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Morphological and Cytological Studies on Two Species of Tomato (Solanum lycopersicum L. and Solanum pimpinellifolium L. (Solanaceae)

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ABSTRACT

Background and objective: Tomato is one of the most important vegetable crops worldwide, valued for its nutritional, economic, and breeding significance. Understanding morphological and cytological variations among cultivated and wild relatives is essential for crop improvement and genetic conservation. This study investigated the morphological and cytological characteristics of two tomato species, Solanum lycopersicum L. and Solanum pimpinellifolium L., with the aim of assessing their taxonomic similarities, differences, and evolutionary relationships.

Materials and Methods: Morphological traits were studied by careful observation and measurement of vegetative and reproductive characters at different growth stages. Parameters examined included growth habit, plant height, stem thickness, leaf shape and size, number of branches, flower structure, fruit size, fruit weight and overall plant vigor. Cytological studies were conducted using standard squash techniques to examine mitotic and meiotic chromosomes.

Results: S. lycopersicum exhibited an erect growth habit with thicker stems, broader leaves, fewer but larger fruits, and greater fruit weight. In contrast, S. pimpinellifolium showed a semi-prostrate to spreading growth habit, slender stems, smaller leaves, numerous small round fruits, and relatively lower fruit weight. Despite these differences, both species shared similar floral morphology, indicating close taxonomic affinity. Cytological result revealed that both species possess a diploid chromosome number of 2n = 24 and a haploid number of n = 12, confirming chromosomal stability and conservation within the tomato plant. Chromosome pairing and segregation during meiosis were normal in both species, with no observable abnormalities, suggesting genomic compatibility and the possibility of successful interspecific hybridization.

Conclusion: The combined morphological and cytological findings indicate that although S. lycopersicum and S. pimpinellifolium differ markedly in vegetative and fruit characteristics, they are cytologically similar and closely related thus their inclusion in the same genus. The integration of morphological and cytological data demonstrates clear phenotypic differences alongside strong chromosomal similarities between the two species. These findings provide valuable insight into their taxonomic relationship and highlight the potential of wild relatives as valuable genetic resources in tomato improvement programs.

Keywords: Morphological, Cytological, Solanum lycopersicum, Solanum pimpinellifolium, Crop improvement, taxonomic relationship

1. Introduction

The tomato belongs to the Solanaceae family under the genus Solanum. It is one of the most important fruit crops in the world. It is regarded second in importance only to the potato in several nations[1]. The species of Solanum are indigenous to Ecuador, Peru, and the Galapagos Islands; however, most evidence indicates that Mexico was the primary site of domestication[2,3]. The tomato is incredibly flexible; it can be found in thousands of cuisines across Europe, from pizza to bloody marys, and from ketchup to chowder[4]. In Nigeria, tomatoes are a vital ingredient in the cuisine of both affluent and impoverished individuals. Tomato stew is particularly enjoyed on Sundays and during festive occasions. Tomatoes also have therapeutic and nutritional qualities. It is necessary to offset the acids produced when meat and other fatty acids are digested[5]. This roughage is advantageous since it improves digestion and helps with constipation[6]. Tomatoes provide carbohydrates, fats, proteins, vitamins, and minerals, which can enhance the brightness of the eyes more effectively than cosmetics when consumed[7,8]. It can be processed and packaged for both industrial and economic uses. Furthermore, it can also be employed for gardening purposes[5].

Solanum lycopersicum and Solanum pimpinellifolium are two of the several tomato (Solanum) species that are the focus of this study. According to research, fruit size QTLs are the main determinant of tomato production, however locule and soluble solids may have additional influence on the overall expression of tomato output[9].

S. lycopersicum is a species characterized by medium to large fruit size, commonly grown in contemporary agriculture, and usually yields 2 to 4 locules. Nevertheless, it possesses a small to moderate lycopene content, shows low resistance to diseases, and displays a light red hue along with minimal levels of soluble solids in the fruit[3]. It is a herbaceous plant that can be classified as either an annual or a short-lived perennial; it generally grows upright, possesses weak stems, and frequently displays sprawling or vining traits. The stem is soft and covered in hair, adorned with glandular trichomes, green in hue, and weak in structure, although it becomes woody at the base as the plant matures. The leaves are pinnately compound, arranged alternately, featuring irregularly lobed edges, a hairy surface, and release a strong tomato aroma when crushed. The root system comprises a taproot with numerous lateral roots; when grown in the field, the roots develop a fibrous and extensive nature. The flowers are small, ranging from 1 to 2 cm, yellow in color, pentamerous (consisting of five petals and five sepals), actinomorphic, and grow in clusters referred to as racemes; they are bisexual and primarily self-pollinating, although cross-pollination may also take place. The fruit is a fleshy berry that can be round, oblong, or pear-shaped. Depending on the cultivar, its smooth skin is green when it is young and turns red, orange, pink, yellow, or purple when it is mature. The kidney-shaped, tiny, flat seeds are covered in fine hairs and vary in colour from pale yellow to brown[10].

S. pimpinellifolium, conversely, is a wild species distinguished by its small fruits that possess 2 locules. It is noted for its exceptional resistance to diseases, high concentrations of soluble solids in its fruit, increased lycopene levels, and a rich red hue[11,12]. These characteristics hold immense importance for the tomato sector. Lycopene, the compound chiefly accountable for the red hue of tomato fruit, serves as a vital marker of fruit quality and is an essential element in the manufacturing of premium processed tomato products[6,13]. The characteristics of soluble solids and lycopene have garnered significant attention from tomato geneticists and breeders, resulting in considerable initiatives directed towards improving these traits in new cultivars[11,13]. This is a herbaceous plant that can be categorized as either an annual or a short-lived perennial; it initially grows erect but ultimately becomes sprawling or viny, extending to lengths of about 3 meters. The stem is slender and green, with a thickness of 8 to 11 millimeters at the base, and is sparsely adorned with various types of glandular and non-glandular trichomes. The leaves are imparipinnate, measuring between 4 to 12 centimeters in length, and consist of 2 to 4 pairs of lateral leaflets along with a terminal leaflet. The fruits are quite diminutive, roughly 1 centimeter in diameter, vibrant red, and spherical in shape, featuring two seed chambers. At first, they have a hairy texture but gradually become smooth; they are consumable yet not grown on a commercial scale. The seeds are small, ranging from 2 to 3 millimeters, light brown in hue, and have fine, silky hair-like extensions[14].

The morphological traits of plants are easily observable and accessible, which makes them the most frequently employed in taxonomic studies[15]. The data obtained from external morphology acts as the essential language for the characterization, identification, classification, and relationships of plants[16]. It is now broadly recognized by taxonomists that morphological characteristics should not be the only criteria taken into account in the systematic classification of plants[17]. Cytology has demonstrated significant advantages in resolving specific taxonomical challenges by providing additional characteristics[17,18]. The number of chromosomes and their homology predominantly affect the pairing behavior observed during meiosis, which in turn partially governs the fertility levels of hybrids, consequently influencing breeding behaviors and variation patterns within populations. The chromosomal count acts as a crucial and commonly employed taxonomic feature, and it is, in fact, almost the only biosystematic evidence that is consistently recorded in standard floras and the like[19].

Although the importance of these two species is well acknowledged, there have been few comparative studies aimed at systematically assessing their morphological and cytological characteristics together. This gap in knowledge hinders the effective application of S. pimpinellifolium in tomato breeding programs, as breeders require a thorough understanding of its cytogenetic compatibility and morphological traits in relation to the cultivated tomato. Therefore, a comprehensive analysis concentrating on the morphology and cytology of S. lycopersicum and S. pimpinellifolium is crucial to furnish essential information that can support genetic enhancement initiatives, conservation strategies, and sustainable tomato farming. The aim of this study was to evaluate the morphological and cytological studies on two species of tomato (Solanum Lycopersicon and Solanum pimpinellifolium (Solanaceae).

2. Materials and Methods

2.1. Area of study

Seeds of the species were germinated in polythene bags behind the Botany laboratory premises, Awka, and subsequently taken to the laboratory for cytological and morphological examination at maturity.

2.2. Collection and identification of plant materials

S. lycopersicum and S. Pimpinellifolium were bought from Oshe market Onitsha, Anambra State. Plants identification was done by plant taxonomist at Botany department, Nnamdi Azikiwe University, Awka. The voucher specimens were deposited at the Botany department herbarium, Nnamdi Azikiwe University, Awka.

2.3. Morphological studies

Observations regarding vegetative and reproductive characteristics, including plant habit, leaf structure, flower morphology, fruit size, shape, and seed traits, were conducted using samples obtained from mature plants. These attributes were measured, documented, and analyzed to identify similarities and differences between the two species.

2.4. Cytological studies

The materials listed below were employed for the study of

- **Mitosis and meiosis:** S. lycopersicum and S. Pimpinellifolium root tips, immature flower buds, photomicroscope, reagents and stains used were Carnoy's fluid, 1:3(v/v) glacial acetic acid and 95% ethanol, 70% ethanol, 18% hydrochloric acid, F.L.P. orcein, distilled water and 0.002m, 8-hydroxy-quinoline.
- **Procedure for mitotic chromosome studies:** The root tip squash technique was employed for the studies of mitotic chromosomes.

The roots of newly germinated plant species under investigation were gathered and subjected to a pretreatment of 5 hours with 8-hydroxyquinoline, followed by fixation in Carnoy's fluid (1:3 (V/V) glacial acetic acid and 95% ethanol) for a duration of 24 hours. Subsequently, the roots underwent hydrolysis using 18% HCl for 5 minutes to facilitate the loosening of the cementing substance between cells, thereby allowing the cells to spread during the squashing process. The hydrolyzed roots were then rinsed in 70% alcohol to avert the crystallization of the stain caused by the acid. A mounted needle was utilized to excise the apical 1mm segment of the root tip onto a clean slide. A single drop of FLP (formic, lactic, and propionic acids) orcein was applied to the specimen. A thin cover slip was then placed over the specimen and gently squashed by briskly tapping the cover slip with the blunt end of a biro. This tapping continued until the material was adequately spread and became nearly invisible. To enhance the spreading of the cells, the slide was positioned between a large fold of filter paper on a smooth yet firm surface, and thumb pressure was cautiously applied on top of the cover slip. Excess stain was absorbed with filter paper, and the slide was subsequently placed under the microscope for chromosome observation. This procedure is as outlined by[20].

2.5. Procedure for meiotic studies

For meiotic investigations, the young flower buds of the two species being examined were gathered between 8 am and 12 noon and subsequently fixed in Carnoy's fluid, which is composed of glacial acetic acid and 95% ethanol in a 1:3 ratio for a duration of 24 hours. The Pollen mother cells (PMC) were then extracted from the anthers in a drop of acetic orcein stain, covered with a cover slip, and gently squashed by tapping the cover slip briskly with the blunt end of a biro. This tapping was continued until the material was adequately spread and became nearly invisible. To enhance the spreading of the cells, the slide was positioned between a large fold of filter paper on a smooth yet firm table surface, and thumb pressure was applied carefully on top of the cover slip. Excess stain was removed using filter paper, and the slide was then mounted on the microscope for chromosome observation. This procedure is as outlined by[20].

2.6. Statistical analysis

Analysis of variance was used to examine quantitative morphological data. The significance of the Duncan's multiple range test was employed to examine treatment differences. Results were presented in Mean ± Standard Deviation.

3. Results

3.1. Morphological result

3.1.1. Root morphology: S. lycopersicum and S. pimpinellifolium exhibit a taproot system characterized by numerous lateral roots. The roots of S. lycopersicum are moderately thick and possess fewer lateral branches, in contrast to the thinner and more extensively branched roots of S. pimpinellifolium. The root system of S. lycopersicum is shallow to moderately deep, featuring noticeable adventitious roots at the base of the stem, while S. pimpinellifolium has a deeper and more spreading root system with a dense arrangement of lateral roots. In terms of root hair distribution, S. lycopersicum shows a moderate presence, whereas S. pimpinellifolium displays an abundance of root hairs (Plate 1-2).

3.1.2. Stem morphology: Both S. lycopersicum and S. pimpinellifolium have branching, upright, herbaceous stems. S. pimpinellifolium had a narrower, tougher, and slenderer stem than S. lycopersicum, which was comparatively thicker, softer, and more succulent. While both species displayed pubescent stems, S. pimpinellifolium had denser trichomes than S. lycopersicum. S. pimpinellifolium had a greener, slightly tougher stem with longer internodes than S. lycopersicum, which had a green stem with a weakly woody base **(Figures 1-2)**.

3.2. Leaf morphology and phyllotaxy

Solanum lycopersicum and Solanum pimpinellifolium exhibit alternate phyllotaxy, with leaves appearing singly at each node. Both species possess compound and pinnately lobed leaves. Nevertheless, the leaves of S. lycopersicum are larger, broader, and more expanded compared to those of S. pimpinellifolium, which are smaller and narrower. The average leaf length of S. lycopersicum varies from 15 to 25 cm, with a width ranging from 8 to 15 cm, while the leaves of S. pimpinellifolium measure between 5 and 12 cm in length and 3 to 6 cm in width.

Both species exhibited petiolate leaves; however, S. lycopersicum had a longer petiole, averaging between 4 and 7 cm, whereas S. pimpinellifolium had a shorter petiole, measuring from 2 to 4 cm. The leaf margins of both species were irregularly lobed, with S. pimpinellifolium displaying deeper and more pronounced lobing. The surfaces of the leaves were pubescent in both species; nonetheless, the trichomes were more plentiful in S. pimpinellifolium compared to S. lycopersicum. In both species, the leaf apex was acute to acuminate, while the leaf base was unequal and slightly decurrent on the petiole. Both species exhibited petiolate leaves; however, S. lycopersicum had a longer petiole, averaging between 4 and 7 cm, whereas S. pimpinellifolium had a shorter petiole, measuring from 2

to 4 cm. The leaf margins of both species were irregularly lobed, with S. pimpinellifolium displaying deeper and more pronounced lobing. The surfaces of the leaves were pubescent in both species; nonetheless, the trichomes were more plentiful in S. pimpinellifolium compared to S. lycopersicum. In both species, the leaf apex was acute to acuminate, while the leaf base was unequal and slightly decurrent on the petiole **(Figures 3-4)**.

3.3. Morphology of inflorescence and flower

Both Solanum lycopersicum and Solanum pimpinellifolium exhibit cymose inflorescences. The inflorescence of S. lycopersicum is typically simple or exhibits weak branching, in contrast to the more extensively branched inflorescence of S. pimpinellifolium, which supports a higher quantity of flowers. On average, S. lycopersicum has 4 to 8 flowers per inflorescence, while S. pimpinellifolium inflorescences contain between 10 and 20 flowers. The flowers of both species were small, pedicellate, and actinomorphic. However, the flowers of S. lycopersicum were comparatively larger, measuring approximately 1.5-2.5 cm in diameter, whereas those of S. pimpinellifolium were smaller, with a diameter ranging from 0.8 to 1.2 cm. In both species, the flowers exhibited bisexual and pentamerous characteristics. Both species featured five green sepals that formed a persistent calyx. The corolla in both species was yellow and comprised five petals that were fused at the base; however, the corolla lobes were broader in S. lycopersicum and narrower in S. pimpinellifolium. The androecium included five stamens in both species, with yellow anthers arranged in a cone around the style. The pistil was singular, characterized by a superior ovary, a slender style, and a terminal stigma in both species **(Figures 5-6)**.

3.4. Morphology of fruit and seed

S. lycopersicum and S. pimpinellifolium yield fleshy berry fruits. The fruits of S. lycopersicum are larger, typically globose to slightly oblong, and smooth, whereas those of S. pimpinellifolium are smaller, round, and more consistent in shape. The average diameter of S. lycopersicum fruits ranges from 4 to 8 cm, while the fruits of S. pimpinellifolium measure approximately 0.8 to 1.5 cm in diameter. At maturity, the fruit color is red for both species, although the fruits of S. pimpinellifolium exhibit a brighter red hue.

Both species produce many-seeded fruits; however, S. lycopersicum has fewer seeds per fruit in comparison to S. pimpinellifolium. The average seed count per fruit varies from 100 to 300 in S. lycopersicum and from 200 to 500 in S. pimpinellifolium. The seeds of both species are small, flat, and ovate. The seeds of S. lycopersicum are slightly larger, measuring about 2.5 to 3.5 mm in length, while those of S. pimpinellifolium are smaller, with lengths ranging from 1.5 to 2.5 mm. The seed color is cream to light brown in both species, and the seed surface is slightly pubescent **(Figures 7-10)**.

3.5. Habit

S. lycopersicum and S. pimpinellifolium are erect, branching herbaceous plants. S. lycopersicum is taller and more robust, while S. pimpinellifolium is shorter, slender and more delicate **(Figures 1-2)**.

3.6. Habitat

S. lycopersicum commonly occurs in cultivated fields and gardens under managed conditions, while *S. pimpinellifolium* grows naturally in open, disturbed soils and marginal areas. *S. pimpinellifolium* shows greater adaptability to dry and nutrient-poor soils compared to *S. lycopersicum* **(Figures 1-2)**.

Figure 1: *S. lycopersicum* Plant.

Figure 2: *S. pimpinellifolium* Plant.

Figure 3: *S. lycopersicum* showing its leaves.

Figure 4: *S. pimpinellifolium* showing its leaves.

Figure 5: *S. lycopersicum* showing its Inflorescence.

Figure 6: *S. pimpinellifolium* showing its Inflorescence.

Figure 7: *S. lycopersicum* showing its fruits.

Figure 8: *S. pimpinellifolium showing its fruits.*

Figure 9: *S. lycopersicum* showing its seeds.

Figure 10: *S. pimpinellifolium* showing its seeds.

3.7. Cytological result

Cytological result revealed that both species possess a diploid chromosome number of 2n = 24 and a haploid number of n = 12.

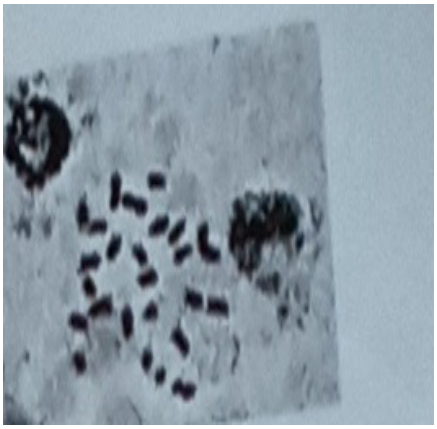

Figure 11: *S. lycopersicum* showing its diploid (2n) chromosome number.

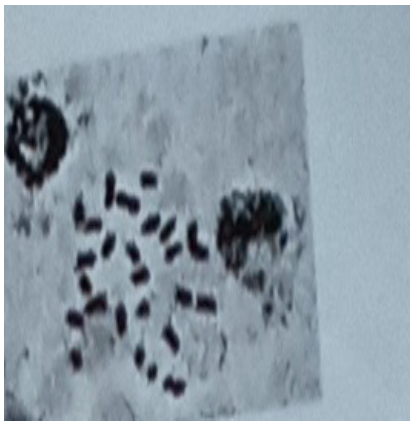

Figure 12: *S. pimpinellifolium* showing its diploid (2n) chromosome number (24).

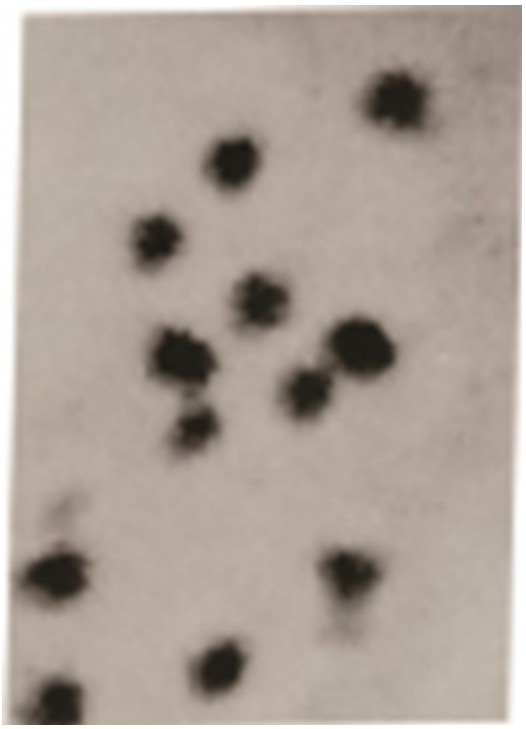

Figure 13: *S. lycopersicum* showing its haploid (n) chromosome number (12).

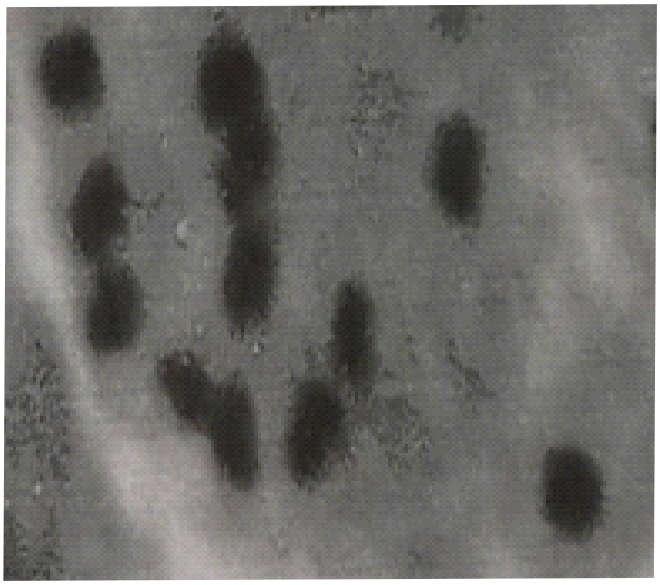

Figure 14: *S. pimpinellifolium* showing its haploid (n) chromosome number (12).

4. Discussion

The current research investigated the morphological and cytological features of Solanum lycopersicum L. (cultivated tomato) and Solanum pimpinellifolium L. (wild currant tomato), which are both species of the Solanaceae family. The findings from the research indicated both similarities and differences in the morphological characteristics of S. lycopersicum and S. pimpinellifolium, as members of the genus Solanum. The commonalities consist of having a taproot system accompanied by lateral roots, akin to dicotyledonous plants, a herbaceous growth form, the existence of branched stems, simple compound leaves featuring petioles, and the development of flowers that exhibit similar structures and arrangements. Furthermore, both species have similar floral characteristics and luscious berry fruits, suggesting a close taxonomic relationship **(Figures 1 and 2)**. Nevertheless, there were notable distinctions in their growth habits; for instance, S. lycopersicum exhibited a more upright growth pattern characterized by thicker stems, larger leaves, and considerably larger fruits, whereas S. pimpinellifolium displayed a more spreading or semi-prostrate growth habit with slender stems, smaller leaves, and very small fruits **(Figures 1 and 2)**. These traits primarily stem from the processes of domestication and the artificial selection aimed at enhancing agronomic performance[11,21]. The leaves tend to be bigger and less dissected than those of their wild counterparts, suggesting a selection process aimed at enhancing photosynthetic efficiency and yield performance[2]. Variations in plant height, leaf size, stem thickness, and fruit dimensions were also noted, with S. lycopersicum typically exhibiting greater values compared to S. pimpinellifolium **(Table 1)**. These variations can be ascribed to the influence of domestication and selective breeding in S. lycopersicum aimed at enhancing agronomic performance, in contrast to the wild characteristics of S. pimpinellifolium[11,21]. This aligns with the findings of Lester et al.[22], which indicate that the leaves of plants belonging to the genus Solanum are simple compounds with petioles and an herbaceous growth habit. In contrast, the cultivated tomato displays a broader variety of fruit shapes and sizes, while its wild relatives consistently yield small, round fruits[23]. These differences may explain the differentiation in genus. The qualitative and quantitative characteristics observed strengthen the interspecific relationships and can be employed to enhance precise taxonomic classification and identification of plant species with considerable economic value. Morphological characteristics are essential for species delimitation and taxonomic categorization in plants[3].

The cytological findings revealed that both S. lycopersicum and S. pimpinellifolium have a diploid chromosome count of 2n = 24, along with a meiotic chromosome count of n = 12, which are preserved throughout the tomato clade[24]. The publication of the tomato reference genome has validated the structural integrity of the tomato genome and has uncovered significant synteny between cultivated tomatoes and their wild counterparts[24]. Comparative genomic studies indicate that S. pimpinellifolium exhibits considerable chromosomal similarity with S. lycopersicum, reinforcing their close evolutionary relationship. The meiotic behavior observed in tomato species demonstrates consistent chromosome pairing and segregation, which implies structural similarity and genomic compatibility[24]. The absence of polyploidy in cultivated tomatoes further suggests that the processes of speciation and diversification within the tomato lineage primarily occurred through gene-level mutations and structural variations, rather than through whole-genome duplication. Comparative genome sequencing of S. pimpinellifolium has uncovered greater genetic diversity compared to cultivated tomatoes, especially concerning genes related to stress response and disease resistance. This diversity illustrates the genetic bottleneck that took place during the domestication of S. lycopersicum[2]. However, the preserved chromosome count and genome architecture clarify the reasons behind the successful interspecific hybridization between the two species, which is extensively employed in breeding initiatives[11].

5. Conclusion

This study revealed that Solanum lycopersicum L. and Solanum pimpinellifolium L. share important morphological and cytological characteristics that confirm their close taxonomic and evolutionary relationship within the genus Solanum. Both

species exhibited similar vegetative and reproductive structures and maintained a conserved diploid chromosome number of 2n = 24 (n = 12), indicating chromosomal stability and genomic compatibility.

However, distinct differences were observed in growth habit, stem thickness, leaf size, and particularly fruit size, with S. lycopersicum showing traits associated with domestication and selection for improved agronomic performance, while S. pimpinellifolium retained typical wild characteristics. Overall, the combined morphological and cytological evidence demonstrates that although the two species are closely related, they are clearly distinguishable at the interspecific level. The conserved genome structure explains the success of interspecific hybridization and highlights the importance of the wild relative as a valuable genetic resource for tomato breeding and crop improvement programme.

6. Authors' Contributions

This work was carried out in collaboration between all authors. Author CVI designed the study and author IEO wrote the first draft of the manuscript and managed the literature searches. Authors IEO and OKK managed the analyses of the study and Author CVI supervised the work

7. Competing Interests

Authors have declared that no competing interests exist.

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