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Application of the yeast comet assay in testing some food additives for genotoxicity by comet assay in yeast

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Abstract

Food additives of various types are frequently used in the food sector. Their use is justified by the need to preserve, color, or sweeten a variety of foods. Despite the fact that some additives have been demonstrated to be cytotoxic, they are nevertheless utilized in practice. The effects of monosodium glutamate (MSG), sodium benzoate (SB), and saffron on several yeast haploid knockout strains were studied in this study (YKO). We used the Comet test method to find the optimum amounts at which this set of dietary additives could cause DNA damage. Three regularly used dietary additives were found to efficiently damage DNA. We also evaluated the sensitivity of higher eukaryotic cells to the genotoxic effects of these chemicals with yeast. The comet assay exhibited a better sensitivity of yeast cells, which was undeniably confirmed. The genotypes of haploid (knockout) yeast strains were chosen based on the (Clustal Omega Multiple Sequence Alignment EMBL-EBI) alignment of human and yeast gene sequence homology.

Keywords: YKO, MSG; saffron, SB, Comet assay, genotoxic

1. Introduction

Yeast is a type of organism that lives Saccharomyces cerevisiae is an essential microbial bioreactor for the generation of added-value compounds and biofuels, and it remains a highly relevant experimental model in toxicogenomics. Purir is at the forefront of systems and synthetic biology, as an instrumental tool for obtaining mechanistic insights into the response to multiple toxicants and in the event of strong industrial strains, thanks to its deep functional characterization combined with the straightforward exploitation of Omic approaches and metabolic engineering.

Various additives have been used in the food industry for hundreds of years. Originally, they were only used to preserve food by pickling (with vinegar), salting, or adding sulphur dioxide (as in some wines). After that, some chemicals and supplements are used to improve the style and appearance of the dish. Several chemicals have been shown to be harmful and have been banned from use as food preservatives, such as furylfuramide (AF-2), which has been banned since 1974 after being found to be carcinogenic in both experimental animals [10] and humans [25]. Similarly, numerous radical chemicals, including Butter yellow, were discovered to have undeniable genotoxic and carcinogenic activity in experimental animals [6]. Despite the fact that food additives are dangerous, they are still used.

Surprisingly, several chemicals and prescription medications have modest toxicity, but at low doses, they can cause certain genotoxicity [18]. Genotoxins may alter DNA through direct cleavage or chemical group modifications in DNA. The majority of polymer damages are unquestionably harmful to both the cell and the organism as a whole. They have the potential to cause cell death or to enhance carcinogenic processes [1].

Several approaches for detecting genotoxicity have been developed [12,13]. However, none of them found any damage to the DNA structure per se. Furthermore, many of these procedures are time-consuming, costly, and necessitate a large number of cells.

Single Cell Gel Electrophoresis (SCGE), often known as the Comet Assay, has grown in popularity

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as a genotoxicity test [17, 21]. The technique allows for the imaging and measurement of deoxyribonucleic acid damage at the single-cell level in a simple, rapid, and repeatable manner [5]. Wherever fragmented deoxyribonucleic acid exits the remaining nucleoid and produces an extraterrestrial comet tail, electrical is applied to cells implanted in agarose during this assay. The resulting can be easily quantified using either visual scoring of the comets or software [27].

The Comet Assay method was modified to work with yeast cells. Saccharomyces cerevisiae cells were shown to be more sensitive to the action of alkyl paraffin salt and oxide than class cells [16]. The Comet Assay's findings of increased sensitivity of S. cerevisiae to irradiation [4], oxidative damage during replicative ageing [15], and Cr-(III)-organic compounds [3] were further confirmed [20, 2].

The goal of this study was to see if three food additives (mono sodium glutamate (MSG), sodium benzoate (SB), and saffron) had any mutagenic or carcinogenic effects on yeast haploid knockout strains (YKO).

2. Materials and Methods

2.1. Materials

2.1.1. Yeast Saccharomyces cerevisiae (haploid strain)

Knock out haploid yeast strains (Mat-A) Complete Set (Cat. no. 95401.H2), were bought from Invitrogen company (1600 Faraday Avenue PO Box 6482, Carlsbad CA, 92008 United States).

2.1.2. Food additives

Three differing kinds of food additives MSG, SB and Saffron were employed in this study [obtained from Sigma Chemical Company, St. Louis, USA].

2.2. Methods

2.2.1. Yeast Comet assay (YCA)

The in vitro Comet assay was performed using the first procedure published by [23]. We used yeast culture media with the optimal MSG dosage of 0.5 mg/ml. SB was added in triplicates at the optimal dosage of 0.05 mg/ml, and saffron was added at the optimum concentration of 1 mg/ml. A medium without chemical components was also employed as an untreated control. 1 g of crushed materials were placed in a one-cubic-centimeter container with cold PBS.

This suspension was filtered after five minutes of stirring. 100 g of cell suspension was combined with 600 g of low-melting agarose (0.8 percent in PBS). On pre-coated slides, one hundred percent of this mixture was spread out. For fifteen minutes, the coated slides were immersed in lyses buffer (0.045 M TBE, pH 8.4, containing a pair of.5% SDS). The slides were placed in an activity chamber with a same TBE buffer but no SDS. Two V/cm for two minutes and one hundred mA were the activity conditions. Ethidium bromide 20g/m1 staining at 4^{o}C.

The polymer fragment migration patterns of one hundred cells for each exposure level were analysed with a visible radiation magnifier while the samples were still moist (With excitation filter 420-490nm [issue 510nm]). The tail lengths of extraterrestrial objects were measured from the nucleus to the top of the tail with a 40x increase to count and measure the comet's size. Observations of Gel Red-stained polymer were done using a 40x objective on a fluorescence magnifier to visualise polymer damage. By measuring the length of polymer migration and the proportion of migrated polymer, an extraterrestrial object five image analysis code developed by Kinetic Imaging, Ltd. (Liverpoo1,UK) connected to a CCD camera was used to assess the quantitive and qualitative extent of polymer injury within the cells. The program then estimated the tail moment. In most cases, fifty to one hundred randomly selected cells were evaluated per sample. This study looked at the effects of three popular food additives on yeast cells: MSG, SB, and saffron.

2.2.2. knockout yeast strains of choice

Two plates containing two haploid knockout strains with completely distinct genotypes were used in this investigation, and the sequences of each strain were chosen and aligned with human sequence information in NCBI (The National Center for Biotechnology Information). four genes aligned with cancer-related human genes were chosen to correspond with the yeast genes used in this investigation (Table 1).

Table (1): Selected yeast genes which matched with cancer related human

Selected strains	Selected genes of yeast strains (genotypes)	Homologous genes in human
YMR035W	IMP2	IGF2BP2
YMR190C	SGS1	RECQL
YMR167W	MLH1	MLH1
YER095W	RAD51	RAD51

2.2.3. selection of yeast haploid strains deficient in genes similar to human cancer genes

The genotypes of yeast haploid (knockout) strains were chosen based on (Clustal Omega Multiple Sequence Alignment EMBL-EBI) alignment between human and yeast sequence sequence similarity, as well as the question cowl and E-value Table (1).

2.2.4. Protein-protein interaction prediction

In accordance with the sequence, the interaction network was used. MANIA (http://www.genemania.org) is a flexible, user-friendly web interface for evaluating gene function hypotheses, examining sequence lists, and prioritising genes for specific experiments.

Sources of information

Co-expression information from the organic phenomenon Omnibus (GEO); physical and genetic interaction information from Bio GRID; foretold macromolecule interaction information supported by orthology from I2D; and pathway and molecular interaction information from Pathway Commons, which includes data from Bio GRID, Memoria, and Pathway Commons.

Yeast protein-protein interaction network , Human protein-protein interaction network

3. Results and Discussion

3.1. Toxicity to (YKO) strains tested with monosodium glutamate by extraterrestrial object assay

All three chemicals displayed varying degrees of yeast significant genotoxic effects on YKO in accordance with the extraterrestrial object assay. At the optimum dose of zero.5 g/ml, monosodium glutamate revealed its genotoxic effect (recommended by FDA). The genotoxic effects of the IMP2, RAD51, and SGS1 genes were severe, whereas the genotoxicity of the MLH1 sequence was low. The distribution of the share of determined comets for monosodium glutamate was shown in Table (2).

It should be noticed that the yeast predicted significantly more comets than the various management for each of the four tested genes (Fig. 1), indicating that the tested drug caused a large number of identified desoxyribonucleic acid damages. To be more precise in contrast to the strategies, we have a tendency to accept the optimal concentration as a live of sensitivity at that twice a lot of comets appear in treated culture as compared to management.

The cells that had been exposed to twenty-five metric linear units of monosodium glutamate were each given a pre-treatment with a concentration of zero.5 g/ml of monosodium glutamate. It was clear that monosodium glutamate therapy caused significant damage to each of the four genes evaluated.

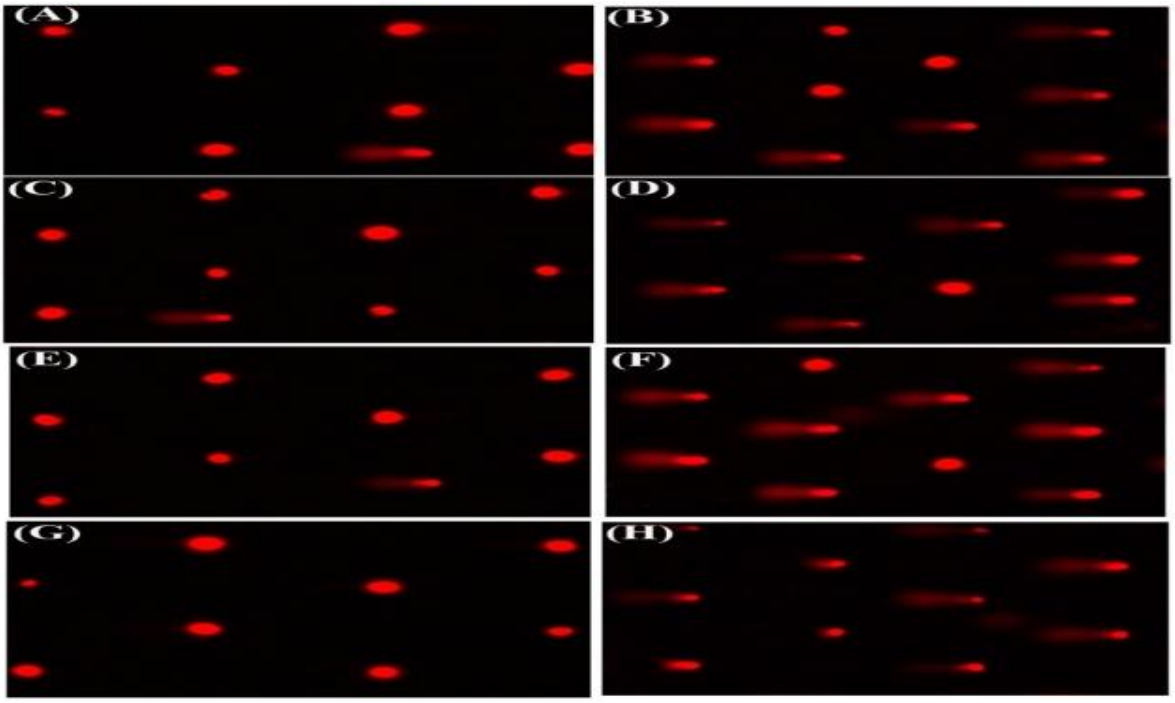

Fig. 1: Photomicrographs showing DNA damage in yeast strains using the Comet assay and MSG at a dose of 0.5 (g/ml). Control cells A: control MLH1 gene; B: treated MLH1 gene; C: control IMP2 gene; D: treated IMP2 gene; E: control RAD51 gene; F: treated RAD51 gene; control SGS1 gene; H: treated SGS1 gene; control SGS1 gene.

Table (2): Image analysis of comet assay parameters in cells of all groups after MSG therapy.

Group	Tailed %	Untailed %	Tails length µm	Tail DNA%	Tail moment
Control MLH1	2.5	97.5	2.02±0.21c	1.81	3.66
MLH1	18	82	6.93±0.53 b	5	34.65
Control IMP2	2	98	1.73±0.17d	1.72	2.98
IMP2	24	76	8.37±0.79a	6.8	56.92
Control RAD51	3	97	2.19±0.23c	1.9	4.16
RAD51	26	74	8.82±0.84a	7.45	65.71
Control SGS1	1.5	98.5	1.32±0.12d	1.48	1.95
SGS1	29	71	9.5±0.90a	8.17	77.62

Different superscript letters in the same column of tail length showed significance difference at P< 0.05.

3.2. Toxicity of SB to (YKO) strains as determined by extraterrestrial object assay

At the optimum dose of zero.1 g/ml, SB revealed its genotoxic action. Table 1 shows that the IMP2 sequence has a significant genotoxic impact, while the MLH1, RAD51, and SGS1 genes have moderate genotoxicity (3). It was discovered that the yeast predicted significantly more comets than the various management for each of the four examined genes, when the tested drug caused a lot of desoxyribonucleic acid damage (Fig. 2). Each of the management and treated cells that had been exposed to twenty-five metric linear units of SB received an additional pre-treatment with zero.1 g/ml of SB. It was clear that SB therapy caused significant harm to all four genes evaluated.

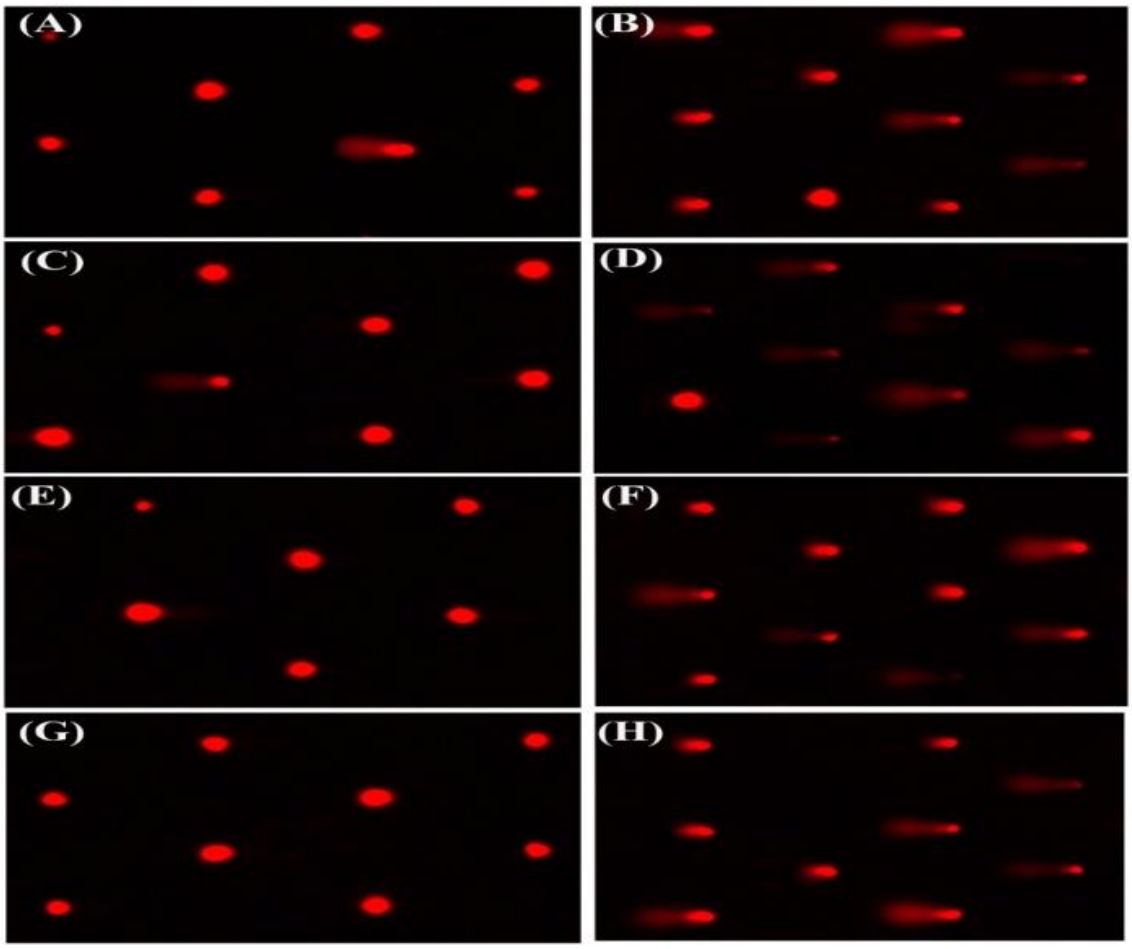

Fig. 1: Assay of a comet Photomicrographs demonstrating DNA damage in yeast strains after treatment with SB at a dose of 0.1 (g/ml). Control cells A: control MLH1 gene; B: treated MLH1 gene; C: control IMP2 gene; D: treated IMP2 gene; E: control RAD51 gene; F: treated RAD51 gene; control SGS1 gene; H: treated SGS1 gene; control SGS1 gene.

Table (3): Image analysis of comet assay parameters in cells from all groups following SB treatment.

Group	Tailed %	Untailed %	Tails length µm	Tail DNA%	Tail moment
Control MLH1	3	97	2.21±0.21c	1.97	4.35
MLH1	15	85	6.34±0.39 b	4.42	28.02
Control IMP2	2.5	97.5	2.09±0.15c	1.86	3.89
IMP2	25	75	8.6±0.58 a	7.11	61.15
Control RAD51	1.5	98.5	1.29±0.10c	1.42	1.83
RAD51	16	84	6.51±0.42 b	4.6	29.95
Control SGS1	1	99	1.03±0.06c	1.17	1.20
SGS1	17	83	6.72±0.41 b	4.81	32.32

Different superscript letters in the same column of tail length showed significance difference at P< 0.05.

3.3. Saffron revealed its genotoxic action at the optimum dose of one g/ml when tested with (YKO) strains using an extraterrestrial object assay.

The SGS1 sequence had a significant genotoxic effect, whilst the MLH1 and IMP2 sequences had mild genotoxicity and the RAD51 gene had quantifiable genotoxicity (Table 4). For each of the four genes evaluated, the yeast predicted much more comets than the various management. Moreover, Fig. (3) revealed that the tested chemical caused a significant amount of desoxyribonucleic acid damage. To be more precise in comparison to the strategies, we have a tendency to accept the optimum concentration as a live of sensitivity at that double a lot of comets appear in treated culture compared to management, untreated culture compared to management. The cells that had been exposed to twenty-five metric linear units of Saffron and the cells that had been exposed to one (g/ml) of Saffron were each pre-treated with one (g/ml) of Saffron. It was proven that each of the four genes studied had unmasked significant harm as a result of Saffron treatment.

Table (4): Image analysis of comet assay parameters in cells of all groups following saffron therapy.

Group	Tailed %	Untailed %	Tails length µm	Tail DNA%	Tail moment
Control MLH1	3.5	96.5	2.31±0.20d	2.10	4.86
MLH1	15	85	6.56±0.40b	4.51	29.59
Control IMP2	4	96	2.51±0.19d	2.13	5.35
IMP2	17	83	6.80±0.44b	4.79	32.57
Control RAD51	4	96	2.62±0.23d	2.17	5.69
RAD51	12	88	5.35±0.46c	3.93	21.03
Control SGS1	3.5	96.5	2.32±0.21d	2.17	5.03
SGS1	24	76	8.45±0.57a	6.82	57.629

Different superscript letters in the same column of tail length showed significance difference at P< 0.05.

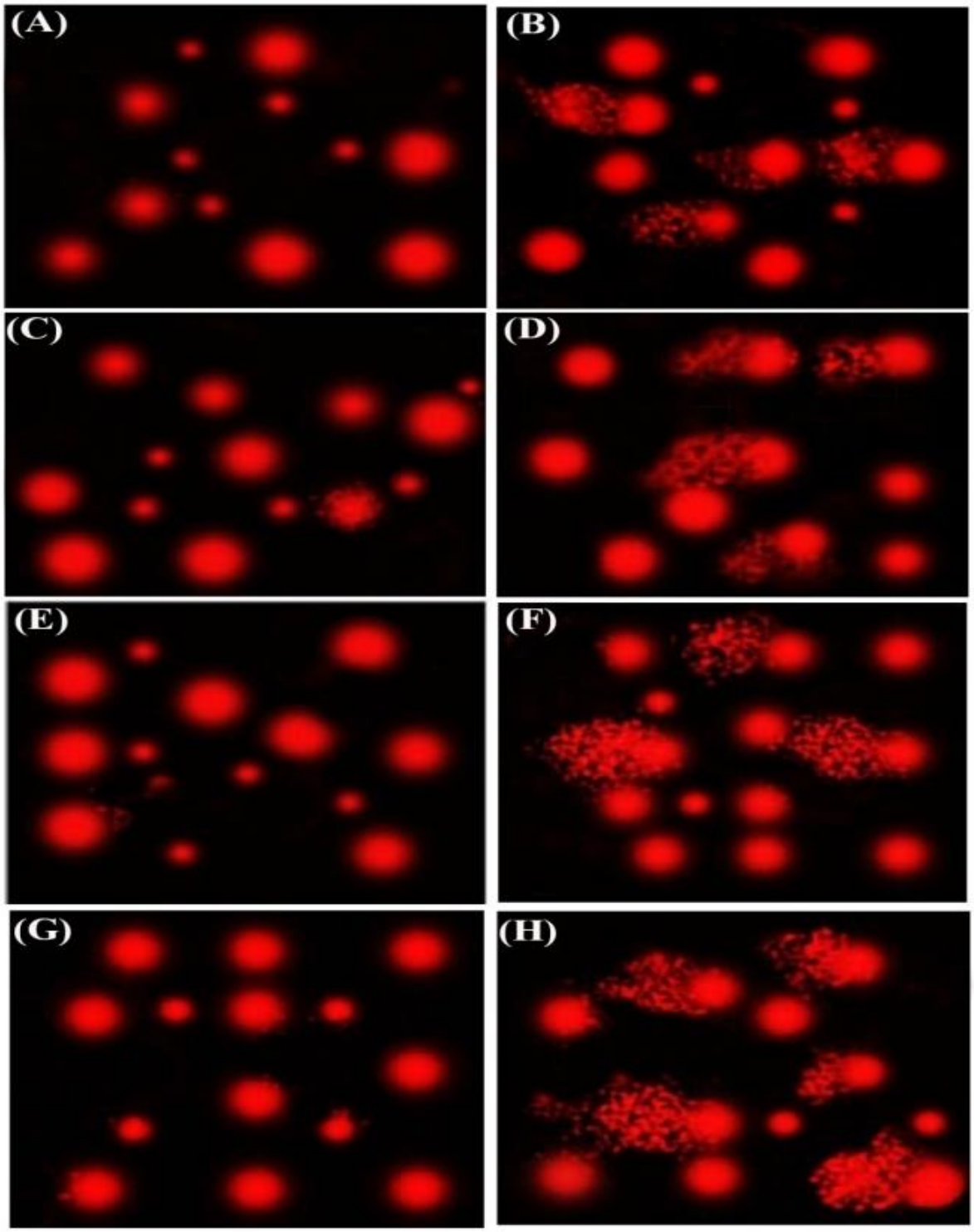

Fig. (3): Shows a comet assay. Photomicrographs demonstrating DNA damage in yeast strains after treatment with saffron at a dose of 1 (g/ml). Control cells A: control MLH1 gene; B: treated MLH1 gene; C: control IMP2 gene; D: treated IMP2 gene; E: control RAD51 gene; F: treated RAD51 gene; control SGS1 gene; H: treated SGS1 gene; control SGS1 gene.

3.4. Selection of yeast haploid strains devoid of genes similar to specific human cancer genes in vitro.

Genotypes of yeast haploid (knockout) strains were determined based on sequence similarity results between human and yeast sequences (E-value and question covery). Figure (4) depicted the results of an alignment between the human IGF2BP2 and the yeast IMP2 sequences, with a question cowl of hour and an E-value of 3e-150. IMP2 was found to play a role in the chemical change monetary unit of the mitochondrial inner membrane enzyme complex; [14] it is required for the maturation of mitochondrial proteins in the inter membrane space; [11] it has two chemical change subunits (Imp1p and Imp2p, which differ in substrate specificity) [19]**.**

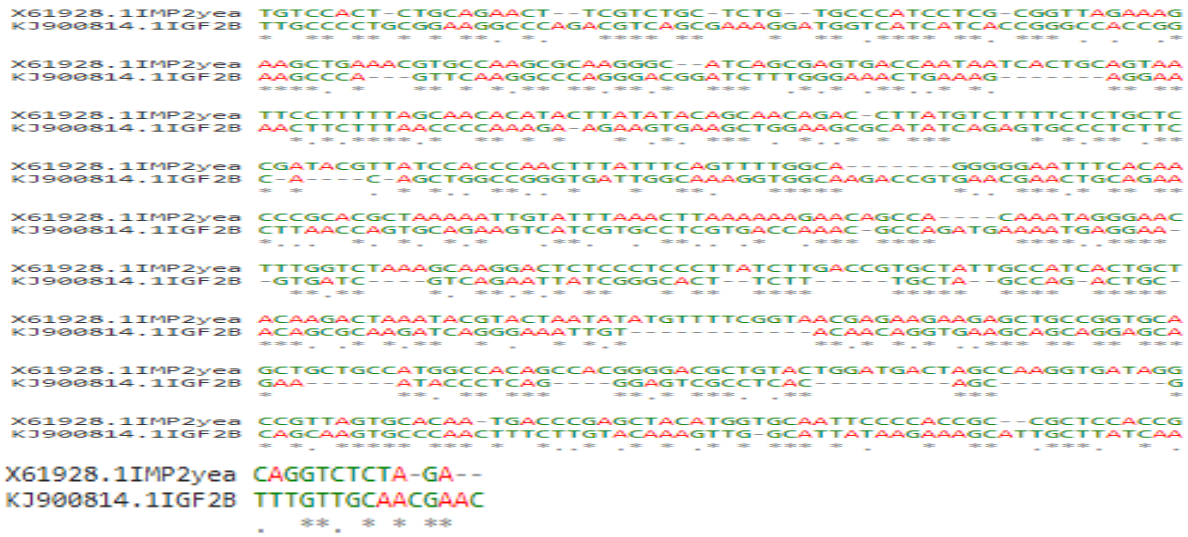

Fig. (4): Gene alignment between human gene IGF2BP2 and yeast gene IMP2 in the Clustal Omega web site ('*' indicates identical between two aligned, '-' indicates gaps missing of one) and ('.' indicates low similarity, ':' indicates more similarity used to denote the level of similarity that are not identical) at position

Figure (5) depicted the results of an alignment between the human and yeast MLH1 sequences, with a question cowl of 99% and an E-value of 7e-170. According to its own role in the sequence, MLH1 is required for pair repair in cellular division and meiosis; it is also required for biological processes during meiosis; it forms a posh with Pms1p and Msh2p-Msh3p during pair repair; and its human homolog is linked to hereditary non-polyposis carcinoma [24]. Pair repair defects produce increased spontaneous mutation rates and parabolic instability of simple repeating sequences, whereas mutations in human pair repair genes cause hereditary nonpolyposis body part malignancies [8].


DQ356646.1MLH1y TTTTTCAATATTCCTTCTAGATTAAGGGCTTTGAGGTCCCATAATGATGAATACTCTAAA
KR709638.1MLH1H TTTTACAACATAGCCACGAGGAGAAAAGCTTTAAAAAATCCAAGTGAAGAATATGGGAAA
DQ356646.1MLH1y ATATTAGATGTTGTCGGGCGATACGCCATTCATTCCAAGGACATTGGCTTTTCTTGTAAA
KR709638.1MLH1H ATTTTGGAAGTTGTTGGCAGGTATTCAGTACACAATGCAGGCATTAGTTTCTCAGTTAAA
DQ356646.1MLH1y AAGTTCGGAGACTCTAATTATTCTTTATCAGTTAAACCTTCATATACCGTCCAGGATAGG
KR709638.1MLH1H AAACAAGGAGAGAC--AGTAGCTGATGTTAG-GACACTACCCAATGCC-TCAACCGT---
DQ356646.1MLH1y ATTAGGACTGTGTTCAATAAATCTGTGGCTTCGAATTTAAT----TACTTTTCATATCAG
KR709638.1MLH1H ----GGACAATATTCGCTCCATCTTTGGAAATGCTGTTAGTCGAGAACTGATAGAAATTG
DQ356646.1MLH1y CAAAGTAGAAGATTTAAACCTGG---AAAGAGTTGATGGAAAGGTGTGTAATTTGAATTT
KR709638.1MLH1H -GATGT-GAGGATAAAACCCTAGCCTTCAAAATGAATGGTTACATATCCAATGCAAACTA
DQ356646.1MLH1y CATATCCAAAAAGTCCATTTCACCAATTTTTTTCATTAATAATAGACTAGTGACATGTGA
KR709638.1MLH1H CTCAGTGAAGAAGTGCA--TCTTC-TTACTCTTCATCAACCATCGTCTGGTAGAATCAAC
DQ356646.1MLH1y TCCTCTAAGAAGAGCTTT--GAACAGTGTTTACT-CCAATTATCTGCCAAAGGGCAACAG
KR709638.1MLH1H TTCCTTGAGAAAAGCCATAGAAACAGTGTATGCAGCCTATT---TGCCCAAAAACACACA
DQ356646.1MLH1y ACCTTTTATTTATTTGGGAATTGTTATAGATCCGGCGGCTGTTGATGTTAACGTTCACCC
KR709638.1MLH1H CCCATTCCTGTACCTCAGTTTAGAAATCAGTCCCCAGAATGTGGATGTTAATGTGCACCC
DQ356646.1MLH1y GACAAAGAGAGAGGTTCGTTTCCTGAGCCAAGATGAGATCATAGAGAAAAT-----CGCC
KR709638.1MLH1H CACAAAGCATGAAGTTCACTTCCTGCACGAGGAGAGCATCCTGGAGCGGGTGCAGCAGCA
DQ356646.1MLH1y AATCAATTGCACGCCAAATTATCTGCCATTGATACTTCACGTACTT--TCAAGGCTTCTT
KR709638.1MLH1H CATCGAGAGCAAG-CTCCTGGGCTCCAATTCCTCCAGGATGTACTTCACCCAGACTT-TG
DQ356646.1MLH1y CAATCTCAACAAACAAGCCAGAGTCATT---GATACCATTTAATGACACCATAGAAAGTG
KR709638.1MLH1H CTACCAGGACTTGCTGGCC----CCTCTGGGGAGATGGTTAAAT----CCACAACAAGT-
DQ356646.1MLH1y ATAGGAATAGGAAGAGTCTCCGACAAGCCCAAGTGGTAGAGAATTCATATACGACAGCCA
KR709638.1MLH1H -------CTGACCTCGTCTTCTACTT---CTGGAAGTAGTGATAAGGTCTATGCCCACCA
DQ356646.1MLH1y -ATAGTCAACTAAGGAAAGCGAAAAGACAAGAGAAT-AAACTAGTCAGAATAGATGCTTC
KR709638.1MLH1H GATGGTTCGTACAGATTCCCGGGA--ACAGAAGCTTGATGCATTTCTG--CAGCCTCTGA
DQ356646.1MLH1y ACAAGCTAAAATTACGTCATTTCTATCCTCAAGTCA----------ACAG----TTCAA
KR709638.1MLH1H GCAAACCCCTGTCCAGTCAGCCCCAGGCCATTGTCACAGAGGATAAGACAGATATTTCTA
DQ356646.1MLH1y CTTTGAAGGATCGTCTACAAAGCGA-CAACTGAGTGAACCCAAGGTAACAAATGTAAGCC
KR709638.1MLH1H GTGGCAGGGCTAGGCAGCAAGATGAGGAGATGCTTGAACTCCCAGCCCCTGCTG-AAGTG
DQ356646.1MLH1y ACTCCCAAGAGGCAGA-----AAAGCTGACACTAAATGAAAGCGAACAACCGCGTGATGC
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DQ356646.1MLH1y CAATACAATCAATGATAATGACTTGAAGGATCAACCTAAG-AAGAAACAAAAGTTGGGGG
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KR709638.1MLH1H ATTCTGATGTGGAAATGGTGGAAGATG-----------ATTCCCGAAAGGAAATGAC-
DQ356646.1MLH1y GGTATATTAGAGTA-CCTAAGGAGCGAGTTAATGTTAATCTTACGAGTATCAAGAAATTG
KR709638.1MLH1H TGCAGCTT---GTACCCCCCGGAGAAGGATCA--TTAACCTCACTAGTGTTTTGAGTCTC
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KR709638.1MLH1H CAGGAAGAAATTAATGAGCAGGGACATGAGGTTCT-CCGGGAGATGTTGCATAAC--CAC
DQ356646.1MLH1y TACGTTG-GGGTTGTAGATGAGGAAAGAAGATTAGCCGCTATTCAGCATGACTTAAAGCT
KR709638.1MLH1H TCCTTCGTGGGCTG----TGTGAATCCTCAGTGGGCCTTGGCACAGCATCAAACCAAGTT
DQ356646.1MLH1y TTT--TTTAATAGATTACGGATCTGTGTGCTATGAGCTATTCTATCAGATTGGTTTGACA
KR709638.1MLH1H ATACCTTCTCAACACCACCAAGCTTAGTG--AAGAACTGTTCTACCAGATACTCATTTAT
DQ356646.1MLH1y GACTTCGCAAACTTTGGT------AAGATA----AACCTACAGAGTACAAATGTGTCAGA
KR709638.1MLH1H GATTTTGCCAATTTTGGTGTTCTCAGGTTATCGGAGCCAGCACCGCTC---TTTGACC-T
DQ356646.1MLH1y TGATATAGTT---TTGTATAATCTCCTATCAGAATTTGACGAGTTAAATGACGATGCTTC
KR709638.1MLH1H TGACATGCTTGCCTTAGATAGTC------CAGAGAGTGGCTGGACAGAGGAAGATGGTCC
DQ356646.1MLH1y CAAAGAAAAAATAATT-AGTAAAATATGGG---ACATGAGCAGCATGCTAA-----ATGA
KR709638.1MLH1H CAAAGAAGGACTTGCTGAATACATTGTTGAGTTTCTGAAGAAGAAGGCTGAGATGCTTGC
DQ356646.1MLH1y GTACTATTCCATAGAATTGGTGAATGATGGTCTAGATAATGACTTAAAGTCTGTGAAGCT
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DQ356646.1MLH1y AAAATCTCTACCACTACTTTTAAAAGGCTACATTCCATCTCTGGTCAAGTTACCATTTTT
KR709638.1MLH1H GATTGGATTACCCCTTCTGATTGACAACTATGTGCCCCCTTTGGAGGGACTGCCTATCTT

Fig. (5): Gene alignment between human gene MLH1 and yeast gene MLH1 in the Clustal Omega web site ('*' indicates identical between two aligned, '-' indicates gaps missing of one) and ('.' indicates low similarity, ':' indicates more similarity used to denote the level of similarity that are not identical) at position.

Figure (6) shows the results of alignment between human and yeast RAD51 sequences, with an E-value of 8e-176 and a question cowl of 85. RAD51 plays a function in genetic recombination and desoxyribonucleic acid double-strand break repair, according to its own description. RAD51 further processes the product of the organic process between these desoxyribonucleic acid partners to produce nicked circular duplex desoxyribonucleic acid, indicating that RAD51 will shift state strand exchange [26]. RAD51 is involved in baker's yeast mitotic recombination and repair of desoxyribonucleic acid damage, as well as meiosis [22]. It is a homologue of recA from Escherichia coli and plays critical roles in both mitotic and cell division recombination, as well as the repair of double-strand breaks of desoxyribonucleic acid.


X64270.1RAD51Ye TTACTTCTTCTATCTTCCGTAGTTTCCATATACTAGTAGTTGAGTGTAGCGACAAAGAGC
CR536559.1RAD51 ----------
X64270.1RAD51Ye AGACGTAGTTATTTGTTAAAGGCCTACTAATTTGTTATCGTCATATGTCTCAAGTTCAAG
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X64270.1RAD51Ye AACAACATATATCAGAGTCACAGCTTCAGTACGGGAACGGTTCGTTGATGTCCACTGTAC
CR536559.1RAD51 ----------GATGT----------
X64270.1RAD51Ye CAGCAGACCTTTCACAGTCAGTCGTTGATGGAAACGGCAACGGTAGCAGCGAAGATATTG
CR536559.1RAD51 ----------GAAGAAATTG
X64270.1RAD51Ye AGGCCACCAACGGCTCCGGCGATGGTGGCGGATTGCAGGAGCAAGCGGAAGCGCAAGGTG
CR536559.1RAD51 ----------
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CR536559.1RAD51 ----------GAAGAAGCTG-----GATTC----------
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CR536559.1RAD51 ----------CATA
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X64270.1RAD51Ye AATTCAGGACAGGTAAGTCCCAGCTATGTCACACTTTGGCCGTGACATGCCAAATTCCAT
CR536559.1RAD51 AATTCCGAACTGGGAAGACCCAGATCTGTCATACGCTAGCTGTCACCTGCCAGCTTCCCA
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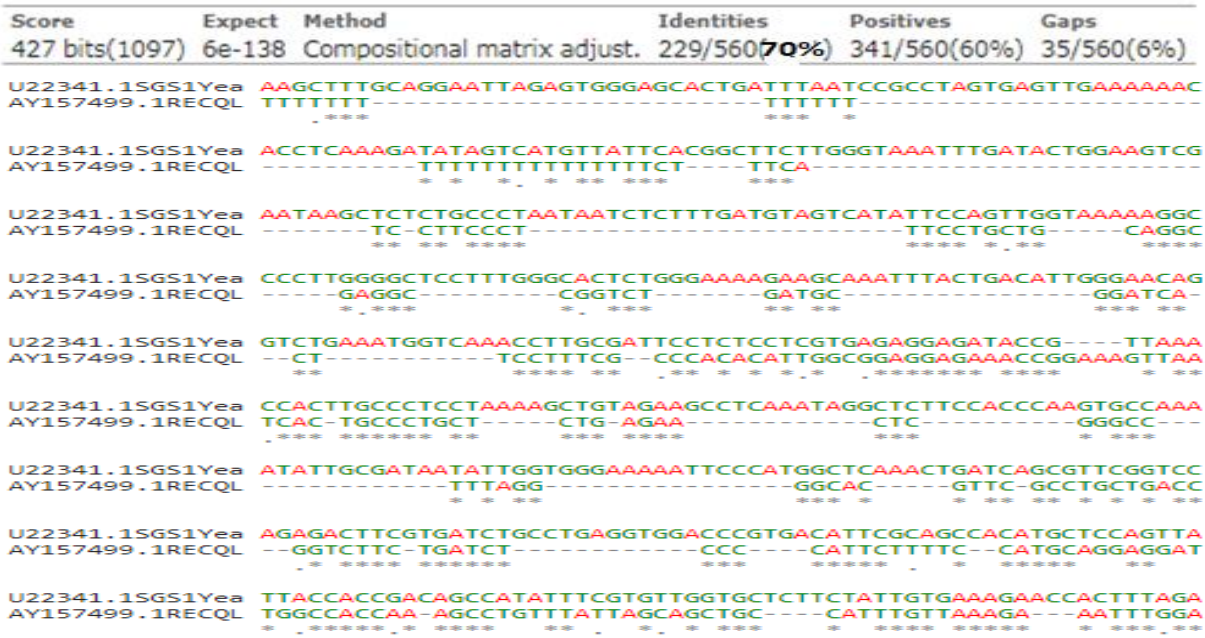
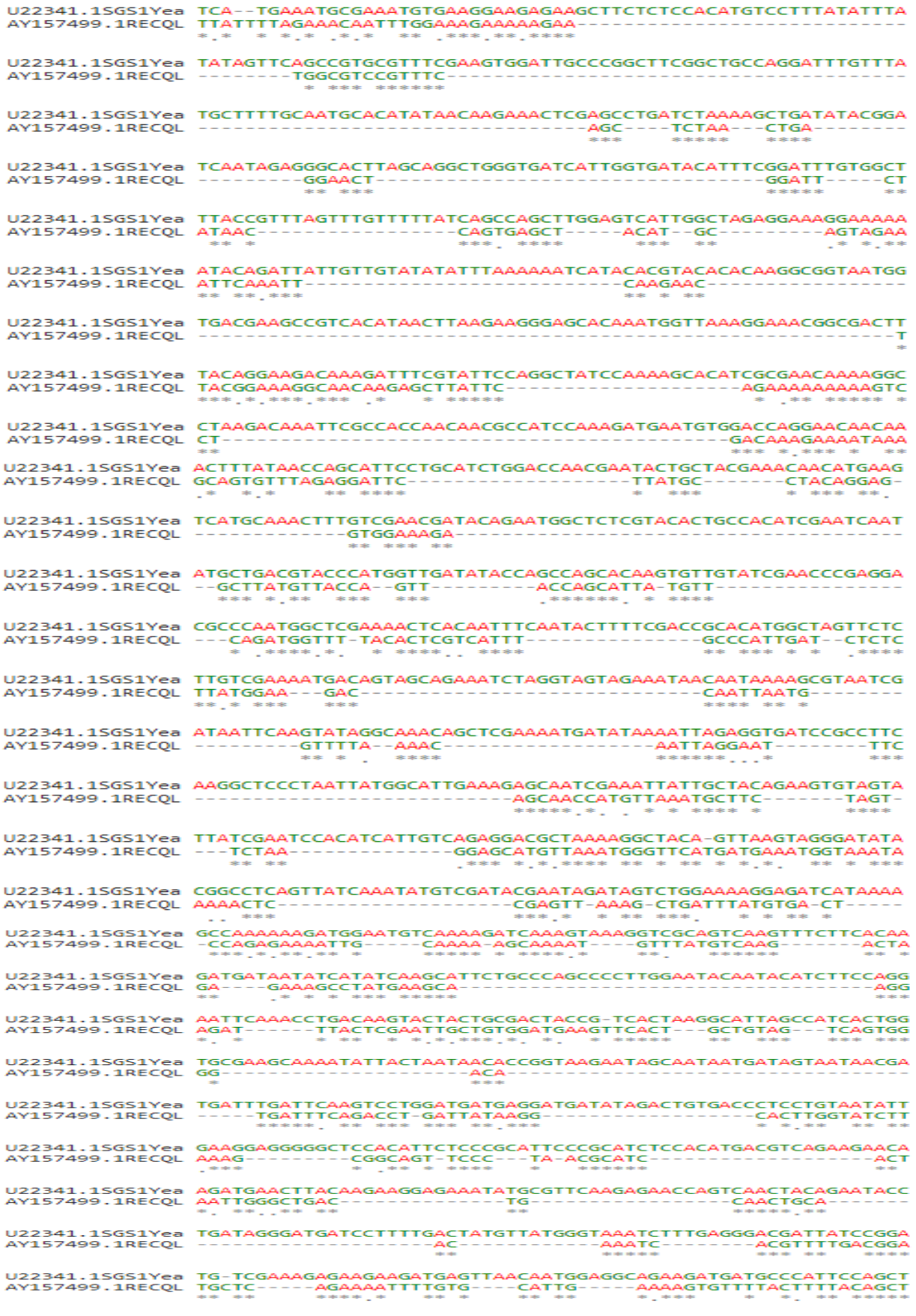
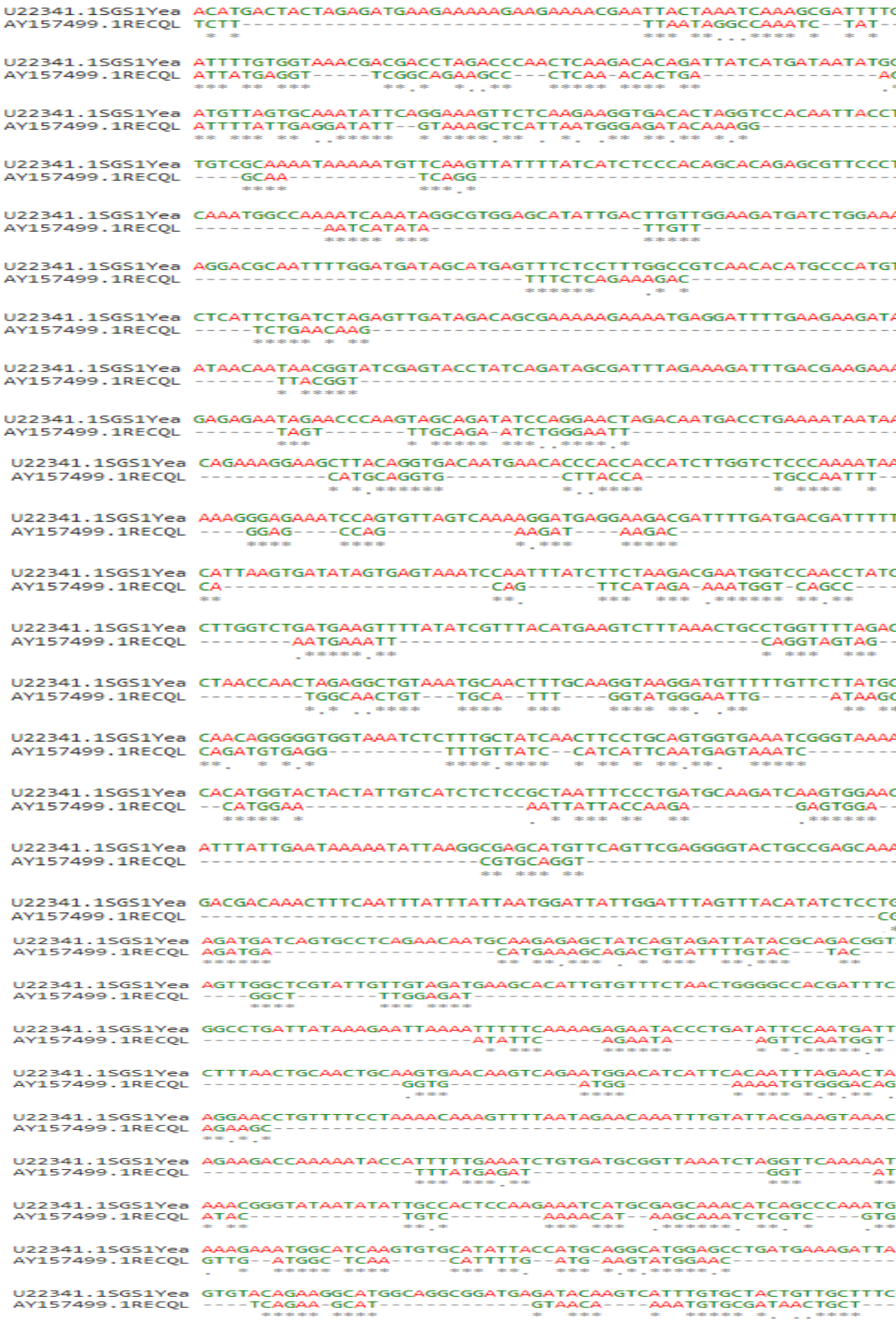
CR536559.1RAD51 TTGACCGGGGTGGAGGTGAAGGAAAGGCCATGTACATTGACACTGAGGGTACCTTTAGGC
X64270.1RAD51Ye CGGTAAGATTGGTATCCATAGCTCAGCGGTTCGGATTAGACCCGG---ATGATGCTTTGA
CR536559.1RAD51 CAGAACGGCTGCTGGCAGTGGCTGAGAGGTATGGTCT---CTCTGGCAGTGATGTCCTGG
X64270.1RAD51Ye ACAACGTTGCGTATGCAAGAGCCTATAACGCCGATCATCAGTTAAGACTTCTGGAT---G
CR536559.1RAD51 ATAATGTAGCATATGCTCGAGCGTTCAACACAGACCACCAGACCCAGCTCCTTTATCAAG
X64270.1RAD51Ye CTGCTGCCCAAATGATGAGCGAGTCTCGGTTTTCCTTGATTGTGGTCGATTCTGTTATGG
CR536559.1RAD51 CATCAGCC---ATGATGGTAGAATCTAGGTATGCACTGCTTATTGTAGACAGTGCCACCG
X64270.1RAD51Ye CTCTATACCGTACGGATTTTTCTGGTCGTGGTGAACTAAGCGCAAGGCAAATGCATTTAG
CR536559.1RAD51 CCCTTTACAGAACAGACTACTCGGGTCGAGGTGAGCTTTCAGCCAGGCAGATGCACTTGG
X64270.1RAD51Ye CCAAATTTATGCG--TGCTTTGCAAAGGCTGGCCGACCAATTTGGTGTTGCAGTCGTCGT
CR536559.1RAD51 CCAGGTTTCTGCGGATGCTT--CTGCGACTCGCTGATGAGTTTGGTGTAGCAGTGGTAAT

Figure (6): Gene alignment in the Clustal Omega web site between human gene RAD51 and yeast gene RAD51 ('*' indicates identical between two aligned, '-' indicates gaps missing of one) and ('.' indicates low similar, ':' indicates more similar used to denote the level of similarity that are not identical) at position.

With a question cowl of thirty-eight and an E-value of 6e-138, **Figure (7)** depicted the findings of alignment between human sequence SGS1 and yeast sequence RECQL. RECQL family nucleolar desoxyribonucleic acid helicase; was involved in ordering integrity maintenance, body organic process, cell division joint molecule/crossover formation; stimulates activity of Top3p; forms nuclear foci in response to desoxyribonucleic acid replication stress in a Rrd1p-dependent manner; yeast SGS1 enhances mutations in human homolog BLM Bloom and Werner syndromes, which are linked to genomic instability and cancer risk, are linked to RECQL helicases [28]. Genes in each creature performed in exactly the same way.






U22341.1SGS1Yea
AY157499.1RECQL


U22341.1SGS1Yea
AY157499.1RECQL

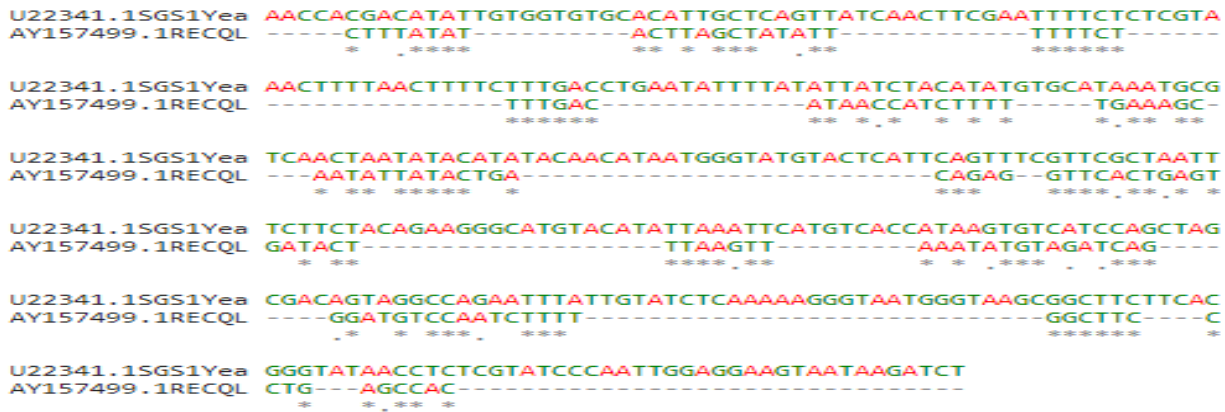


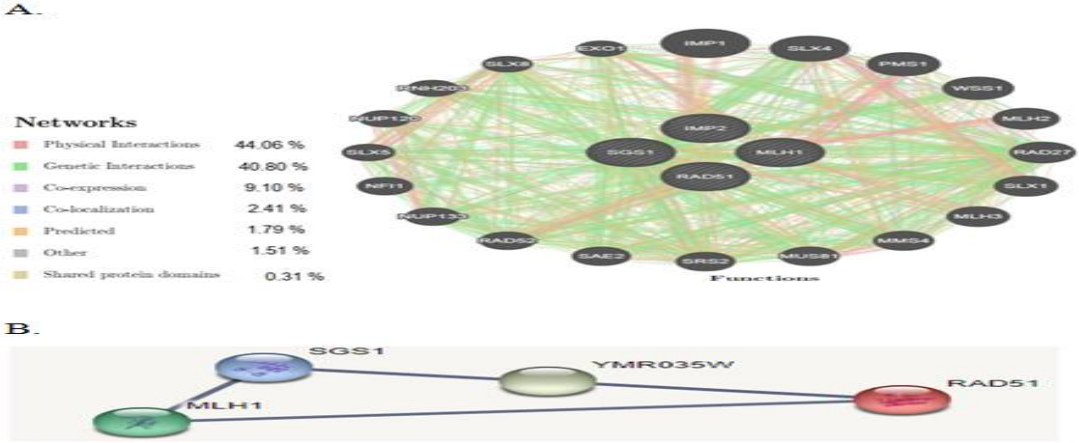
Figure (7): Gene alignment in the Clustal Omega web site between human gene RECQL and yeast gene SGS1 ('*' indicates identical between two aligned, '-' indicates gaps missing of one) and ('.' indicates low similar, ':' indicates more similar used to denote the level of similarity that are not identical) at position.

3.5. Yeast protein-protein interaction prediction (Networking).

Predicting protein-protein interactions in yeast and humans could lead to a determination of the degree of deliberate resemblance between two animals' genes linked to cancer (Figures eight,9).

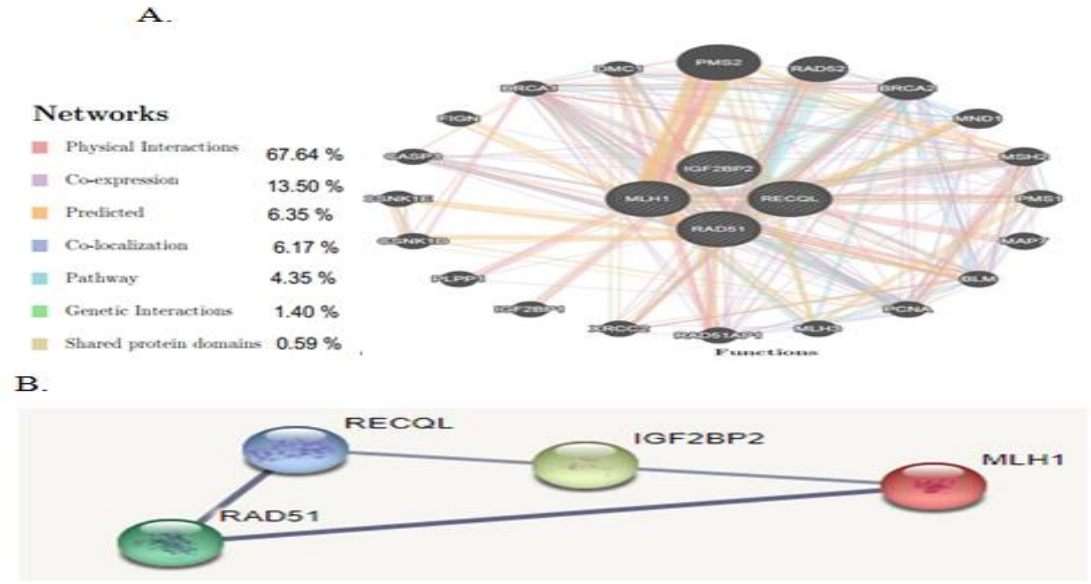
Gene MANIA displays the prognostic value of each data set selected for the inquiry. Currently, two organisms (Homo sapiens and Saccharomyces cerevisiae) are supported in addition to or above distinct sequencing execute prediction methodologies on yeast and humans. The GeneMANIA prediction algorithmic program's excellent accuracy, Associate in Nursing intuitive computer programme, and vast knowledge make sequence MANIA a helpful tool for any scientist [7].

The results of four yeast inquiries are displayed in Gene MANIA (8A, B). The resulting networks are completely different, with various totally different absolutely different relationships and four separate relevant genes in yeast that are linked by a pathway to the query list. Physical interaction (44.06 percent), Genetic interaction (40.80 percent), Co-expression (9.10 percent), Co-localization (2.41 percent), foretold interaction (1.79 percent), other (1.51 percent), and Shared macromolecule domains were the other degrees of question customization (0.31 percent). The impact of knowledge set selection on topology. The results of gene queries are displayed in GeneMANIA. (A) The default inquiry about the yeast cell cycle, with all default parameters mistreated. (B) Mistreatment default network weight methodology for the yeast cell cycle default inquiry. On yeast and mouse benchmarks, the GeneMANIA algorithmic programme outperforms or outperforms alternative sequence perform prediction algorithms, according to [7]. GeneMANIA is a beneficial gadget for any man of science because of the high accuracy of the GeneMANIA prediction algorithmic programme, Associate in Nursing intuitive computer programme, and enormous information.



Figures (8A and B): (A) The yeast cell-cycle default query with all default parameters. (B) The yeast cell-cycle default query with all default parameters. (B) Using the default network weighting approach, the yeast cell-cycle default query.

Four yeast inquiries are being displayed by GeneMANIA (Fig. 8A, B). The networks that result are completely distinct, with various totally different absolutely different relationships and four separate relevant genes in yeast that are linked by a pathway to the query list. Physical interaction (67.64%), co-expression (13.50%), foretold interaction (6.35%), co-localization (6.17%), route (4.35%), genetic interaction (1.40%), and common macromolecule domains are some of the other levels of question customisation (0.59 percent). GeneMANIA displays the results of gene queries in Effects of Knowledge Set Choice on Topology. (A) Mistreatment of all default parameters in the human default question. (B) Mistreatment default network weight approach, which is a human default question. YKOs lacking genes that are similar to cancer genes in humans were chosen. The ability to predict protein-protein interactions in yeast and humans could lead to an assessment of the degree of deliberate similarity of some cancer-related genes between the two organisms.



Figures (9A and B): (A) The human default query, using all default parameters. (B) The human default query, using default network weighting method.

4. Conclusions

Extraterrestrial object assay was used to assess desoxyribonucleic acid harm in yeast deletion strains when treatment with suggested concentrations of food additives to YKO strains resulted in a significant increase in desoxyribonucleic acid harm (P zero.05) as measured by a rise in tail length, tail microchip, and tail moment when compared to traditional management strains. These data indicated that food additives MSG, SB and saffron decreased cell viability in malignant and non-malignant cells as well as confirmed the occurrence of their cytotoxic effects according to a pervious study [22] and determine the possible genotoxic effects of three food additives (MSG, SB and Saffron) on human cell lines by Flow cytometric and RT-pcr to a pervious study [23].

5. Conflicts of interest

"There are no conflicts to declare".

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