

Available on <https://www.joarps.org>
Journal of Applied Research in Plant Sciences
(JOARPS)
ISSN: 2708-3004 (Online), 2708-2997 (Print)



Natural and Artificial Sweeteners a Blessing or Curse: A Phyto-genotoxic Study in *Allium cepa* L. Root Tip Cells

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Article Received 20-05-2026, Article Revised 19-06-2026, Article Accepted 25-06-2026

ABSTRACT

The study investigated the comparative phyto-genotoxic impacts of natural and artificial sweetener brands using the *Allium cepa* L. (onion) assay to predict potential phyto-genotoxic associated with long-term intake. Onion bulbs were exposed for 48 hours to 1% aqueous solutions of natural and artificial sweeteners (Honey, Date, Jaggery, Canderel, Sucral, Stiva and Refined sugar) and distilled water was used as the negative control. All the tested sweeteners exhibited significant variations at $LSD \leq 0.05$ for all phytotoxicity and genotoxicity parameters. The results phytotoxicity, characterized by a marked failure of root emergence and reduction in root length and Mitotic index (M.I.%). Genotoxic analysis showed that both sweetener categories induced a substantial increase in Abnormality Index (A.I.%), escalating from 14.2% at the lowest concentration to 47.2% at the highest. The most prevalent chromosomal aberration was DNA fragmentation in all the tested sweeteners and while occurrences of sticky metaphase and micronuclei were also recorded in low number. Stiva induced nuclear lesions, and impaired chromosome condensation. These two are checkpoints of carcinogenesis in addition to chromosomal aberrations. The high frequency of nuclear damage and cellular membrane irregularities suggests that exposure to 1% solutions of both natural and artificial sweeteners can induce measurable genetic instability. Overall results suggest that Stiva followed by Canderel, Sucral and refined sugar were severely phyto-genotoxic, while honey was moderately phyto-genotoxic and Dates followed by jaggery were the mild phyto-genotoxic. It is concluded that sweetener intake must be minimized to avoid possible cellular and DNA damages. A dose dependent animal-based study must be conducted to estimate cancer risk to humans. Reactive oxygen species generation, lipid peroxidation profiling is also recommended.

KEYWORDS: Sweeteners, Phytotoxicity, Genotoxicity, *Allium cepa* L. Oxidative damages, Carcinogenesis

INTRODUCTION

Since early ages, humans are fond of eating sweets. In early days they have started consuming sugar from natural sources like honey and dates. Later, they have started cooking sweet rice with raw sugarcane juice. With the passage of time love of sweet increased they started to store sugarcane juice by slight processing in solid form called jaggery. As jaggery alter color of, the cosine to dull, refining of jaggery was started by adding much more chemicals turning it to white crystals, called table sugar or refined sugar. **Natural sweeteners** are either plant-based or less processed including honey, dates and jaggery and highly processed form is refined sugar. These are rich in glucose and fructose, vitamins, micro macro nutrients and immunity-boosting compounds like antioxidants and phenols in general (Ibrahim *et al.*, 2020; Mongi and Ruhembe, 2024; Ogwu and Izah, 2025; Zahadi *et al.*, 2026).

The overuse of natural sweeteners often leads to diabetes. Diabetes Mellitus is extremely common all over the world and Pakistan has one of the highest rates

in the world with over 33 million patients (Magliano and Boyko, 2024). Natural sweeteners might seem safe, but your body still treats them like regular sugar. In low intake, Bee's Honey (Banerjee *et al.*, 2018) and Jaggery (Hirpara *et al.*, 2020) provide medicinal benefits, aiding in digestion, liver health. Honey is reported as anticancer and antiviral due to its cytotoxic abilities (Abdelbaky *et al.*, 2023). Dates contain 2.8 calories per gram which leads to weight gain and high potassium content can lead to Hyperkalaemia (Ismail *et al.*, 2025). While natural Stevia plant tea is reported to have beneficial cytotoxic effects against cancer cell lines (damaging cancer cells), research by Nurcayani *et al.* (2025) warns that over-intake by a healthy human may result in cytotoxicity that leads to altered physiology. Refined sugar is tied to obesity and neurodevelopmental issues (Arshad *et al.*, 2022).

Such sweet lovers do not compromise eating sweets after getting sick rather switched to artificial sweeteners directly or in products as sugar free drinks. **Artificial sweeteners** brands commonly used in Pakistan are Canderel and Sucral contains lab-

synthesized chemicals like Saccharin, Aspartame and glycosides, which often lack nutrients and present metabolic risks (Sachan and Singh, 2026). High consumption of artificial sweeteners among diabetic patients is a serious health concerns as some literature is available regarding cellular as well as genetic toxicity.

Long-term use of artificial sweeteners is generally linked to gut dysbiosis, stroke, and coronary heart disease (Meenatchi and Vellapandian, 2024). Sodium saccharin and cyclamate used in tetra pack juices are reported to induce mitotic alterations and DNA damage (Bezerra *et al.*, 2016; Oliveira *et al.*, 2017). The utilization of Canderel specifically has been linked to a growing number of health problems. Helal *et al.* (2019) demonstrated that aspartame-based product “Canderel” induces disturbances in liver enzymes, kidney markers, and hormones. Furthermore, higher consumption of aspartame is associated with a 13% increased risk of overall cancer (Debras *et al.*, 2022). Several studies have reported sucralose as cause of metabolic, genomic alterations and oxidative stress highlighting the need for biological safety of sucralose because its resistance to degradation, as it consumed continuously in sugar free products (Tkach *et al.*, 2025). Stiva a recently introduced product in Pakistan contains 2g of Erythritol and stevia leaf extract (amount not mentioned on label). One sachet weigh 2g and if it contains 2g of Erythritol then it means either it does not contain stevia leaf extract, or the quantities added are unknown. These facts make this product questionable. As crude Stevia leaf extract is not recommended by U.S. Food and Drug agency rather purified steviol glycosides are approved for daily intake (4 mg/kg of body weigh) (Perrier *et al.*, 2018; EFSA FAF Panel, 2021). As Erythritol is associated with high risk of cytotoxicity due to its oxidative properties (Berry *et al.*, 2025).

Therefore, screening of natural as well as artificial sweeteners for phyto-genotoxicity is extremely important. The current work is vital to provide awareness to the common man regarding cancer risks due to the utilization of processed, un-proceeded natural sweeteners and artificial sweeteners. It aims to provide baseline information for researchers, specifically for those conducting animal studies, to help them understand the epigenetic shifts and DNA damages leading to carcinogenicity.

Therefore, the present study was designed to evaluate the phyto-genotoxic potential of selected sweeteners in *Allium cepa* L. and to guide safer consumption.

MATERIAL AND METHODS

Current work was performed in the Genetics Research Laboratory, Institute of Plant Sciences, University of Sindh, Jamshoro, during February to March 2026. The experiment was conducted in a Complete Randomized Design (CRD) with three replications.

Assay plant and its germination: *Allium cepa* L. was used as standard higher plant. Onion bulbs of approximately medium size were obtained from the local market of Jamshoro, Sindh. Dried outer scales were carefully removed, leaving the ring of root primordia intact by using the blunt side of a knife. Bulbs were washed and initially kept in distilled water (dH₂O) for 48 hours to initiate rooting.

Sweeteners used for the study: Current work comprises of phyto-genotoxicity testing of 7 sweeteners (Honey, Date, Jaggery, Refined sugar, Canderel, Sucral, Stiva) (Figure 1). All the sweeteners were applied as 1% aqueous solutions for 48h and dH₂O grown roots were used as negative control

Sweetener solution preparation and treatment: To prepare the 1% aqueous solutions, each sweetener was processed according to its physical properties to ensure uniform integration into distil water. The Canderel and Sucral tablets were first crushed into a fine powder using a mortar and pestle before being dissolved, while the Stevia powder was added directly from the sachets. For the natural sweeteners, the dates were sliced into small pieces and processed in a blender to achieve a smooth, homogenous paste, whereas the honey and sugar were simply stirred into the water until the honey was fully incorporated and the sugar crystals had completely vanished. Finally, a sample of pure distilled water was maintained as a control group to provide a baseline for the study. Pre-germinated bulbs of onion in dH₂O (48h) were shifted to the 1% solutions of sweeteners for 48 hours. The negative control bulbs were maintained in dH₂O. Solutions for each treatment were changed daily throughout the process until the collection of root tips (Figure 2).

Germination and growth assay to monitor phytotoxicity: Root number was simply counted, and root length was measure in cm with a scale before and after the 48-hour treatment with sweeteners as well as for negative control and expressed as mean.

Fixation and storage of the roots: The root tips of the onion bulb were cut and kept in Carnoy’s solution 1 for 24 hours to stop and stabilize cell division of mitosis and then shifted to 70% alcohol at room temperature 25C°



Figure 1: Sweeteners evaluated for phyto-genotoxicity and product labels showing artificial sweetener composition

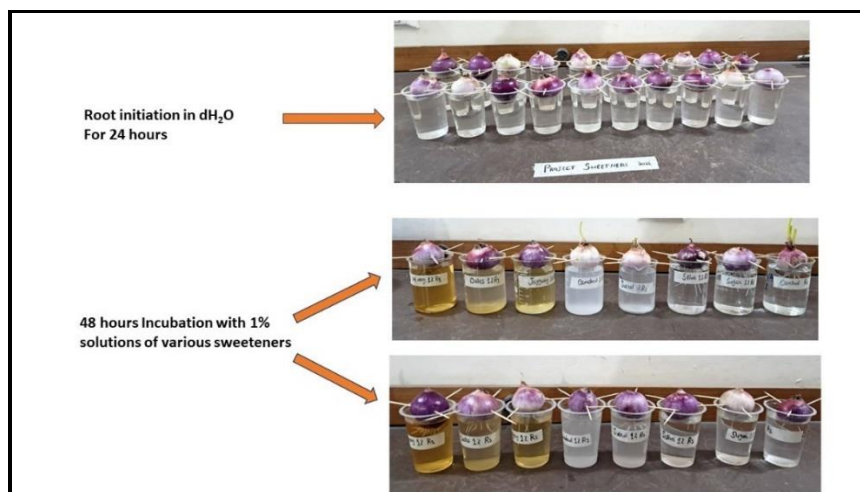


Figure 2: Initial growth of Onion bulbs in dH₂O and shifting to sweetener solutions for 48h for phyto-genotoxicity evaluation

Slide preparation: Collected roots of *Allium cepa* L. were spread by using the squash technique, slides were stained using acetocarmine (2%) prepared in 45% Glacial acetic acid (Channar *et al.*, 2025; Sadia *et al.*, 2026). Three slides per treatment and 250 cells per slide were observed to score the aberrations with the help of Olympus 52x inverted microscope with digital

$$M.I = \frac{\text{Total number of dividing cells} \times 100}{\text{Total cells analyzed}}$$

camera. (Mitotic index, abnormality index, cell cycle abnormalities, oxidative stress) Slides examined and photographed at 400 magnifications.

Mitotic index (%): The following equation was used to calculate percentage of total dividing cells (Racuciu, 2009):

Chromosomal aberration assay to monitor Genotoxicity: Genotoxic effects were assessed by abnormality index and types of chromosomal abnormalities.

$$\text{A.I} = \frac{\text{Total number of abnormal dividing cells} \times 100}{\text{Total number of dividing cells}}$$

Types of chromosomal abnormalities: Cells were simply counted in each type of chromosomal aberrations observable under microscope with simple staining and expressed as number of cells.

Oxidative damages: Oxidation is the most common mechanism of degradation of DNA, lipid and proteins leading to nuclear and cell membrane damage and culminating in the formation of ghost cells. Occurrence of oxidative damages were recorded by cytology as suggested by (Firbas and Amon, 2014; Qureshi et al. 2015; Channar et al., 2025; Sadia et al., 2026).

Statistical Analysis

Least Square Difference (LSD) test at $p \geq 0.05$ was used to compare treatment mean for root number, root length (cm), M.I. (%), A.I. (%), and oxidative damages using computer software statistix 8.1.

RESULTS AND DISCUSSIONS

Phytotoxicity analysis:

Root number and Length (cm): The results of root number showing significant variations at $\text{LSD} \leq 0.05$ are compiled in Table 1 and Figure 3. The multiplication of roots over a 48-hour period the natural sweeteners showed the least activity; Honey and Date resulted in zero new roots. While Jaggerly and the Negative Control showed a slight increase, the Artificial Sweeteners again proved to be more stimulatory. Canderel and Sucral showed moderate increases in root count, but Sativa (Stiva) was the clear outlier, producing the highest increase of all tested groups with 6.5 new roots. Interestingly, in this specific metric, Sativa significantly outperformed pure Sugar, which only showed a 1.5 increase. These findings suggest that Sativa contains properties that specifically trigger root branching or initiation more effectively than standard carbohydrates or other natural sweeteners. This may also be due to reflect mutagenic properties resulting in increased level of gene expression of growth harmons.

Significant variations at $\text{LSD} \leq 0.05$ for root length (cm) (Table 1 and Figure3) was observed for natural sweeteners and artificial sweeteners as compared to negative control. Un processed natural treatments like honey and jaggerly showed minimal growth increase, while date resulted in no increase in root length (0 cm). In contrast, the artificial sweeteners brands Canderel and Sucral showed moderate increases. The most significant growth was observed in Stiva (1.107 cm) and refined sugar treatment (1.134 cm). The results suggests that the artificial sweeteners present in sugar free brands and refined sugar more aggressively

Abnormality index (%): Using the following formula, the percentage of abnormal dividing cells was determined (Racuciu, 2009):

promotes longitudinal root extension. This may be due to increase in rate of replication.

Mitotic index (M.I.%): The results regarding cell division reveal that different sweeteners have a significant impact ($\text{LSD} \leq 0.05$) on cellular activity (Table 2, Figure 4). Stiva, Honey, and Dates exhibited the highest M.I. (%), with Stiva leading at 98%, followed by Honey at 89.2% and Dates at 88.6%. This indicates that these substances promoted the most active cell division among the tested groups. Conversely, brands containing artificial sweeteners resulted in the lowest rates of cell division, specifically 74% for Sucral and 76% for Canderel.

High level of phytotoxicity found in current work is supported by many researchers working on natural and artificial sweeteners in different assay systems (Table 1 & 2). Banerjee et al. (2018) working with Huh-7 human liver cancer cells found Bee's honey as potent cytotoxic leading to apoptosis. High cytotoxicity found in product stiva may be due to higher Erythritol content (2g/sachet) that is associated with nerve cell damage (Berry et al., 2025). The second component of Stiva is stevia plant leave extract reported as cytotoxic in human cell line (Ibrahim et al., 2020; Kulkarni et al., 2021; Selvapriya et al., 2021; Orellana-Paucar, 2023). The ethyl acetate fraction of stevia leaf extract at 1000 ppm totally inhibits weed germination and growth, whereas chloroform fraction was cytotoxic at 700 ppm in the brine shrimp assay (Nuryandani et al., 2024). High phytotoxicity observed in refined sugar, Canderel and sucral is supported by the findings of Bezerra et al. (2016), Oliveira et al. (2017) and Prakash et al. (2021); who found Polyphenol-rich sugarcane extract, Sodium saccharin and sodium cyclamate and other artificial sweeteners used in powder fruit juices extremely phytotoxic in Onion Mouse and human cancer cell lines.

Genotoxicity analysis

Abnormality Index (A.I.%): Mean comparison at $\text{LSD} \leq 0.05$ revealed significant variations between the tested sweeteners as compared to negative control for A.I% (Table 2, Figure 4.). Our findings revealed refined sugar as the most DNA damaging substance among the sweeteners, reaching an A.I. of 84.5%. While Stiva (70.5%), Canderel (69%), and Sucral (69%) also showed high aberration rates, Honey appeared to be the most stable sweetener with the lowest abnormality rate (46.1%).

Table 1: Difference in mean root number and root length before and after treatment of sweeteners at LSD (≤ 0.05)

S.No	Treatment	Root Number			Root length (cm)		
		Before	After	Differences	Before	After	Differences
1	Honey	13.5	13.5	00 ^C	1.006	1.24	0.234 ^B
2	Date	20	20	00 ^C	0.47	0.47	0 ^C
3	Jaggery	12.5	14 ^B	1.5 ^B	1.568	1.64	0.072 ^B
4	Canderel	21.5	24	2.5 ^B	0.637	1.04	0.034 ^B
5	Sucral	16	19.5	3.5 ^B	1.334	1.78	0.446 ^B
6	Sativa	11	17.5	6.5 ^A	1.663	2.77	1.107 ^A
7	Sugar	16	17 ^B	1 ^C	1.88	3.014	1.134 ^A
8	-ve control	6	7.5	1.5 ^B	0.933	1.47	0.537 ^B

(Critical value =2.093; Means with same alphabet are not significantly different)



Figure 3. Onion bulbs before and after 48h treatment of 1% sweeteners aqueous solutions showing varying number and length of roots

Table 2. Number of non-dividing, normal and abnormal dividing cells in *Allium cepa* L. to calculate mean M.I. and A.I. (%) (LSD (≤ 0.05) induced by -tive control and 1% sweeteners aqueous solutions

S#NO	Treatment	Non-Dividing Cells	Normal Dividing cells	Abnormal dividing Cells	M.I. (%)	A.I. (%)
1	Honey	27	120	103	89.2 ^B	46.1 ^B
2	Dates	28.5	93.5	128	88.6 ^B	57.7 ^B
3	Jaggery	50	64	136	80 ^B	68 ^B
4	Canderel	97	125	128	76 ^C	69 ^B
5	Sucral	65	57	128	74 ^C	69 ^B
6	Stiva	80	50	120	98 ^A	70.5 ^B
7	Refined Sugar	91	24.5	134.5	78 ^C	84.5 ^A
8	-tiv Con.	10	220	20	96 ^A	8.3 ^C

(Critical value =2.093; Means with same alphabet are not significantly different)

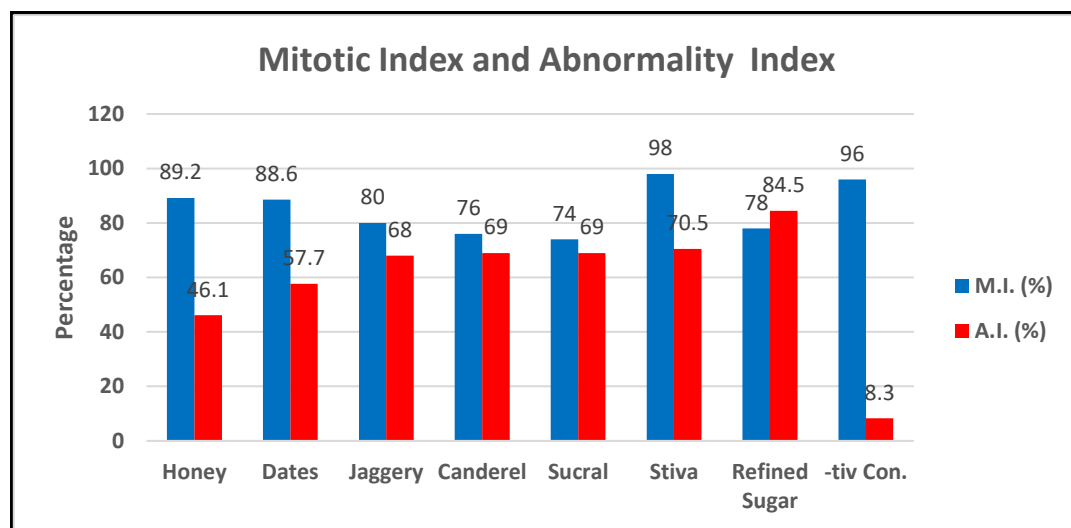


Figure 4. Effect of 48h treatment of 1% sweeteners aqueous solutions on M.I. and A.I. of *Allium cepa* L. root tip cells

Types of chromosomal abnormalities in sweeteners:

The data for natural sweeteners (Table 3 and 4, Figure 5 and 6) illustrates specific types of structural damage observed during different stages of the cell cycle. Jaggery was particularly notable for inducing a high number of scattered nuclei (75) during the metaphase stage. During the interphase stage, Dates were associated with significant occurrences of micro-nuclei (77.5) and di-nuclei (33.5). Cellular fragmentation was a widespread issue among all unprocessed natural sweeteners, with Honey (70) and Jaggery (65) showing particularly high counts. While the negative control maintained very low levels of aberrations, Honey showed a notable increase in micro-nuclei (26) compared to the control (4). Ultimately, Jaggery recorded the highest total number of abnormal cells within the natural sweetener group (136).

Processed natural refined sugar induced severe DNA damage. Competing with artificial sweetener brands (Table 3 and 4, Figure 5 and 6). The most prominent defect across this entire group was fragmentation, with Stiva recording the highest frequency at 120, followed by 100 each for Canderel, Sucral, and Refined Sugar. This suggests that these substances are highly likely to cause the double stranded breaks in chromosome during division. Other aberrations, such as micro-nuclei, were observed in Canderel (20), Sucral (20), and Refined Sugar (10). Refined Sugar caused the most diverse range of damage, showing instances of scattered nuclei (13.5), imbalance during anaphase (1), and multi-nuclei (10). In this specific category, Refined Sugar ended up with the highest total number of abnormal cells (134.5).

Higher number of aberrant cells (Table 2, 3 & 4) especially fragmented cells induced by all artificial sweeteners is supported by the findings of Debras *et al.* (2022) who conducted huge population of French adults and found that higher consumption of aspartame and acesulfame-K is associated with 13% increased

risk of cancer. Natural fruit juices are also associated with higher cancer incidence and mortality in a study conducted by Eshaghian *et al.* (2023).

Epigenetic changes: A nuclear lesion describes a structural abnormality; it frequently results in or from epigenetic alterations (a change in the "locks" on the DNA). Nuclear lesions and incomplete condensation were found only in Stiva treated roots of *Allium cepa* L. (Figure 7).

Nuclear lesions induced by Stiva clearly reflect its carcinogenic potential. As changes in the nuclear envelope, lamina, and chromatin organization (nuclear lesions) can affect how genes are packaged (Lancôt *et al.*, 2007; Cavalli and Misteli, 2013). Tightly packed heterochromatin is often associated with silenced genes, while loose euchromatin is active reflected by incomplete condensation. Cancer cells often show "nuclear anarchy," with abnormal nuclear shapes and composition. This structure alteration is driven by and drives epigenetic alterations, such as DNA hypermethylation and histone modifications (Zink *et al.*, 2004).

Oxidative damages: The measurement of oxidative damage revealed significant variations at $LSD \leq 0.05$ (Table 5). Stiva caused the most intense oxidative damages, leading to the highest numbers of ghost cells (150), nuclear membrane (180) and cytoplasmic membrane (180) evident by irregular shapes and cellular tube-like connections called proliferation pilli. Canderel also exhibited high and uniform levels of damage, recording 125 (cells) in all three measured categories (Figure 8, 9 and 10).

Table 3: Types of abnormalities induced by negative control and natural sweeteners (1%)

Stage	S#No	Abnormalities	-tive Cont.	Honey	Dates	Jaggery
Metaphase	1	Scattered nuclei	12	5	0	75
	2	Sticky Metaphase	0	0	0	0
Anaphase	3	Laggard	0	1	5.5	0
	4	Translocation ring	2	0	5.5	1
	5	Imbalance anaphase		1	1	0
Interphase	6	Fragmentation	0	70	27.5	65
	7	Micro-nuclei	4	26	77.5	0
	8	Di-nuclei	0	0	33.5	0
	9	Multi-nuclei	2	0	7.5	0
		Total Abnormal cells	20	103	128	136

Table 4: Types of abnormalities induced by negative control and artificial sweeteners (1%)

Stage	S#No	Abnormalities	Canderel	Sucral	Stiva	Refined Sugar
Metaphase	1	Scattered nuclei	0	5	0	13.5
	2	Sticky Metaphase	0	0	0	0
Anaphase	3	Laggard	0	0	0	0
	4	Translocation ring	0	0	0	0
	5	Imbalance anaphase	0	0	0	1
Interphase	6	Fragmentation	100	100	120	100
	7	Micro-nuclei	20	20	0	10
	8	Di-nuclei	0	0	0	0
	9	Multi-nuclei	8	3	0	10
		Total Abnormal cells	128	128	120	134.5

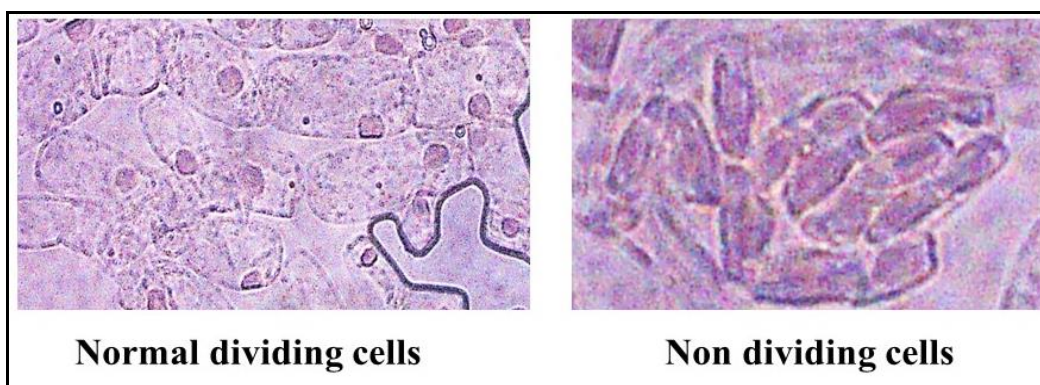


Figure 5. Normal dividing cells and non-dividing cells induced by 48h treatment of 1% sweeteners aqueous solutions

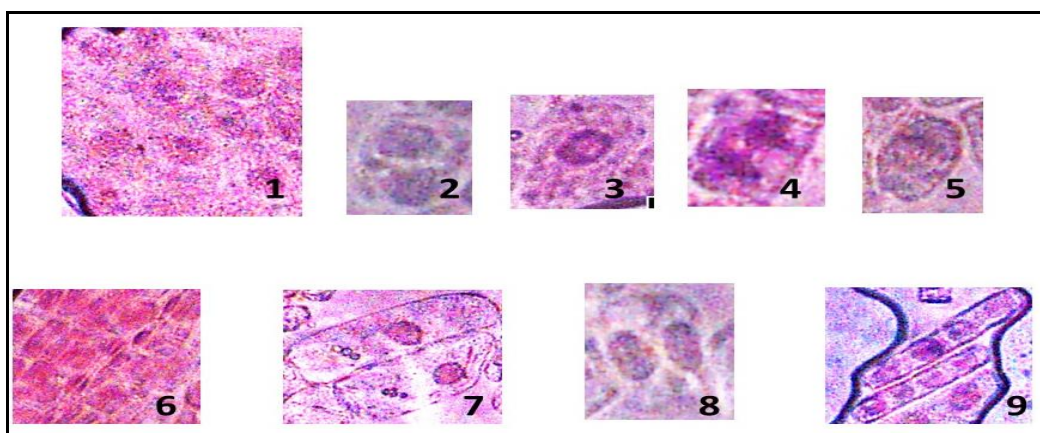


Figure 6. Aberrant cells induced by 48h treatment of 1% sweeteners aqueous solutions (1=Scattered nuclei, 2= laggard, 3= Translocation ring, 4= chromosomal bridge, 5= Imbalanced anaphase, 6= Fragmentation, 7= micro-nuclei, 8= Di-nuclei, 9= multi-nuclei)

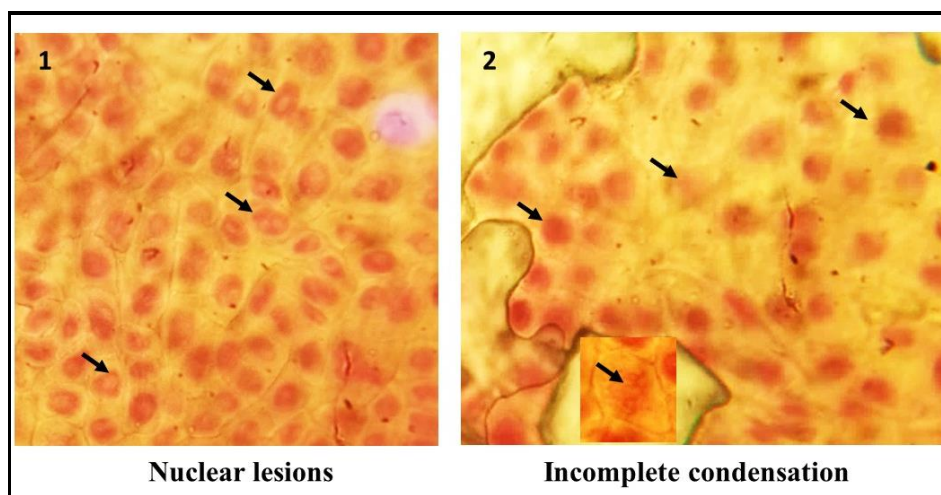


Figure 7. Nuclear lesions and incomplete condensation of chromosomes induced by 48h treatment of 1% Stiva aqueous solutions

In comparison, natural options like dates and honey resulted in much lower levels of membrane damage, indicating they are less physically aggressive toward the cells. While refined sugar caused significant damage to the cell membranes (110 for N.M. and 113.5 for C.M.), it produced fewer ghost cells (50) than the artificial sweeteners. The negative control consistently showed the lowest levels of damage (10 or less), providing a clear contrast to the stress induced by the sweeteners (Table 5; Figure 8, 9 and 10). High degree of oxidative stress (Table 5) found in artificial sweeteners in current study is supported by Xu *et al.* (2025), who concluded that aspartame, acesulfame potassium, sucralose, and saccharin has ability to induce oxidative stress to skeletal muscle tissues Tkach *et al.* (2025) and Lui *et al.* (2026) also found hepatotoxic effects and glycolipid metabolism disruption mechanism in the zebrafish liver. ACE significantly inhibited antioxidant enzyme (SOD, CAT) activities, induced reactive oxygen species (ROS) accumulation, leading to lipid peroxidation and

DNA damage. Meanwhile, chronic high-concentration exposure exacerbated cell apoptosis. Oxidative properties of Aspartame, saccharin, sucralose, and stevia in plant and animal-based studies has been reported earlier (Atalay *et al.* (2025)). Stevia leaf extract primarily operates as an antioxidant. However, in specific cancer cell lines, certain concentrated Stevia extracts can indeed induce cytotoxicity by temporarily spiking intracellular Reactive Oxygen Species (ROS) and promoting oxidative stress, leading to cell death (apoptosis) (Wisal *et al.*, 2025). Other component of Stiva is Erythritol that is reported to increase oxidative stress (Berry *et al.*, 2025). Overall results suggest that product Stiva followed by Canderel, Sucral and refined sugar were severely genotoxic, while honey was moderately cyto-genotoxic and Dates followed by jaggery were the mild cyto-genotoxic sweeteners. A dose dependent animal-based study must be conducted to estimate cancer risk to humans. Reactive oxygen species generation, lipid peroxidation profiling is also recommended.

Table 5. Mean comparison for oxidative damages (LSD $p \leq 0.05$) induced by 48h treatment of sweeteners aqueous solutions (1%)

S#No	Treatment	Ghost cell	N.M. Damage	C.M. Damage
1	Honey	60 ^B	64.5 ^{BC}	60 ^C
2	Dates	25 ^C	30 ^B	30 ^{CD}
3	Jaggery	40 ^B	40 ^B	40 ^C
4	Canderel	125 ^A	125 ^A	125 ^A
5	Sucral	50.5 ^B	48.5 ^B	78 ^{BC}
6	Stiva	150 ^A	180 ^A	180 ^A
7	Refined Sugar	50 ^B	110 ^A	113.5 ^B
8	-tiv Con.	10 ^C	3 ^C	3 ^D

(N.M. Damage= Nuclear membrane Damage; C.M. Damage= Cell membrane Damage; Critical value =2.093; Means with same alphabet are not significantly different)

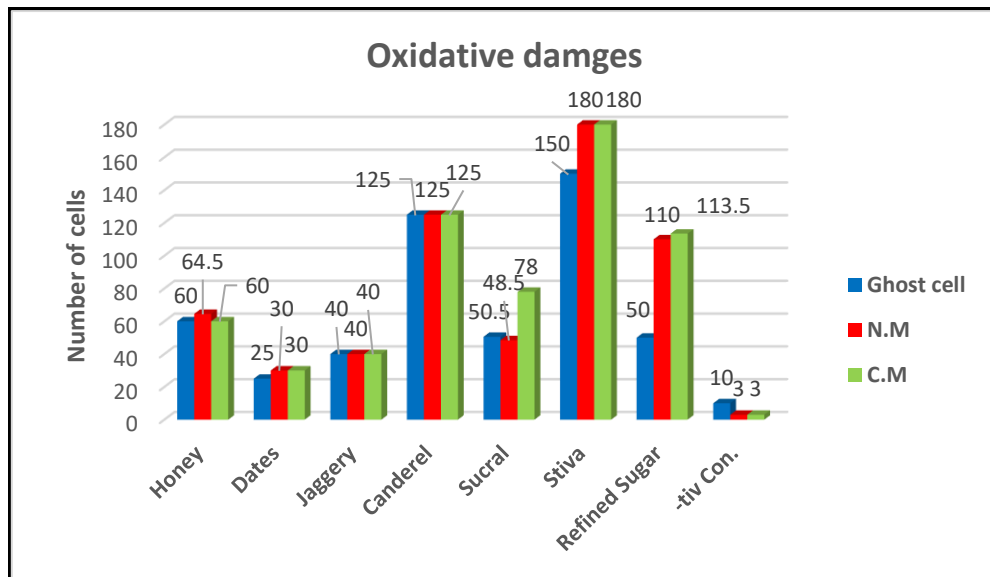


Figure 8. Oxidative damages induced by 48h treatment of 1% Stiva aqueous solutions

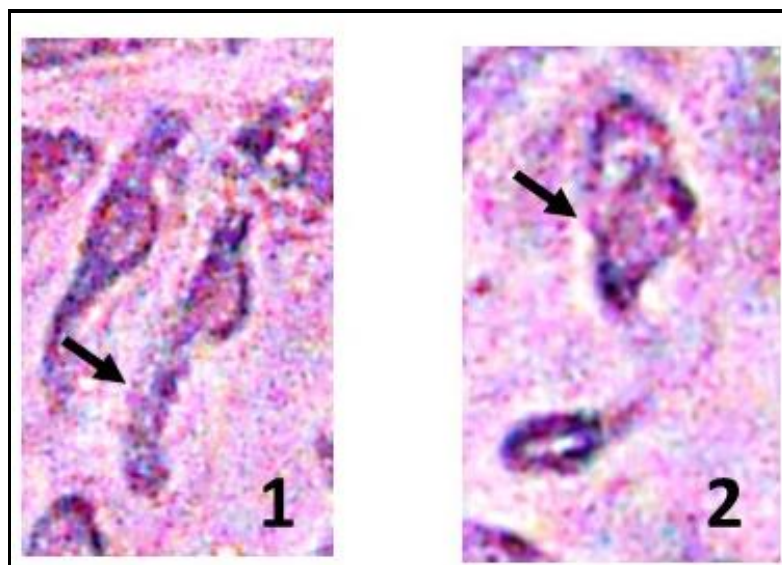


Figure 9: Membrane damage observed in sweetener treated root tip cells indicated by arrows (Damaged nuclear and cell membranes)

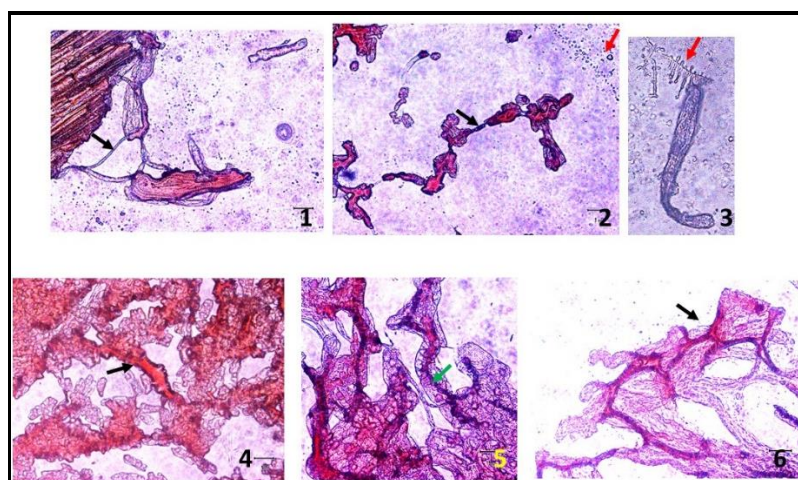


Figure 10. Ghost cells observed in sweeteners with Proliferation Pilli (Cell 1, 2, 3 & 4) and connected nuclei (Cell 5 & 6) indicated by arrows

CONCLUSION

All the tested sugars exhibited different levels of cyto-genotoxicity; therefore sweetener intake must be minimized to avoid possible cellular and DNA damages.

CONFLICT OF INTEREST

No conflicts of interest are declared by the authors.

AUTHOR CONTRIBUTION

S.T. Qureshi conceived, designed research, conducted statistical analysis and revised manuscript and approved final version of manuscript. S. Zahid edited and revised discussion of manuscript, M R Laghari, S A Rajper, S A Jarwar, S Bheel, KA Arian performed experiments, interpreted results of experiments, prepared figures and drafted manuscript.

REFERENCES

- Abdelbaky, A.S., Tammam, M.A., Ali, M.Y., Sharaky, M., Selim, K., Semida, W.M., Abd El-Mageed, T.A., Ramadan, M.F., Oraby, H.F., Diab, Y.M. (2023). Antioxidant and Anticancer Assessment and Phytochemical Investigation of Three Varieties of Date Fruits. *Metabolites*, **13** (7), 816. DOI: 10.3390/metabo13070816.
- Arshad, S., Rehman, T, Sail, S., Rajoka, M.S.R., Ranjha, M.M.A.N., Hassoun, A., Cropotova, J., Trif, M., Younas, A. and Aadil, R.N. (2022). Replacement of refined sugar by natural sweeteners: focus on potential health benefits. *Heliyon*, **8**(9), e10711. DOI: org/10.1016/j.heliyon.2022.e10711.
- Atalay, E.C., Celep, A.G.S., Catanzaro, R. and Marotta F. (2025). Aspartame, saccharin, sucralose, and stevia: Oxidative stress pathways and functional food perspectives. *Bioactive Compounds in Health and Disease*, **8**(11), 474-48.
- Banerjee, A., Das, D., Maji, B.K., and Mukherjee, S. (2018). An in vitro cytotoxic activity of Bees' honey in Huh-7 cell line. *Biotechnology and Microbiology*, **9** (1), 555751. DOI: 10.19080/AIBM.2018.09.555751.
- Berry, A., Ruzzene, S., Izaia, J., Holzer, J., Stone, M., Orozco-Fersiva, N., Greiner, W.V.J., Garcia V.P. and DeSouza, C.(2025) Erythritol Increases Oxidative Stress and Reduces Nitric Oxide Production in Human Cerebral Microvascular Endothelial Cells. *Physiology*, **40**, (1) https://doi.org/10.1152/physiol.2025.40.S1.1054
- Bezerra, M.D.S., Malaquias, G.D.S., Castro e Sousa, J.M.D. and Peron, A.P. (2016). Cytotoxic and genotoxic potential of powdered juices. *Food Science and Technology*, **36**(1), 49-55. DOI: 10.1590/1678-457X.0006.
- Cavalli, G. and Misteli, T. (2013). Functional implications of genome topology. *Nature Structural and Molecular Biology*, **20**(3), 290-299.
- Channar, A.P., Sadaf Tabasum Qureshi, S.T., Soomro, A.N., Saddar A.N., Sadia (2025). DNA damaging effects of lotion vs electric mosquito repellent assessed via chickpea chromosomal aberration assay. *Journal of Applied Research in Plant Sciences*, **6** (2), 143-154. https://doi.org/10.38211/joarps.2025.06.323.
- Debras, C., Chazelas, E., Srouf, B., Druet-Pecollo, N., Esseddik, Y., Edelenyi, F.S.D., Agae, C., Sa, A.D., Lutchia, R., Gigandet, S., Huybrechts, I., Julia, C., Kesse-Guyot, E., Alles, B., Andreeva, V.A., Galan, P., Hercberg, S., Deschasaux-Tanguy, M.I., Touvier, M. (2022). Artificial sweeteners and cancer risk: Results from the NutriNet-Sante' population-based cohort study. *PLoS Medicine*, **19** (3), e1003950. DOI: 10.1371/journal.pmed.1003950.
- EFSA FAF Panel (EFSA Panel on Food Additives and Flavourings). (2021). Scientific opinion on the safety evaluation of steviol glycoside preparations, including rebaudioside AM, obtained by enzymatic bioconversion of highly purified stevioside and/or rebaudioside a stevia leaf extracts. *EFSA Journal*, **19**(8), 6691. DOI:10.2903/j.efsa.2021.6691.
- Firbas, P., and Amon, T. (2014). Chromosome damage studies in the onion plant *Allium cepa* L. *International Journal of Cytology, Cytosystematics and Cytogenetics*, **67** (1), 25-35.
- Helal, E.G.E., Abdelaziz, M.A., El-Gamal, M.S. (2019). Effects Of CANDEREL® and the ameliorative role of stem cell enhancer on some physiological parameters in male albino rats. *The Egyptian Journal of Hospital Medicine*, **76** (3), 3702-3708.
- Hirpara, P., Thakare, N., Kele, V.D., and Patel, D. (2020). Jagger: A natural sweetener. *Journal of Pharmacognosy and Phytochemistry*, **9**(5), 3145-3148.
- Ibrahim, E.S., Ragheb, E.M., Yousef, F.M., Abdel-Aziz, M.F., and Alghamdi, B.A. (2020). Nutritional value, cytotoxic and antimicrobial activities of *Stevia rebaudiana* leaf extracts. *Journal of Biochemical Technology*, **11**(12), 108-115.
- International Diabetic Federation (I.D.F). 2024. <https://idf.org/our-network/regions-and-members/middle-east-and-north-africa/members/pakistan/>.
- Ismail, I., Zakaria, N.F., Yong, J.H. Y., Ismail, N. (2025). Lessons From Practice: Severe Hyperkalemia and life-threatening events after dates consumption during Ramadan in end-stage renal failure patient. *Malaysian Journal of Emergency Medicine*, **7**(1): 41-43.
- Kulkarni, A.S., Karkhanis, D., Barve, S.S. and Sharma, S. (2021). Antioxidant and cytotoxic study of extract of vegetative and flowering stages leaves *Stevia rebaudiana*. *Journal of Pharmacognosy and Phytochemistry*, **10** (4), 303-306.
- Lancôt, C., Cheutin, T., Cremer, M., Cavalli, G. and Cremer, T. (2007). Dynamic genome architecture in the nuclear space: regulation of gene expression in three dimensions. *Nature Reviews Genetics*, **8**(2), 104-15. DOI: 10.1038/nrg2041.
- Magliano, D.J., Boyko, E.J. (2021). IDF Diabetes Atlas 10th edition scientific committee. 10th edition. Brussels: International Diabetes Federation; 2021. Chapter 3, Global picture. Available from: <https://www.ncbi.nlm.nih.gov/books/NBK581940/>.
- Meenatchi, M., Vellapandian, C. (2024). Exploring the Long-Term Effect of Artificial Sweeteners on Metabolic Health. *Cureus*, **16**(9), e70043. DOI: 10.7759/cureus.70043.
- Mongi, R.J. and Ruhembe, C.C. (2024). Sugar profile and sensory properties of honey from different geographical zones and botanical origins in Tanzania. *Heliyon*, **10** (18), e38094. doi: 10.1016/j.heliyon.2024.e38094.
- Nurcayani, N., Barliana, M.I., Dandan, K.L. (2025). Cytotoxic effects of tea (*Camellia sinensis*), stevia (*Stevia rebaudiana*), moringa (*Moringa oleifera*) leaves, and their potential as functional beverages for cancer patients. *Bioactive Compounds in Health and Disease*, **8** (8), 292 – 307. Doi.org/10.31989/bchd.v8i8.1690.
- Nuryandani, E., Kurnianto, D., Jasmadi, J., Sefrienda, A.R., Novitasari, E., Apriyati, E., Wanita, Y.P., Indrasari, S.D., Sunaryanto, R., Tjokrokusumo, D., Yani, A., Suryaningtyas, I.T., and Andriana, Y. (2024). Phytotoxic and Cytotoxic Effects, Antioxidant Potentials, and Phytochemical Constituents of *Stevia rebaudiana* Leaves. *Scientifica*, **2024**, 2200993. <https://doi.org/10.1155/2024/2200993>.
- Ogwu, M.C. and Izah, S.C. (2025). Honey as a Natural Antimicrobial. *Antibiotics*, **14** (255), 1-29. <https://doi.org/10.3390/antibiotics14030255>.
- Oliveira, V.A.D., Oliveira, V.M.A.D., Oliveira, T.W.N.D., Damasceno, A.N.C., Silva, C.E.D.O., Medeiros, S.R.A., Soares, B.M., Silva, F.C.C.D., Aguiar, R.P.S.D., Islam, M.T., MeloCavalcante, A.A.D.C., Peron, A.P., and Sousa, J.M.D.C.E. (2017). Evaluation of cytotoxic and mutagenic effects of two artificial sweeteners using eukaryotic test system. *African Journal of Biotechnology*, **6** (11), 547-551. <https://doi.org/10.5897/AJB2016.15695>.
- Orellana-Paucar, A.M. (2023). Steviol Glycosides from *Stevia rebaudiana*: An Updated Overview of Their Sweetening Activity, Pharmacological Properties, and Safety Aspects. *Molecules*, **28**, 1258. <https://doi.org/10.3390/molecules28031258>.

- Perrier, J.D. Mihalov, J.J., Carlson, S.J. (2018). FDA regulatory approach to steviol glycosides. *Food and Chemical Toxicology*, **122**, 132-142. <https://doi.org/10.1016/j.fct.2018.09.062>.
- Prakash, M.D., Stojanovska, L., Feehan, J., Nurgali, K., Donald, E.L., Plebanski, Flavell, M., Kitchen, B., Apostolopoulos, V. (2021). Anti-cancer effects of polyphenol-rich sugarcane extract. *PloS One*, **16** (3), e0247492. [Doi.org/10.1371/journal.pone.0247492](https://doi.org/10.1371/journal.pone.0247492).
- Qureshi, S. T., Chandio, P., Noman, A., Parveen, A., and Soomro, Y. (2015). Cytotoxic and genotoxic and oxidative effects of aqueous extract of some frequently used medicinal plants in Pakistan. *Botany Research International*, **8** (1): 29-35.
- Racuciu, M. (2009). Effect of radiofrequency radiation on root tip cells of Zea mays. *Romanian Biotechnological Letters*, **14** (3): 4365-4369.
- Sachan, M. and Singh P. (2026). Artificial Sweeteners: Chemistry, Health Impacts and Regulatory Perspectives. *Asian Journal of Pharmaceutical Research and Development*, **13**(5), 39-43. DOI: 10.22270/ajprd.v13i5.1624.
- Sadia, Qureshi, S.T., Soomro, A.N., Malik, S. Punar, Z. (2024). Mutational and carcinogenic potential of Amaltas fruit via oxidation. *Journal of Applied Research in Plant Sciences*, **5**(2), 269-280. <https://doi.org/10.38211/joarps.2024.05.276>.
- Selvapriya, S., Priyadarshini, R., Kumar, S.R., and Sinduja, P. (2021). Anti-Inflammatory and Cytotoxic Effect of Stevia and Neem Based Herbal Formulation. *Journal of Pharmaceutical Research International*, **33**(59(A)), 150-160. DOI: 10.9734/JPRI/2021/v33i59A34259.
- Tkach V.V., Morozova, T.V., Gaivão, I.O., Gomes de Miranda, N., Ivanushko, Y.G., Martins J.I.F., Barros, A.N. (2025). Sucralose: A review of environmental, oxidative and genomic Stress. *Nutrients*, **17**(13), 2199. doi: 10.3390/nu17132199.
- Wisal, A., Asad-Ullah, Khan, B., Sıcak, Y., Öztürk, M., Muhammad, A., Anwar, W., Begum, S., Zafar, S., Carvalho, P.V.S.D., Hassan, S.S., Halim, S.A., Khan, A. (2025). The cytotoxic, antioxidant, and anticholinesterase activity of steviol and isosteviol analogues, together with structure-activity relationships and molecular docking studies. *Journal of Molecular Structure*, **1345**, 43049.
- Xu, X., Wand, Q., Li, B., Liu, C., Cu, C., Yi, M., Zhai, L., Wong, R.M.Y., Zhang, N. and Cheung, W.C. (2025). Effect of artificial sweeteners on the Musculoskeletal system: A systematic review of current evidence. *Nutrients*, **17**(21).3489. <https://doi.org/10.3390/nu17213489>.
- Zahadi, N.A. Berahim, Z. and Zainuddin, S.L. (2026). Evaluating the effect of Tualang honey on the viability and migration of human gingival fibroblasts. *Journal of Orofacial and Health*, **7**(1), 28-39. <https://doi.org/10.31436/ijohs.v7i1.425>.
- Zink, D., Fischer, A.H. and Nickerson, J.A. (2004). Nuclear structure in cancer cells. *Nature Reviews Cancer*, **4**(9), 677-87. DOI: 10.1038/nrc1430.

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