GENOTOXIC EFFECT OF CADMIUM IN PROKARYOTE AND EUKARYOTE

H. H. Ahmed

Microbial Biotechnology Dept., GEBRl. USC.

Corresponding author: hanan.mohamed@gebri.usc.edu.eg

ABSTRACT

This study evaluates the genotoxic effects of cadmium (Cd) at a toxic concentration of 300 ppm/L on prokaryotic organisms (*Escherichia coli* and *Bacillus subtilis*) and the eukaryotic plant *Vicia faba*. Using DNA profiling, protein pattern analysis, and superoxide dismutase (SOD) enzyme activity assays, the study highlights significant Cd-induced genomic and proteinic change. Treated by Cd resulted in novel genomic DNA bands in *E. coli* (1 band), *B. subtilis* (2 bands), and *V. faba* (2 bands), indicating DNA mutation. Protein profiling resulted in the disappearance of native bands one band in *E. coli*, one band in *B. subtilis* and one band in *V. faba*.G40 and the appearance of novel bands in *V. faba*.G40 compared to control. *V. faba* showed the greatest effect. Induced SOD activity, showed additional bands, confirmed by the organisms' stress response mechanisms. Overall, *V. faba* showed higher sensitivity to Cd-induced genotoxicity compared to *E. coli* and *B. subtilis*. These results provide understanding the molecular mechanisms of Cd toxicity, with indications for environmental observing and biotechnological applications.

Keywords: Cd, *E. coli*, *B. subtilis*, *V. faba*, DNA, protein pattern, SOD.

Introduction

The Cd sources, it used in chemical batteries, alloys, stabilizers, solar cells, paints, stains and in nuclear reactors where it absorbed neutron (Satarug *et al*., 2010 and 2012, Rani *et al*., 2014).

$CdCl_2$ is a highly toxic salt soluble in water. The minimum toxic and lethal dose of Cd were (30 ppm- 1.5 g) respectively. The highly Cd concentrations are found in liver and kidneys. Cd accumulates through lifetime; its biologic half-life may be reach 38 years. The reason is that urinary Cd excretion is slow. The most source of Cd is through eating food, especially shellfish, kidney, and liver. Fish accumulate cadmium from water. Plants absorb cadmium from soil. Long-term exposure to Cd through soil, air, water, and food leads to cancer and organ system toxicity such as heart and blood vessels, skeletal, urinary, reproductive, central peripheral nervous and. respiratory systems, Cd levels can be determined by measuring in the blood, urine, nail, hair, and saliva patient's samples. (Mehrdad *et al*., 2017) (U.S. E. P. A.).

Cd is nonessential heavy metals and cytotoxic and genotoxic for human genome and toxic for human health and eukaryotic. Cd also leads to mitotic inhibition in eukaryotic cells and inhibition growth in bacteria (Subhajit *et al*., 2018).

Cd genotoxic effects by interfering with cellular processes, disrupting DNA integrity, and altering protein synthesis. Prokaryotes like *E. coli* and *B. subtilis* are widely used in genotoxicity assays due to their rapid growth and ease of genetic manipulation. Similarly, the eukaryotic plant *V. faba* serves as an effective model for studying heavy metal-induced genotoxicity, owing to its genetic and physiological responsiveness to environmental stressors and as carries the toxic in food becouse it traditionally eaten in many countries all around the world in different ways. (Munawar 2016). Faba bean elevated, it is one of the rich source of carbohydrates, protein, vitamin and minerals (Duc, 1997). 1n agricultural soils the heavy metals like cadmium (Cd) and other pollutants lead to bioaccumulation of several toxicants in plants; (Nagajyoti *et al.*, 2010).

Plant cells accumulate 19 elements, including C, O, H, S, N P, K, Mg, Cd, (macro elements) and B, Cu, Zn, Mn, Fe, Mo, Ni, Co, Cl, Br (micro elements) for their vital metabolic functions. These macro and micro elements are necessary for several biochemical and physiological functions in plants, such as photosynthesis, carbohydrate synthesis, chlorophyll biosynthesis, nucleic acid synthesis, protein modification, nitrogen fixation, oxidation and reduction reactions. Several heavy metals are used as microelements, other heavy metal elements, like Cd, Al, Cr, Pb, Hg create toxic results for plants, like reduce production of biomass, photosynthesis reduction, altered water neutralize and nutrient sucking. These agents lead to plant growth suppression and loss of yield (Singh *et. al*., 2016).

Cd prevented cells entering cell division phases. The main action of Cd on mitotic spindle helped spindle-correlated aberrations like bridges and lagging chromosomes during cell division. The mitotic inhibition caused by heavy metals blocks to prevent several cells entering the prophase and locking the mitotic phase of the cell cycle (Seth *et. al.*, 2008).

The *Phanerochaete chrysosporium* (*P. chrysosporium*) (eukaryotic) and *E. coli* (prokaryotic) exhibit bioaccumulation and cytotoxicities of Cd. The bioaccumulation amounts by *P. chrysosporium* and *E. coli* due to the smaller particle size and fewer negative surface charges of Cd. Gradually decreased in Plasma membrane fluidities and membrane H^+-ATPase activities of *P. chrysosporium* and *E. coli* with the increasing concentrations of Cd^2. Viabilities and intracellular reactive oxygen levels indicated that the enhance cytotoxicity in eukaryote and prokaryote (Liang *et. al*, 2019).

Plants organisms have developed some unique strategies to environmental stress *Vicia faba* treatment with cadmium chloride blockers, DNA repair Cd-induced adaptive response in plants undergoing genotoxic stress. (Manoswini and Anita, 2021).

The present research aim to compare the genotoxic effects of Cd on *E. coli*, *B. subtilis*, and *V. faba*, focusing on genomic DNA damage, protein pattern changes, and SOD enzyme activity, by clarify these effects, hoping to better understand impacts of

cadmium on living systems and inform strategies for lessen heavy metal toxicity in the environment.

Materials and Methods

Plant samples and growth-conditions

Three different varieties of *V. faba* were used in present work G1, G2 and G40. G40 was selected because it's the most tolerant Cd dose used in this study.

Dry seeds of *V. faba* (G1, G2 and G40) sterilized by 70% ethanol for 5 min., washed by sterilized distilled water 3 times (5 min. for each time), saturated in water (control) and Cd 300 ppm/ L (treated) for 5 hours .and used for sowing 5 seeds were transplanted into pots (25 cm) at 2 cm depth, containing washed three times with (0.1% HCl), sterilize, dried and distributed (one kilo) peat moss and sand (l-l) V/V. The seeds irrigated and drenched in (pH 6) (to avoid Cd precipitation, to stimulate and accumulation of Cd in soil), irrigated 3 times a week for l6 days by equal volume of water (control) and (Cd) concentration 300 ppm/L.(6 pots for each until 16 days).

Bacterial media and growth conditions

Six isolates of bacteria were used in this study. Three isolates of *E. coli* and three isolates of *B. subtilis*. The isolates were isolated from agriculture wastes, animal wastes and soil. The isolates were inoculated and grown at 28^{o}C on Luria Bertani medium with agar (LB medium) (control) and LB medium containing (Cd) concentration 300 ppm/L for 48 hours. After 48, the highest number plates and largest colonies were selected. This step repeated 8 times (for l6 days) to collect the most tolerant isolates**.**

LB medium

(NaCl 10g; Tryptone 10g; Yeast extract 5g and Agar 15g.) per liter completed to 1 liter with distilled water, pH (6 -7). The medium was sterilized at 121^{o}C for 20 min.at autoclave.

Genomic DNA isolation from plant

The *V. faba* plant after l6 days of sowing were used to extract DNA by CTAB protocol according to (Dolye and Dolye 1990).

Genomic DNA isolation from bacteria

Total genomic DNA was isolated from *B. subtilis* according to (Desomer *et. al*; 1991).

Total genomic DNA was isolated from *E. coli* according to (Fanglian 2011).

Determination of total genomic DNA extraction concentration

The total genomic DNA extraction concentration determined by DNA measuring the absorbance at 260 nm wave length (Spectrophotometer).

Detection of genotoxic on DNA by *Hindlll* on agarose gel electrophoresis

Gel Electrophoresis for DNA samples The same DNA solution concentration were used (digested by *HindIII* at 37°C for 30 min. the volume of 30 µL contained l µL from restriction enzyme 7 unit/µL, 1 µL from RNase 100 µg/mL, 3 µL from restriction enzyme buffer 1X., 5 µL from genomic DNA 10 ng/µL, 20 µL from Deionised water (DI) on l% agarose with 1µg/ mL Ethidium Bromid (ETBr), the gel was visualized by UV light (Sharp *et.al.*, 1973).

Totat protein estimation from plant

The total protein extracted from plant according to (Hurkman and Tanaka.1986).with modification

- Ground 1 g of plant with liquid nitrogen using a mortar. Transfer the sample to lock centrifuge tubes and add 1 mL of extraction buffer it contained per 15 mL (7.5 mL Tris HCl (1M, pH=7.5), 1.5 mL KCl (lM), 1.5 ml EDTA (0.5M, pH=8.0), 3.6 g Sucrose (0.7 M), 300 µL *B*-mercaptoethanol 2% (v/v) It must be added to the extraction buffer, just before using and should be homogenized, 5.22 mg Phenylmethylsulfonylfluoride (PMSF) 2 mM (It must be dissolved in 300 µL of isopropanol, before been added to the extraction buffer), 0.15 g polyvinylpolypyrrolidone (PVPP) 1% It is added as powder (1%), to the extraction buffer, just before using and should be homogenized, Deionised water (DI water) up to l5 mL,Vortex. and shaker (200 rpm) the samples for 20 min. at 4ºC. Add 1 mL of equilibrated phenol with 10 mM deTris-HCl (pH=8.0), shaker (200 rpm), for 20 min. at 4ºC. Centrifuge (12.000 *g),* for 25 min. at 4ºC, the supernatant, transfer it to a new tube, add (2 mL) of extraction buffer. shaker (200 rpm), for 20 min. at 4ºC. Centrifuged (12000 g) for 25 min. at 4ºC. Add 2 mL of the wash buffer it contain per 100 mL (l00% methanol + 0.1 M Ammonium acetate (0.78 g)) to the supernatant cold at -20ºC overnight. Centrifuged (14000 g) for 40 min. at 4ºC. Discard the supernatant without disturbing the pellet. Keep the tube in the freezer (-20ºC), (minimum) for l hour: and centrifuge (14000 *g*) for 40 min. at 4ºC. Discard the supernatant, add 2 mL of wash buffer, cold (-20ºC), ovide disturbing the pellet. Keep the tube in the freezer (-20ºC), for 1 (minimum) hour, add 1.6 mL of precipitation buffer (100% acetone) cold (-20ºC), ovide disturbing the pellet. Keep the tube in the freezer (-20ºC), for (minimum) 1 hour. Centrifuged (14000 g) for 40 min. at 4ºC. Dry the pellet. Close and keep at 4ºC, overnight. Suspend it in 500 µL of solubilisation buffer it contain per 10 mL (Urea 7.0 M 4.2 g, Tiourea 2.0 M 1.52 g, DTT 10 mM (= 0.01M) 15.4 mg, Triton X-100 0.01% (w/v) l mg, DI water up to 10 mL. Centrifuge (14000 *g*), for 20 min., at 4ºC, to remove the residual. Transfer the supernatant to eppendorf tubes. Keep the samples at -80°C until used.

Total protein estimation from bacteria

Total proteins extracted from bacteria were carried out according to (Michiels *et al;* 1994) with modification add 10 mg/mL lysozyme and incubate for 1 hour at 37ºC while shaking when extracting proteins from G^+ bacteria.

Total extracted proteins were assayed by Folin-lowry method separated by denatured PAGE. Gel was stoned by comassie blue and gel was photographed. (Laemmli 1970). The same protein solution concentration was used.

Superoxide dismutase SOD patterns

Crude extract of plants were prepared by ground 1 g of fresh plant with liquid nitrogen using a mortar. To eliminate particulate matter transfer the sample to lock centrifuge tubes and add 500 µL of potassium phosphate 0.05 M pH 7.8 Vortex and shaker (200 rpm) the samples for 3 min. at 4ºC then Centrifuge (10000 *g),* for 15 min. at 4ºC. The supernatant transfer into a new tube.

Crude extract of bacteria were sonicated 5 times for 20 Sec. at 40 pluses in 200 µL of 0.05 M K_3PO_4 (pH 7.8) then centrifuge (15000 g), for 10 min. at 4ºC. The supernatant transfer into a new tube

Crude extracts used for SOD enzyme activity as per (Beauchamp and Fridovich 1971) separated on 10% polyacrylamide gel and stain with nitroblue tetrazolium choloride (NTC) to visualize band (clear area) blue dye color was absent, gel was photographed.

Results and Discussion

Effect of Cd on Plant

In the present study three different varieties of *V. faba* were used Gl, G2 and G40. The effect of the processing, on germination of the seeds and the efficiency of the processed seeds comparison to control were tested. G40 was selected because it tolerant Cd dose used in this study (most seeds were sowing in Cd concentration 300 ppm/L). In this study found that Cd induced a reduction in Faba bean in comparison with control.

Table (l): The effect of treated of Faba bean seeds with Cd on germination compared with control (C).

Cultivars	No of germination seeds	Germination % control	No of germination seeds	Germination % treated (Cd)
Gl	60	l00%	48	80 %
G2	60	l00%	51	85%
G40	60	l00%	57	95%

Effect of Cd on bacterial growth

Six isolates of bacteria, three isolates of *E. coli* and three isolates of *B. subtilis* were grown on L.B media plates control and L.B media with 300 ppm/L for 48 hours. After 48, the highest number of colonies and the largest colony from each was selected. This step repeated 8 times (for 16 days) to collect the most tolerant isolates.

The colonies mean number of bacteria isolated from agriculture wastes, animal wastes and soil decreased when treated with Cd in *E. coli* 17%, 23% and 28% and *B. subtilis* 22%, 19%, 30% respectively. So the *E. coli* isolate isolated from agriculture wastes and *B. subtilis* isolate isolated from animals wastes used in this study because these isolates more tolerant when treated by Cd.

Generally, Cd inhibits the bacterial growth in comparison to control. The isolate showed the highest number of colonies for all the repeated test (this step 8 times for l6 days) treatment, on the other hand the isolate was greater than all the other isolates which indicated that it is more tolerant to the Cd concentration compared to the control was selected for this study. One isolate of *E. coli* and one isolate of *B. subtilis* were selected for this study.

Table (2): The colonies mean number of different bacterial isolates from different sources treated with Cd compared with control (C).

Bacterial isolates	Agriculture wastes (C)	Animals wastes (C)	Soil (C)	Agriculture wastes treated (Cd)	Animals wastes Treated (Cd)	Soil Treated (Cd)
E. coli	59	66	54	49	5l	39
B. subtilis	65	75	51	5l	6l	36

Detection of genotoxic effect on DNA from plant by *HindIII*

The agarose gel electrophoresis of the DNA in control and treated with Cd to study the induction change in genomic DNA by *HindIII*. The results are presented in figure (l) showed the photograph of the electrophoretic pattern of total genomic DNA bands isolated from *E. coli* bacterial isolate control and treated with Cd. Cd enhanced change in appearance one band in genomic DNA pattern comparison with control. *B. subtilis* bacterium isolate control and treated with Cd. Cd enhanced change in appearance two band in genomic DNA pattern comparison with control. Cd induced change in genomic DNA profiles comparison with control were induced and appearance two bands in *V. faba* plants treated with Cd. The changes caused DNA mutation due to chemical genotoxic by Cd. Cd accumulation in cells with DNA change which following cell division will produce mutations.

The results in table (3):.showed the reduction in DNA concentrations treated with Cd were 0.52µg/mL compared with control 0.59 µg/mL in *E. coli*. In *B. subtilis* were 0.61 µg/mL compared with control 0.71 µg/mL and in *V. faba* 0.72 µg/mL compared with control 0.85 µg/mL.

Table (3):.showed the reduction in DNA concentrations treated by Cd were compared with control

DNA µg/ mL concentrations	*E. coli*	*B. subtilis*	*V. faba*
control	0.59	0.71	0.85
treated with Cd	0.52	0.61	0.72

The structure and function of the genomic DNA were related to the genotoxicity of Cd (Unyayar *et al*.; 2006).

The change in DNA genome resulted by genotoxic chemical (Savaa 1998).

Disappeared bands referred to DNA damage but appearance of additional bands indicated mutation to DNA (Ateinzar *et al*., 2000)**.**

DNA replicates before cell division genotoxicity can cause mutations that often show their effects after cell division. Mutations in germ cells may carriage a risk to reproduction. DNA damage lead to the loss of genetic information which also causes in cells malignancy (Roger *et al;* 2019).

In *V. faba* demonstrated that reactive oxygen species (ROS) caused oxidative degradation of membrane lipids, induce significant percentage of DNA double strand breaks and genome instability were induced by high level of Cd accumulation has been shown (Lin *et al;* 2007)

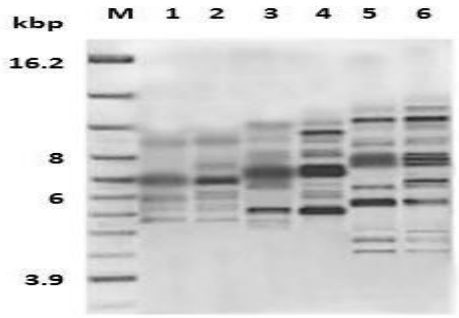

Fig. (1): Genomic DNA *HindIII* digest of *E. coli* (1, 2), *B. subtilis* (3, 4*) and V. faba* (5, 6) Control and treated respectively and marker (M)

Protein profiles

The polyacrylamide gel electrophoresis of the protein in control and treated with Cd to study enhanced change in gene expression for protein profiles. The results are presented in figure (2) showed the bacterial protein bands of total protein profiles isolated from *E. coli*, *B. subtilis* and *V. faba*.G40 There are different in profiles protein bands were induced in treated with Cd. Protein pattern showed disappearance of one natural band in *E. coli*, disappearance of one natural band in *B. subtilis* and in *V. faba*.G40 disappearance of one natural band and presence of novel band compared to control. *V. faba* plant protein profile more affected than *E. coli* and *B. subtilis*. In this

study generally protein pattern showed quantitative, qua1itative, band intensity, bands appearance and bands disappearance compared to the control reverse metabolic change due to change in gene expression by gene damage and mutation. There was correlation between alteration in genomic DNA and protein bands pattern. The change proteins concentration were The results in table (4): showed reduction in protein concentrations treated with Cd compared with control so it was 0.40 µg/mL compared with control. 0 46 µg/mL in *E. coli, B. subtilis* were 0.52 µg/mL compared with control 0.6l µg/mL while in *V. faba* were 0.69 µg/mL compared with control 0.83 µg/mL.

Table (4): showed protein concentrations treated with Cd were compared with control

DNA µg/mL concentrations	*E. coli*	*B. subtilis*	*V. faba*
control	0 46	0.6l	0.83
treated with Cd	0.40	0.52	0.69

Metal stress lead to increase protein on *Aryza sativa* (Liu *et al.,* 2007).

Protein bands pattern on SDS-PAGE by RAPD-PCR of Alyssum L. (Brassicaceae) species showed change included appearance of novel bands and variations in band mobility (Babaoglu *et al.,* 2004).

When Cd (II) interacts with proteins, it can lead to unfolding and then accumulation on the protein, altering other binding interactions, and also lowering protein stability. This led to induced-suitable mechanisms in which the unfolded states are stabilized. The function of transcription factors can be affected by unfolded proteins binding to specific DNA sequence (Martina 2012).

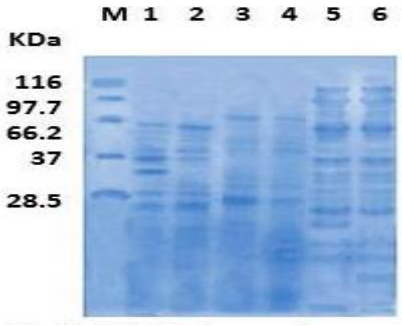

Fig. (2): PAGE of protein pattern of *E. co1i* (1, 2) , B*. subti1is* (3, 4*)* and *V. faba* (5, 6) Control and treated respectively and marker (M).

SOD pattern

The non denuatreated polyacrylamide gel showed the electrophoretic of the SOD enzymes in control and treated with Cd to study enhanced change in gene expression for SOD isozymes. The results are presented in figure (3) showed the bacterial SOD activity isozymes bands number of total SOD pattern isolated from *E. coli*, *B. subtilis* isolates and *V. faba* control and treated with Cd. Cd enhanced change in SOD isozymes

pattern in *E. coli* induced one band, in *B. subtilis* appearance (one band) and in *V. faba* G40 induced appearance two bands controlled by genes were obtained. That represent enhanced enzyme activity were induced in plants treated with Cd. SOD enzyme activity in *V. faba* plant more effected than *E. coli* and *B. subtilis* isolates treated with Cd compared to control.

Organisms developed protective recover, reactive oxygen spiecies (ROS), Catalase (CAT), to defense themself from the effects of heavy metals. Organisms generate antioxidant enzymes such SOD and CAT to protect themselves from toxic radicals (Okamoto *et al*., 2001, Daz *et. al*, 2006).

Genotoxicity of Cd, the sensitivity of faba beans (*V. faba* L.) and (*Allium cepa* L.) to Cd stress was examined. Lipid peroxidation and the antioxidative stress enzymes catalase and guaiacol peroxidase both occur concurrently and showed that in both *A. cepa* and *V. faba*, exposure to Cd caused a dose-dependent increase in chromosomal aberrations, DNA fragmentation, and micronucleus frequency. The activity of the enzymes catalase (CAT) and guaiacol peroxidase (GPX) rose in both plants, although they were greater in *V. faba*. At all Cd concentrations, there was a positive connection found between GPX and CAT activity. *V. faba* was more sensitive plant than *A. cepa* to the toxicity of Cd (Arya and Mukherjee 2014).

Root cytotoxicity and genotoxicity caused by Cd. By measuring the effects of Cd stress on metal accumulation, lipid peroxidation, protein contents, hydrogen peroxide production, and the activities of superoxide dismutase (SOD), catalase (CAT), and guaiacol peroxidase (GPX) after seven days of Cd stress in the seedling roots of *V. faba*, the genotoxic damaging effects of Cd were detected. The effects of Cd stress included alterations in SOD activity, H_2O_2 generation increased, lipid peroxidation generated by Cd, loss of membrane lipid content and quantity, and an increase in CAT and GPX activities on *V. faba* (Issam *et al*; 2019).

Cd decreased photosynthesis, genes expression, production of proteins, activity of antioxidant enzymes, influenced on plant metabolism to high emission of hydrogen peroxide (H_2O_2) (Zhaowen *et al*; 2023)

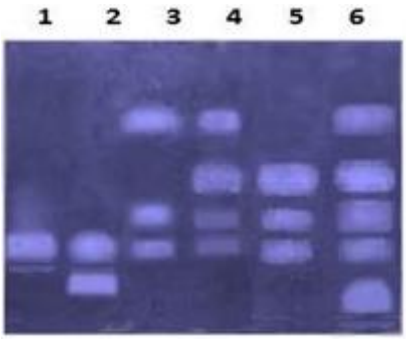

Fig. (3): Photograph of SOD enzyme of *E. coli* (1, 2), *B. subtilis* (3, 4), *V. faba* (5, 6) Control and treated respectively.

Conclusion

The genotoxic effects of Cd Metabolic ions on both prokaryotic *(E. coli*, *B. subtilis*) and eukaryotic organisms *V. faba* indicate the highest sensitivity. Cd induced genomic DNA mutations, altered protein profiles, and induced SOD enzyme activity in all tested organisms, reflecting their stress response mechanisms. The results highlight the correlation between genomic alterations, proteinic changes, and SOD enzyme activity as a fused response to Cd toxicity.

The differential tolerance levels observed suggest that *E. coli* and *B. subtilis* may have greater efficiency mechanisms for lessen Cd-induced damage compared to *V. faba*. These perceptions are essential for developing bioremediation strategies and evaulating environmental risks associated with heavy metal pollution. Future studies could expand on the molecular pathways in Cd tolerance and investigate potential applications in biotechnology and environmental science.

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تأثير السميه الجينيه للكادميوم على بدائيات النواة وحقيقيات النواه

د/ حنان حسن

قسم البيوتكنولوجيا الميكروبيه- معهد بحوث الهندسة الوراثيه والتكنولوجيا الحيويه - جامعة مدينه السادات

البريد الالكتروني للباحث الرئيسي : hanan.mohamed@gebri.usc.edu.eg

الدراسة الحالية تمت لمقارنة التأثير السام للكادميوم (Cd) بجرعة سامة تركيز ٣٠٠ جزء في المليون/لتر في كل من البكتيريا سالبة لجرام. *Escherichia coli (E. coli)* بكتيريا موجبة لجرام *Bacillus subtilis* (*B subtilis*)؛ ونبات الفول *Vicia faba (V. faba)* . تم تطبيق اختبارات مختلفه على الحمض النووي (DNA) ونمط البروتين وإنزيم سوبر أكسيد ديسميوتيز (SOD) لتأكيد التحمل والتأثير السام للجينات في التعبير الجيني.

وخلصت هذه الدراسة إلى أن التغيرات الجينية في الحمض النووي (DNA) المستحثة بواسطة الكادميوم في بدائيات النواه وحقيقيات النواه. أدى الكادميوم إلى زيادة التغيير الجينومي في *E. coli* في ظهور حزمه واحده جديده، وفي *B. subtilis* أدى التغيير في ظهور حزمتين جديدتين، وفي *V. faba* G40 أدى الكادميوم إلى التغيير في ظهور حزمتين (DNA) جديدتين مقارنة بـالكنترول .تسببت التغييرات في حدوث طفرة في الحمض النووي بسبب السمية عن طريق تراكم الكادميوم في الخلايا. فى البروتين تسبب الكادميوم فى اختفاء حزمه بروتين واحدة في *E. coli* ، واختفاء حزمه بروتين واحدة في *B. subtilis*. واختفاء حزمه واحده ووجود حزمه جديده في *V. faba.G40* وذلك مقارنة بالكنترول .أظهر نمط البروتين تغييرات في عدد الأشرطة الكمي والنوعي وشدة الأشرطة. تأثر البروتين في *V. faba* أكثر من *E. coli* و *B. subtilis* المعاملين بالكادميوم. نشاط SOD يتأثر بالكادميوم، يوجد اختلاف في ظهور حزمه واحده في *E. coli* و ظهور حزمه واحده *B. subtilis* وفي *V. faba* .يوجد اختلاف في ظهور حزمتين. وذلك مقارنه بالكنترول . هذا التركيز غيرت الكائنات الحية موضع الدراسه نمط الحمض النووي ونمط البروتين وتنتج أيزوزيم SOD عالية لحماية نفسها من التعرض للسموم.

الكلمات المفتاحيه: الكادميوم ، بكتريا اشريشيا كولاى، بكتريا باسلس ساتلس، الفول، نمط الحمض النووي، نمط البروتين وانزيم سوبر اكسيد ديسميوتيز.