in the sieve cells runs through the whole active phase, which can be regarded as a sign of the beginning and end of the active phase. The time of division and differentiation of phloem parenchyma cells and ray parenchyma cells was about half a month later than that of sieve cells.

A small number of functional sieve cells were found in *Taxodium ascendens* all year round. It is speculated that some of the sieve cells formed at the end of the last long season remained mature through the winter and became the earliest functional sieve cells in the next spring, until the new sieve cells began to differentiate, at which point they lost their contents and enter into programmed death. The resin droplets of phloem parenchyma cells were highly dispersed in the active phase and uniformly distributed along the cell wall during the dormant period. The nucleus and abundant cytoplasm of sieve cells, phloem parenchyma cells, and phloem ray cells were all observed in the active phase. In the late active phase, the cytoplasm of sieve cells began to condense, the contents of the cells decreased sharply, and a large number of substances were consumed. On the contrary, ray parenchyma cells accumulated a large number of inclusions in the late active phase, and it was not until the middle of dormancy that oil droplets and starch grains in some ray parenchyma cells were consumed. It can be inferred that most of the ray parenchyma cells can survive for multiple growing seasons and store nutrients during the dormant period.

Climatic factors have a great influence on the partial formation of cambium and phloem. Wuhan belongs to the temperate zone, and the activity of tree cambium has obvious periodicity. Some studies have shown that the division peak and differentiation rate of cambium cells are affected by the average rainfall in the rainy season. Rainfall has a great influence on the division and differentiation of cambium, while temperature has little effect on it (Konrad *et al.* 2019). In this experiment, it was found that the phloem of *Taxodium ascendens* in Wuhan began to split in mid-late April. In this experiment, it was found that the phloem of *Taxodium ascendens* began to divide and differentiate earlier in 2019, and the newly differentiated young sieve cells could be observed in late March. Combined with climatic factors, it was speculated that it was caused by intensive precipitation from early March to the end of April in the Wuhan area; the cambium activity was advanced and phloem cells differentiated earlier.

At present, the understanding of phloem structure, whether in ontogeny or phylogeny, is difficult to achieve in-depth and comprehensive understanding of the xylem, especially at the ultrastructural level. The main reasons are as follows: (1) There are many kinds of cells in the phloem, and the arrangement is more complex; (2) Some or all of the phloem is easy to destroy and disappear during senescence; (3) They change during ontogeny and are not as easy to understand as the xylem; (4) The protoplasts of sieve elements are very sensitive; the cell walls of all kinds of parenchyma cells are very fragile and sensitive; the contents are complex; and the cells vary in length. Therefore, the original state is often changed in the process of fixation, especially in the radial section and tangential section, and the images under a different electron microscope are often obtained because of different fixing methods and slice angles.

V. CONCLUSION

- 1) The newly differentiated young sieve cells were observed in late March. The cell walls of the newly differentiated young sieve cells were thickening during the developmental period (April to June), and S plastids could be observed in June. Some sieve cells went into programmed death at the end of August. In the process of sieve cell programming, P plastids condensed, and the nuclear changes were very obvious. The nucleus in the sieve cells showed pyknotic degeneration, chromatin aggregates, nucleoli disappeared, and nucleoplasm decreased. At the same time, along with the gradual reduction of nuclear volume, the sieve cells reach their functional stage when they stop functioning and become phloem sieve cells without dredging function. The protoplasts also died, and finally, the whole cell was squeezed and deformed.
- 2) The phloem parenchyma cells of *Taxodium ascendens* belong to secretory cells, which contain a lot of resin. Phloem parenchyma cells began to divide and differentiate in mid-April. At the initial stage of cambium activity, that is, from mid-April to early May, the resin was distributed in large droplets, and at the peak of cambium activity (from the end of May to

the end of June). The resin was highly dispersed in the cell, and then the resin droplets gathered. During the dormant period, resin droplets were evenly distributed along the inner wall of the cells.

- 3) In mid-April, newly differentiated young ray parenchyma cells were observed in the phloem near the cambium. From mid-late April to early June, the young phloem ray cells developed to mature and remained in this state until the middle of November. After that, the cells entered programmed death, and most of the ray parenchyma cells died in dormancy in December, and there were only a large number of oil droplets and empty vesicles in the cells.
- 4) The development and maturation of phloem fibers occur in all periods. Phloem fibers can be seen in any period of phloem development. The proportion of mature and developing phloem fibers differs slightly in each period. The development of phloem fiber can be divided into three periods: the transition period between nonfunctional sieve cells and phloem fiber, the phloem fiber development period, and the phloem fiber maturity period. In the nonfunctional sieve cell-phloem fiber transition period, the sieve cell is extruded and the intercellular layer is thickened; at the phloem fiber development stage, the secondary wall is greatly thickened; at the phloem fiber mature stage, the mature phloem fiber cell lumen is very small.

AUTHOR CONTRIBUTIONS

Conceptualization, Y. X; Methodology, Validation, Formal analysis, Y. X and H.W; Investigation, H.W., and J.T.; resources, H.L.; data curation, Y.X. and H L; writing—original draft preparation, H.W. and Y.X; writing—review and editing, supervision and funding acquisition, Y.X.; project administration and visualization, Y.X and H L.; All authors have read and agreed to the published version of the manuscript.

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CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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Production Technology and Edaphological Anticipations of a Promising Soil Amendment: Vermicompost

Yatharth Ratan Gondwana

B.Sc. (Honours) Agriculture Graduate, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi, U.P.- 221005, INDIA

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Abstract— Vermicompost, an enriched compost made by involving earthworms in the preparation process, is one of the most important products in sustainable agriculture today. It has the potential to promote significantly more plant growth than other composts, prevent soil degradation, remediate it, and naturally improve overall soil health and quality. It provides cultivators with numerous opportunities, not only in terms of social and economic advancement, but also for maintaining and increasing the productivity potential of agricultural land. The dynamic nutritional composition and soil-enhancing properties of vermicompost are unmatched by any other available alternative.

Keywords— Black gold, earthworms, soil health, vermicomposting bed and vermicompost.

I. INTRODUCTION

Composting is a viable method for converting biodegradable solid wastes into beneficial organic soil amendments for environmentally friendly agricultural production systems. Earthworms are known as ecological engineers and farmers' best friends because they help to promote the composting process, improve nutrient value, and speed up the formation of stable organic end products. Vermicomposting involves using earthworms to create enriched compost, and the finished product is known as vermicompost. Earthworms contribute to the decomposition process in three ways: physically by serving as an aerator, crusher, and mixer; by chemical means as a degrader; and biologically as a stimulator. Earthworms feed on biomass (organic matter which is in the process of decaying) and excrete in digested form as worm casts/ worm manure.

II. SUITABLE EARTHWORM SPECIES

Earthworms that live on the surface are frequently used in vermicomposting, namely *Eisenia fetida* (redworm) and *Eudrilus eugeniae* (African nightcrawler). They can convert a wide range of biodegradable waste into worm casts or worm manure, also known as "black gold," and are highly engaged in the reproduction process and effective in organic material recycling



PIC 1[©]: *Eisenia fetida* (redworm), a popular choice among vermicompost cultivators.

III. REQUIRED MATERIALS FOR VERMICOMPOSTING

- a. A Cement pit or portable vermicomposting bed (made of silpaulin) of dimensions of 12×4×2 feet.
- b. Approximately 750-800 kg of cattle manure, at least 1-1.5 months old (pre-digested).
200-250 kg of dry matter and shredded pre-digested organic waste. Fresh green waste has a considerable nitrogen content and is susceptible to overheating compost thus, pre-digested waste is encouraged. The ratio of dry matter (high carbon content) to organic waste (high nitrogen content) ought to equal 3:1.

TABLE 1
RECOMMENDED CARBON-RICH DRY MATTER & NITROGEN-RICH ORGANIC WASTES.

Dry matter	Organic wastes
Peat moss	Chopped green vegetable peels/ waste
Shreds of newspaper	Sawdust
Corrugated cardboards	Softwood chips (Fine)
Shrub trimmings	Shredded barks of softwood

Source: Chanu et al. (2018).

- c. Since paddy straw has a C:N ratio of 150:48, or nearly 3:1, it can be used as both dry matter and organic waste. One should also refrain from using diseased plants and pungent kitchen wastes like onions, garlic, eggshells, etc. because earthworms are unable to consume these things and the quality of the finished product will suffer.
- d. Approximately 800-815 earthworms (350-360 worms/m³ of bed or pit volume).
- e. A shovel and/or similar garden tool, along with a sieving net.



PIC 2[©]: Portable silpaulin vermicomposting bed.

IV. SITE OF PREFERENCE

Vermicomposting can be carried out anywhere that features shade, a high humidity level, and a cool climate. Cattle sheds, unused buildings, and open areas with artificial shading are all possible locations.

V. METHODOLOGY

After installing or erecting a vermicomposting bed or pit in a desired location, fill it with the following cross-section of its content and release earthworms at its upper surface as they eventually make their way inside.

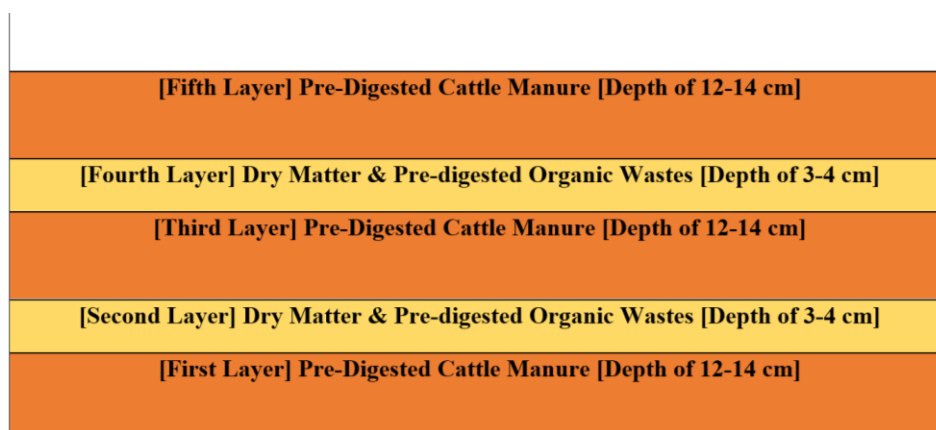


FIGURE 1[©]: Cross-section of the make-up of pre-digested (at least one month old) cattle manure, dry matter(s), and/or pre-digested organic wastes inside vermicomposting bed/pit. *Source: Author's own depiction of cross-section of vermicomposting bed.*



PIC. 3[©], 4[©] & 5[©] (Top left, top right & bottom): Application of cattle manure at the bottom of the bed after its installation, application of dry matter (sawdust) above the bottom layer of cattle manure, & application of earthworms over the bed.

VI. MANAGEMENT

- Cover the top portion of the bag or pit using jute bags or paddy straws to avoid predatory creatures like birds from eating the earthworms.
- Throughout the composting period, regular watering, turning, and subsequent monitoring are recommended to maintain the suggested percentages of moisture (around 60%) and temperature (18 °C- 35 °C).
- Moisture content can be manually determined by placing a small amount of vermicompost in your own palm; it should adhere to your palm but not drip liquid over your fingers.
- To ensure adequate air circulation and decomposition, the material inside the bag or pit should be turned upside down once every 18 to 20 days with a shovel or a similar tool.



PIC 6[©]: Turning of vermicombed contents (Upside down).

VII. HARVESTING & STORAGE

- a. The final product (vermicompost) accounts for approximately 75% of the initial input.
- b. Depending on management, vermicompost would be ready in 75 to 90 days on average.
- c. The final product would be made up of tiny granular fragments similar to tea granules, would not smell bad, and would have a dark brownish-black colour.
- d. Watering should be stopped a week prior to the anticipated harvest date.
- e. Before harvesting, manually check the moisture content of vermicompost by placing some of it in your palm; it should stick to the palm of your hands but not drip any water.
- f. To make certain that only a tiny percentage of earthworms perish during the compost harvest, use the sieving net gently.
- g. The harvested vermicompost must be stored in a cool, dark place that is not exposed to direct sunlight.
- h. If moisture is maintained at 40%, vermicompost could be stored for an entire year without losing quality.



PIC 7[©]: Harvesting of vermicompost (by manual means) using sieving net.

VIII. VERMICOMPOST: A MEANS FOR IMPROVING SOIL HEALTH AND ENHANCING CROP PRODUCTIVITY

Vermicompost contains a substantial amount of pathogenic microbe-inhibiting agents, useful soil microbial flora, and essential plant nutrients. As a result, the organic by-products produced by earthworms inherit the majority of black gold's advantageous characteristics. When used as an organic soil amendment, vermicompost enhances soil quality by improving its chemical, physical, and biological properties. It also has earthworm cocoons, which increase earthworm activity and population growth in the soil. It reduces nutrient losses and increases the efficiency of chemical fertilisers. Vermicompost reduces pests and

diseases, accelerates the decomposition of organic material in the soil, is void of pathogens, toxic substances, weed seeds, and other contaminants, and incorporates beneficial vitamins, enzymes, and hormones such as auxins and gibberellins.

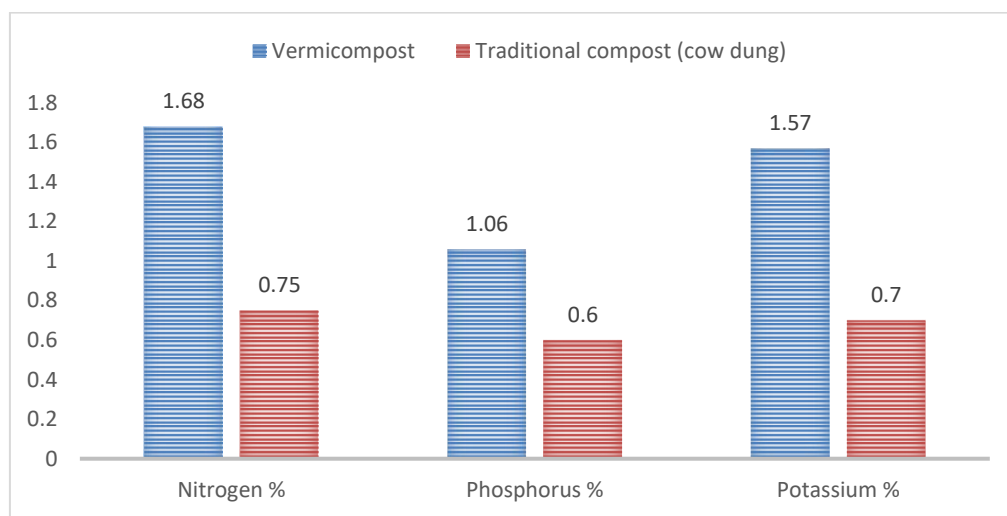


FIGURE 2: Comparison of contents of primary nutrients (N, P, K) in vermicompost & traditional compost (cow dung).

Source: Author's own graphical depiction.

In plant nurseries, vermicompost outperforms other synthetically grown media. It can be used in nurseries as a rooting medium and to establish saplings, and it has excellent impacts on plant growth in general, promoting the formation of new shoots and leaves and improving the quality and shelf life of the produce. It preserves the quality of the produce while improving its taste, lustre, and appearance. Immobilized enzymes such as protease, lipase, amylase, cellulase, and chitinase in vermicompost continue to break down agricultural residues in the soil itself. It does not smell bad, unlike traditional manure and decomposing organic waste. Vermicompost is becoming more popular as a key component of the organic farming system due to its excellent nutritional value as well as its effectiveness as an environmentally friendly soil amendment.

TABLE 2
AVERAGE NUTRITIVE VALUE OF VERMICOMPOST.

Nutrient	Content
Organic Carbon (OC)	9.15- 17.98%
Nitrogen (N)	1.5- 2.1%
Phosphorus (P) & Potassium (K)	1.0- 1.5% & 0.6%
Calcium (Ca) & Magnesium (Mg)	22.0-70.0 meq/ 100g
Sulphur (S) & Copper (Cu)	128- 548 ppm & 100 ppm
Iron (Fe) & Zinc (Zn)	1800 ppm & 50 ppm

Source: Chanu et al. (2018).

Application of vermicompost for different crops: The type of crop grown in the field or nursery determines how much vermicompost is applied. Vermicompost should be used as part of integrated nutrient management systems to improve crop production. The recommended amount of vermicompost after transplanting and last ploughing for major field crops like rice and maize is 2.5 tons/ha; for oilseeds and pulses, it is between 2.5 and 4.5 tons/ha; and in case with the vegetables, it is roughly 3-3.5 tons/ha.

IX. LIMITATIONS

It is to be noted that vermicompost, despite being a dynamic and diverse compost, has limitations, primarily due to its inability to compete individually over the primary nutrients (nitrogen, phosphorus, and potassium) of industrially produced fertilizers, such as urea (46% nitrogen), diammonium phosphate (18% nitrogen and 46% phosphorus) and muriate of potash (60 % potassium), which far outweighs the N, P, K composition of vermicompost.

X. CONCLUSION

Vermicompost has the potential to transform conventional agriculture into a more sustainable form of farming, and it should be used as a part of integrated nutrient management to increase crop productivity rather than relying solely on it, as it cannot supplement the requirements of conventional fertilizers but can work in conjunction with them, thus leading to a prolonged retention of soil quality and health.

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I would like to extend my sincere appreciation to Dr. B.K. Sarma, Professor, Department of Mycology and Plant Pathology, Institute of Agricultural Sciences, Banaras Hindu University, for his benevolent guidance during my studies and for providing me with the opportunity to extend myself beyond the classroom and into the practical arena, which ultimately motivated me in the domain of research.

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Agronomic Characteristics of Three Soybean Varieties at Different Levels of Genistein

Aman Suyadi¹, Totok Agung Dwi Haryanto², Ismangil³, Ponendi Hidayat⁴

¹Department of Agrotechnology, Faculty of Agriculture and Fishery Universitas Muhammadiyah Purwokerto, Indonesia

²Doctoral Program in Agricultural Sciences, University of Jenderal Soedirman Purwokerto, Indonesia

^{3,4}Department of Agrotechnology, Faculty of Agriculture University of Jenderal Soedirman Purwokerto, Indonesia

*Corresponding Author

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Abstract— This research aimed to identify the agronomic characteristics of three soybean varieties at varied genistein concentrations. Three soybean varieties (V1 = Deja1, V2 = Denal, V3 = Demas1) and four levels of genistein concentration (G0=0ppm, G1=60ppm, G2=120ppm, G3=180ppm) were the two components that made up the plot design in this research. A total of three times were used for the treatment combination. Genistein concentrations were used in the split plot's design, while the variety was applied to the main plot. The observed factors include the amount of genistein in the leaves, the number of root nodules, the amount of chlorophyll, the amount of leaf area, and the pace of plant growth.

The study's findings demonstrate that the variety Demas1 produced the most chlorophyll. 32.04 mg/L, 0.269 mg/g dried leaves of leaf genistein, and 47.88 grains of root nodules. The Deja 1 variety yielded the widest leaf area, measuring 33.26 cm², and the fastest plant growth rate, measuring 0.853 g/m²/week. According to the study's findings, the best genistein concentration up to the age of 49 days after planting is 180 ppm, which can result in leaves with an area of 32.3 cm², chlorophyll content of 34.21 mg/L, a genistein concentration of 0.032 mg/g in leaves, 43.60 grains of root nodules, and a growth rate of 0.736 g/m²/week in plants. The best agronomic characteristics are produced by the Demas1 soybean variety, which has 180 ppm of genistein.

Keywords—Soybean (*G. max (L.) Merr.*), genistein, Deja1, Denal, Demas1.

I. INTRODUCTION

Due to ozone layer depletion, UV radiation will be more prevalent on Earth's surface. UV-B light (280–320 nm) can be hazardous because it damages cells. Plants are vulnerable to increasing UV-B radiation because several biological components, including nucleic acids, proteins, lipids, and quinones, can directly absorb UV-B radiation. According to Kakani et al. (2003), increased UV-B exposure causes plants to photosynthesize slowly. This effect is caused by extracting photosynthetic gene expression, reduced Rubisco activity, altered thylakoid ion membrane permeability, and changes in chlorophyll and carotenoid levels. (Gaberscik et al., 2002).

According to Diffey (1991), the annual UV-B radiation flux decreases with increasing distance from the equator. Crop productivity is higher in mid-latitude nations like China, Korea, and Japan since UV-B radiation exposure is lower than in Indonesia. As compared to China's soybean production of 12.943 million tons (Myers, 2014), which was more significant than Indonesia's national soybean production of 0.63 million tonnes in 2020, Korea's soybean production climbed to 154,067 MT in the 2013/2014 planting season, up 31,548 MT (26%) from the previous year. (Ministry of Agriculture, 2021)

UV light and exposure duration mainly contribute to Indonesia's low soybean productivity (Sumarno et al., 2007). UV-B radiation's photobiological effect generates free radicals and damages cells (Baumann and Allemann, 2009). Free radicals can harm proteins, amino acids, protective enzymes, and the structure and function of cells (Refdi, et al., 2014). They can also diminish the performance of substances in the body (Fisher, 2002). Biomolecules like DNA, RNA, proteins, and membranes can become damaged when exposed to UV-B radiation at high fluences or over extended periods. (Alsekn et al., 2020).

The formation of flavonoid molecules is a natural response of Leguminosae plants to UV-B exposure. In addition, flavonoids show protective effects against biotic and abiotic stressful situations, such as pathogen infections, exposure to UV-B and high-fluence white light, drought, cold, salt, and herbivore attacks (Falcone Ferreyra et al., 2012). There are 12 isoflavones in soybeans, including three aglycones called genistein, daidzein, and glycerin, all produced in reaction to UV light and herbivorous insects (Zavala et al., 2015, Piubelli et al., 2005). The primary, secondary metabolite of soybeans, genistein, is a natural isoflavonoid with several beneficial properties. (Polkowski, 2000). It takes a lot of energy to produce secondary metabolites, which inhibits pod filling and growth (Xing et al., 2014). The energy required to synthesize isoflavones can increase soybean plant growth and production by reducing the effects of UV-B radiation.

Exogenous isoflavones can boost soybean production in milk, tofu, flour, protein concentrate, soy protein isolate, and other processed soy foods that are not fermented (Coward et al., 1998). Exogenous isoflavones are given as sunscreen to lessen exposure to UV-B rays. A thin layer of isoflavones can tolerate most UV-B radiation because of one of the characteristics of UV-B light, which has deficient penetrating power. Reduced UV-B exposure can inhibit the generation of endogenous isoflavones, allowing soybean plants to grow and produce more using that energy instead of producing endogenous isoflavones.

Deja1 is a water stress-tolerant soybean variety created by a single cross between the Tanggamus variety and Anjasmoro (Balitkabi) (Anggraini, 2020). Deja1 has more trichomes than Derap1, Detam1, Detam2, and Dering1 types (Pandjaitan, 2021). Argomulya and IAC were crossed to create the shade-tolerant soybean variety known as Dena1 (Balitkabi). Dena1 has the highest antioxidant activity (80.23% DPPH inhibition), total phenolic content (4.18 mg GAE/g), and total flavonoid content (4.23 mg QE/g) (Aurelia, 2022). The excellent soybean variety Dena1 is resistant to pests and diseases (Balitkabi, 2015). In addition, Dena1 has the highest concentrations of chlorophyll a, b, and the chlorophyll a/b ratio (Noya et al., 2019) Demas1 is a variety made by crossing Mansuria and SJ that can survive in dry, acidic soils (Balitkabi). Demas-1 soybeans grow better in acidic soils because they are genetically resistant to acid. (Selvia et al, 2019).

This research aimed to present scientific data on the agronomic characteristics of three soybean varieties at different genistein concentrations. Experiments were done to see how each type would react to genistein concentrations. The study's findings are anticipated to be applied as a recommendation to administer genistein to three soybean types to boost production.

II. MATERIALS AND METHODS

2.1 Study Area

The research was conducted in the experimental garden of the Faculty of Agriculture and Fisheries, Muhammadiyah University, Purwokerto, Karangasari Village, Kembaran District, Banyumas Regency (coordinates 07° 23' 56" South Latitude and 109° 16' 53" East Longitude, altitude ± 85 m above sea level (asl), Agrotechnology Laboratory Applied, Laboratory of Basic Agrotechnology, Faculty of Agriculture and Fisheries, and Laboratory of Pharmaceutical Biology, Faculty of Pharmacy, Universitas Muhammadiyah Purwokerto. The research started from July to October 2021.

2.2 Procedures

The research involved a factorial experiment with polybags set up in an entirely random pattern. The genistein administration concentration (G) and the soybean varieties (V), specifically V1 = Deja1, V2 = Dena1, and V3 = Demas1, were evaluated. G0 = 0 ppm, G1 = 60 ppm, G2 = 120 ppm, and G3 = 180 ppm were the control factors. Three repeats were done, with 108 plants in each repetition. Observational variables include leaf area, chlorophyll content, leaf genistein concentration, number of root nodules, and plant growth rate.

The research entailed creating the planting media, a mixture of manure and loose dirt obtained by sieving three millimeters of soil (1:1 ratio). Two soybean seeds are contained in each polybag as they are planted by hand using a shovel. Genistein was provided with concentrations by the therapy from 0 days after planting (DAP) to 50 DAP at intervals of 10 days. At zero hours, genistein was administered by watering the plant with 30 ml each. Genistein spray volume per plant, aged ten days: 180 ml, 20 days: 200 ml, 30 days: 320 ml, 40 days: 330 ml, and 50 days: 330 ml.

2.3 Data Analysis

Observational data were analyzed using the 5% level DMRT test after the F test at the 95% level of confidence if they were found to be substantially different.

III. RESULT AND DISCUSSION

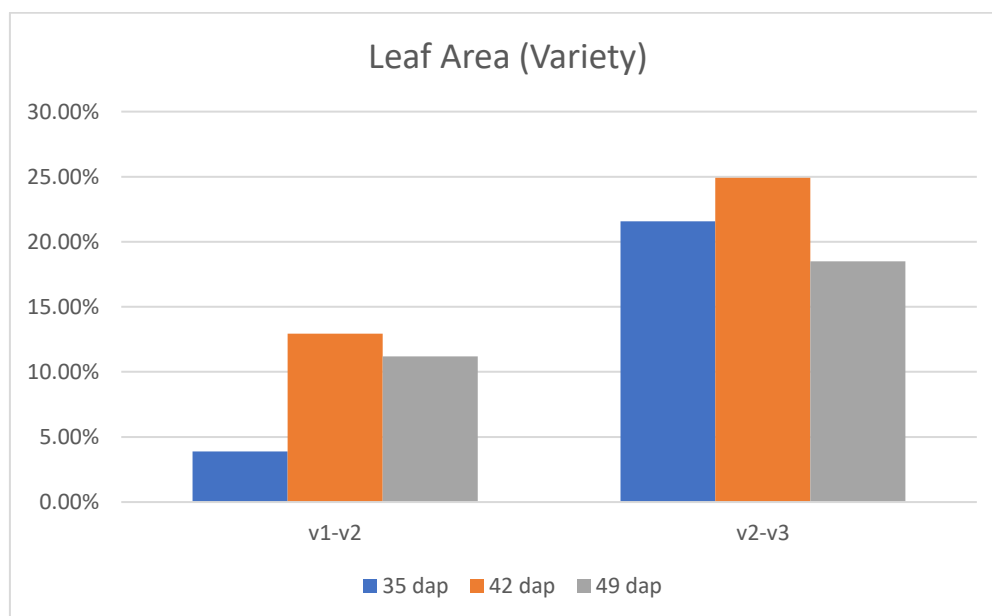
3.1 Leaf Area

TABLE 1
RESULTS OF THE VARIABLE LEAF AREA (cm²) ANALYSIS OF 3 SOYBEAN VARIETIES WITH GENISTEIN ADDITION

Treatment	35 days after planting	42 days after planting	49 days after planting
variety			
V1	25.81a	31.99a	33.26a
V2	24.84a	28.32b	29.91b
V3	21.23b	25.61c	28.07c
genistein			
G0	20.91c	26.29c	28.27c
G1	24.18b	28.43b	30.26ab
G2	24.92ab	29.26b	30.92ab
G3	25.82a	30.58a	32.3a

Information: V = soybean variety, V1 = Deja1, V2 = Dena1, V3 = Demas1, G = genistein concentration, G0 = 0ppm, G1 = 60ppm, G2 = 120ppm, G3 = 180ppm

The type of soybean and the amount of genistein had a big impact on the leaf area. Table 1 demonstrates that at age 35 days after planting, treatment V1 (Deja) had the widest leaf area, measuring 25.8 cm², 3.9% wider than treatment V2 (Dena1), and 21.57% wider than V3 (Demas1). 42 years old. The V1 treatment (De ja 1) had the widest leaf area at 31.99 cm², 12.95% wider than the V2 treatment (Dena 1), and 24.91% wider than the V3 treatment (Demas1). 49 days after planting old The V1 treatment (Deja1) has the widest leaf area, measuring 33.26 cm², 11.2% wider than the V2 treatment (Dena1), and 18.49% wider than the V3 treatment (Demas1).

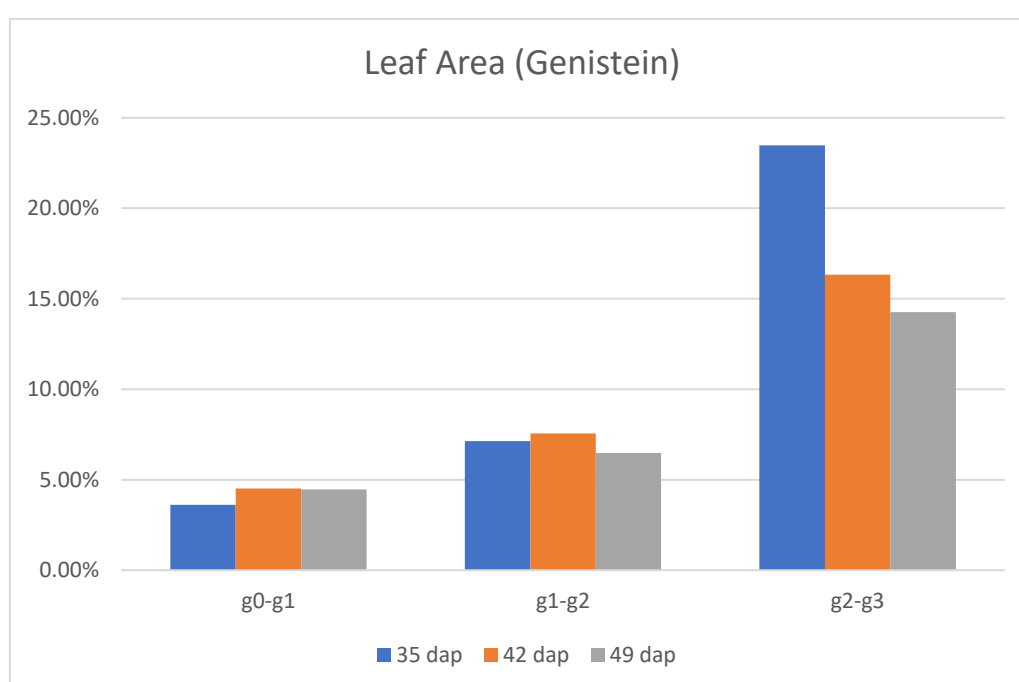


GRAPHIC 1: The percentage of leaf area increases in the various treatment

These results show that the Deja1 variety was able to produce the largest leaf area at each observational stage. It is believed that the Deja1 variety is a light-sensitive kind. Thus it must adapt to be able to absorb more sunshine. The leaves extend as a result of adaptation. The Deja1 (V1) has more wide and thin leaves, which are supposed to represent a defense mechanism against low light (Khumaida, 2002). According to Sopandie et al. (2002), shade-tolerant soybean varieties have higher

chlorophyll an in their genetic characteristics to increase photon absorption. According to Evans and Porter (2001), the shadow avoidance reaction of plants under light intensity stress is carried out by maximizing light capture by altering the structure and morphology of leaves for effective photosynthesis, with thinner leaves so that the distribution of photosynthetic results to light harvesting network becomes faster. The Dejal variety is characterized by wide, thin leaves that increase the effectiveness of photosynthetic processes by absorbing lighter and swiftly conveying it to the lower leaves. The Dejal type can also slow down transpiration since it has more trichomes. The reduced rate of transpiration satisfies the requirement for photosynthetic water. (Pandjaitan, 2021).

According to Table 1, the leaf area tends to grow weekly. The age of the leaf area was considerably impacted by genistein concentration at 35, 42, and 49 days after planting. At the age of 35 days after planting, G3 (180 ppm) produced the widest leaf area, measuring 25.82 cm², 23.48% wider than G0 (0 ppm), 7.14% wider than G1 (60 ppm), and 3.61% wider than G2 (120ppm). At the age of 42 days after planting, G3 (180ppm) produced the widest leaf area, which was 30.58cm², 16.32% wider than G0 (0ppm), 7.56% wider than G1 (60ppm), and 4.51% wider than G2 (120ppm). The best treatment is G3 (180ppm) at the age of 49 days after planting, which produced the widest leaf area, or 33.30², 14.26% wider than G0 (0ppm), 6.74% wider than G1 (60ppm), and 4.46% wider than G2 (120ppm).



GRAPHIC 2. The percentage of leaf area increases in the genistein treatment

Genistein can prevent plant cell damage brought on by UV light exposure. According to Muchtadi (2012) and Cahyati et al. (2013), genistein is a secondary metabolite that protects cells from free radical damage. Without genistein protection, plants will keep growing in the presence of ultraviolet radiation, giving up their chloroplasts to protect other cells, which interferes with photosynthesis. (Barufi *et al.*, 2010). According to Ai and Banyo (2011), chlorophyll is the primary plant pigment that contributes to photosynthesis, particularly when solar energy is used to start CO₂ fixation, which produces carbohydrates and energy. Proteins, lipids, nucleic acids, and other organic compounds are created from the carbohydrates generated during photosynthesis. Supplying genistein 180 ppm (G3) is likely to lessen the effects of exposure to UV-B rays by preventing and reversing chloroplast damage, allowing the photosynthesis process to function properly, and the growth and development of soybean plant leaves.

Genistein is often ingested from plants in the form of the glycoside genistin. Genistin is hydrolyzed into genistein (the bioactive aglycone) by phlorizin hydrolase, a small intestine brush-border lactase, before being absorbed or further changed by enteric bacteria (Walsh et al., 2007). (Mattison et al, 2014). Like other polyphenols, genistein has a 10% oral bioavailability. (Hu *et al.*, 2007). Genistein can be passively carried into intestinal cells due to its poor absorption potential, lipophilic nature, and low molecular weight (Liu et al., 2002). This results in post-absorption metabolism. The accumulation of various flavonoids, such as flavonols, flavones, and anthocyanins, helps plants solve a variety of biotic and abiotic stresses. Phenolic chemicals, particularly flavonoids, are essential plant secondary metabolites. (Mouradoy et al 2014, Jiang et al 2016).

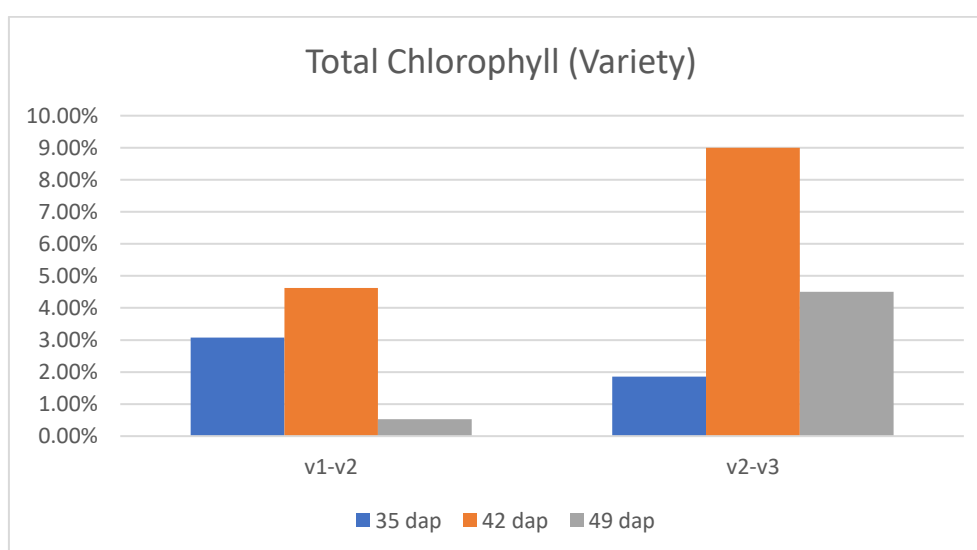
3.2 Total Chlorophyll

TABLE 2
THE RESULTS OF THE ANALYSIS OF THE VARIABLE AMOUNT OF CHLOROPHYLL (MG/L) OF THE ROOTS OF 3 SOYBEAN VARIETIES WITH THE ADDITION OF GENISTEIN

Treatment	35 days after planting	42 days after planting	49 days after planting
variety			
V1	23.38a	28.6ab	30.66b
V2	23.66a	27.45b	31.87b
V3	24.10a	29.92a	32.04b
genistein			
G0	21.58c	27.087c	30.66b
G1	23.39b	28.06bc	31.87b
G2	23.60a	29.11a	32.04b
G3	26.15a	30.47a	34.21a

Information: V = soybean variety, V1 = Deja1, V2 = Dena1, V3 = Demas1, G = genistein concentration, G0 = 0ppm, G1 = 60ppm, G2 = 120ppm, G3 = 180ppm

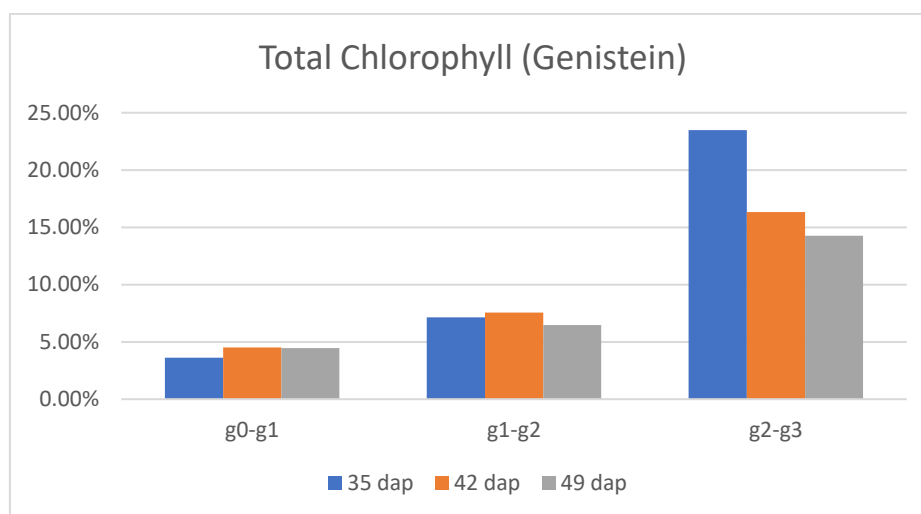
Table 2 demonstrates that although the soybean varieties did not significantly change the quantity of chlorophyll at 49 days after planting, they did significantly affect the amount of chlorophyll in leaves aged 35 days after planting and 42 days after planting. Treatment V3 (Demas1), which had a leaf chlorophyll concentration of 24.10 mg/L at age 35 days after planting, had the highest level, 3.08% higher than treatment V1 (Deja1) and 1.86% higher than V2 (Dena1). Treatment V3 (Demas1), which had a leaf chlorophyll concentration of 29.92 mg/L at age 42 days after planting, had the highest level, 4.62% higher than treatment V1 (Deja1) and 9.00% higher than V2 (Dena1). Treatment V3 (Demas1), which had a leaf chlorophyll concentration of 32.04 mg/L at age 49 days after planting, was 4.50% higher than treatment V1 (Deja1) and 0.53% higher than V2 (Dena1). These findings show that the V3 (Demas1) treatment may effectively adapt to changing environmental conditions and can lessen the harmful effects of UV-B radiation, protecting against and reversing chloroplast damage. Plants become resistant and tolerant to UV-B radiation by shielding their cells. (Levit, 1980). Cotton plants' adaptive process to exposure to UV-B rays is carried out by decreasing leaf thickness and raising the stomata index, palisade layer, and wax content. (Kakani V., G., *et al* , 2002).



GRAPHIC 3: The percentage of total chlorophyll increases in the various treatment

Table 2 shows that the genistein concentration dramatically affects the amount of chlorophyll in leaves aged 35 days after planting, 42 days after planting, and 49 days after planting. At the age of 35 Days after planting, G3 (180ppm) produced the

highest amount of leaf chlorophyll, namely 26.15 mg/L, 10.8% more than G0 (0ppm), 11.80% more than G1 (60ppm), and 10.81% more than G2 (120ppm). G3 (180 ppm) was the most effective treatment at 42Days after planting, producing 30.47 mg/L of leaf chlorophyll, 4.67% more than G0 (0 ppm), 8.59% more than G1 (60 ppm), and 12.49% more than G2 (120 ppm). G3 (180 ppm) was the most effective treatment at 49Days after planting, producing 34.21 mg/L of leaf chlorophyll, 6.77% more than G0 (0 ppm), 7.34% more than G1 (60 ppm), and 11.58% more than G2 (120ppm). These data suggest that genistein can protect soybean plants from the effects of exposure to UV-B radiation since it increases the amount of chlorophyll the higher the genistein concentration given which can decrease the amount of chlorophyll, it was discovered that *Phaseolus vulgaris* leaves grown under UV-B stress had lower levels of chlorophyll a and b (Michaela et al., 2000). Flavonoid compounds serve as UV filters by absorbing a wide spectrum of UV-B rays. Anthocyanin-containing flavonoids in corn plant DNA prevent DNA from being damaged by UV light. According to Cohen et al. (2001), flavonoids play a role in plants' defense mechanisms against UV-B exposure. Supplying exogenous flavonoids is hypothesized to be capable of providing the photosynthetic organs with a layer of protection, allowing the photosynthetic process to proceed generally while suppressing the formation of endogenous flavonoids. The next consequence is the ability of soybean plants to conserve energy and improve growth and bean pod filling through photosynthetic activity.



GRAPHIC 4: The percentage of total chlorophyll increases in the genistein treatment

3.3 Leaf Genistein Concentration

TABLE 3

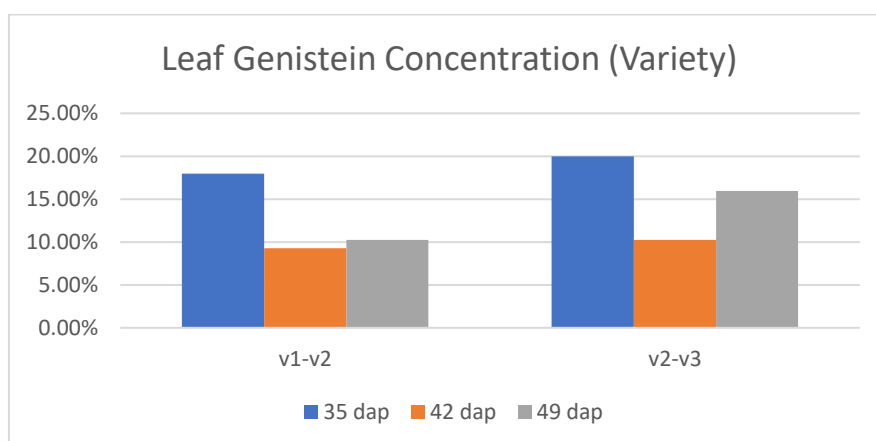
THE RESULTS OF THE ANALYSIS OF THE VARIABLE GENISTEIN LEAF CONCENTRATION (MG/G DRY LEAVES) OF THREE SOYBEAN VARIETIES WITH GENISTEIN ADDITION

Treatment	35 days after planting	42 days after planting	49 days after planting
variety			
V1	0.175b	0.215b	0.244b
V2	0.178b	0.216b	0.232b
V3	0.210a	0.237a	0.269a
genistein			
G0	0.101d	0.150d	0.179d
G1	0.163c	0.192c	0.214c
G2	0.217b	0.251b	0.278b
G3	0.271a	0.289a	0.322a

Information: V = soybean variety, V1 = *Deja1*, V2 = *Dena1*, V3 = *Demas1*, G = genistein concentration, G0 = 0ppm, G1 = 60ppm, G2 = 120ppm, G3 = 180ppm

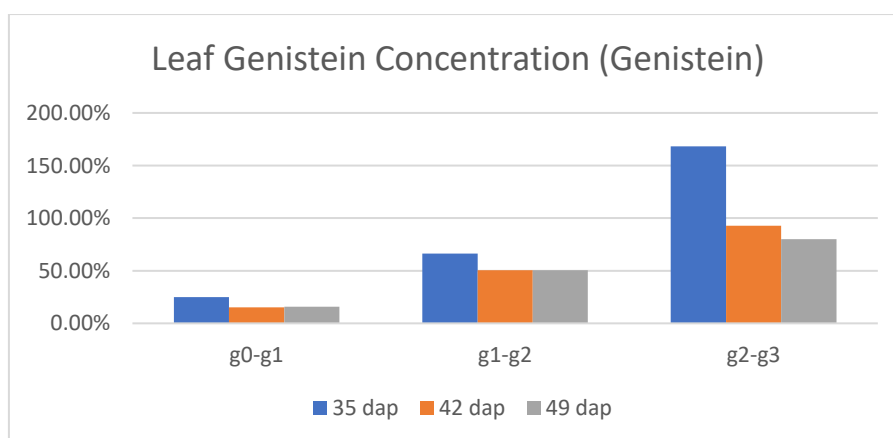
According to Table 3, soybean varieties significantly affect the number of genistein leaves aged 35, 42, and 49 days after planting. At 35 days after planting, the V3 (*Demas1*) treatment had the highest level of genistein leaves, 0.210 mg/g, 20%

higher than the V1 (Deja1) treatment and 17.98% more than the V2 treatment (Dena1). Treatment V3 (Demas1) had the greatest level of genistein leaves at age 42 days after planting, with 0.237 mg/g, 10.23% more than treatment V1 (Deja1) and 9.72% more than V2 (Dena1). Treatment V3 (Demas1) had the most genistein leaves at age 49 days after planting, with 0.269 mg/g, 10.25% more than treatment V1 (Deja1), and 15.95% more than V2 (Dena1). According to the data, the V3 (Demas1) treatment could prevent chlorophyll damage from UV-B light exposure. According to Moghaddam et al. (2011), leaves generate flavonoid compounds as a coating for photosynthetic pigments that prevent plant damage to lessen UV-B radiation's impact. Isoflavones are synthesized by plant cells under conditions of growth stress, such as exposure to UV-B rays. Sulistyowati et al. (2018) indicated that each genetic diversity is directly proportional to the response of plant growth and development and that variety affects the genistein content of soybean plants.



GRAPHIC 5. The percentage of leaf genistein concentration increases in the various treatment

According to Table 3, the genistein concentration greatly affects the amount of genistein in leaves aged 35 days after planting, 42 days after planting, and 49 days after planting. The most effective treatment is G3 (180ppm), which was able to produce the most leaf genistein at the age of 35 Days after planting, namely 0.271 mg/g, 168.32% more than G0 (0ppm), 66.26% more than G1 (60ppm), and 24.88% more than G2 (120ppm). The most effective method at 42Days after planting produced 0.289 mg/g of genistein in leaves, 92.67% more than G0 (0ppm), 50.52% more than G1 (60ppm), and 15.14% more than G2 (120ppm). The most successful treatment at 49Days after planting was G3 (180ppm), which was able to produce 0.322mg/g of leaf genistein, 79.89% more than G0 (0ppm), 50.47% more than G1 (60ppm), and 15.83% more than G2 (120ppm). These findings suggest that genistein can be applied to soybean plant leaves to form a residue that acts as a UV radiation absorber. 90% to 99% of UV light is absorbed by epidermal cells. The main UV absorption agents are believed to be waxes and flavonoid chemicals. UV exposure negatively impacts plants, including stunted development and morphological changes. Genistein and daidzein glycosides were primarily used to study the uptake and metabolism of isoflavones, making them both the most representative phytoestrogens for nutraceuticals. (Franke *et al.*, 2004; Heinonen *et al.*, 2003).



GRAPHIC 6. The percentage of leaf genistein concentration in the genistein treatment

According to Ho et al. (2002), isoflavone glycosides and derivatives predominate in soybean seeds, as opposed to Kaempferol glycosides are the primary flavonoids in soybean leaves. However, Piubelli et al. (2005) confirmed that soybean leaves of

various genotypes contained substantial levels of rutin (Quercetin 3-O-rhamnosyl glucoside) and genistin (genistein 7-O-glucoside). Genistein and rutin, two flavonoids, were found to be correlated with lower *Anticarsia gemmates* larval growth after several soybean varieties were characterized.

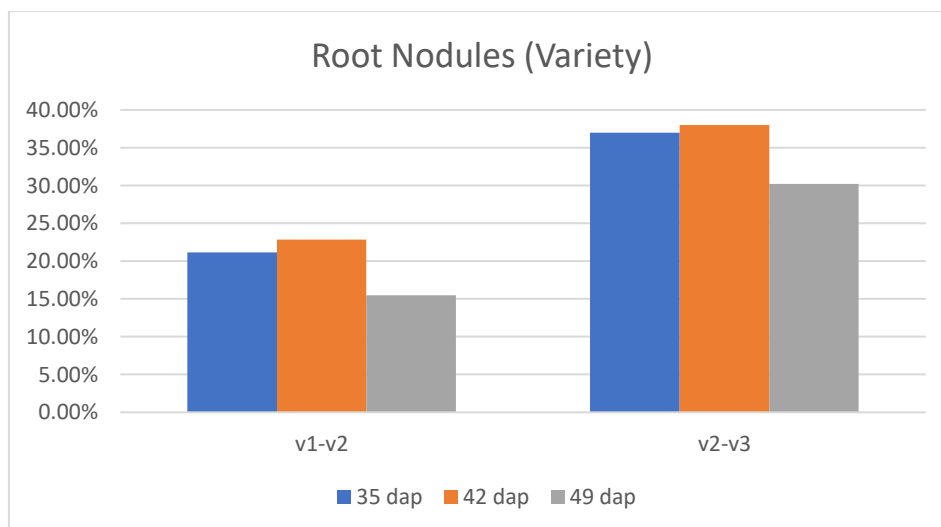
3.4 Number of Root Nodules

TABLE 4
THE RESULTS OF THE ANALYSIS OF THE VARIABLE NUMBER OF ROOT NODULES (GRAINS) OF THE THREE TYPES OF SOYBEAN VARIETIES WITH THE ADDITION OF GENISTEIN

Treatment	35 days after planting	42 days after planting	49 days after planting
variety			
V1	26,66c	29.91a	36.77c
V2	30,22b	33.60b	41.47b
V3	36.61a	41.27a	47.88a
genistein			
G0	29d	31.58d	40.70d
G1	30.81c	33.59c	41.44c
G2	31.63b	36.11b	42.40b
G3	33.22a	38.18a	43.60a

Information: V = soybean variety, V1 = Deja1, V2 = Dena1, V3 = Demas1, G = genistein concentration, G0 = 0ppm, G1 = 60ppm, G2 = 120ppm, G3 = 180ppm

According to Table 4, V3 (Demas1) produced the most nodules at 35 days after planting, 42 days after planting, and 49 days after planting. At 35 DAP, the V3 (Demas1) treatment produced the most root nodules, 36.61 grains, 37% more than the V1 (Deja1) treatment and 21.14% more than the V2 treatment (Dena1). The V3 (Demas1) treatment produced the most root nodules at 42 DAP, with 41.27 grains, 37.98% more than the V1 (Deja1) and 22.82% more than the V2 treatment (Dena1). At 49 Days After Planting, the V3 (Demas1) treatment produced the most root nodules, or 47.88 grains, which is 30.21% more than the V1 (Deja1) treatment and 15.46% more than the V2 treatment (Dena1).

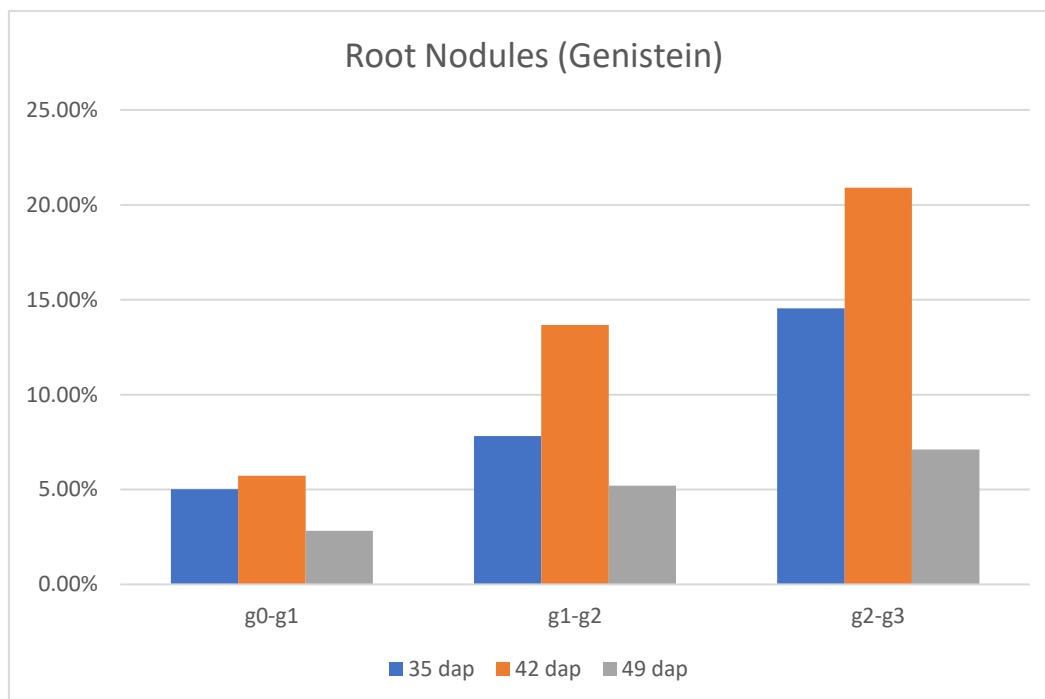


GRAPHIC 7. The percentage of root nodules (grains) increases in the various treatment

The Demas1 variety showed the most significant potential to form a symbiotic relationship with *Rhizobium* bacteria, as evidenced by an increase in the number of root nodules. According to numerous studies, the ability of plants to morphologically alter their root architecture, form root symbioses, activate high-affinity phosphate (Pi) transporters, increase internal phosphatase activity, and release organic acids and phosphatases into the rhizosphere are all factors that contribute to their ability to adapt to acid soil. (Vance et al. 2003; Gahoonia and Nielsen 2004). The efficient use of phosphorus is connected to the importance of the soybean root system in acidic soil. Uguru et al. (2012) evaluated soybean tolerance to acid soil. They

concluded that root length, root weight, and the number of root nodules were adaption characteristics of soybean in the acid soil. Using all three of these characteristics, they successfully mapped the amount of genotype tolerance of soybean to low pH. The Demas1 variety produced the most root nodules, which led to a high N fixation rate.

The number of root nodules increased when genistein was administered, and the number of root nodules grew as genistein concentration increased. Most root nodules were formed by the G3 treatment (180 ppm) 35 days after planting, 42 days after planting, and 49 days after planting. The Days after planting G3 treatment at age 35 (180 ppm) produced the most root nodules, 33.22 grains, 14.55% more than G0 (0 ppm), 7.82% more than G1 (60 ppm), and 5.02% more than G2 (120ppm). The G3 treatment (180 ppm) aged 42 Days after planting produced the most root nodules, 38, 18 grains, 20.9% more than G0 (0 ppm), 13.67% more than G1 (60 ppm), and 5.73% more than G2 (70 ppm) (120ppm). The G3 treatment (180 ppm) aged 49 Days after planting, produced the most root nodules, 43.60 grains, 7.1% more than the G0 (0 ppm), 5.21% more than the G1 (60 ppm), and 2.8302% more than the G2 treatment (120ppm).



GRAPHIC 8. The percentage of root nodules (grains) in the genistein treatment

An increase in root nodules suggests that genistein concentration significantly promotes *Bradyrhizobium japonicum*'s symbiotic relationship with soybean plants, particularly as an inducer of multiple bacterial nod gene systems (Dolatabadian et al., 2012; Lang et al., 2008). Flavonoid concentrations have been found to directly impact legume nodulation and N₂ fixation. It has been demonstrated that giving genistein to soybean seeds with *Bradyrhizobium* inoculation increases the quantity and dry weight of nodules as well as soybean N fixation at low temperatures. (Zhang and Smith, 1996).

The control treatment (G0), which produced the fewest root nodules compared to the other treatments, was thought to be less successful at N₂ fixation than the root nodules of soybean plants. Active root nodules determine the ability of soybean cultivars to fix N₂. The greater the ability of soybeans to fix N₂ will be achieved, the more active the root nodules are. 2019 (Purwaningsih et al.). When producing primary metabolites in soybean plant cells involves too much carbon, genistein, a flavonoid molecule, is created. (Collin and Edward, 1998). Sugiyama et al. (2008) suggest that soybean plants secrete signaling molecules in the form of flavonoids from root tissue that will attract rhizobia while living in symbiosis with *Rhizobium* bacteria. Genistein isoflavone acts as a signal molecule for chemical communication between *Bradyrhizobium japonicum* and soybeans. It is found as an exudate. The number of nodules that formed due to the application of genistein may have increased due to an increase in the number of infections that were started or the percentage of infections that progressed to nodule development. Genistein stimulates the production of bacterial Nod factor by the nod genes. (Loh et al. 2002). According to Vollmann et al., the size and number of chlorophyll leaves on soybean plants have a positive correlation with the root nodules, which in turn affects the rate of photosynthesis, the amount of nitrogen fixed, the number of pods per plant, the weight of 100 seeds, the protein content, and the oil content of the seeds.

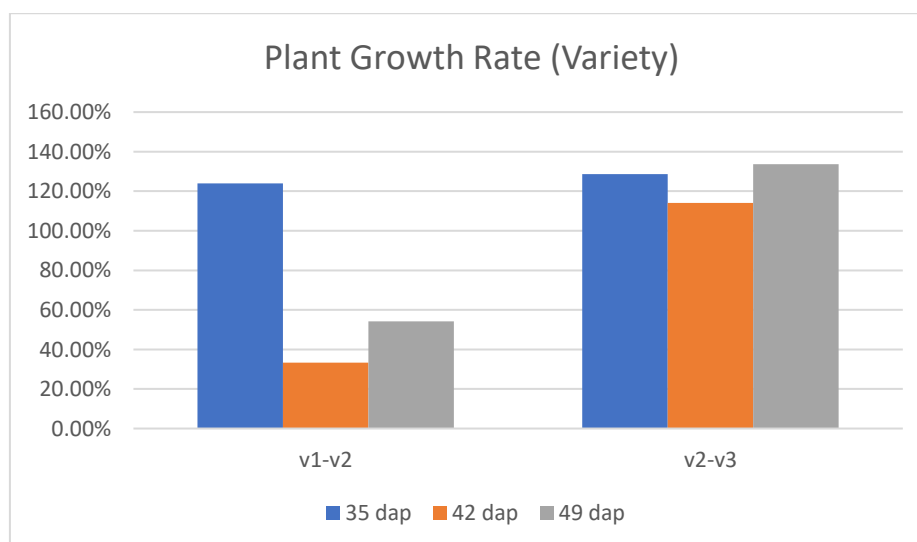
3.5 Plant Growth Rate

TABLE 5
PLANT GROWTH RATE VARIABLES (g/ m²/week) OF THREE TYPES OF SOYBEAN WITH THE ADDITION OF GENISTEIN

Treatment	35 days after planting	42 days after planting	49 days after planting
variety			
V1	0.098b	0.359a	0.853a
V2	0.224a	0.269a	0.553b
V3	0.1b	0.167b	0.365c
genistein			
G0	0.113	0.228	0.492
G1	0.144	0.277	0.564
G2	0.156	0.276	0.569
G3	0.15 0	0.28 0	0.736

Information: V = soybean variety, V1 = Deja1, V2 = Dena1, V3 = Demas1, G = genistein concentration, G0 = 0ppm, G1 = 60ppm, G2 = 120ppm, G3 = 180ppm

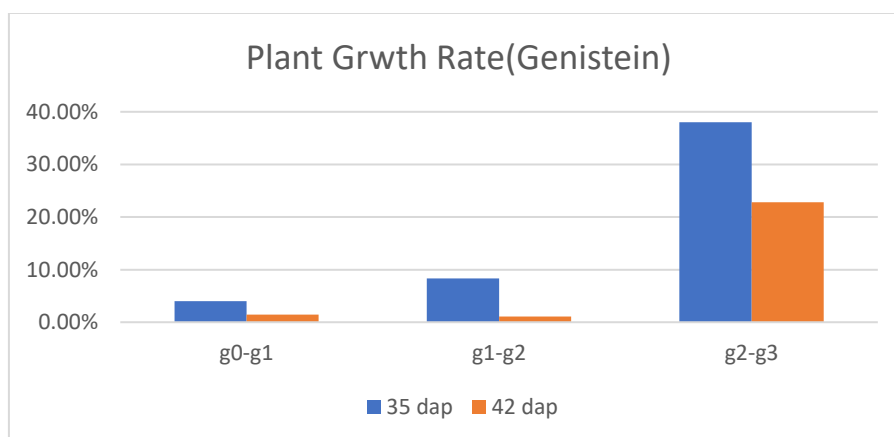
According to Table 5, the treatment of soybean varieties considerably impacted the growth rate of plants between the ages of 14 and 56 days after planting. The maximum Plant Growth Rate (LPT) was produced by Treatment V2 (Dena1), 0.22g/m²/week, 128.57% faster than Treatment V1 (Deja1) and 124% faster than Treatment V3 (Demas1). This indicates that Treatment V2 (Dena1) grew optimally in the vegetative to the generative growth transition period, which is assumed to be related to the concentration of genistein and the amount of chlorophyll. The genistein concentration of 0.17 8 mg/g dry leaves, capable of producing photosynthates for the best vegetative growth of soybean plants, exhibited the ability of the leaves to protect chlorophyll by absorbing UV-B rays. Early generative growth phase measurements of plant growth rate (LPT) revealed that treatment V1 (Dena1) produced the highest LPT, 0.359g/m² / week, 33.36% quicker than treatment V1 (Deja1), and 114% greater than V3 (Demas1). Plant Growth Rate (LPT) results from the final generative growth phase showed that treatment V1 (Dena1) produced the highest LPT, 0.853g/m² / week, 54.250% faster than treatment V2 (Deja1), and 133.699% faster than treatment V3 (Demas1). This suggests that treatment V1 (Dena1) had the best growth in the early and late generative growth phases, which is thought to be related to the leaf area of treatment V1 (Dena1) in that phase which reached 31.99 cm² and 33.26 cm². Soybeans have an indeterminate growth type, meaning plants keep growing even after entering the generative phase. The distribution of photosynthate by the soybean plant promotes the growth of its vegetative tissue and the filling of its pods. The accumulation of dry matter is a reflection of plants' ability to use sunlight energy for photosynthesis and their interactions with the surrounding environment. (Sumarsono, 2008).



GRAPHIC 9. The percentage of plant growth rate increases in the various treatment

The amount of genistein present substantially impacts the rate of plant growth. The LPT of soybean plants in the vegetative growth to generative growth transition phase revealed that the G2 treatment (120 ppm) produced the highest LPT, 0.156g/m² / week, 38.05% faster than the G0 treatment (0 ppm), 8.33% faster than the G1 treatment (60 ppm), and 4.0% faster than the treatment G3 (180 ppm). This demonstrates that the administration of 120 ppm genistein can be effectively controlled to protect chlorophyll from harm caused by exposure to UV-B rays during the transitional period of vegetative to generative growth. According to Table 3, the G2 treatment (120 ppm) was able to leave a genistein residue in the leaves of 0.217 mg/g dry leaves. This residue protected the chlorophyll from UV-B exposure so that it reached 23.60 mg/L and was sufficient for processing agriculture to produce wide leaves of 24.92 cm² and the highest soybean LPT. Early generative growth of soybean plants revealed that treatment G3 (180 ppm) produced the highest LPT, namely 0.280g/m²/week, which was 22.81% greater than treatment G0 (0 ppm), 1.08% higher than G1 (80 ppm), and 1.45% higher than G2 (120ppm). The final generative development phase of the soybean plant growth rate demonstrated the G3 treatment (180 ppm). These findings suggest that plants need significant energy to add and fill pods during the early and late generative growth phases.

Consequently, a high genistein concentration is required to protect the leaf chlorophyll, prevent damage, and ensure optimal photosynthesis. At the early and late generative growth periods, the G3 treatment (180 ppm) generated 30,47 mg/L and 34,21 ml/L of chlorophyll, as well as 30,58 cm² and 32.2 cm² of leaf areas, respectively. This data generated the best LPT of soybean plants during the filling and pod-formation phases. According to Hamid et al. (2020), photosynthesis can improve photosynthesize. The distribution of photosynthate among the various sections of the soybean plant favours the leaves more than the other plant components. This demonstrates that the source (leaf) is still active and impacts the sink's capacity.



GRAPHIC 10: The percentage of plant growth rate in the genistein treatment

IV. CONCLUSION

The study aimed to investigate the agronomic characteristics of three soybean varieties with varying concentrations of genistein. The findings suggest that the variety Demas1 produced the most chlorophyll and root nodules, while Deja1 had the widest leaf area and fastest plant growth rate. The optimal genistein concentration was found to be 180 ppm, which resulted in leaves with a large area, high chlorophyll content, significant root nodules, and fast plant growth rate. The Demas1 variety produced the best agronomic characteristics with 180 ppm of genistein. The study highlights the importance of understanding the impact of genistein on soybean agronomics and the potential trade-offs between genistein content and desirable agronomic traits. Further research is needed to explore the relationship between genistein and soybean agronomics to improve soybean production and genistein content.

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Assessment of soil erosion in the Autonomous District of Abidjan, Côte d'Ivoire

Kinakpefan Michel TRAORE^{1*}, Nambahigué Mathieu BAKARY², Awa Timité TAMBOURA³

^{1,3}Department of Geography, Jean Lorougnon Guédé University of Daloa (Côte d'Ivoire),

²Institute of Tropical Geography (IGT), Félix Houphouët-Boigny University (Abidjan-Côte d'Ivoire),

*Corresponding Author

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Abstract— *In a context of global changes and climatic uncertainties, this study poses the problem of the occurrence of water erosion induced by natural constraints and uncontrolled and speculative anthropisation of soils in the Autonomous District of Abidjan. The aim is to propose a decision-making tool that identifies the areas of the Autonomous District of Abidjan (ADA) most prone to soil loss and the risk of water erosion. More specifically, the aim is to evaluate, prioritise and map the levels of susceptibility to the occurrence of this hydroclimatic risk in the ADA. The mapping approach is based on the aggregation of factors such as climatic aggressiveness, soil erodibility, topography, soil cover and anti-erosion practices from the Revised Universal Soil Loss Equation (RUSLE). The exploitation of these data reveals that soil loss in the ADA oscillates between a minimum value of 0 t/ha/year largely observable in the south of the lagoon system to a maximum value of 107.53 t/ha/year which concerns the plateau areas in the north. The average of this series of 0.48 t/ha/year translates into an overall low level of water erosion in the ADA over nearly 95% of the ADA territory. This soil degradation, which impacts human settlements induced by the morphology of the site, is amplified by human activities. In a context of rapid urban expansion linked to a remarkable demographic growth coupled with climatic uncertainties, development policies must better integrate sustainable soil management.*

Keywords— *Autonomous District of Abidjan, Water erosion, RUSLE model, Land use, Land cover, Vulnerabilities.*

I. INTRODUCTION

In sub-Saharan Africa, water erosion is a major risk with more than 20% of land degraded, affecting more than 65% of the population (FAO, 2015). For Chafai and al., (2022), this phenomenon appears as a warning signal of the imbalance between the soil environment and its exploitation system. This risk can affect infrastructure such as roads, riverbanks, crop areas and urban land (Morgan, 2005; Sourlamtas, 2019). A direct product of complex interactions between natural and anthropogenic factors (Phinzi and Ngetar, 2019), it impacts on food production, drinking water quality, ecosystem services, landslides, eutrophication, biodiversity and declining carbon stocks (Boardman and Poesen, 2006; Panagos, 2015). This is a triptych due to firstly the extraction of particles from the soil surface, secondly the transport of these particles by water and thirdly the deposition of the soil when there is no more energy to move the particles (Renard and al., 1997; Morgan, 2005; Sourlamtas, 2019). The management of this major risk by restoring degraded soils is essential for the sustainability of territories. With this in mind, in May 2022, Abidjan, the economic capital of Côte d'Ivoire, hosted the fifteenth session of the Conference of the Parties (COP15) of the United Nations Convention to Combat Desertification (UNCCD). Adopted on 17 June 1994 and entered into force on 26 December 1996, this convention has 197 States and Parties. More generally, it aims to combat all forms of land degradation, understood as the reduction or disappearance of land resources as a result of land use, one or more phenomena caused by human activity and settlement patterns. Land degradation is defined as the reduction or loss of land resources as a result of land use, human activity and settlement patterns, such as soil erosion caused by wind and/or water, deterioration of

the physical, chemical and biological or economic properties of soils, and the long-term loss of natural vegetation (United Nations, 1994).

This article is in line with this desire for sustainable soil management. It is intended as a decision-making tool for soil restoration and the anticipatory and preventive management of crises inherent in the risk of erosion in the Autonomous District of Abidjan (ADA). The choice of this area is justified not only by its morphology, which predisposes it to this hydroclimatic risk, but also by its pre-eminence in the urban framework of Côte d'Ivoire. Indeed, the ADA is home to around 42% of the urban population of Côte d'Ivoire (INS, 2021) and polarises almost 80% of the economic activities of Côte d'Ivoire (World Bank Group, 2019). This demographic and economic concentration is at the root of a rapid artificialisation of the land at a rate of almost 7% per year (Traoré, 2022). These mostly informal and spontaneous changes expose soils initially protected by plant cover to the kinetic energy of raindrops, to the ablation of particles from the upper horizons and increase the exposure and vulnerability of territorial issues to the risk of water erosion (Sourlantas, 2019).

Through modelling based on the integration of the main soil loss factors of the Revised Universal Soil Loss Equation (RUSLE) model into a Geographic Information System, this study prioritises the susceptibility of occurrence of water erosion in the ADA. Specifically, it (i) maps the main erosive factors, (ii) maps soil loss susceptibility and (iii) identifies the soils most prone to erosion risk in the ADA.

II. MATERIAL AND METHODS

2.1 Study area

The Autonomous District of Abidjan is located on the coast of Côte d'Ivoire between 5°10'0" and 5°40'0" North Latitude and 4°30'0" and 4°60'0" West Longitude (Figure 1).

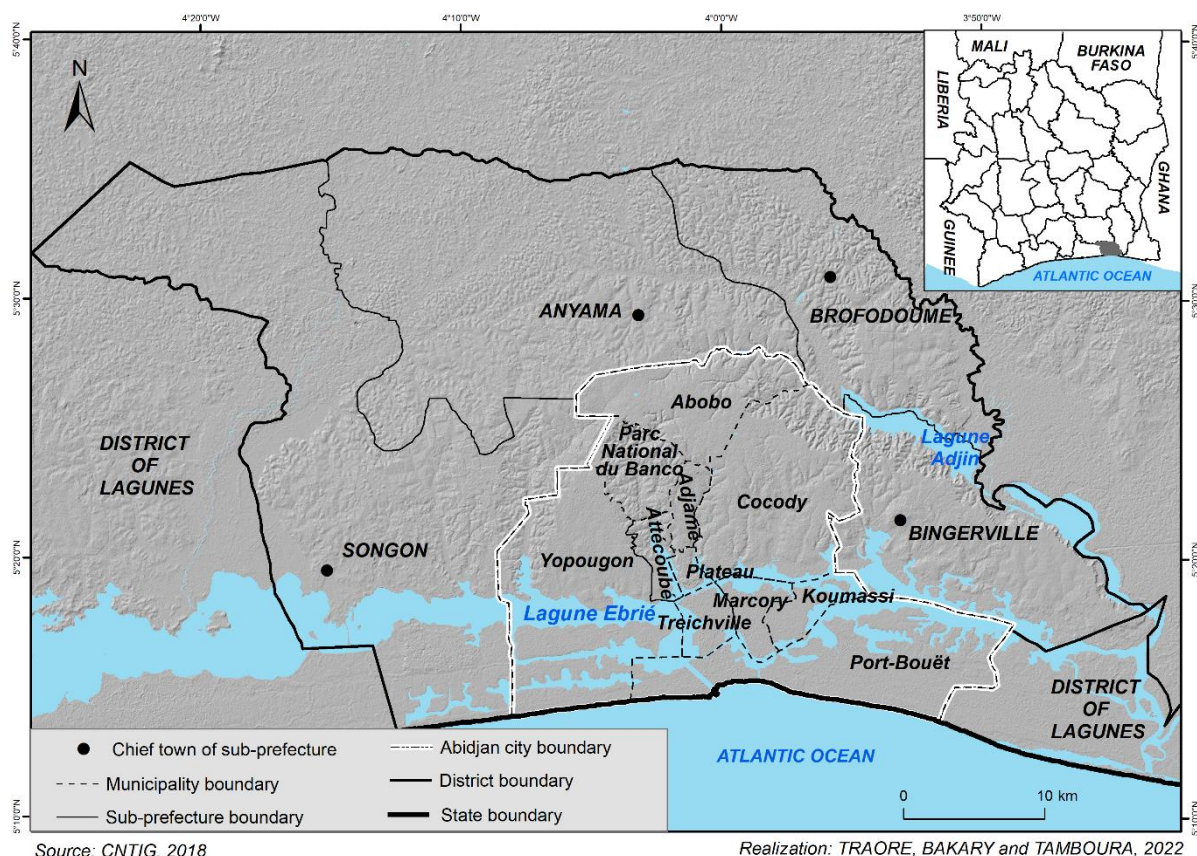


FIGURE 1: Location of the study area

In addition to the city of Abidjan, the ADA has four Sub-Prefectures (Anyama, Bingerville, Brofoudoumé and Songon). It covers about 2,034 km² with an estimated population of 4,707,404 in 2014 (SDUGA, 2015). The ADA is located on the sedimentary basin which covers a substratum of Birrimian formations made up of rocks (flysch, schists, micaschists, quartzites, etc.). Geological evolution has given the site a morphology divided between coastal plains and inland plateaus separated by the Ebré lagoon system (Figure 1). Altitudes vary from 0 to 5 m on the coastal plains and 173 m on the inland plateaus. The

southern part corresponds to an area of coastal sediment cover belonging to the Cretaceous. The northern part corresponds to an area of fairly thick cover of sediments belonging to the Tertiary. The dominant forms are plateaus made up of more or less ferruginised sandstones, clayey sediments, and shales and grauwackes in the north (BNEDT, 2008).

This area is dominated by four types of soil cover. These are ferrallitic soils, hydromorphic soils, podzolic soils and soils that are not very advanced on marine sands. Ferrallitic soils with a sandy-clay texture cover more than three quarters (77.6%) of the area (E. Roose et al, 1966).

This territory is located in the intertropical zone characterised by average temperatures that remain above 20°C. The ADA region has a humid equatorial climate with four seasons. The average annual rainfall is well over 1200 mm. The entire area was once covered by dense evergreen rainforest, which now exists only as a relic, such as the Banco National Park.

2.2 Input data

In this study soil loss and erosion risk were assessed and mapped according to the Revised Universal Soil Loss Equation (RUSLE). The RUSLE model, based on the empirical equation of Wischmeier and Smith proposed at the Seventh Congress of Soil Science in Madison (1960), is based on the statistical analysis of more than 10,000 annual results from various stations in the American Great Plain (Roose, 1975). The revised version RUSLE (Revised Universal Soil Loss Equation) was produced by Renard et al (2011). It estimates the long-term average annual loss of soil through sheet erosion and gully erosion (Wischmeier and Smith, 1978; Panagos et al, 2015). This equation (equation 1) integrates five erosive factors of water erosion which are: climatic aggressiveness, soil erodibility, the topographic factor integrating slope inclination and length, land use and anti-erosion practices. It is written :

$$A = R * K * LS * C * P \quad (1)$$

- A (t/ha/yr): Soil loss per unit area per year;
- R: Rainfall index characterising the climatic aggressiveness;
- K: Erodibility index in relation to soil resistance to sheet and channel erosion;
- LS: Topographic index combining both the effect of length L and slope steepness S;
- C: Land cover index and
- P: Erosion control index.

Although designed in the United States, this equation is one of the most suitable models for the annual estimation of water erosion (Payet et al, 2011). These erosive factors were determined from input data (Table 1).

TABLE 1
DATA USED

N°	Data	Factor	Format	Sources	Dates	Resolution
1	Climatological	R	NC	crudata.uea.ac.uk/cru/data/hrg	2011-2020	0,5° x 0,5°
2	Pedological	K	Shapefile	FAO (DSMW)	Jan. 2003	0,5° x 0,5°
3	Topographic	LS	GEOTIFF	ALOS PALSAR –R.T.C. (https://search.asf.alaska.edu/#/)	18/07/2007 04/08/2007	12,5 m
4	Soil cover	C	TIFF	Sentinel 2	01/01/21- 31/12/21	10 m
5	Supporting or protective practice	P	GEOTIFF	ALOS PALSAR –Radiometric Land Correction	18/07/2007 04/08/2007	12,5 m

2.3 Modelling processes for soil loss and erosion risk

While in the literature there are several empirical, semi-empirical and physical approaches to assess soil loss, Nacishali Nteranya, (2021) notes that the direct measurement approach remains practically the most reliable. However, due to prohibitive difficulties linked to the enormous cost in terms of time and money for data collection, especially in southern countries, the modelling approach using a GIS makes it possible to give a first approximation of the risk of erosion at the scale of a territory (Ganasri and Ramesh, 2015; Nacishali Nteranya, 2021; Adamou and al., 2022).

In this study, using GIS and remote sensing tools, the five parameters (R, K, LS, C and P) of the RUSLE model were determined and a hierarchy of soil loss and erosion risk was established. However, these five parameters relate to different realities. Indeed, while the R, K, L and S factors relate to the physical characteristics of the site, i.e. precipitation, pedology and topography, the C and P factors emanate from anthropogenic soil use and protection practices. In this study, soil loss is therefore a combination of both physical processes, the so-called "potential" erosion factor, and anthropogenic processes, the so-called "induced" erosion factor. Soil loss and consequently the risk of erosion is the product of these two processes which are in fact complementary and interact in a holistic manner. These parameters are not independent but for the convenience of the analysis they have been treated separately before being aggregated (Roose, 1977).

2.3.1 Potential Erosion Risk in the ADA

Potential Erosion (PE) is modelled on the basis of the universal soil loss equation, taking into account only the factors R, K and LS (De Figueiredo and Fonseca, 1997; Pradhan et al., 2012; Le Van et al., 2014; Karamage et al., 2016, 2017; Nacishali Nteranya, 2021).

2.3.1.1 R-factor of climatic aggressiveness

Rainfall data between 2011 and 2020 with a resolution of $0.5^\circ \times 0.5^\circ$ (Table 1) was used to determine the average rainfall amounts. From these average rainfall amounts, the climatic aggressiveness index R was determined. It should be recalled, however, that the original design of the USLE model requires rainfall intensity data to calculate the R-index (Wischmeier and Smith, 1978; Nacishali Nteranya, 2021). This condition is generally met in the absence of rainfall data. Authors then proposed mathematical models that make it possible to correlate R and average annual rainfall (P) (Nacishali Nteranya, 2021). Thus, in his work on soil losses in Adiopodoumé (Côte d'Ivoire), E. Roose came to the conclusion that rainfall alone does not explain erosion phenomena and that the relationship between rainfall and erosion only becomes highly significant when the maximum intensities are measured over more than 20 minutes for erosion and 10 minutes for runoff (Roose, 1973, 1975, 1977). For the West African zone, the author's observations established a theoretical relationship ($R / P = 0.50$) between the mean annual climatic aggressiveness index (R) and the mean annual rainfall (P). From this simple relationship, the climatic aggressiveness parameter R was derived through equation 2:

$$R = P * 0.5 \quad (2)$$

From this equation developed for Côte d'Ivoire and Burkina Faso (Roose, 1977), the R index expressed in MJ.mm/ha.h.year was determined.

2.3.1.2 Soil erodibility factor K

The erodibility index K is expressed in t/ha/h and varies from 0 for less sensitive soils to 1 for soils highly prone to water erosion (Ganasri and Ramesh, 2015; Nacishali Nteranya, 2021). Soil erodibility is defined as its susceptibility to erosion (chuma and Rosseau, 1978) or its resistance to erosion (Khemiri and Jebari, 2021). It expresses the vulnerability of the soil to be eroded by rain and is inherent to the texture, structure, organic matter content of the upper horizons and permeability of the soil (Wischmeier and Smith, 1978; Nacishali Nteranya, 2021).

In practice, there are several methods and equations for determining the K-factor. These different approaches depend on the availability of soil data (Benavidez, 2018). In this study, the K-factor was calculated from FAO data (Table 1) and the rates of sand, silt, clay and organic matter in the upper horizons of the soils that are most exposed to the effects of rainfall. This calculation was made through the equation (equation 3) of J. R. Williams: (Williams, 1995; Eitsch, 2000; Waver et al, 2005). It is written.

$$K = fcsand * fcl-si * forgc * fhisand \quad (3)$$

With:

$$fcsand = 0.2 + 0.3 * \exp \left[-0.256 * ms * \left(1 - \frac{msilt}{100} \right) \right]$$

$$fcl - si = \left(\frac{msilt}{mc + msilt} \right)^{0.3}$$

$$forgc = 1 - \frac{0.036 * orgC}{orgC + \exp [3.72 - (2.95 * orgC)]}$$

$$f_{hisamd} = 1 - \frac{0.7 * (1 - \frac{ms}{100})}{(1 - \frac{ms}{100}) + \exp[-5.51 + 22.9 * (1 - \frac{ms}{100})]}$$

2.3.1.3 Topographical factor LS

In the process of soil erosion the parameter LS is a topographical factor which takes into account the length (L) and angle (S) of the slope. In fact, the slope typology influences the surface runoff velocity and plays an important role in erosion (Biswas and Pani, 2015; J. Nacishali Nteranya, 2021; Adamou et al, 2022). Roose (1975) points out that many authors have shown that soil loss increases exponentially with slope steepness. The LS factor was calculated according to the equation (equation 4) developed by (Stone and Hilborn, 2012):

$$LS = [0.065 + 0.0456 (\text{Slope}) + 0.006541 (\text{Slope})^2] * (\text{Length of slope} \div \text{Constant})^{NN} \quad (4)$$

Where:

Slope = slope inclination in %.

Slope length = length of slope in m (Flow accumulation)

Constant = 22.1 metric units

NN = see Table 2

TABLE 2
VALUES OF NN

S	< 1	1 ≤ Slope < 3	3 ≤ Slope < 5	≥ 5
NN	0.2	0.3	0.4	0.5

Source : <http://www.omafra.gov.on.ca/english/engineer/facts/12-051.htm>

The different calculations were carried out using the Arc Hydro extension tools that work in the ArcGIS environment. The slope raster (in percent) obtained was reclassified according to the values in Table 2 in order to deduce the NN value according to the dominant slope class. After these operations, it appears that more than 60% of the ADA territory has a slope value higher than 5%. Thus, the NN value retained is 0.5. Finally, using the Map Algebra Raster calculator, the LS factor was calculated by numerically applying the equation of STONE and HILBORN, (2012) as follows:

$$LS = (0.065 + 0.0456 * "Slope" + 0.006541 * ("Slope")^2) * (("FlowAcc" * 30 / 22.1) ^{0.5})$$

From these factors the Potential Erosion (PE) was determined. PE refers to the power of a soil to produce sediment as a result of the degradation of rainfall droplets in its morphological environment (Tahiri and al, 2017). It results from the factors R of climatic aggressiveness, K of soil erodibility and LS of length and slope angle and is written as follows (equation 5):

$$EP = R * K * LS \quad (5)$$

2.3.2 Induced Erosion Risk in the ADA

Potential erosion due to climatic, pedological and topographical factors can be mitigated or exacerbated by human development choices and practices. Indeed, man, through his actions, causes the destruction, total or partial, of the surface structure of the soil and, consequently, accentuates the erosive phenomenon (El Hage Hassan, 2018). This erosion process due to soil artificialisation is referred to in this study as "Induced Erosion". It is determined by equation 6.

$$IE = C * P \quad (6)$$

2.3.2.1 Cover factor C

Vegetative soil cover plays a vital role in erosion prevention (Sourlantas, 2019). It appears to be the most active factor in controlling the risk of water erosion as it provides the soil with mechanical protection against climatic aggressiveness and ablation by reducing the energy released by raindrops (El Hage Hassan, 2018). The C-factor is therefore intrinsically linked to

land use and land cover patterns. As the definition of land use classes varies from one area to another, from one country to another and from one zone to another depending on the landscape units (Nacishali Nteranya, 2021), in this study, the NDVI (Normalized Difference Vegetation Index) approach was preferred.

Calculated from the red and near infrared channels ($NDVI = \frac{NIR-RED}{NIR+RED}$) (equation 7), this index is sensitive to vegetation variation and quantity. The NDVI values and map were determined from 2021 Sentinel 2 images on the Google Earth Engine platform and exported to One Drive and ArcGIS. From the NDVI raster, the C-factor was calculated using the CrA method of Colman (2018) adapted from Durigon et al (2014). This method is applied according to equation 8:

$$CrA = 0.1 \left(\frac{-NDVI+1}{2} \right) \quad (8)$$

The CrA is based on the NDVI for areas under tropical climate with intense rainfall.

2.3.2.2 Support practice factor P

The support practice factor (P), or protection factor, is considered to be one of the most uncertain parameters in the RUSLE model and thus in the assessment of water erosion risk (Haan and al, 1994; Alloi and al, 2022). This uncertainty arises from the multitude of approaches and methods used to determine the values of the P-factor. Many of them have a universality problem as their predictive capacity is limited only to the regions for which they have been developed (De Vente and Poesen, 2005; Phinzi and. Ngetar, 2019); Alloi and al., 2022). In fact, like the C-factor, the most common approach to estimating P-factor values uses in situ observation data and/or visually interpreted images (Phinzi and Ngetar, 2019). From this approach P-factor values are derived either from remote sensing image classification, land use and land cover maps, previous studies or expert knowledge (Panagos and al, 2015); Alloi and al, 2022).

However, following the literature, this classification approach using remote sensing methods requires very high resolution images and feedbacks that are not always freely available (Panagos et al, 2015b; Phinzi and Ngetar, 2019). This issue of availability and free availability of satellite imagery with adequate spatial resolution to detect soil conservation practices with certainty is more pronounced in southern territories like the ADA. From the above, in this study, we opted for equation 9 developed by Wener (1981) in Kenya.

$$P = 0.2 + 0.03 \times S \quad (9)$$

with **S** expressing the value of the slope in percentage.

This empirical equation has been used by various researchers in different areas, notably in the Lake Victoria catchment (Lufafa et al, 2003), in Italy (Terranova, 2009; Napoli, 2017) and in China (Fu et al., 2005).

From these C and P factors, the Induced Soil Loss (IL) from land use and land cover patterns was determined. Finally, the susceptibility of occurrence of water erosion risk in the ADA was determined by crossing the Potential Erosion raster related to physical factors and the Induced Erosion raster related to land use and development choices. All these calculations were performed with the Map Algebra Raster Calculator tool, from the Spatial Analyst Tools group in ArcToolbox. For the sake of accuracy of the statistical results, the lagoon water body was removed from the DEM.

III. RESULTS AND DISCUSSION

3.1 Potential Erosion Risk: Favorable physical conditions

3.1.1 R-factor

The R-factor values determined vary between 704 and 751 MJ.mm/ha.h.yr (Figure 2).

These values belong to the interval [500 - 1400] determined by Roose (1973, 1975) in Côte d'Ivoire. The mean of this statistical distribution is 723 MJ.mm/ha.h.yr. The standard deviation of 10.767 shows a relatively large dispersion of values around the mean. The R-index evolves crescendo along a south-east and north-west axis.

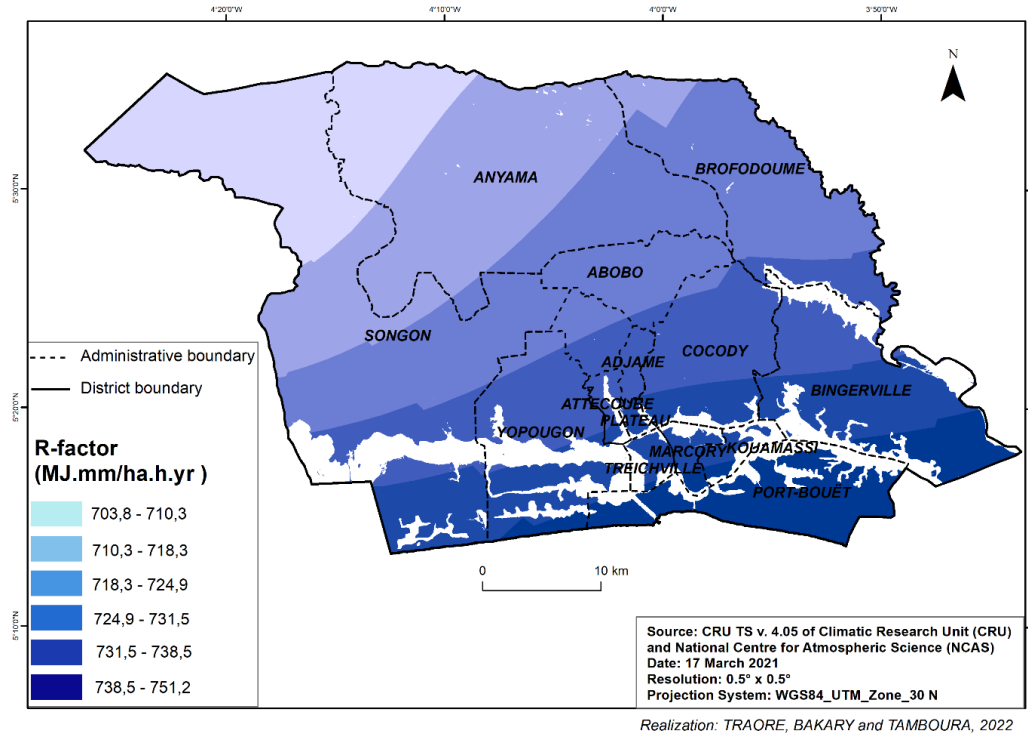


FIGURE 2: R-factor of climatic aggressiveness in the ADA

The low values of the R factor cover more than 60% of the District. The high levels mainly concern the southern districts of the city of Abidjan and almost a third of the communes of Bingerville, Cocody, Plateau and Yopougon.

3.1.2 Soil erodibility factor K

The values of the K index vary between 0.07 and 0.17 t/ha/h (Figure 3).

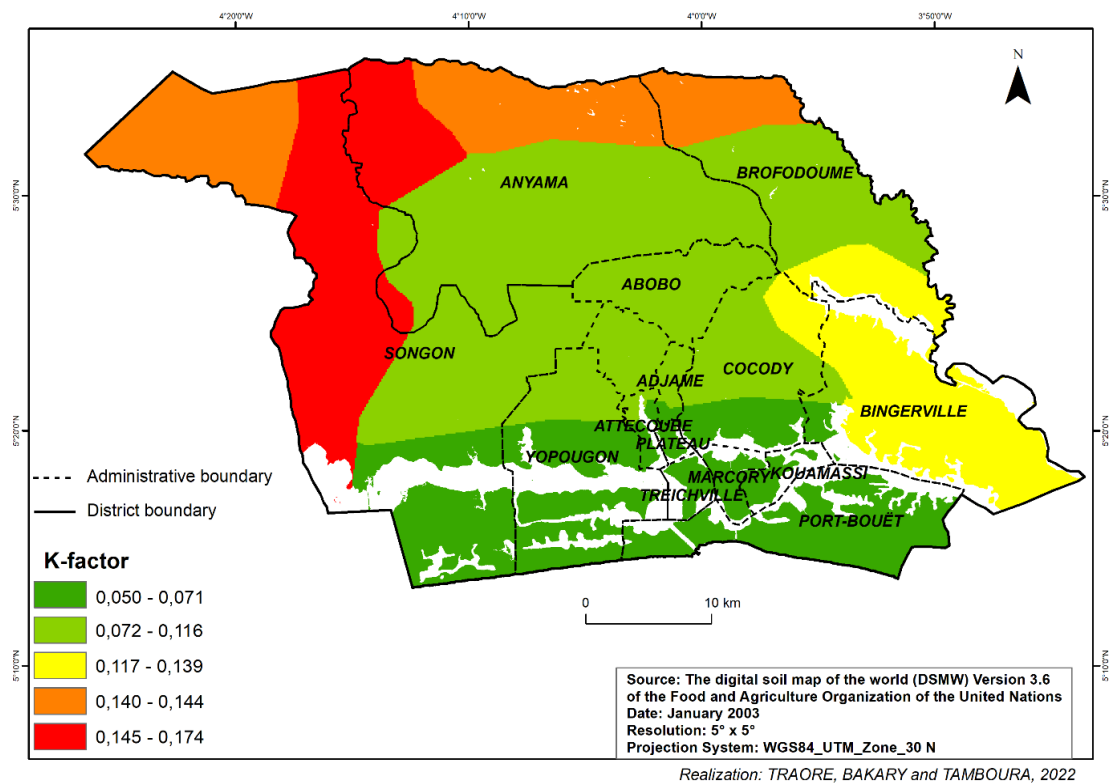


FIGURE 3: Distribution of soil erodibility K-factor values

Despite the difference in approaches, these values remain within the limits of 0.05 and 0.18 t/ha/h defined by E. J. Roose in his work on erosion and runoff in West Africa (Roose, 1972; 1975; 1977). The standard deviation of a value of 0.04 reflects a small dispersion of the values of the distribution around the mean of 0.10.

The spatial distribution of soil erodibility in the ADA according to the K-factor indicates that the high levels of erodibility related to soil texture and structure are mainly found in the extreme north and northwest of the sub-prefectures of Brofodoumé, Anyama and Songon. With K-factor values above 0.140, these are ferrallitic soils that are most often reworked or evolved. In contrast, low levels of erodibility are found in the south and centre of the ADA on typical ferrallitic soils, podzolic soils or soils that are not very developed, with K-factor values between 0.071 t/ha/h and 0.116 t/ha/h. Statistically, more than three-fifths of the ADA area has a low erodibility level of 41.27%.

3.1.3 Topographical factor LS

The values of the LS index vary between 0 and 32.3984 (Figure 4). The mean of this statistical distribution is 0.3137 with a standard deviation of 0.8125. This higher standard deviation value reflects a certain dispersion of the LS factor values around the mean. The mapping results show a strong dominance of the classes [0 - 0.381] and [0.382 - 1.271]. These two classes with small slope gradients cover 79.83% and 13.59% of the area respectively.

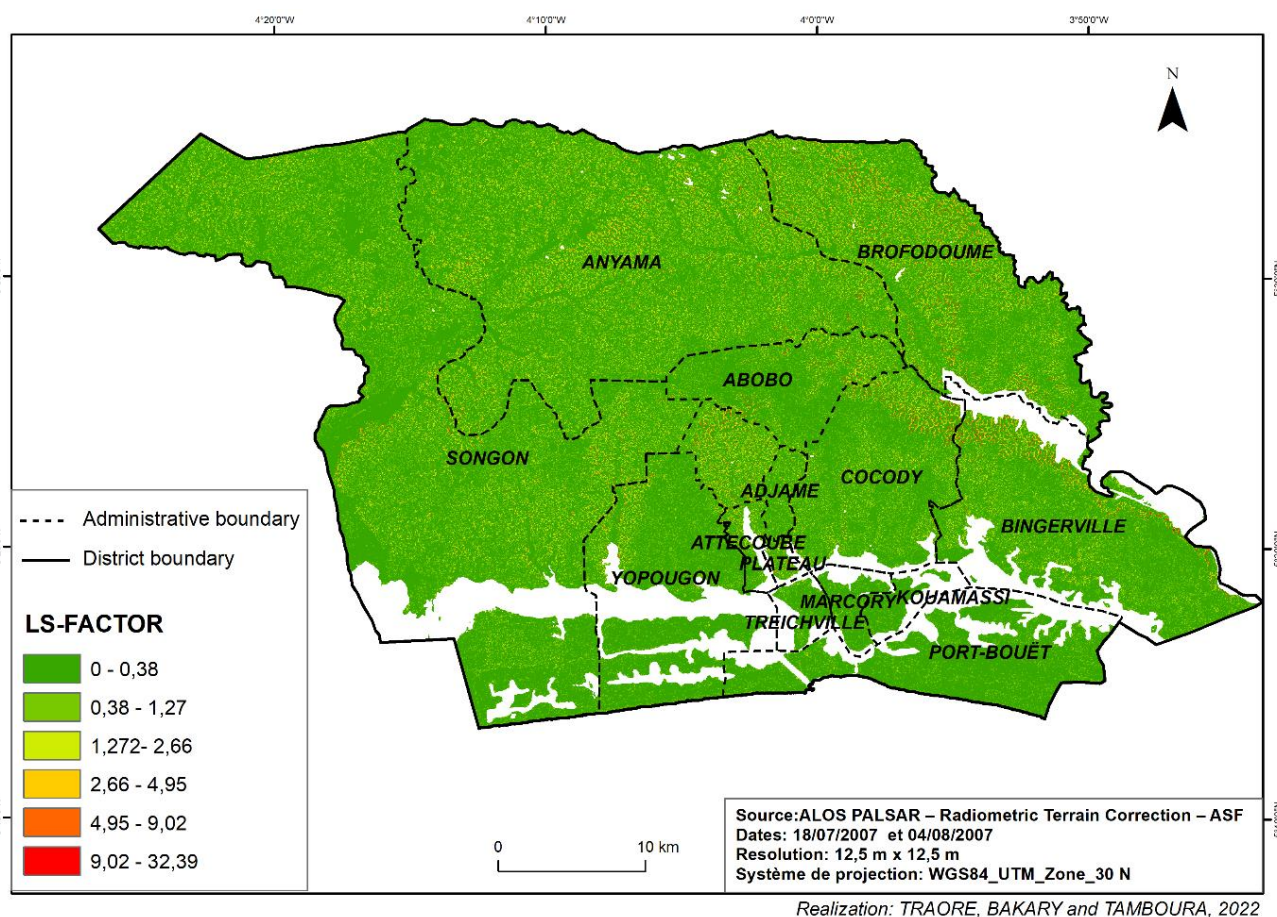


FIGURE 4: LS factor

The values resulting from the intersection of climatic (R), edaphic (K) and topographic (LS) factors have made it possible to map the risk of Potential Erosion (figure 5).

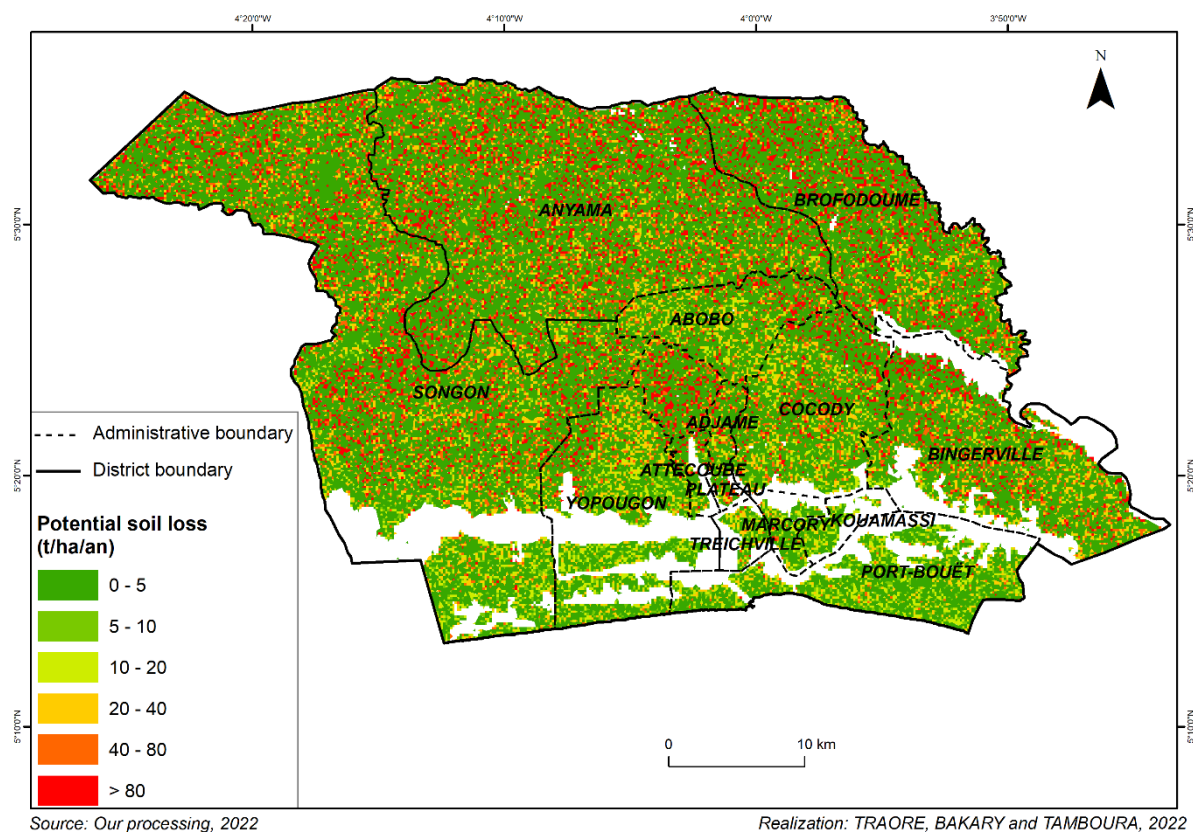


FIGURE 5: Potential soil loss and erosion risk in the ADA

These three factors: climatic aggressiveness, soil erodibility and topography were crossed and a potential erosion risk map was generated

The values are between 0 (Xmin) and 1943.35 (Xmax). The average of this statistical series is 29.51 for a standard deviation of 78.15 which reflects a strong dispersion of values.

These values are significantly influenced by the climatic aggressiveness index, which is higher than 700. Thus, as Roose (1977) observed, the serious erosion damage observed locally in Africa is due above all to the aggressiveness of tropical rains (whose energy is two to six times higher than in temperate zones) rather than to a hypothetical fragility of tropical soils.

The spatial analysis shows a rather heterogeneous distribution of Potential Erosion levels. On the whole, these levels are low over more than two thirds of the territory (68.30%). Thus, due to the relatively low slopes of our study area and the better resistance of ferralitic soils, potential water erosion is largely influenced by the R factor of the aggressiveness of the climate marked by the height, intensity and duration of rainfall (Roose, 1977).

As in Brazil, according to the findings of Panagos (2015), in tropical climates, it is the energy of raindrops that is the main factor in erosion in Africa and particularly in the ADA. Rainfall, through its intensity and energy, is considered the 'main causal agent' of land loss because its action amplifies the driving forces necessary to pull soil particles out of the ground (El Hage Hassan et al, 2018). As rain falls, the soil begins to become saturated and additional rainfall runs off its surface as runoff (Sourlamtas, 2019).

This potential vulnerability of soils to aggressive rainfall and water erosion will undoubtedly increase with climate change, which is now manifested by increasingly heavy rainfall with high intensity. In Petropolis, Brazil, for example, the rainfall that caused the massive flooding and landslides in February 2022 was equivalent to a month's worth of rain falling in just 6 hours. This is also the case in the Durban area of KwaZulu-Natal province in South Africa, where the weather system that triggered the floods in April 2022 resulted in more than 300 mm of rainfall over a 24-hour period, or about 75% of the country's average annual rainfall. These two hydroclimatic events, in addition to the extensive material damage, caused more than 110 and 440 deaths respectively. Thus, intensity is the main parameter that links rainfall to erosion through the momentary saturation of soil

porosity, as runoff can only occur when the flow of rain exceeds the possibilities of absorption by the soil pores, the threshold of which is lowered as the rainfall is prolonged (Roose, 1977).

The risk of soil erosion will be affected by climate change (Nearing, 2004; Colman, 2019) as changes in temperature and precipitation impact plant biomass production, infiltration rates, soil moisture, land use and crop management (Li et al, 2016; Colman, 2019).

In addition to these climatic disturbances, this potential erosion remains influenced by human development choices and practices. Indeed, man, through the practices he carries out, causes the total or partial destruction of the soil structure and, consequently, accentuates the erosive phenomenon (Hage Hassan, 2018). An inevitable natural phenomenon, water erosion becomes a serious environmental and socio-economic problem when it is accelerated by human activities (Meliho, 2016; Lal, 1998; Del Mar López and 1998; Yameogo, 2020).

3.2 A steadily increasing risk of Induced Erosion

3.2.1 The coverage factor C

The values of C determined from the NDVI vary between 0.02 and 0.06 (Figure 6). The average of this series is 0.04. The standard deviation of 0.006 indicates a very low dispersion of these values. The values of parameter C are inversely proportional to those of NDVI. This evolution of the two variables in opposite directions implies that when the biomass is of good quality, it effectively protects the soil from erosion.

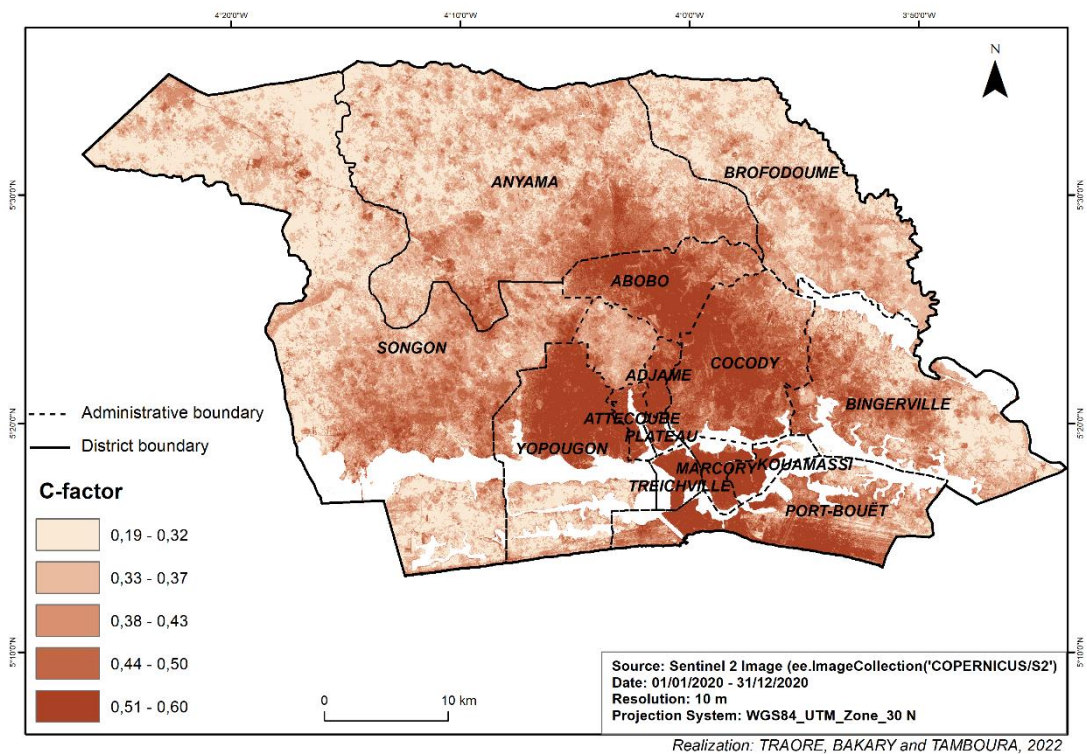


FIGURE 6: Spatial distribution of coverage C-factor

This high propensity for erosion is all the more worrying as these areas are home to most of the population (93%) and economic activity (80%). Also, the C factor is more important in areas where biomass is declining. These are naturally built-up areas, bare areas or, to a lesser extent, areas with severely degraded vegetation. They correspond globally to the ten communes of Abidjan (figure 6).

3.2.2 P-factor

The values of the soil erosion parameter P in the ADA range from 0.20 (Xmin) to 2.99 (Xmax) (Figure 7).

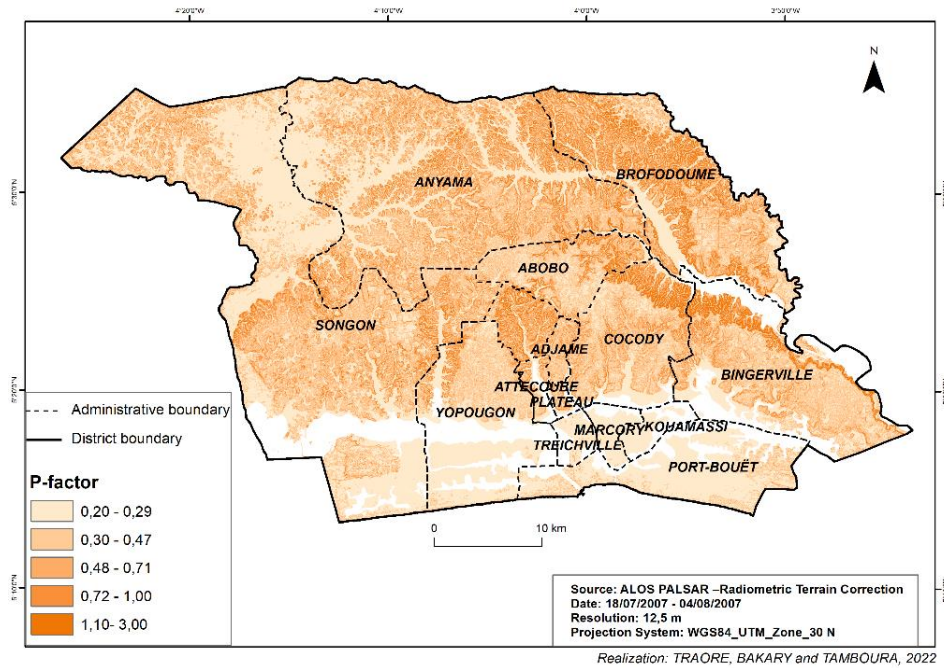


FIGURE 7: P-factor

Statistically, almost two thirds of the territory (65.22%) are of "Low" and "Very low" erodibility level while only 16.39% are of "Somewhat high", "High" and "Very high" level. The average erodibility level covers just under one fifth of the District.

The results of the product of the C and P factors resulted in Induced Erosion values between 0.004 and 0.141 for a mean of 0.014 and a standard deviation of 0.008 which translates into a low dispersion of values and a certain homogeneity of the series. In contrast to Potential Erosivity, the spatial distribution of Induced Erosivity seems more homogeneous. The significant soil losses induced by development choices and human practices are globally concentrated between the lagoon system in the south and the marshy areas in the north-west of the ADA (Figure 8).

This high susceptibility to induced erosion particularly concerns the communes of Plateau, Adjamé and Attécoubé, the sub-prefectures of Brofodoumé, Bingerville and Anyama. The communes of Yopougon, Aboobo and Cocody, which are home to nearly one in three (58%) of Abidjan's inhabitants, are also subject to this significant susceptibility to induced erosion. Statistically, more than two thirds (67.23%) of the territory have an overall low induced erosion risk.

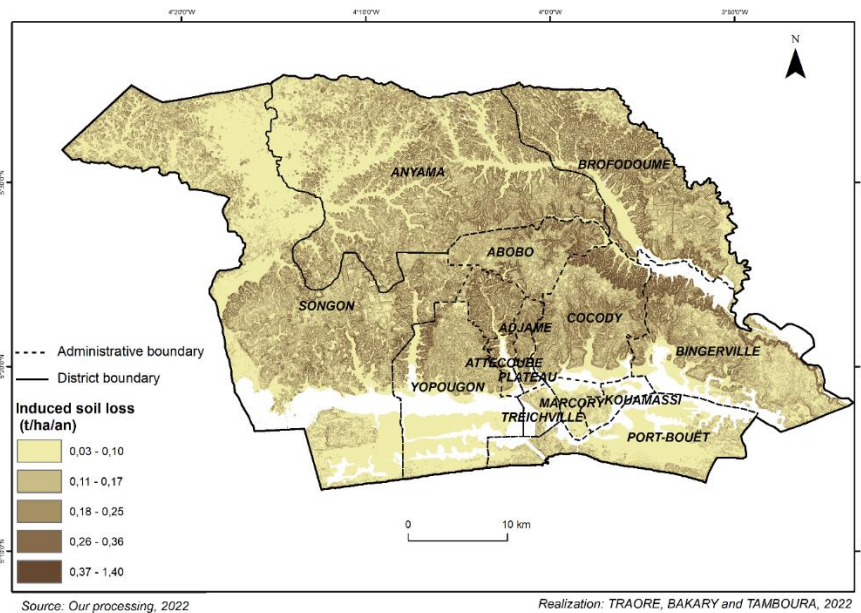


FIGURE 8: Induced soil loss and induced water erosion risk in the ADA

3.3 Susceptibility of occurrence of water erosion risk in the ADA

Crossing the Potential Erosion and Induced Erosion raster generated the raster of soil losses in the ACD. These values range from a minimum (Xmin) of 0 t/ha/yr to a maximum (Xmax) of 107.53 t/ha/yr. Using the RUSLE model in Burkina Faso, in the same West African spatial context, Ouédraogo et al (2019) found soil loss rate values between 0 - 36.35 t/ha/yr. Compared to the results obtained in our research, these soil losses can be considered low. The average distribution of 0.48 t/ha/year is lower but close to the average found by Yaméogo et al (2020) in Burkina Faso, which is 1.22 t/ha/year. However, these averages remain well below the 24 t/ha/yr and 76.59 t/ha/yr found respectively by Chuma et al (2022) and (Nacishali Nteranya, 2021) in the Democratic Republic of Congo. This difference is probably explained by the variation in edaphic, topographic and climatic conditions that can modify the values of the erosivity parameters.

It should be noted that with a Q1 equal to 0, 25% of the values are lower than 0, i.e. no soil loss. In addition, half of the soil losses are less than 0.42 t/ha/year, i.e. the median value (Q2). Three quarters of the annual soil losses are below 1.69 t/ha/year which is the Q3 value. Only 25% of the soil losses are above 1.69 t/ha/year. Based on the classification proposed by Yameogo et al. (2020), a discretization of the values of the soil loss series A allowed the generation of the raster in Figure 9.

Visually, this figure corroborates the quartile analysis which showed a predominance of low values of annual soil loss in the ADA. These low losses occur throughout the ADA but particularly in the areas south of the lagoon system.

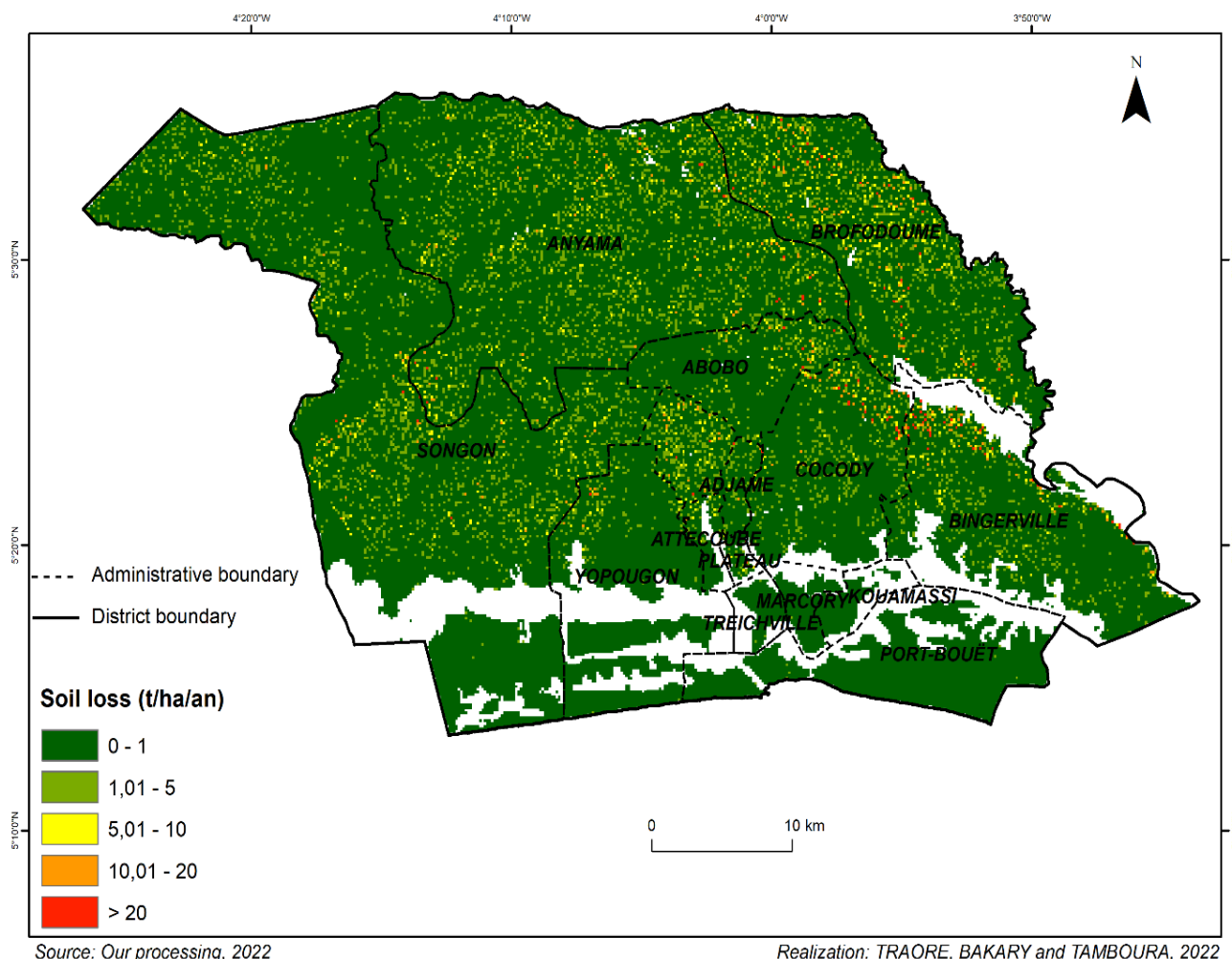


FIGURE 9: Erosion risk in the ADA

This situation could probably be explained by the very gentle relief (slopes of less than 2%) encountered in these areas. Even though the high values are not evenly distributed, they are mainly found on the Gbébouto plateau in the centre of Abidjan, somewhat in Songon, Anyama and along a south-east and north-west axis between Bingerville and Brofodoumé (Figure 9).

Statistically, this discretization results in five soil loss classes ranging from "Low" severity (0-1- t/ha/ha) to "Extremely high" severity (> 20 t/ha/ha) (Table 3).

TABLE 3
EROSION RISK IN THE ADA

Soil loss (t/ha/year)	Severity class	Area (ha)	Frequency (%)	Freq CC	Freq CD	Erosion risk
0 - 1	Low	161 808.7	90.47	90.47	100.00	Low
1 - 5	Fairly severe	13 794.7	7.71	98.18	9.53	Fairly high
5 - 10	severe	2 131.6	1.19	99.37	1.82	High
10 - 20	very severe	839.9	0.47	99.84	0.63	Very High
> 20	Extremely severe	283.1	0.16	100.00	0.16	Extremely high

Source: Our processing, 2021

These soil loss classes correspond to erosion risk levels ranging from "Low" to "Extremely high" (Table 3). Areas with a low level of erosion risk are the most prevalent. They cover more than 90% of the ADA territory, i.e. 161 808.7 ha. Soil losses greater than 5 t/ha/year, for an erosion risk ranging from "High" to "Extremely high" by our model, cover 17,049.3 ha, i.e. a little less than 10% of the District (table 3). This low spatial distribution of the significant risk of water erosion could be explained by the presence of a globally homogeneous relief with more than half of the territory (56.98%) on gentle slopes of less than 5%. To this must be added the use of land and the occupation of soils which are still mainly the preserve of nature reserves, orchards and plantations. These woody landscapes, which still occupy more than two thirds (68.17%) of the territory, contribute to the fixation and protection of the soil against the aggressiveness of the violent tropical rainfall. Runoff coefficients are obviously low on vegetated plots and high on bare soil and steep slopes (Sbai and Mouadili, 2021).

However, this soil stability conferred by the vegetation cover is gradually being eroded by the artificialization of the soil due to the agglomeration and urbanization phenomena that began when Abidjan was chosen as the capital in 1934 and the Port of Abidjan was created in 1950. Between 1932 and 2021, this agglomeration effect has seen land use indices such as built-up and bare land progress very significantly to the detriment of natural and planted forests, marshes, fields and formations dominated by grasses or fallow land. These indices increased by 97.95% from 1,178.57 ha to 52,494.0612 for an average annual growth rate over the period of 4.42%. The proportion of built-up area, which was only 6.19% in 1980, reached 10.89% in 2000, 25.81% in 2020 and 29.5% in 2021 (Traoré, 2022).

Today, the trend of water erosion is generally exacerbated by demographic growth, which increases the artificialisation of land. As in Burkina Faso (Yameogo et al, 2020), the extension of cultivation areas, farming techniques and above all urbanisation have profoundly modified the organisation of the ADA landscape. This is also the case in the Democratic Republic of Congo, where the change in land use due to population growth has favoured the extension of agricultural and urban areas to the detriment of forest areas (Nacishali Nteranya, 2021).

Dans les zones de plus en plus urbaines, l'érosion du sol est un problème important en raison de la perte de la végétation. En fait, comme l'indique Sourlamtas (2019) les surfaces urbaines plus résistantes à l'eau du fait du bâti et de l'asphalte ; recueillent les flux d'eau et les transportent vers les sols exposés.

In increasingly urban areas, soil erosion is a significant problem due to the loss of vegetation. In fact, as Sourlamtas (2019) points out, urban surfaces that are more resistant to water due to buildings and asphalt collect water flows and transport them to exposed soils.

In the District of Abidjan, as in the El Abed basin in Morocco, the significant occurrence of water erosion is therefore the result of natural predispositions but also and above all of anthropic actions reflected by changes in land use and land occupancy patterns (Sbai and Mouadili, 2021). In tropical climates such as that of the ADA, this erosion occurs mainly in the slick with selective removal of fine particles, which accelerates the physical, biological and chemical degradation of the surface soil horizons (Bwandamuka et al, 2021). This occurrence of hydroclimatic risks most often concerns the least affluent populations (Traoré, 2016; Traoré et al, 2018).

However, it should be noted that the rainfall, soil, topographic and land use data used from global and regional databases may contain biases (Payet et al, 2011). These may be related particularly to the high spatial resolutions and a certain imprecision of the information.

Rainfall data from the Climatic Researcher Unit database, for example, may have deviations from local data, which themselves are often biased by metrological and reading errors. The FAO soil data, on the other hand, remain globally general and may inevitably differ on a large scale. The same applies to the DEM and the Sentinel 2 image, which have a resolution of 12.5 and

10 m respectively. They probably minimise the differences in level and the values of certain slopes. The quality of the biomass could also be affected.

The probable biases and inaccuracies of the input data are likely to have influenced the output raster (Adamou et al., 2022). Also, more accurate input data could lead to quite different results. For example, LIDAR images with a finer resolution of 1 m would have led to possibly different results. The water erosion susceptibility layer generated from the RUSLE model applied in a GIS environment therefore deserves to be confronted with empirical results in situ.

Notwithstanding these limitations, this model remains a robust decision-making tool that assists in the use of spatial information, soil loss modelling and soil erosion risk management (Sourlamtas, 2019). This model has shown that water erosion despite its significant occurrence on only 2% of the territory remains a major risk in the ADA, as it affects more than one in four inhabitants. With an average annual growth rate of around 6.8%, this mostly informal land artificialisation combined with the effects of climate disturbances will inevitably increase these vulnerabilities. Political awareness of these threats and anticipatory and preventive management of immanent crises are a guarantee for preserving the environment, preserving human lives and promoting the social and economic development of the ADA.

IV. CONCLUSION

The objective of this study was to model water erosion in the Autonomous District of Abidjan. The approach integrates the parameters of the RUSLE model into a Geographic Information System. It allowed to discretize and map the levels of erosion risk occurrence susceptibility. On analysis, the ADA has an average erosive potential of 0.48 t/ha/year. The map of erosive potential has enabled the identification of the most vulnerable areas on which to concentrate efforts and direct investments aimed at restoring soils and protecting them against water erosion for preventive management of crisis situations.

In a research and development context, this tool is important for land use planning in order to achieve sustainable development objectives. In fact, in semi-urban areas where agriculture and urban areas are intermingled as in the ADA, soil erosion can be a significant problem due to exposed agricultural land and urban surfaces heavily waterproofed by developments. This significant vulnerability of human and material stakes to the risk of water erosion will thus crescendo in view of the development prospects of the ADA.

This model, in spite of some limitations linked in particular to the data, seems to be well adapted in the ADA in order to define the relative influence of the factors which condition water erosion.

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Effect of Mineral and Organic Fertilizer on the Performance of Cassava (*Manihot esculenta* Crantz) in the Pissa region of the Central African Republic

Kosh-Komba E.^{1*}, Gougodo De Mon-Zoni L. J.², Omenda J.A.³, Zaman M.⁴, Mingabay-Bendima B.⁵, Batawila K.⁶, Akpagana K.⁷

^{1,2}Plant and Fungal Biodiversity Laboratory, Faculty of Sciences, University of Bangui, Central African Republic.

¹Laboratory of Biological and Agronomical Sciences for Development, Faculty of Sciences, University of Bangui, Central African Republic.

^{1,2,5}Central African Institute for Agricultural Research (ICRA), Bangui, Central African Republic.

³Department of Water & Agricultural Resources Management, University of Embu, Kenya

⁴International Atomic Energy Agency (IAEA), Vienna, Austria

^{6,7}Laboratory of Botanical and Ecological Plant, Faculty of Sciences, University of Lome, Togo.

*Corresponding Author

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Abstract— *Inefficient and unbalanced use of fertilizer is some of the plausible reasons contributing to the significant cassava yield gaps in Sub-Saharan Africa. However, there is limited research regarding the responses of cassava to organic and inorganic fertilizers used in these agrological settings. We conducted a study in the Pissa areas with the participation of members of an agropastoral group to improve the cassava yield in rural areas of the Central African Republic. The experimental layout was a randomized complete block design (RCBD) with four treatments replicated four times and comprising 16 elementary plots of 25m² (5m by 5m). The treatments were randomly assigned from 1 to 4 (T1, T2, T3, and T4). According to the treatments, the variabilities between cassava's growth, yield, and economic productivity in pure culture were measured and quantified. The data were subjected to Analysis of Variance (ANOVA) using a linear model of R statistical software version 3.1.2. We performed Principal Component Analysis (PCA) on several parameters. This study shows that treatments T4 (cassava + NPK + cow manure) and T3 (cassava + NPK) resulted in plants that have better growth in heights and diameters, unlike T1 (control) and T2 (peasant practice), which have the lowest values. The PCA confirmed that the variability between the treatments is up to 52.12% on the two axes (1 and 2) and affirms that the yields obtained during the study show a highly significant difference. The treatment T4 (cassava + NPK + cow manure) gave the highest yield and generated a good profit compared to other treatments. There was no correlation between treatments in terms of growth and productivity parameters. The T4 treatment proposed by the International Atomic Energy Agency (IAEA) performed better on all the evaluations. Therefore, organo-mineral fertilization can contribute to the improvement of cassava production.*

Keywords— *Cassava, Fertilizers, Yield, and Cow manure.*

I. INTRODUCTION

Cassava (*Manihot esculenta* Crantz) is ranked the third most crucial tropical food crop contributing 32% of the world's food root and tuber production after potatoes, contributing 45% of the total to food security (FAOSTAT, 2016). This plant is widely cultivated and is the staple food of households in tropical and subtropical regions FAO (2014). Its starch-rich terrified roots are consumed. Also, the leaves are consumed as a vegetable in some countries (Celcos et al. (2012). Cassava is also suitable for industrial utilization and can be processed into flour, chips, starch, ethanol, and other non-food products (Balagopalan, 2009). In the last century, cassava has transitioned from a subsistence to a more commercial crop (Onyenwoke & Simonyan, 2014). Cassava is highly adaptable and can grow in different agroecological settings, and it can also grow on low-fertility soils and

has the advantage of flexibility in harvest periods (El-Sharkawy, 2014). It can withstand prolonged droughts and erratic rainfall (Okogbenin et al., 2013). Therefore, it is considered a highly resilient crop in the face of current climatic changes (Jarvis et al., 2012). Thus, cassava can be crucial in tackling food insecurity and hunger in the face of climate uncertainty (Biratu et al., 2018).

In Africa, cassava is the second most important food source in terms of calories consumed per capita (Roothaert & Magado, 2011). Sub-Saharan Africa is the largest cassava producer globally: For instance, out of the 277 million tons produced worldwide in 2013, 158 million tons were from Africa (Bennett, 2015). Cassava is also the first food crop in the Central African Republic (CAR), which plays a vital role in household food security, especially in rural and urban areas. It is the staple food of more than 95% of Central Africans (Zinga et al., 2006). Central African agriculture remains the heart of economic development, employing 75% of the active population and contributing 45% of the Gross Domestic Product (GDP). The CAR wonderland enjoys a myriad of natural resources, comprising of vast arable land, covering about 15 million hectares, of which only 800,000 hectares are annually cultivated. Also, the average area cultivated per agricultural worker is 0.53 hectares; it has five zones within which cassava and other crops are cultivated, but the productions are not the same since each zone has specific characteristics. Despite all its resources, there is a considerable yield gap between actual productivity in farmers' fields and the potential cassava productivity (Ezui et al., 2016). The yield gaps between farmer and researcher-managed trials remain large; for instance, In Nigeria, Adiele et al. (2020) reported a root dry matter yield of 35 t ha⁻¹ compared to 2.51 t ha⁻¹ obtained on farmers' fields in Africa (De Souza et al., 2017).

Given its tolerance to various climatic stresses (El-Sharkawy, 2005), cassava is usually grown in nutrient-depleted soils with little or no input of fertilizer (Howeler, 2002), thus the low yields. Trials in Africa, Brazil, and India showed increased cassava root yield production with fertilizer application (FAO, 2013). Research in DR Congo (Munyahali et al., 2017) and Zambia (Biratu et al., 2018) has shown that fertilizer can improve cassava yield. Hence, bridging the yield gap lies on a better understanding of soil organo-mineral fertilization.

Further, application of soil amendments at appropriate doses does not only increase crop yield but also preserves the soil's productive capacity during crop's growth cycle (Palm et al., 2001; Akanza et al., 2001). Agricultural soils in some CAR regions have a low level of intrinsic fertility. This notwithstanding, the use of mineral and organic fertilizers on food crops in rural areas remains insignificant due to the low purchasing power of farmers and input scarcity in the market (Akanza and Yao – Kouamé, 2011). Organic resources play an essential role in soil fertility management, for instance, animal manure, a locally available resource, improves soil structure for easier root penetration and development. It can also contribute to long-term soil organic matter formation and improve soil biological properties (Blanchet et al., 2016). Additionally, nutrients contained in organic manure are released more slowly and are stored for a longer time (Rani et al., 2022). However, cassava is an exhausting plant, given the mineral mobilizations that its cultivation entails at the end of the cycle (Pouzet, 1988; Raffailac & Nedelec, 1984; Troupa & Koné, 2003). Therefore, sustaining cassava tuber production, which has been on the decline (IRAD, 2013), is dependent on the use of both organic and mineral fertilizers.

Previous studies conducted on cassava show that mineral elements such as N, P, K, Ca, and Mg are essential in increasing cassava tuber yield (Ngome et al., 2013). This calls for mechanized farming systems that use fertilizer among other inputs. On the contrary, little attention has been given to the potential of organo-mineral fertilization in combating soil fertility problems in cassava fields (Biratu et al., 2022). In the current study, we report on the potential of integrated management practices in improving cassava yield. The central hypothesis is that cow manure and mineral fertilizer will achieve higher cassava yields than the sole application.

II. MATERIALS AND METHODS

2.1 Study area, trial site history, and land clearing

The study was conducted in the municipality of Pissa. The trial was initiated near M'baiki in the Prefecture of Lobaye (4.046°N, 18.166°E) south of CAR (Figure 1). The study was conducted in collaboration with lead farmers who were members of the Groupement Agro-Pastoral Pissa 2 (GAP2) with a participatory approach. The climate is Guinean forest characterized by nine

months of the rainy season with annual rainfall amounts ranging from 1600 to 1800mm with a mean annual temperature of between 23 and 26 °C. With a maximum annual temperature of 29°C and a minimum of 18°C, the average relative humidity is 77%. The predominant vegetation is of the forest type. Given these climatic divergences, the municipality of PISSA is found in the agroecological zone No. 1. Cassava is the main food crop grown and an important cash crop grown by the majority of farmers. Other major crops grown in the area are; coffee, cocoa, and banana. The predominant soil is clay-sandy soil near the national road. It is a fallow of more than five years dominated by fertility indicator plants, namely *Pueraria javanica* (Roxb.) Benth and *Chromoleana odorata* (L.).

This study used rudimentary tools such as a hoe and a machete to open the plot. A total area of 1369m² (37 m by 37m) was delimited and cleared; we piled up the waste on the plot's edges and proceeded to plough finally to make the soil loose for the establishment of culture. The plot was divided into small elementary plots with a dimension of 25m² (5m by 5m); the milestones were cut to two meters (2m), and the stakes of one meter long were used; the area was divided into four blocks, and each block had four elementary plots whose treatments were randomly assigned, the plot was separated by two meters of the driveway between the blocks and one meter between the elementary plots and a border to avoid the edge effect and predators.

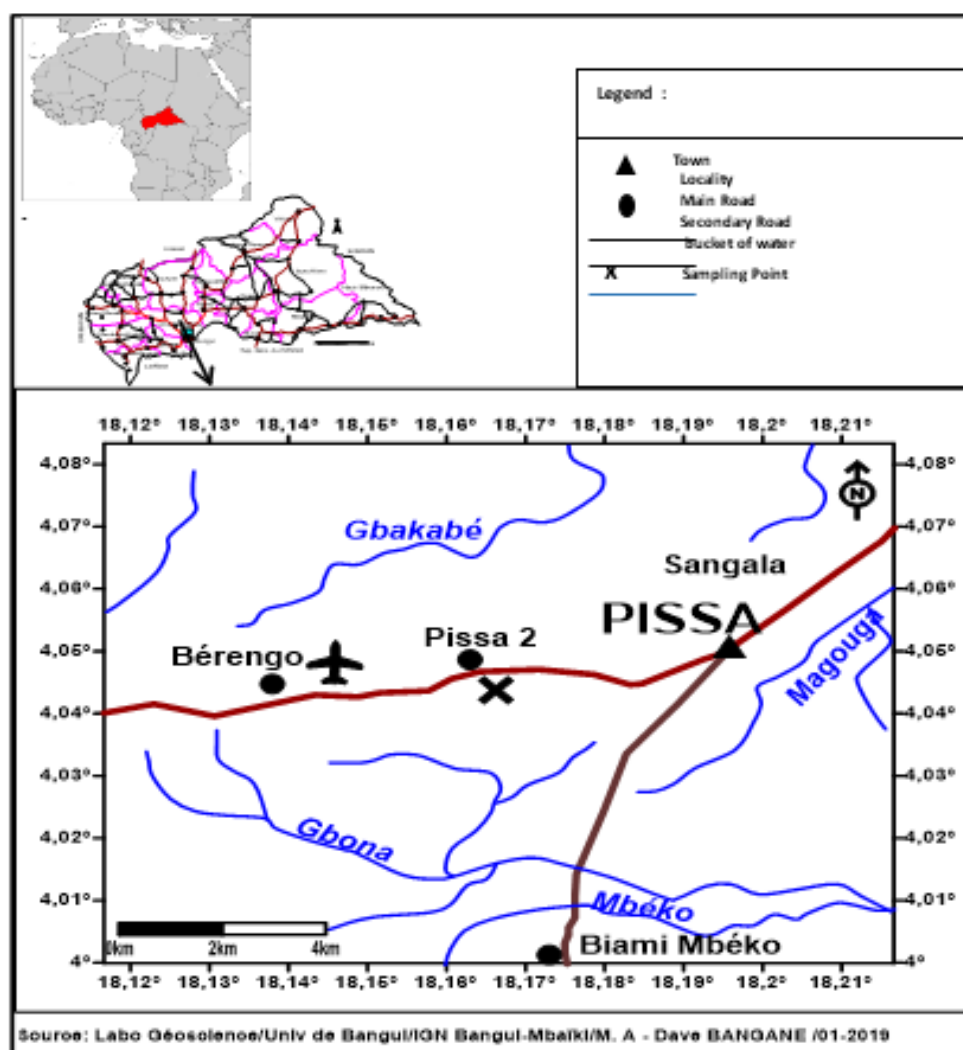


FIGURE 1: Study site

2.2 Trial design

The experiment adopted a randomized complete block design (RCBD) with four treatments and four replications comprising 16 elementary plots of 25m² (5m by 5m). The four treatments were randomly assigned from 1 to 4 (T1, T2, T3, and T4) (Figure 2). Where; T1 is the Control plot; T2 is the Peasant practice; T3 is the practice recommended by the University of Bangui (80kg N, 40kg P and 120kg K) and T4 is the practice recommended by the IAEA; 90 kg N, 30 kg P, and 180kg K. For the

fertilizer application N and K were divided into three 1/2 doses of N, K, and 1/2 in the third month and the rest in the fifth month, but the full P 2/2 was applied in the first month. The mineral fertilizers used consist of urea ($\text{CO}(\text{NH}_2)_2$) containing 46% N, potassium sulphate (K_2SO_4) containing; 50% K, and triple superphosphate, TSP ($\text{Ca}(\text{H}_2\text{PO}_4)_2$) containing 46% P. The rates for these three fertilizers were 80, 40, and 120kg/ha or 90, 30, and 180 kg/ha. Plants in their ammoniacal form do not directly assimilate urea, with 46% nitrogen in granular form. It is hydrolyzed into ammonium for 2-3 days in the soil before use by the absorbent hairs of the roots.

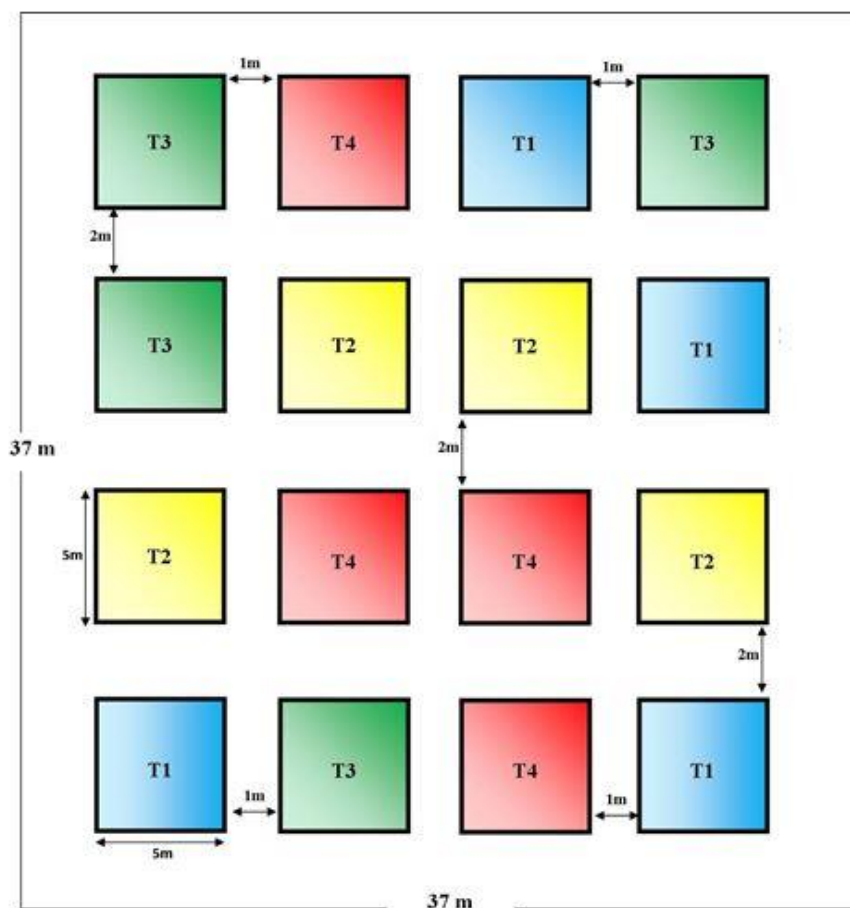


FIGURE 2: Experimental design

2.3 Trial establishment and crop husbandry

A total area of 1369 m² (37 m × 37 m) was divided into four blocks, each of which has four elementary plots of 25 m² (5 m × 5 m). The cuttings used (12 months old) were cut into small pieces of 15-20cm. The plant material consists of a local cassava cultivar, "Gabon" (Figure 3). This cultivar has been identified because of its resistance to the African cassava mosaic (CMD), its agronomic performance, from the point of view of yield, it is highly appreciated by cassava producers (Zinga et al., 2012). The variety is also available in the region (Pissa2). Cassava cuttings were collected from the field of the agropastoral group.

The cuttings were sown in five rows and five columns with 25 cuttings per elementary plot. The total number of cuttings in the entire area was 400. The cutting points measured 1 m between and on the line (1m by 1m). The gaps between the blocks were 2m. Each block had a cultivable area of 100 m² (5 m x 5 m x 4); the plot had an area of 400 m² or 0.04 ha. A 4 m border was laid out around the plot. The experimental plots were maintained weed-free throughout the season with no herbicides. During the experiment, no phytosanitary treatment was administered.

Urea and potassium sulphate was applied twice, and phosphorus and cow manure were simultaneously applied (Table 1). First, 1/3 of the amount of urea and potassium was applied one month after cassava planting, another 1/3 after 3 months, and finally, the last third in the fifth month of planting. The total amount of phosphorus was applied during the first treatment (Table 2). After 12 months of testing, cassava tuberous roots were harvested to assess the average yield in fresh weight and number of cassava roots per plant according to treatments.



FIGURE 3: Preparation of cuttings by farmers

**TABLE 1
SUMMARY OF MINERAL AND ORGANIC FERTILIZERS AND RECOMMENDED TREATMENTS BY ELEMENTAL PLOT**

Treatment	Manure Rate (Kg/ha)	Mineral fertilizer (Kg/ha)		
		N	P	K
T1	0	0	0	0
T2	0	0	0	0
T3	0	80(1/3 N+	40(3/3P+	120(1/3K
T4	3000	90(1/2N+	30(2/2P+	180(1/2P

**TABLE 2
QUANTITIES AND TIMES OF FERTILIZERS APPLICATION**

Duration			
Quantity	Start of cuttings	Three months after cuttings	Five months after cuttings
N	$\frac{1}{3}$ OU $\frac{1}{2}$	$\frac{1}{3}$ OU $\frac{1}{2}$	$\frac{1}{3}$ OU $\frac{1}{2}$
P	$\frac{3}{3}$ OU $\frac{2}{2}$ -		-
K	$\frac{1}{3}$ OU $\frac{1}{2}$	$\frac{1}{3}$ ou $\frac{1}{2}$	$\frac{1}{3}$ OU $\frac{1}{2}$
Cow manure	3000Kg/ha -		-

2.4 Data collection

2.4.1 Soil sampling and analysis

Before the experimental installation, surface soil was sampled using an Edelman auger at 0-20cm depth, bulked together, and harmonized to obtain a composite. The soil was air-dried, passed through a 2mm sieve, and analyzed for texture and chemical properties. The particle size distribution was done using the hydrometer method described by Bouyoucos (1951).

The pH was determined in water at 1.2.5 solution using a pH meter (Okalebo, 2002). Organic carbon determination followed the method of Heanes (1984), and total nitrogen was determined by Kjeldahl digestion and colorimetric analysis (Bremmer,1982). Exchangeable cations, available macronutrients, and the available phosphorus were extracted using Melich 3 method. All the analyses were performed by the Kenya Agricultural and Livestock Research Organisation (KALRO, NARL).

2.5 Parameters to be studied

The 23 descriptors presented below (Table 3) were used during the study to collect field data related to agro-morphological characterization.

TABLE 3
AGRO-MORPHOLOGICAL DESCRIPTORS

Three months descriptors:		
No.	Descriptors	Codes
1	Fundraising rate	TL
2	Plant circumference	CP
3	Height of seedlings	HP
4	Number of leaves 10 cm from the apical part	NF
Six months descriptors:		
5	Leaf retention	RF
6	Length of lobes	LL
7	Width of the lobe	LI
8	Lobe margin	ML
9	Petiole length	LP
10	Flower	F
11	Pollen	P
9-months descriptors:		
12	Foliar Healing	CF
13	Length of internodes	LEN
14	Length of stipules	LS
15	Margin of Stipules	MS
12 months descriptors:		
16	Fruit	F
17	Seeds	S
18	Plant height	PH
19	Branching level	NR
20	Number of roots per seedling	NR/P
21	Number of marketable roots/seedlings	NRC/P
22	Average root weight	MR
23	Disease observation	OM

2.6 Statistical analysis:

The statistical analyses were performed in the R software version 3.6.1 (Team, 2013). Data on cassava growth attributes were checked for normal distribution using the hist function. Principal Component Analysis (PCA) was performed on several agronomic parameters. Data were then subjected to analysis of variance using the generalized linear model to determine the effect of treatments and the means separated using Tukey-Kramer Honest Significant Difference Test at $P \leq 0.05$.

III. RESULTS

3.1 Rainfall distribution and soil physicochemical properties

During the experimental period, the cassava received different amount of rainfall distributed differently over the growing period (Figure 4). Cumulatively rainfall received during the experimental season was 100.4mm. The highest daily rainfall amount was recorded in July 2018 (11mm). The site experienced a meteorological drought (defined as the absence of rainfall for a period of more than 28 days during the growing period) and dry spells (absence of rainfall in periods between 10-28 days

during the experimental period) (Okeyo et al., 2014; Kiboi et al., 2017). During the growing season, the site experienced dry spells of 18, 20, 21, and 23 days with meteorological droughts of 30 and 90 days.

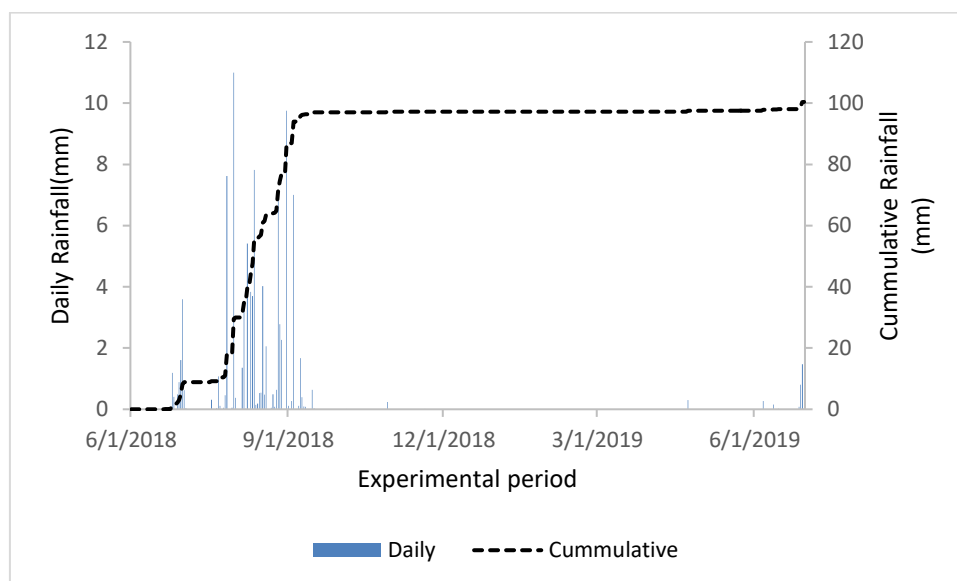


FIGURE 4: Daily and cumulative rainfall during the experimental period (June 2018-June 2019)

Table 4 shows the results of the soil physicochemical analysis of the experimental site before the establishment of the trial. In terms of the soil's physical properties, sandy texture dominated the soil particle size distribution (76-78%) with low levels of silt (4-6%) and clay (16-20%). The soil reaction in the experimental site was 5.5, which can be rated as strongly acidic (Hazleton & Murphy, 2016). Based on the nutrient supply capacity, the soils are deficient in organic carbon (0.8%) and total nitrogen (0.06%) with very low CEC (3.37 cmol (+) /kg) and low in basic cations (Ca^{2+} , Mg^{2+} , K^+ , $\text{N}^{\text{a}+}$).

TABLE 4
SELECTED PHYSIOCHEMICAL SOI PROPERTIES OF TOPSOIL (0-20) BEFORE PLANTING

Parameter	Soil analysis
pH (Water)	5.5
Organic carbon (%)	0.8
Total nitrogen (%)	0.06
C/N	13
Available P(mg/Kg)	1.54
CEC (cmol(+)/Kg)	3.27
Exchangeable bases (Cmol(+)/Kg)	
Ca	1.64
Mg	0.56
K	0.15
Na	0.05
Micronutrients (mg/Kg)	
Zn	0.5
Cu	1.45
Fe	34
Particle Size (%)	
Sand	76
Silt	4
Clay	20
Texture Class	Sandy Loam

3.2 Plant height

The animal manure and mineral fertilizer significantly ($P = 0.001$) influenced plant height at 12 months. Plant height significantly increased under T4 (7.32m) and T3 (2.13m) compared with T1 (1.82m), T2 (1.13m) (Figure 5).

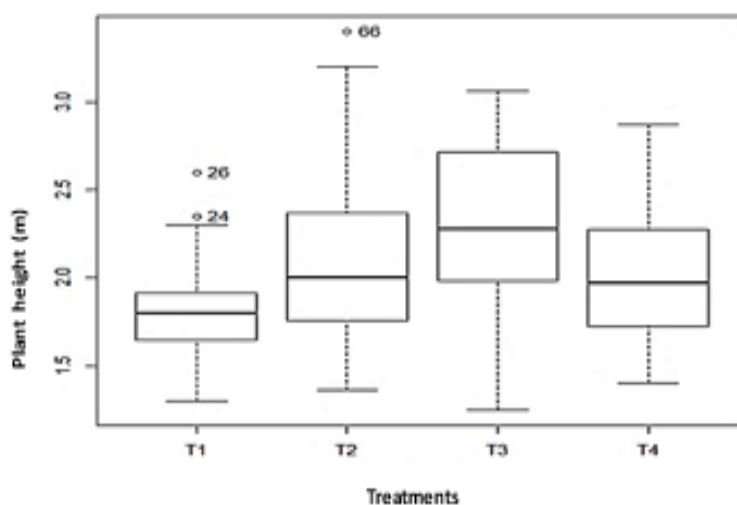


FIGURE 5: Plant height at 12 months

3.3 Plant circumference

There was a significant ($P = 13e-12$) treatment effect on cassava plant circumference. The plant circumference was in the descending order of T4, T3, T1, and T2 (Figure 6).

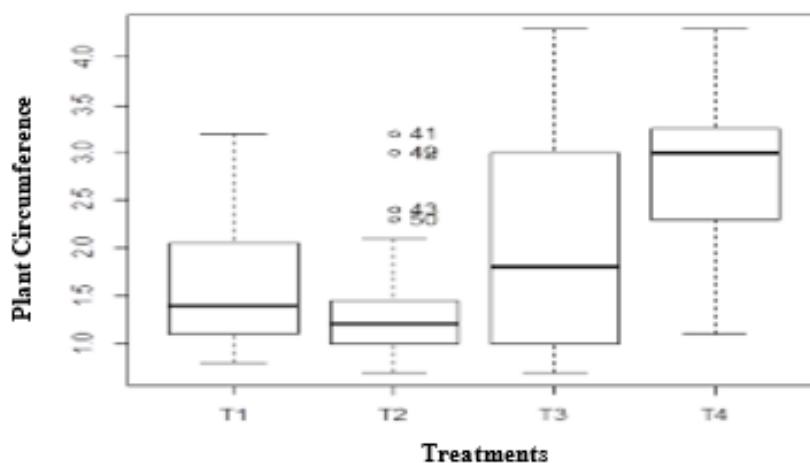


FIGURE 6: plant circumference based on different treatments

3.4 Agro-morphological characterization

The nature and degree of divergence of descriptors were assessed using the Principal Component Analysis (PCA) with quantitative and qualitative variables (Figure 7). The two axes revealed 52.13% of the total variability—Axis 1 with 34.91% variability and 17.22% for axis 2. Based on axis 1, petiole length, root weight per plant, leaf retention, marketable roots per plant, and the total number of roots per plant were highly correlated. However, on axis 2, the strongly correlated parameters were the plant's height, the length of the nodes, and the plant circumference. On the other hand, there was no correlation between the parameters of axis one and those of axis 2. Based on these results, it is clear that the treatments did not impact the morphological characteristics of accessions but influenced growth and productivity, as shown by the strong correlation between these parameters.

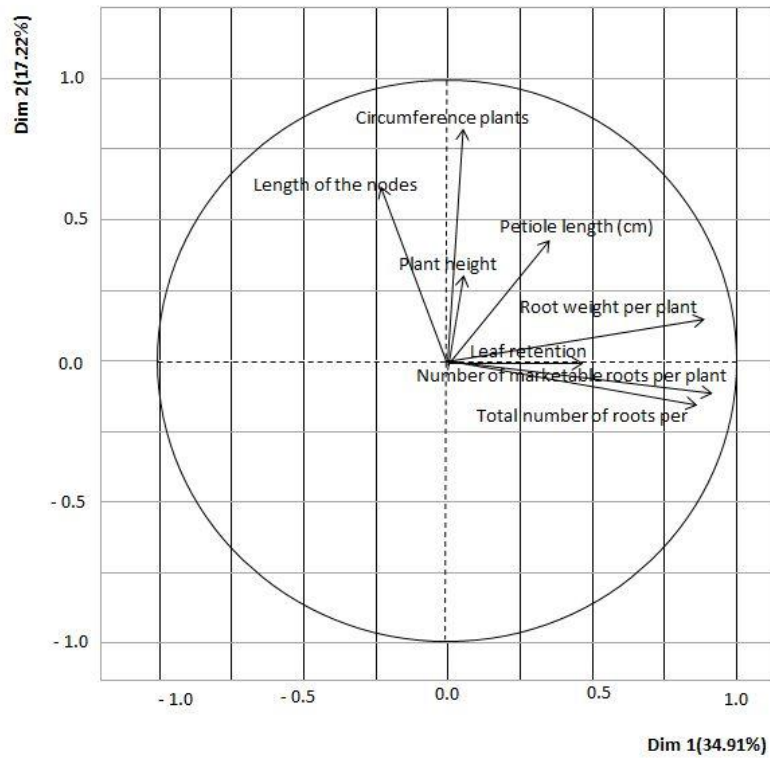


FIGURE 7: Agro morphological characterization

3.5 Evaluation of production and root yield

At 12 months, treatments significantly ($P = 0.001$) influenced the number of tubers per plant (NT/P) and the number of marketable tubers (NTRC/P) (Fig. 8). The total number of roots per plant was in the descending order of T3, T4, T2, and T1. Manure and mineral fertilizer use significantly influenced ($p= 0.004$) the number of marketable roots per plant. The highest number of marketable roots was obtained with T4, which was not statistically different from T3. The treatments under T2 and T1 gave the lowest marketable roots per plant.

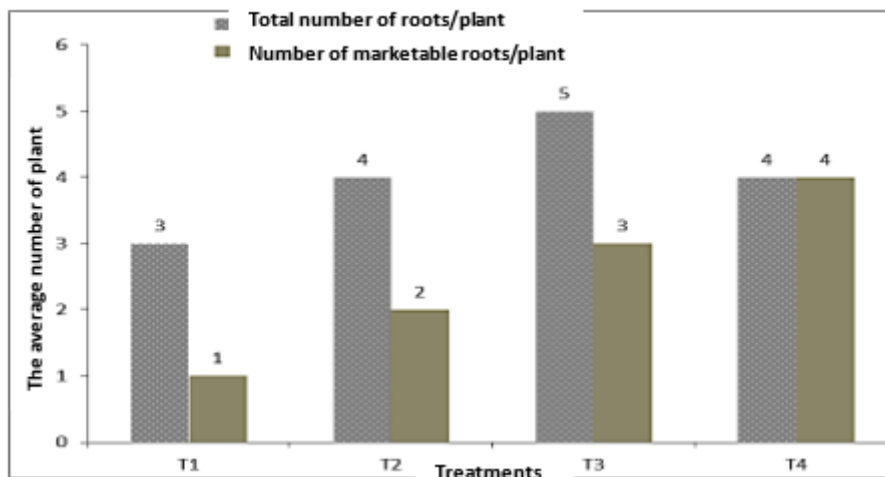


FIGURE 8: Evaluation of production and root yield

3.6 Average root weights

The use of animal manure and mineral fertilizer significantly ($p = 4.01e - 16$) influenced the mean root weight. The higher mean weight was obtained with T4 (5kg/plant) followed by T3 (3.2kg/plant). There was no statistical significance between T1(1.7kg/plant) and T2(1.8kg/plant) (Figure 9).

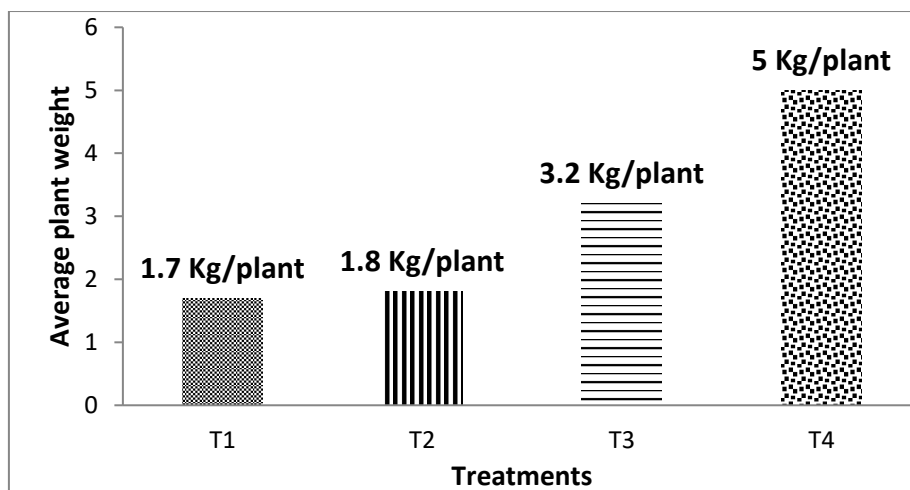


FIGURE 9: Average root weight under different treatments

3.7 Economic evaluation

The results relating to income and profits generated following different treatments are in figure. (10). The T4 treatment was the best in terms of yield, with a total production cost is (7870,22 USD/ha) and a total profit of (4486,03 USD/ ha), followed by T3 treatment with a total production cost of (5036,94 USD/ha) but with a profit of 2203,66 USD/ha. Farmers can successfully adopt T4 and T3 treatments. On the other hand, given the yield obtained by T4 and T3 from the growth parameters to the harvest, production can be doubled or tripled with mineral and organic fertilizers (cow manure). Economically, these treatments are more cost-effective with higher and more incomes than T1 and T2 treatments at the end of the cycle. Provided there is access to the necessary inputs at the proper doses; this increases the yield of cassava tubers while considering the benefit at the end of production.

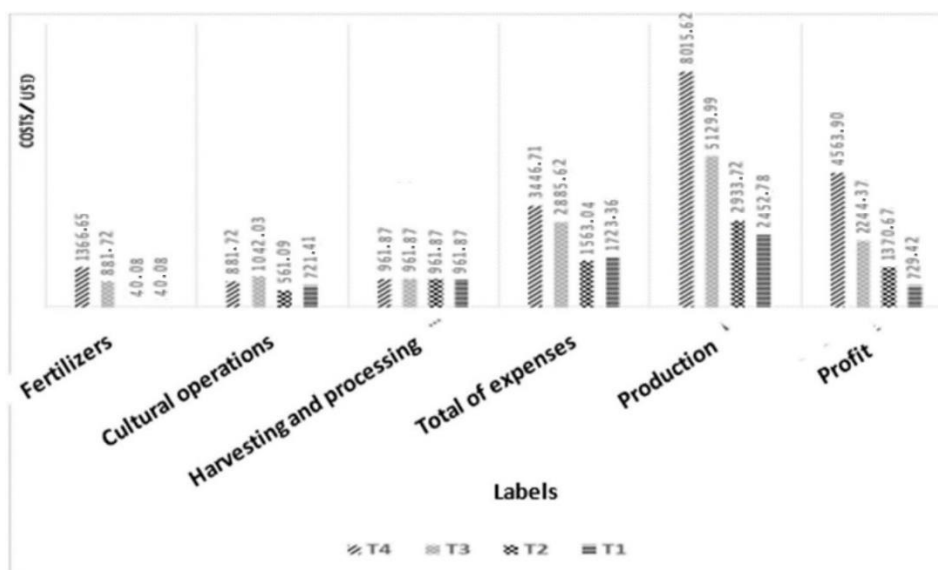


FIGURE 10: Economic valuation

IV. DISCUSSION

Using animal manure and mineral fertilizer significantly ($P = 0.001$) enhanced the cassava growth attributes: plant height, plant circumference, root yield, and the average weight of tubers. The plant height was highest in the T4 and T3 treatments. Increased plant height could be attributed to the readily available nutrient in the mineral fertilizer and the many secondary micro-nutrients in animal manure that enhanced plant growth (Biratu et al. 2022). These findings agree with Uwah et al. (2013), whose findings relate to our present study outcome. Similarly, a study carried out in the savannah zone in CAR by Ballot (2016) found that the use of organo-mineral fertilizers contributed to the enhanced growth of cassava. However, they conducted the study in an agro-climatic zone different from ours. The nature of organic manure, mineral fertilizers, and the study environment

characterized by environmental factors (pedo-climatic, soil, temperature, wind, rainfall, and vegetation) specific to each environment inevitably influences cassava's growth. In the Niari Valley in the Democratic Republic of Congo, Mombo-Tsimba (2008) and Nyombo et al. (2014), in a maize study, reported that the combination of chicken manure and mineral fertilizer had better performance compared to other treatments. Hence, it reinforces the importance of organic fertilizers for crop growth.

There was a significant ($P = 13e-12$) treatment effect on the cassava plant diameter. Plots with treatment T4 (9.7 cm) had the largest diameter, followed by T3 (4.8 cm), T2 (3.6 cm), and T1 (3.04 cm). Munyahali et al. (2017) reported similar observations in their study conducted in the Democratic Republic of Congo. In their study, they observed upsurges in both height and cassava stem diameter with the use of NPK fertilizer. The rates of the organo-mineral fertilizer of T4 further impacted the growth of cassava in diameter compared to T3, where only mineral fertilizer was used at lower doses. Berger's (1996a) work on the impact of fertilization on crop production reported that the associated use of organic and mineral manure could give a satisfactory agronomic response to crop production. Similar results were also observed by Gnahoua et al. (2017), who reported a significant positive effect on stem diameter using mineral fertilizer.

The Principal Component Analysis (PCA) carried out using quantitative and qualitative variables made it possible to assess the variability between agro-morphological parameters and fertilization regimes. The analysis revealed that the two axes have values greater than one and express 52.13% of the total variability. Parameters such as leaf retention, marketable roots per plant, and the total number of roots per plant are highly correlated. However, plant height, length of the nodes, and plant circumference are the parameters strongly correlated on axis two. Kosh-Komba et al. (2014) in the Central African Republic and Djaha et al. (2017) in Côte d'Ivoire, in their study carried out on the agro-morphological characterization of different cassava accessions, obtained respectively an overall variability of 55% and 63.84% higher than within accessions for the first six axes cumulated with a partial contribution of 15 descriptors out of 27 in CAR against 08 out of 24 in Côte d'Ivoire. These authors pointed out that there is significant variability between accessions. Although the previous studies on different scales have similarities, like the descriptors, the results obtained differ under the variability within the parameters subjected to the analyses.

The overall response of cassava to mineral fertilizer and animal manure resulted in significant upsurges in the average weight of tubers and root yield. Treatments with mineral fertilizer and cow manure (T4) performed significantly higher than treatments with sole manure application (T3). The unfertilized treatment provided a baseline to which responses of all treatments were compared. The findings of this study parallel the observations from previous work (e.g., Molina & El-Sharkawy (1995); Fermont, 2009; Uwah et al., 2013; Biratu et al., 2018; Biratu et al., 2022) reported rises in cassava tuber weights with the usage of different fertilization regimes. The response shown by yield parameters to N and K could be associated with; acute photosynthesis surface and improved physiological activities leading to the creation of more assimilates that are moved and used in rapid tuber growth. N and K are essential for cassava root initiation and increase in tuber size and numbers (Uwah et al., 2013). Similar observations were made by Uwah et al., 2013 who reported that nitrogen surges the leaf chlorophyll content, thus promoting the photosynthesis capacity of plants and is responsible for plants' high yield. However, the observations in the present study are opposite to the reports of Lema et al., 2004 who observed no significant effect of mineral fertilizer on tuber yield with the use of both an improved and local cassava variety. Reports by Berger (1996) and Clermont (1998) pointed out that the nutrients provided have only a short-term effect on mainly sandy ferritic soils. Thus, these nutrients are quickly leached without organic matter, and the plants benefit only partially.

These results confirm the interest in the organic fertilizer used in T4, which has a good performance on the yield of cassava tubers. The contribution of manure enriches the soil with primary nutrients (NPK) and secondary trace elements that play an essential role in the physiology and metabolism of the plant, allowing for good growth and better returns. In addition, Dupriez et al. (1983), Feller (1995), and Nyembo et al. (2014) claimed that organic manure provides nutrients to the soil and improves its structure. These results are consistent with those of Anderson and Wood (1983), Akanza et al. (2002), and Compère et al. (1991). Who reported that a contribution of 20 t ha⁻¹ of manure is advisable to establish a viable and sustainable production system. Therefore, organic manure is an essential component of T4 treatment.

Based on the economic analyses, the T4 (cassava + N P K + cow manure) gave the best profit starting from the expenses made to the tune of 7870,22 US per hectare and generated a total profit of 4486,03 US/ha, followed respectively by T3 (2203,66 USD/ha); T2 (1345,81 USD/ha) and T1 (716,19 USD/ha). These findings agree with another study conducted in Western Kenya and Uganda (Fermont, 2009) that reported returns to investments using different fertilization regimes. Similarly, Ballot et al. (2016), in their work on cassava in the savannah area of CAR, confirm the economic gain generated by using different fertilization regimes. Even if the cost of fertilizers remains a significant challenge for producers compared to the country's

isolation, the results show there is always a benefit if adequately used. Using fertilizer increases producers' incomes and contributes to food security, as confirmed by James et al. (2000) and Oti et al. (2010) in their studies. The cassava sector is a necessary means to improve incomes if the actors consider the appropriate technical routes to add the efficient use of organo-mineral fertilizers per the results obtained.

V. CONCLUSION

The study evaluated the effect of the combined application of cow manure and mineral fertilizer on cassava growth, root biomass yield, and profitability. We concluded that cassava responds more to the combined use of organic and inorganic fertilizers than the sole use of inorganic fertilizers. The combined use of fertilizer also resulted in higher profitability than other treatments. Thus we recommend using 3000Kg/ha manure in combination with 90N-30P-180K Kg/ha in regions similar to forest areas (municipality of Pissa) in the Central African Republic.

CREDIT AUTHORSHIP CONTRIBUTION STATEMENT

Kosh-Komba: Investigation, data collection, writing- original draft, supervision, and methodology, **Gougodo De Mon-Zoni:** Conceptualization, methodology, data collection, writing-review and editing, **Omenda J.A:** Writing- review and editing, **Zaman M.:** Proposal of treatments for trials and protocol for data collections and the protocol for data collection, **Mingabaye-Bendima:** Data collection, **Batawila K:** data analysis and validation of results, **Akpagana K:** Comceptualizationn

DECLARATION OF COMPETING INTEREST

The authors declare no conflict of interest.

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Little Millet: An Indigenous Grain

Biswa Sankar Das^{1*}, Susanta Sekhar Choudhery², Pulaka Ranjan Nayak³

Department of Seed science, Odisha Millets Mission, Odisha
Watershed Support Services and Activities Network, Hyderabad, India

*Corresponding Author

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Abstract— Little millet is a small seeded cereal grain defined very familiar due to its climate resilient smart crop with encouraging for its essential elements composition and nutritional benefits such as Low calorie content with high fiber helps in weight loss, High Protein content helps for muscle growth, Rich in antioxidants and minerals which improves immunity. Little millet have abundance of calcium, iron, zinc, dietary fiber up to 38.0 %, Protein 10-11 % with polyphenol content provides them an edge over staple cereal crop. This article covers Data in a strategic manner, History and Origin, Taxonomy, Crop management with Indigenous to Improved Practices, Weeds and management, Processing techniques and Value addition process techniques. Millets are a small seeded cereal crop have been playing as a staple food in many parts of world. These are of many different types which all have unique characters, unique traits, unique health benefits, Wherever Little millet is a millet type has been used across many parts of world like, China, East Asia, India and Malaysia, where as it is native to India and called as “Indian Millet”. The Species name is based on a Specimen collected from Sumatra (Indonesia) (deWet et al.1983) . it is used to make many more value added food products, fermented foods, healthy foods, which are stores in large supermarkets throughout the country and it’s widely used as a Alternative to Rice replacement.

Keywords— Crop management, Little millet, Processing, Traits, Weeds.

I. INTRODUCTION

The Scientific name of Little millet is “Panicum sumatrense” is a species of millet in the family “Poaceae”. Little millet is an annual herbaceous plant that is cultivated as a cereal crop in the Arid to Semi- arid regions of Asia, East Asia, India. It is a tetraploid and Self-pollinated species that most likely descended from Indian peninsula (Weber and Fuller, 2007) . There are two races of Little millet “Panicum sumatrense” and “Panicum milicare” (House et al.2000) .

Little millet is grown throughout India up to altitude of 2100m.Although the Origin of Little millet is not clearly evidenced much favors for Indian origin because Endemic nature to India.

1.1 Taxonomic Rank:

- Kingdom – Plantae
- Clade - Tracheophytes
- Clade - Angiosperms
- Clade - Commelinirs
- Order - Poales
- Family - Poaceae
- Genus – Panicum
- Species – P.sumatrense

II. HISTORY

History: Little millet (Panicum sumatrense) (Roth.ex Roem & Schult) is all so known as “Miliare”. It is widely cultivated as a cereal across India, Neapal, and western Myanmar. It is particularly important in Eastern ghats of india. where it forms an important part of tribal agriculture practice.

2.1 Origin, Domestication and Botanical Description:

Little millet (*Panicum sumatrense*) was originated in Indian peninsula (Weber and Fuller 2007) . The genus *Panicum* has two species which usually reported as Tetraploid in nature with $2n$ (Diploid chromosome set) is 36 (Hamoudetal: 1994: Moulik, 1997), although Chen and Renvoize (2006) report a hexaploid nature of *Panicum sumatrense* with $2n=54$ chromosome number. *Panicum sumatrense* is a domesticated species of Little millet cultivated mainly in India and usually reported as tetraploid with $2n = 36$, distributed in India, Nepal, Pakistan, Srilanka, Eastern Indonesia, Western Myanmar. There are two races of Little millet, namely “nana” and “robusta” (House et al; 2000), in the “nana” have constitute a height of 60 -70cm., the inflorescence is 14-15cm., erect, open and highly branched. The branches sometimes droop at maturity. Plants in the race “robusta” have constitute 120 -190 cm. of plant height (tall), the inflorescence is 20-45cm. long, opening compact, and highly branched. It is primarily a self-pollinated crop that grown throughout India in a altitude of 2100meter.

2.2 Growing Regions in India:

The Little millet growing states in India are Odisha, Gujarat, Maharashtra, Karnataka, Andhra Pradesh, Madhya Pradesh, Tamil Nadu, over an area of 2.34 lakh ha with total production of about 1.27 lakh tones with productivity 544 kg / ha during the year 2015 -16 (IIMR Research Bulletin) . As a result of the multi – prolonged approach, Little millet is cultivated over an area of 3837 hector, with total production about 21.20 tonnes, with productivity 511 kg / ha during kharif 2021-22 of Odisha.

TABLE 1
LOCAL NAMES OF LITTLE MILLET

Language	Local name
Hindi	Kutki, Thoda Bajara
Bengali	Sama
Kannada	Same
Gujarati	Gajro, Kuri
Telugue	Samalu
Tamil	Samai
Marathi	Gajro
Odiya	Suan, Guruje suan

TABLE 2
STATE WISE VARIETIES OF LITTLE MILLET

Sl.no	State	Varieties
1	Odisha	OLM -203, OLM-208, OLM-217
2	Madhya Pradesh	JK – 4, JK- 8, JK -36
3	Andhra Pradesh	Olm-203, JK -8
4	Tamil Nadu	Paiyur 2, TNAU 63, CO-3, OLM -203, CO-4, K-1, OLM -20

2.3 Climate Requirement:

Little millet originated in south-east Asia and is now a days it is grown throughout india, particularly in Madhya Pradesh, Odisha, Jharkhand, Andhra Pradesh, Tamil Nadu and Uttar Pradesh in mostly kharif season.

In the temperate zones of Asia: China, East Asia, also in tropic continent of India, Indo china, Malaysia are major crop grown area which can withstand both drought and water logging situations as it is known as climate resilient crop. it can be cultivated up to 2000 meter above sea level. So, it have concluded that, Little millet is best for tropical and sub-tropical regions with a long growing season.

2.4 Soil:

The best soil for Little millet are alluvial, loamy, sandy soil with good drainage capacity, less water holding capacity, deep loamy fertile soil rich in organic matter are preferred for best growth. It can also withstand salinity and alkalinity to some extent. As in other small millets are cultivated in rusticated to tribal areas with marginal lands. Little millet is a drought tolerant crop that can grow in various soils in a PH range 5.5 to 6.5, with good source organic matter is preferred the best soil and retains the adequate moisture content.

2.5 Temperature:

Little millet requires a minimum temperature of 20* C and it prefers a range of 25*C to 30*C. It can tolerate temperature up to 35*C, but higher temperature may reduce crop yield.

2.6 Rainfall:

Little millet requires a minimum rainfall of 750 -800 mm (30 -32 inches) in annually, with a consistent water supply throughout the growing season. Drought conditions can severely impacts on crop yields.

2.7 Season:

- Odisha – Middle of June
- Tamil Nadu – June and September –October
- Karnataka, Madhya Pradesh and South Bihar – Last week of June to first week of July

III. PACKAGE OF PRACTICES

- Seed rate:
 - 8 kg / ha for line sowing
 - 12 kg / ha for broadcasting
- Spacing: 20 -22.5 cm between rows and 10 cm between plants within a row
- Manures and Bio-fertilizer:
 - Manures is needed Five tons of farm yard manure per one hector during 2-3 weeks before sowing.
 - Bio –fertilizer: Seed inoculation with Agrobacterium radiobacter and Aspergillus awamori improves production and quality of seed yield.

IV. GROWING PRACTICES

Basically the farmers used the landraces in tribal areas in where it cultivated, or the local seeds available. There are four types of sowing methods adopted by farmers in respect to their indigenous cultivation practice.

4.1 Broadcasting:

Seeds are immediately sown in the field during the first phase of sowing time. This approach is frequently used because it is simple and does not call for a specialized mechanism. The average seed rate is 15 kg / ha, or slight more than that due to the mind setup of farmers to maintain proper plant population.

4.2 Line sowing:

This is a better than the broadcasting in where the crops and weeds are easily distinguish, which also facilitates weed management. When using this technique the lines should be 22.5 – 25 cm. distance. The average seed rate is 8kg / ha.

4.3 Row by Row drilling:

Using a direct seed drill, seeds are drilled directly in to unrented soil. The average seed rate is 8 kg / ha.

4.4 Transplanting:

The seedlings are raised in nursery beds with appropriate specification. At the age of 25 days old seedlings should be transplant in main field with spacing of 22.5cm x 10cm (R x P), or 25cm x 10cm (R x P) according to season specific. The required seed rate is 10 kg / ha.

V. CROPPING MODEL

It constitutes the Cropping system and the Cropping sequence as preferred by the farmer.

5.1 Cropping system:

A cropping system refers to the type of crops grown in a combined approach and practices used for growing them. Cropping systems have been traditionally structured to maximize crop yields.

- For Odisha – Little millet + Black gram (2:1) row ratio
- For Madhy Pradesh – Little millet + Sesamum / Pigeon pea (2:1) row ratio

5.2 Cropping sequence

A cropping sequence refers to growing two crops in the same field, one after the other in the same year.

- For Odisha – Little millet (short duration) – Horse gram / Black gram, or Green gram.
- For South Bihar – Little millet – Niger

VI. CROP CARE MANAGEMENT

6.1 Water management

Little millet is majorly a rainfed crop, drought tolerance, climate resilient crop that does best where there is an average rain fall less than 1000 mm (750 -800 mm) of annually to complete the crop cycle. Where as in irrigated condition it requires the first irrigation at 25 -30 Days after transplanting, and second irrigation at 45 – 50 Days after transplanting.

6.2 Manure and Fertilizer management:

6.2.1 Manure management

A well decomposed farm yard manure of 5 tones / ha requires before sowing / transplanting as a basal dose.

6.2.2 Fertilizer management

On the basis of soil structure and soil profile a recommended dose of fertilizer in the form of N: P₂O₅: K₂O requires for good production,

For Andhra Pradesh: 20: 20: 0 kg / ha

For Bihar & Odisha: 20: 10: 0 kg / ha

For Other states: 20: 20: 0 kg / ha

VII. WEEDS AND THEIR MANAGEMENT

7.1 Important weeds

Among grassy weeds, Echinochola colonum, Echinochola crusgulli, Dactyloctenium acgypticum, Elusine indica are the major weeds found in little millet crop field.

7.2 Weed Control

Two inter –cultivation and one hand weeding in line sown crop and two hand weeding in broadcasted crop are necessary for effective weed control. In transplanted crop field three times of mechanical weeding by use cycle weeder is most effective for better crop yield.

VIII. MAJOR DISEASE AND THEIR MANAGEMENT

TABLE 4

Common name & Scientific name	Casual organism	Nature of Damage	Management
Grain smut (Macalpinomyces mycessharmae)	Ustilago paradoxa	The Ovary is converted in to smut sorus	Adopting resistant cultivars (OLM-208, CO2), Seed treatment with Carbendazim (contact fungicide) @2gm /kg, Delayed sowing
Brown spot (Seedling blight or Leaf blight)	Helminthosporium	Appearance of brown to Dark brown spot on leaf lamina especially found in older plants. Under high humidity condition fungus lesion formation seen	Seed treatment with contact fungicide (Carbendazim) @ 2gm / kg seed

IX. MAJOR PEST AND THEIR MANAGEMENT

TABLE 5

Common name & Scientific name	Casual organism	Nature of Damage	Management
Shoot fly	Atherigonia pulla	It affect on centrally shoot which shows the dead heart formation, Damaged tillers are all so formed	Using higher seed rate Using fish trap Using carbofuron 3G @1.5 Kg / ha Spraying quinolphos @2ml / lit effectively reduced shoot fly infection

X. HARVEST AND HARVESTING TIME

Kharif season crop: September to October

Basically the harvesting procedure at crop maturity stage is followed by single harvest either the panicle, or the entire plant body is harvested.

XI. THRESHING

Threshing in Little millet refers to separating the grains from plants stem to panicles and chaffs. It can be done manually by beating the plants with a stick, or using a machine such as thresher.

XII. YIELD

The Yield of Little millet depends on various factors such as soil fertility, soil moisture content, cultivar used, weather condition and crop care management. the optimum yield will 12 -15 qt / ha (5.0 – 6.5 qt / acre) .

XIII. STORAGE

For Grain Purpose: After threshing the produce (seed) will thoroughly dry to a moisture level up to 10 -15 % and stored in gunny bags which will maintain its self-life up to two years. No fungal contamination will seen.

For Seed Purposes: Once cleaned, free from other inert materials, it is recommended to store at moisture content level 10 -12 % to prevent spoilage and insect infection. The seeds should be stored in a dry and cool place in a container of gunny bags, earthen pots in order to prevent rancid, or attracting pests. Sealed containers, or gunny bags are the good storage choice for a longer period up to two years will be a good quality of seeds. It is essential to store the seeds in a place free from moisture, as this can cause the seeds to spoil. In some Indigenous seed storage practice the cleaned and well maintained seed materials are mixed with dried neem leaf and pongamia leaf for to prevent seed borne diseases for a longer period.

XIV. PROCESSING

Similar to other millets, Little millet is also required to undergo certain basic steps of primary processing operations such as cleaning, grading, and separation where in removal of unwanted materials like stones, soil particles, stalks, chaffs and grains of other crops. These operations are also important for adding value to the produce from the point of view of getting better returns from sowing.

14.1 Milling

The milling process of Little millet have so many process steps. Like other grains processing it involves three primary tasks:

- Size Grading
- Density Grading

14.2 Hulling Grading (referred to as de husking)

An eccentric or vibrating grader is used to achieve size grading and a destoner is used for density grading and an impact to share huller is used for hulling.

XV. IMPORTANT VALUE ADDITION PROCESS

15.1 Roasting

Customary roasting of grains is utilized to improve Flavor, test, yet different advantages incorporate decrease of antinutritional properties.

15.2 Malting

Malted millet is said to be nutritionally superior to un malted one. The complete process of malting mainly includes four stages viz. soaking, germination, roasting and milling, where the most desired physico –chemical changes occurs during soaking and germination stages (swami et al.2013) .

Malting is the process of steeping germinating and drying grains to convert it into malt in where germination and sprouting involves a number of enzymes to produce the changes from seed to seedlings. The malt producer stops this stages of the process when the required enzymes are optimal.

XVI. HEALTH BENEFITS OF LITTLE MILLET

- Low calorie content and high fiber content which helps in weight loss.
- High protein content for muscle growth.
- Rich in antioxidants and minerals which improves immunity.
- Loaded with B-vitamins, which helps in metabolism and energy production.
- Helps in reducing cholesterol level.
- Little millet is rich in Magnesium which helps to improve heart health.
- It also rich in Niacin which helps lowers the cholesterol level.
- Little millet contains Phosphorus which is important for weight loss, tissue repair and energy production after strenuous workout.
- It helps detoxify the body.

XVII. CONCLUSION

The Little millet having its high enrich high fiber, high protein and rich source of antioxidants have been provided many more medical advantages and additionally it is similarly wealthy in sugar, energy and sustenance making significant element of dietary and wholesome adjusted food sources. Little millet is also a climate resilient, smart crop in rainfed agro ecosystem contributing a major livelihood supports to the farming communities. Little millet farming can be a lucrative and sustainable agricultural venture for farmers. It has a high nutritional values and rich in essential elements making a valuable addition to

the diet of humans. It has a high demand in the market, both domestically and internationally, making it a viable option for farmers looking to sell their produces.

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Zero Budget Spiritual Farming (ZBSF)

Priya Kaur^{1*}, Aparajita Das², Puneet Pathak³, Anup Kalra⁴

Ayurved Research Foundation
4th Floor, Sagar Plaza, Distt Centre, Laxmi Nagar, Vikas Marg, Delhi-110092, India
*Corresponding Author

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Abstract— According to the most recent WHO reports, more than 50% of foods contain natural carcinogens [11]. This allays a grave worry in the agricultural sector. Despite of the fact that pesticides used to boost agricultural Development, are known to modify human chromosomes, they are still utilized rapidly and in excessive amounts only to generate income. But at the end of the day, it becomes mandatory for a novice farmer to share a meal with his family members in these times of rising inflation. Only in India, 200k farmers committed suicide in 2016 as a result of the heavy debt loads, they were forced to take on to pay for the pricey and lethal crop growth enhancers. After witnessing the detrimental consequences of chemical farming, recently a very effective agricultural practice is getting quite trendy among the farmers known as Zero Budget Natural Farming (ZBNF), also known as Zero Budget Spiritual Farming. In Southern India, especially in Karnataka, where it was initially developed, it has achieved widespread success [7]. It is currently expanding so rapidly and actively across India.

Keywords— Natural Farming, Palekar Model, Bijamrita, Jeevamrit, Zero Budget Natural Farming.

I. INTRODUCTION

The philosophy of natural farming is working in close proximity with nature to produce nutritious food, keep ourselves well, and maintain the health of the land. Everything in nature has a use and a function within the larger system of life. This practice is also known as "do nothing farming" because the farmer is merely seen as a facilitator for Nature, who does the actual labor. This farming technique involves no tillage and cultivation without the use of pesticides, inorganic fertilisers, and herbicides. In this case, compared to other farming techniques, actual physical work and labour have been observed to decrease by up to 80%. Avoiding the use of organic amendments like FYM and vermicompost is something that makes natural farming very different from organic farming. Fukuoka pioneered natural farming in Japan by experimenting with nature and by using natural methods of crop multiplication. Without soil erosion, he produced yields comparable to those obtained with chemical farming [1,6,9]. The key to natural farming is reducing the external inputs that harm the natural soil composition on the farm. Initially, he had to produce natural insecticides like pyrethrum, which comes from chrysanthemum roots, and sprayed them on his veggies to stave off pests like cabbage worm and cabbage moths because there wasn't a habitat for many of the insects.

The name "Zero Budget Natural Farming" refers to a farming technique where the cost of cultivating and harvesting the plants is zero. As a result, farmers are freed from the expense of purchasing fertilizers and pesticides to maintain the healthy growth of their crops. The technique calls for using readily available, locally produced, naturally biodegradable materials combined with cutting-edge technology and traditional farming techniques based on biological processes that occur naturally. Shri Subhash Palekar brought this idea to light, for which he was awarded the Padma Shri in 2016. [2]. He was born on February 2, 1949, at Belora, a small village in the Amravati district of Maharashtra, India. He graduated from Nagpur with a B.Sc. degree in Agriculture. After graduating, he experimented because he was committed towards improving the farms in his village and found that regular use of chemicals had left the farm area arid. So, he made the decision to seek out the best answer. The natural systems that operate in forests, have the capacity to develop and nourish them while sustaining many healthy ecosystems, according to Palekar's 1986–88 research on forest vegetation. And after putting in a lot of work in the field, he finally provided the ZBNF equations. Additionally, he published other works, including "The Philosophy of Spiritual Farming and Five Layer Palekar Models".

II. PALEKAR'S SIX YEARS OF THOROUGH INVESTIGATION REVEALED THAT:

1. Only the local Indian cows' excrement can successfully re-enrich the depleted soil. It is less effective to use the dung from Jersey and Holstein cows, as the indigenous Indian cows contain higher amount of calcium, phosphorus, zinc and copper in comparison to the cross-breeds [7]. We can even use the dung from bullocks or buffaloes if the local cows' dung runs out.
2. The black-colored Kapila cow's dung and urine are found to be magical, as some reports say that Kapila is considered to be the best cow breed for "zero-budget farming", as this breed can survive on jungle forage and very less additional fodder also produces high quality dung and urine.
3. We also need to make sure that the cow dung is as fresh as possible and urine as stale as possible to get the maximum use out of them. Since fresh cow dung contains 86% of water content, which helps the soil to remain moist throughout its growing period and stale urine contains more population of multiplied beneficial microbes.
4. Ten kilogrammes of local cow dung must be applied monthly to an acre of soil, provided that the typical cow excretes 11 kg of dung every day, one cow's manure can help fertilize 30 acres.
5. Additives that can be used along with cowdung are urine, jaggery, and dicot flour that can improve the microbial activity in the cow dung.
6. The lesser milk, the cow produces, the more helpful its dung is for revitalizing the soil [3]. Since, lactating cows being in a stressful situation cannot produce high quality, nutrient and microbes rich cow dung.

Our organization, Ayurved Research Foundation, which is a non-profit organization, is already running a 3-year project funded by DBT (Dept. of Biotechnology, Govt. of India) under the program, "SUTRA- PIC (Scientific Utilization through Research Augmentation Prime Products from Indigenous Cows)" based on production of vermicompost developed by the excreta of indigenous cow "Sahiwal Breed". Also, another 2-year project on multilayer farming funded by NABARD, utilizes natural components such as Beejamrit, Jeevamrit and Acchadana to protect the crops against fungal/bacterial/soil borne disease, insect pests, nematodes etc. which aligns with the principles of Palekar model (zero budget spiritual farming). ZBNF is symbiotic in nature and self-nourishing [10]. Peasants cannot access privatized markets, seeds, or inputs because these are too expensive. Indian farmers increasingly find themselves trapped in a debt cycle as a result of their high cost of production, high interest rates on credit, fluctuating agricultural market prices, rising input costs dependent on fossil fuels, and the usage of proprietary seeds. So, they end-up committing suicide. For Indian farmers of all sizes, debt is like a parasite. To avoid such tough circumstances, "zero budget natural farming" promises to eliminate the need for loans and significantly reduce production expenses, breaking the debt cycle for helpless farmers [8]. The following are the four key pillars of ZBNF:

2.1 Jeevamrit: Composition-

TABLE 1

Sl. No.	Ingredients	Quantity
1	Water	200 litres
2	Desi cow dung	10 kg
3	Desi cow urine	5-10 litres
4	Jaggery	2 kg
5	Floor of any pulse	2 kg
6	Handful of soil from farm or forest	-

Jeevamrutha serves as a catalytic agent that encourages earthworm activity and soil microbial activity. It aids in the prevention of bacterial and fungal plant diseases. For making the system self-sustaining, Jeevamrutha is the only thing required, during the first three years of the transition.

2.2 Bijamrita:

Bijamrita is a seed treatment that can shield tender roots from fungus as well as from soil-borne and seed-borne diseases.

TABLE 2

Composition:

Sl. No.	Ingredients	Quantity
1	Water	20 litres
2	Desi cow dung	5 kg
3	Desi cow urine	5 litres
4	One handful of soil/dirt from surface of the field	-
5	Lime	50 gm

2.3 Acchadana (Mulching):

This can be done using live, straw, or soil mulch. It reduces evaporation, preserving soil moisture.

2.4 Whapasa (Moisture):

Irrigation should be done in alternate furrows only at noon and with less frequent irrigation. Palekar refutes the notion that plant roots require a lot of water, arguing that they actually, require water vapour instead. As a result, whapasa is the situation in which both air molecules and water molecules are present in the soil.

In addition to the aforementioned four pillars, intercropping, furrow method of cropping, contouring or bunds system, and the use of native earthworm species are some other crucial natural farming principles. Additionally, Palekar in 2014 provided the pest management equations, which he named as Agniastra, Brahmastra, and Neemastra. Under plant protection, to control insects and diseases, the farmers can prepare home-made pesticides on their own and use it on the crops.

**TABLE 3
FUNGICIDE-I**

Sl. No.	Ingredients	Quantity
1	Five days fermented butter milk	5 litres
2	Water	50 litres

**TABLE 4
FUNGICIDE-II**

Sl. No.	Ingredients	Quantity
1	Desi cow milk	5 litres
2	Black pepper powder	200 gm
3,	Water	200 litres

**TABLE 5
INSECTICIDE- I**

Sl. No.	Ingredients	Quantity
1	Powder of neem seeds/ leaves	20 kg
2,	Water	200 litres

TABLE 6
INSECTICIDE- II

Sl. No.	Ingredients	Quantity
1	Cow dung	5 kg
2	Cow urine	10 litres
3,	Neem leaves	10 kg
4	Water	200 litres

This mixture is specifically effective against sucking pests such as aphids, jassids, mealy bugs and white flies.

TABLE 7
INSECTICIDE – III

Sl. No.	Ingredients	Quantity
1	Neem leaves	10 kg
2	Tobacco powder	3 kg
3,	Garlic paste	3 kg
4	Green Chilli paste	4 kg

The above ingredients should be soaked in cow urine for 10 days. About 3 litres of this mixture is mixed with 100 litres of water and sprayed on crops. In addition to assisting farmers in paying-off debt, this method raises soil fertility, yield, and product quality [10]. Earthworms break down the plants and animals, which adds humus to the soil. By creating tiny and large-scale pores in the soil, it also increases the soil's ability to hold water and to breathe. The pest management technique utilized here not only prevents pest damage but also guards against the humorous side effects of chemicals, such as magnification, pollution, carcinogenic elements, and food poisoning. It does not contribute to soil and water contamination or their erosion, in contrast to chemical fertilizers. Intercropping and crop rotation prevent the soil from becoming depleted of moisture and nutrients. While mulching slows water evaporation and keeps the soil adequately moist. It offers the soil's microorganisms a hospitable environment. The term "quality of product" refers to the absence of undetectable disease-causing substances, which is of great concern today. In conclusion, ZBNF is unquestionably a significant technique from an economic, social, biological, and physiological standpoint.

III. CASE STUDY- (BY THE NEW INDIAN EXPRESS NEWSPAPER ON 22ND JULY, 2021)

A 50-year-old farmer named Mr. Bannur Krishnappa, in a village in Karnataka has proved that "nothingness" can bloom into beautiful forests and farms. He used the natural farming method on his five acres of land to grow everything from teak and mango to coffee, turmeric, ginger, paddy, and sugarcane. He learned about natural farming technique in 2005 when he met a Maharashtrian farmer named Subhash Palekar. On a single acre of land, he began experimenting with natural farming in 2005. After being under the supervision of Mr. Palekar for one year, he entirely moved over to natural farming in the next two years. This resulted in a five-layered, forested-farm spread across an area of five acres, with vegetation of various heights (tall, medium, short, shrubs, and creepers) growing alongside one another and receiving enough sunshine in each canopy. In Krishnappa's opinion, ZBNF can address economic non-viability of agriculture and the debt and despair that stem from it. Farmers these days are quitting agriculture, but if they practise inter-cropping through ZBNF, there will be no losses and they can overcome agriculture-related problems which can also prevent farmer suicides.

IV. CONCLUSION

The injudicious use of chemical pesticides and fertilizers can endanger the ecosystem and soil. Numerous studies have demonstrated the negative impacts of changes in soil characteristics, soil contamination, ground water pollution, and declines in soil microflora, among other things. Studies have shown that natural farming, with least amounts of external inputs and by adding supplements like Jeevamrutha, improves soil fertility by boosting the amount of accessible nutrients and soil microflora.

This approach promotes diversity in micro and macro flora as well as multiple cropping. Production and labor costs are kept to a minimum.

As a result, many may consider it to be sustainable and eco-friendly. However, these studies are still in the early stages, and more research must be done to confirm the advantages for all crops and the effectiveness of native insecticides like Neemastram, Brahmastram, etc., and how long it will take to enrich the contaminated soil. In conclusion, there have been significant cost savings on seeds, fertilizers, and plant protection chemicals. The new system has released the farmers from their debt trap and given them newfound confidence to turn farming into a financially viable endeavour. There is little doubt that Palekar's Zero Budget Natural Farming has permanently changed Indian agriculture. This method has substantially benefited more than 40 lakh farmers around the nation, and this number will undoubtedly rise dramatically in the upcoming years.

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Improvement of Sugarcane Seeds Drought Stress Tolerance by Invigoration using of *Trichoderma* as Bio-Primer

Danie Indra Yama^{1*}, Muhammad Rizal², Muhammad Ali³

Department of Agricultural Technology, Pontianak State Polytechnic, Indonesia

*Corresponding Author

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Abstract— *Trichoderma* used in this study resulted in a density of 1.4×10^{-9} with their morphology had septate mycelium, rounded or oval conidia shape and attached to each other, has a bright green color and branched conidiophores. The biopriming treatment had a significant effect on germination, germination rate, vigor index and total chlorophyll content of sugarcane that was stressed by drought. Soaking for a long time (24 hours) caused a decrease in the percentage of germination in sugarcane seeds by 12.69%, the vigor index of 1.06 and short immersion also caused a decrease in the percentage of germination by 10.61% and a decrease in the average vigor index by 21.74. The 12-hour soaking treatment was a better treatment than control to increasing germination rates under drought stress, it was 12.5% specifically and fast germination rate of 3.76 days. Sugarcane germination rates was reduced if the soaking process is carried out in brief time and vice versa. The highest vigor index value was produced by biopriming treatment for 12 hours, which was 81.74. Biopriming treatment using secondary metabolites of *Trichoderma* was increasing the total chlorophyll content in sugarcane germination by 0.22 g/ml compared to without soaking treatment.

Keywords— *Fungi; Germination; Priming.*

I. INTRODUCTION

Sugarcane growth consists of germination, budding, vegetative growth, and maturity phases. Determination of the next phase is determined by the germination phase which is influenced by seed quality, glucose, nitrogen, water, seed treatment [1]. The problem in the field, especially in West Borneo, availability of superior sugarcane planting material was limited and forcing farmers to buy seeds from overseas. As that result, the seeds fall on deteriorate and the quality of the seeds decreases due to the delivery process which takes several days. In the shipping process, especially in the tropics, temperature fluctuations will occur, with the recalcitrant nature of sugarcane seeds, the seeds cannot withstand storage conditions at 10°C and above 35°C so they will be through chilling injury and reduce their viability [2]. Seeds that have low quality can reduce their vigor and viability, which in turn reduces plant productivity. In West Borneo during the long dry season experienced drought conditions so that water supply was limited. Though sugarcane seeds require a lot of water, one of which is during the germination phase. Drought stress in the germination phase affects the quality, growth in the next phase. Each sugarcane clone produce a different expression in response to drought stress. Research by [3] showed that the PS881 clone was able to adapt to conditions drought stress in the early growth phase in unfavorable seed conditions experiencing storage. The main key in increasing crop production is healthy seeds that have viability and vigor of more than 85%. According to [4] that the highest growth and yield was produced by sugarcane seeds that were not stored.

The existence of the above problems, one of the strategies to solve them can be through biopriming techniques using biological agents *Trichoderma* spp by utilizing the secondary metabolites produced. This technique is often used in food crops, but not many have applied this technique to plantation seeds, especially sugarcane. Biopriming is the immersion of seeds in a suspension of biological agents so as to activate signals to maintain stress and increase vigor [5]. *Trichoderma* was also able to increase resistance to abiotic stresses, one of which is drought, nutrient absorption, and increasing root growth and development. *Trichoderma* produces phytohormones such as Indole Acetic Acid (IAA) which is stimulating root and shoot growth [6].

Trichoderma priming technique gave a significant increase in the content of phenol, proline, stomata conductance, photosynthesis, chlorophyll content of wheat plants under drought stress conditions [7]. This technique had also been increasing the size of the seed germination, vigor index and root-shoot growth [6]. Biopriming of seeds using *Trichoderma harzianum* for 4 hours at 40% concentration resulted in a germination rate of 78%, an index of germination speed of 9.2 in chickpeas [8]. Priming for 24 hours increased vigor, average germination time and increased okra yield up to 55% [9]. In another study, *Azoospirillum* 20% biopriming treatment for 12 hours was the best treatment for maize germination and vigor [10].

Research on the use of secondary metabolites of *Trichoderma* in the invigoration of drought stress tolerant sugarcane clones using biopriming technique can be used to improve the quality of sugarcane seeds after experiencing setbacks due to storage and increasing plant resistance in drought stress conditions and can be further developed in sugarcane cultivation in suboptimal land. This study aimed to analyze the effect of *Trichoderma* biopriming on drought stress sugarcane clone germination, determine the appropriate duration of *Trichoderma* biopriming for drought stress sugarcane clone germination, determine drought stress sugarcane seed vigor index with *Trichoderma* biopriming application.

II. RESEARCH METHODS

The research was carried out at the Plantation Plant Science Laboratory and Greenhouse, Pontianak State Polytechnic, West Borneo, for 3 months

2.1 Materials and Equipment

The materials used were *Trichoderma* spp, potato, dextrose, aquadest, water, aluminium foil, mineral soil, dolomite, cow manure compost, fungicide, West Kalimantan, Indonesia local sugarcane budchips, NaClO. The equipment used were soil sieve, hoe, ruler, thermometer, test tube, Erlenmeyer, beaker glass, stirrer, microscope, preparations, clear nail polish, 21D spectrometer, analytical balance.

The study was structured using a Completely Randomized Block Design with 4 times replications. The treatments used were variations of biopriming, T0: Without immersion, T1: 6 hours, T2: 12 hours, T3: 18 hours, T4: 24 hours.

2.2 Research Implementation

Biopriming treatment by immersing the budset into a suspension of *Trichoderma* with the appropriate time of treatment. Before the biopriming treatment, sugarcane buds had been stored for 7 days were disinfected using 2% NaClO for 5 minutes and rinsed thoroughly with distilled water. *Trichoderma* was propagated using Potato Dextrose Broth (PDB) media and incubated for 5 days until conidia formed [11]. Determination of spore density was calculated using a haemocytometer by dripping a suspension of conidia that had been diluted to 10^8 with 400 times magnification [12]. The spore density had been calculated by the formula:

$$C = \frac{t \times d}{n \times 0,25} \times 10^6 \quad (1)$$

Annotation:

C	= conidia density per ml of solution
t	= number of conidia in the observed box
n	= number of boxes sample
0,25	= small-scale sample box correction factor on haemocytometer
d	= dilution factor
10^6	= conidia constant value

After application, the budset was germinated into a seedling basket with Podsolic soil mixed with compost and then maintained for 30 days. Observations made in the study are as follows:

2.2.1 Seed Germination (SG)

To measure the viability of the observed budset daily.

$$SG = \frac{\sum \text{normal sprouts}}{\sum \text{Total seed planted}} \times 100\% \quad (2)$$

2.2.2 Germination Rate (GR)

To see the ability to germinate speed [13]

$$GR = \frac{N_1T_1+N_2T_2+\dots+N_xT_x}{\Sigma \text{total sprouts}} \quad (3)$$

Annotation:

N = number of budset germinated at day x

T = germination at day x

2.2.3 Vigor Index (VI)

To measure the ability of the budset to grow [14]

$$VI = \frac{\Sigma \text{normal sprouts}}{\Sigma \text{total sprouts}} \quad (4)$$

2.2.4 Total Chlorophyll Content (TCC)

Indicator of plant tolerance in response to drought stress measured using a UV-VIS spectrophotometer with wavelengths of 645 nm and 663 nm [15]

$$TCC = 20,2 A_{645} + 8,02 A_{663} \quad (5)$$

2.2.5 Relative Water Content (RWC)

This observation was carried out with fresh weight (FW), dry weight (DW) and leaf turgor weight (TW) obtained by soaking the leaves in water for 24 hours [7]

$$RWC = \frac{(FW-DW)}{(TW-DW)} \times 100\% \quad (6)$$

2.3 Statistical Analysis

Observational data were analyzed by ANOVA and if there was a significant effect, then further tested with the Least Significant Differences (LSD) test at 5% level.

III. RESULTS AND DISCUSSION

Trichoderma propagation carried out in this study resulted in a density of 1.4×10^{-9} (Figure 1) with a morphology of septate mycelium, conidia shape that is round or oval and one cell attached to each other, has a bright green color and branched conidiophores. *Trichoderma* is a fungus that is generally found in root areas or free-living soil. *Trichoderma* plays a role not only in reducing the negative effects on plants in the face of drought stress but also increasing growth significantly. This fungi has an easily recognizable character and grows fast and matures at 5 days of growth [16]. *Trichoderma* is able to adapt to different living environments, whether in a humid, dark or dry environment by regulating the growth of conidia and enzyme production [17].



FIGURE 1: *Trichoderma* spp.

TABLE 1
AVERAGE OF SEED GERMINATIONS, GERMINATION RATES AND VIGOR INDEX ON SUGARCANE SEEDS DROUGHT STRESS TOLERANCE BY OF TRICHODERMA SECONDARY METABOLITES AS BIOPRIMER.

Bioprimer Treatment	Seed Germination (%)	Germination Rate (day)	Vigor Index
T0 (without soaking)	81.06 c	5.73 a	49.37 b
T1 (6 hours)	87.31 bc	4.98 a	60.00 b
T2 (12 hours)	93.56 ab	3.76 b	81.74 a
T3 (18 hours)	97.92 a	3.58 b	80.11 a
T4 (24 hours)	85.23 bc	3.68 b	80.68 a
Average	89.016	4.34	70.38

Annotation: Numbers followed by the same letter within a column are not significantly different according to the least significant difference (LSD) test.

2.4 Seed Germination

The results of analysis of variance showed that the bioprimer treatment had a significant effect on the germination of sugarcane under stress. The bioprimer treatment for 18 hours (T3) resulted in the highest germination rate of 97.92%, while the bioprimer treatment for 12 hours (T2) slightly decreased the percentage of germination but statistically, it was not significantly different with 18 hours soaking. Sugarcane seeds without soaking resulted in the lowest germination rate of 81.06%, which was not significantly different from soaking for 6 and 24 hours (Table 1). Soaking too long (24 hours) and too short caused a decrease in the percentage of germination in sugarcane seeds with a decrease of 12.69% and 10.61%, respectively. Soaking sugarcane seeds using *Trichoderma* for too long (24 hours) or too short causes a decrease in germination power of 10.61% - 12.69%. This means that the 12-hour soaking treatment is a good treatment to increase the percentage of sugarcane germination under stress. These results are in accordance with research conducted by [6] who said that the *Trichoderma* isolate test could increase the germination percentage of wheat seeds by 87% compared to the control which only got 72.7% of seed germination. The increase in germination was due to *Trichoderma* produce secondary metabolites in the form of Indole Acetic Acid (IAA), which can stimulate meristem cells to trigger the emergence of roots and shoots. IAA is an unstable auxin that functions in root formation and division and enlargement of plant shoot cells.

2.5 Germination Rate

The treatment of immersion bioprimer *Trichoderma* affects the rate of germination of sugarcane seeds under stress. Sugarcane seed germination takes longer if the soaking process is carried out for a short time and will be slightly faster if soaked for a longer time. Sugarcane clones soaked in *Trichoderma* for 18 hours (T3) resulted in the fastest germination rate of 3.58 days, but it was not significantly different from soaking for 12 hours (T2) and 24 hours (T4), while without soaking it produced the longest germination rate of 5.73 days that were not significantly different with 6 hours of treatment (T1) (Table 1). Sugarcane seeds soaked in *Trichoderma* for 12 hours resulted fast germination rate of 3.76 days. The duration was related to the imbibition process. Therefore, 12 hours of soaking the seeds can complete the metabolic process, including starting from the imbibition process of *Trichoderma* suspension. *Trichoderma* can induce various types of signals that can be transmitted to plants. Its secondary metabolites produced in the form of salicylic acid, jasmonic acid or Reactive Oxygen Species (ROS) can trigger plant defense proteins through the induction of systemic resistance [18]. In the priming process, the plant defenses are not activated directly by *Trichoderma*, but the response of plant defenses to environmental stresses is accelerated, besides that *Trichoderma* can encourage plant growth and development. The faster plant growth is influenced by the production of auxin and the activity of the deaminase enzyme which reduces the ethylene content that accumulates during stress. In addition, *Trichoderma* also produces organic acids that can increase the solubility of phosphate, magnesium, manganese so that it can induce roots in the germination process [19]. The presence of *Trichoderma* which is applied to seeds is useful as a barrier to the physical and chemical environmental stresses of the soil in the form of salinity stress caused by lack of water in the soil or drought. In saline soil conditions due to drought *Trichoderma* is able to produce secondary metabolites in the form of acid phosphatase, harzianolides and peptaibols which stimulate root branching and absorption of plant nutrients [20].

2.6 Vigor Index

The vigor index of drought-stressed sugarcane clones was influenced by the immersion of *Trichoderma*. Sugarcane seeds soaked in *Trichoderma* for 12 hours (T2) showed the highest vigor index compared to those without soaking and soaking for 6 hours but not significantly different from 18 hours and 24 hours immersion (Table 1). Short and too long soaking will reduce the average vigor index by 21.74 as well as if sugarcane seeds are soaked in *Trichoderma* for too long it also reduces the vigor index by 1.06. The highest vigor index value was produced by biopriming treatment for 12 hours, which was 81.74. During the priming process for 12 hours there will be activation of enzymes that play a role in the formation of sucrose so that the germination rate increases through decreased lipid prooxidation activity. The priming duration for 12 hours will increase the germination index and decrease the priming duration will cause a decrease in the germination index [21].

2.7 Total Chlorophyll Content (TCC) and Relative Water Content (RWC)

The immersion of *Trichoderma* in sugarcane budset affected the chlorophyll content, while the relative water content was not affected by the biopriming treatment. Soaking for 12 hours produced the highest total chlorophyll but not significantly different from 18 and 24 hours immersion. Meanwhile, the budset that was not soaked by *Trichoderma* produced the lowest total chlorophyll which wasn't significantly different from that of soaking for 6 hours. The biopriming treatment of *Trichoderma* will increase the total chlorophyll content compared to not soaking, but if the immersion time is too long it will decrease the total chlorophyll content by 0.07 g/ml. Chlorophyll is an indicator of drought stress in plants. Plants that experience drought stress will experience chlorophyll damage, both total chlorophyll, chlorophyll a and chlorophyll b due to inhibition of magnesium absorption which is a plant chlorophyll pigment forming. *Trichoderma* spp biopriming treatment can increase the total chlorophyll content in sugarcane germination compared to without soaking treatment. Based on [22] *Trichoderma* inoculation can increase the total chlorophyll content in tomato plants by 15.04%.

TABLE 2
TOTAL CHLOROPHYLL CONTENT (TCC) AND RELATIVE WATER CONTENT (RWC) ON SUGARCANE SEEDS DROUGHT STRESS TOLERANCE BY OF TRICHODERMA SECONDARY METABOLITES AS BIOPRIMER.

Biopriming Treatments	TCC (g/ml)	RWC (%)
T0 (without soaking)	0.48 b	85.16
T1 (6 hours)	0.52 b	86.77
T2 (12 hours)	0.70 a	87.37
T3 (18 hours)	0.75 a	85.98
T4 (24 hours)	0.68 a	83.64
Average	0.626	85.78

Annotation: Numbers followed by the same letter within a column are not significantly different according to the least significant difference (LSD) test.

IV. CONCLUSION

The biopriming treatment of *Trichoderma* had an effect on the germination of sugarcane clones under stress. The duration of biopriming of *Trichoderma* for 12 hours (T2) is the appropriate time for germination of sugarcane clones under stress. Vigor index of sugarcane seeds under stress with the application of biopriming *Trichoderma* for 12 hours is 81.74.

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