# Studying the effect of probiotics on some immune parameters of infected rats Experimentally induced eczema

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#### Abstract

This experiment was conducted in the animal house of the University of Babylon/College of Medicine. 24 white rats of the Albino Rates norvegieus type were used in the study and were obtained from animal fields in the north of Babylon Governorate. The ages of the rats ranged between 5-7 weeks and their weight was 125-190 grams. The animals were divided into three groups, with 8 rats per group:

-The first group (positive control group:(

- These animals were not dosed with any substance, but were fed leech and purchased tap water.

-The second group (negative control group): Eczema was induced experimentally by being injected with formaldehyde at different concentrations.

-In the third group (the treatment group), eczema was induced experimentally by being injected with formaldehyde and dosed orally with the probiotic L.acidophilus and L.plantarum at different concentrations.

The first and second weeks included a concentration of 1 ml, while the third and fourth weeks included a concentration of 1.30 ml) daily to study its effect.

The study included the estimation of (liver function, inflammatory cytokines, and IgE) in the blood serum of the studied samples. The results were obtained from the current research- :

Biochemical study: The results of the current research The weights of organs, including the liver, spleen, and kidneys, increased slightly in the groups induced with formaldehyde and the groups Treated with the bacteria L. acidophilus and Lactobacillus plantarum compared to the positive control group (not induced.(

The study found an increase in the number of L.acidophilus and Lactobacillus plantarum bacteria in the animals' faeces reaching  $43 \times 1010$ 

The activity of liver enzymes (aspartate aminotransferase, in aminotransferase) increased in the blood serum of male rats induced with formaldehyde, and groups treated with the bacteria L.acidophilus and L.plantarum compared to the positive control group (not induced.(

The levels of inflammatory cytokines (interleukin-4, interferon gamma) increased in the blood serum of male rats induced with formaldehyde, and groups treated with Lactobacillus acidophilus and Lactobacillus plantarum compared to the positive control group (not induced.(

The level of IgE increased in the blood serum of male rats induced with formaldehyde and groups treated with bacteria compared to the positive control group (not induced.(

Histological study: The results showed that the group induced with formaldehyde caused the appearance of thick dead tissue that replaced the normal skin, severe skin inflammation and severe degeneration of skin collagen fibres with noticeable infiltration of multicellular cells. As for the groups treated with the bacteria L .acidophilus and L.plantarum, the appearance of a thin line of

newly regenerated epidermis, in addition to mature fibrous connective tissue from the dermis with noticeable formation. For hair follicles in deep skin tissue.

#### -1 Introduction

Atopic eczema (AE) or atopic dermatitis (AD) is an inflammatory skin disease with highly interconnected genetic, immunological and environmental factors. Genetic and environmental factors play the main role in the exacerbation and progression of eczema conditions. Among these environmental factors, climatic conditions are an important factor affecting the appearance and severity of eczema. (Allergy, (2021) Lee, et al. 2016.(

The term eczema or dermatitis is used unusually to describe a similar skin reaction that appears in all types of eczema, but differs according to the spread of the rash and the type of eczema (Akan, et al. 2011). In acute eczema, the rash appears The skin is red and swollen. and the edges are usually indistinguishable, in addition to the presence of pimples. And vesicles, which are often large pimples accompanied by secretions, cracks, and scaly (Amouri, et al. 2011). chronic eczema, refers to all the symptoms mentioned previously, but the secretions and vesicles are less, and thickenings and lichenification are formed in the skin, as well as dry skin with an increase in skin marks as a secondary result of the process of rubbing and scratching. (Abdel Mohsen et al., 2011; Werfel, 2011.(

Epidemiological investigations have found that environmental factors, including high humidity, high temperature, and long-term light, can increase the incidence and severity of eczema.

Studies indicate that continuous exposure to low levels of formaldehyde can have negative effects on human health (Rafael & Taeho, et al, 2016.( The possible causes of developing skin inflammation (atopic dermatitis) are multiple complex. and include and genetic. environmental, immunological and nervous system factors. Atopic dermatitis is thought to arise when the body's immune system reacts to specific substances in the environment (such as dust, insects, or food) abnormally. A role in determining the sensitivity of the body's immune system to these specific substances in the environment. (Kim HJ, et al. 2016.) According to studies, individuals suffering

from atopic dermatitis appear to have changes in certain genes responsible for regulating the function of the immune system and the normal skin barrier. These genetic changes can increase skin sensitivity to environmental factors, leading to an abnormal immune response and the secretion of cytokines and other chemicals that contribute to the development of dermatitis (Lee, et al. 2016.( The Food and Agriculture Organization (FAO) defines probiotics as those living organisms that, when ingested in sufficient quantities, confer a health benefit on the host. (WHO,2016). In recent years, there has been an increasing increase in the demand for therapeutic bacteria through the increased demand for The of functional foods. This is due to the growing awareness of consumers regarding the benefits of therapeutic bacteria health Jose. on human (Leta 2015). Lactobacillus plantarum is considered an important bacteria.

In the field of health probiotics, it plays a prominent role in the manufacture of fermented dairy products and food industry applications. In addition, these bacteria play an important role in the pharmaceutical industries by making a significant contribution to the field of human medicine in a safe and More side-effect-free manner. recently, L.plantarum has been used in medical fields to treat many chronic diseases such as Alzheimer's disease, Parkinson's, diabetes, obesity, cancer, high blood pressure, and liver disorders. (Woo, et al. 2014) and others. Initially, Lactobacillus acidophilus, formerly known as L. acidophilus, was isolated from the human gastrointestinal tract infant faeces in 1900 by Moreau. Approximately 80% of vogurts produced in the United States contain L. acidophilus, which is part of the normal human microbiota. L. acidophilus isolates were cultured from areas such as the mouth, gastrointestinal tract, and vagina.

The current study aims to do the following:

1-Study the effect of probiotics to treat experimentally induced eczema in rats

-2Studying the effect of probiotics on some immune indicators: IgE + interferon (IFN)Y + cytokines IL-4 in rats affected by eczema.

-3Study of the effect of probiotics on some liver function enzymes. ALT+AST in the blood sera of rats with formaldehyde-induced eczema.

4Measuring the weights of some internal organs in the liver, spleen, and kidneys in rats with induced eczema.

-5Measuring the weights of experimental animals and monitoring their health status.

-6 Estimating the number of probiotic bacteria colonizing the animal's intestines.

-7 Conduct a histological study of the skin to determine the effect of probiotics on experimentally induced eczema in rats.

-2Materials and working methods

-Powdered milk (low fat (

from Skimmed milk (low-fat) is dried the Saudi company - Anlene. We take (50gm) (450ml) distilled water, and it is sterilized at  $(121^{\circ}C \text{ for 5 minutes})$  in the autoclave. The milk is left to cool to the temperature (30-32°C). Then the therapeutic probiotic bacteria and L. plantarum + L. acidophilus were added to the milk and the milk was mixed with the bacteria using a spoon and placed in the incubator For a period of (24-48) hours and temperature of (37°C) in order to coagulate the milk.

-Used probiotic bacteria.

In this experiment, a mixture of commercial L.acidophillus and L. plantarum (50mg) bacteria prepared by Vitane Pharmaceuticals, Inc 125 Wells Ave was used in this experiment. Congers, NY 10920 USA), in dried capsule form.

-Preparing the cultural media

The growing media was prepared and a several different compositions of growing media were used during the study. Some of them were prepared according to the instructions of the company that prepared them, and the other was prepared in the laboratory. All of the media were sterilized at a temperature of 121°C and a pressure of 15 pounds/inch2 for 15 minutes using an autoclave.

Medium M.R.S agar

MRS agar medium was prepared according to the manufacturer's instructions, according to the producing company's instructions, by dissolving 13g in 250ml distilled water and and it was used to estimate the total number of L.acidophillus+ Lactobacillus plantarum bacteria (Hassan, etal. 2011).While the glassware was sterilized in an electric oven at 160°C for two hours.

### Medium Broth M.R.S

Broth M.R.S medium was prepared according to the manufacturer's instructions, according to the producing company's instructions, by dissolving 13g m in 250ml distilled water and placing it in an autoclave.according to what was mentioned by Lankaputhra and his group (1996). This medium is used to activate the isolation of each bacteria. Lactobacillus plantarum + L.acidophillus period.( Hassan, et al. 2011.(

A solution of (0.1)% Pepton water

Prepare a peptone water solution by dissolving 1 gm of Bacteriological peptone powder in a volume of distilled water, then transferring it to a volumetric flask with a capacity of 1 liter, then completing the volume to 1 liter, distributing it into decimal dilution tubes, and sterilizing it in an autoclave at a temperature of 121 °C and a pressure of 15 lb/in2. For 15 minutes, it was used to make the necessary dilutions to calculate the living numbers of (Hassan, et al. 2011.(

Activation of the probiotic L.acidophillus and L. plantarum Take 100 ml of distilled water to which powdered from the Saudi company (Anlene) 13 gm is added. Place it at a temperature of 121 °C in an autoclave for 5 minutes and at a pressure of 15 bar, and leave it to cool to a temperature of -37-40 °C.

Then a mixture of probiotic bacteria capsules (L. acidophilus + plantarum (L.)) was added to it, then it was transferred to the incubator at a temperature of  $37^{\circ}$ C for 24-48 hours. (Al-Khazraji, 2005 & Shatti, 2017.(

-Statistical analysis

Data analysis was performed using SPSS version 28 and plotting using Excel 2021. Continuous variables were expressed as mean SD (standard deviation), using one-way ANOVA and comparison between groups with least statistically significant differences (LSD). Differences with p values < 0.05 were considered statistically significant.

-3Results and Discussion-

Study of the effect of L.acidophilus + L. plantarum bacteria on the weight rate in the sera of male rats with experimentally induced eczema.

)Table - 1) shows the average  $\pm$  standard deviation of the weight rate, as it was in the positive control group, the fourth week, the first week, the second week, and the third week (10.6 $\pm$ 171.0.(

 $(152.38\pm12.35)$ ,  $(146.5\pm33)$  and  $(7.44\pm162.25)$ , respectively. As for the negative control group, the average weight in the fourth week, the first the second, and the third week was  $(3.38\pm181.0)$ ,  $(7.67\pm149.75)$ .  $,(11.11\pm156.25)$ ,  $(11.16\pm171.63)$ , respectively. As for the treated group, the average weight was  $(10.45\pm186.4)$ ,  $(6.58\pm151.13)$ ,  $(11.93\pm158.5)$ , and  $(10.7\pm171.88)$  grams, respectively

Average body weig Arithmetic mean ±	Transaction duration Treatment totals			
The fourth week	The third week	The second week	The first week	
10.6±171.0 B	7.44±162.25	12.35±152.38	8.33±146.5	Control
3.38±181.0 A	11.16±171.63	11.11±156.25	7.67±149.75	Induction
			6.58±151.13	Induction and
10.45±186.4 A	$10.7 \pm 171.88$	11.93±158.5		treatment
9.1601 *	10.299	12.277	7.861	LSD
0.007	0.110	0.585	0.467	p-value

(Table - 1) Mean ± standard deviation of average body weight in the blood serum of male rats under study

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The different letters have significant differences below the probability of less than P<0.05. ANOVA test with Thesignificant difference (LSD.(

The results of the current study showed that the average weight decreased in the fourth

In the group treated with bacteria, as in Figure 1

week and the first and second weeks, with an increase in the average weight in the third week compared to the positive control, At the same time while it was found that the average weight decreased in the first and second weeks and increased in the third week.

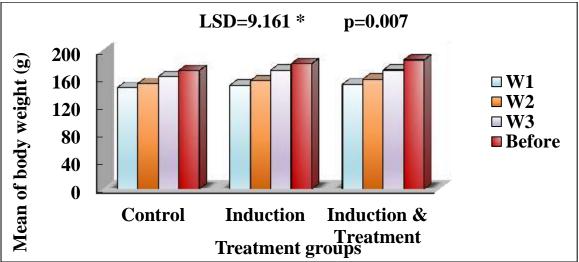


Figure 1- shows the average body weight in the studied groups

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The different letters have significant differences below the probability level less than P<0.05. ANOVA test with least significant difference (LSD.(

The slight increase in weight in the third week may be due to prolonged exposure to formaldehyde, as the results are consistent with the results of (Omar., 2013) and the study (El-Bayoumy., 2001), which explains that formaldehyde has a role in increasing the weight rate, so there is a need for More studies to prove the relationship between formaldehyde and weight.

The results can be explained based on the reactions that occurred in the cytoplasm of cells exposed to formaldehyde, where the of protein chains and the dissolution of working enzymes occurs, leading to a defect in cell growth, increase or decrease, according to the concentrations to which the cells were exposed, and according to the defense mechanism that the body possesses from its use of specific elements. Such as selenium, which is referred to as the safe element that protects cells from damage by these degrading and carcinogenic factors (El-Bayoumy., 2001). A conducted on rats to determine the effect of bacteria on body weight, it was found that eating them for

It has the ability to regulate the level of glucose, which is related to the feeling of hunger. over a long period leads to a reduction in body weight and the amount of fat in the abdominal area. This is not consistent with the results of the current study, especially in the third week. (Mamoru Tanida, 2008.( Or the decrease is due to the difference in bacterial strains and isolates that affect the immune and hormonal factors and the degree to which they change the intestinal tubule, which are factors that affect weight loss (Evrad et al. 2011.(

It was also found, (Shati. 2017) that probiotics work to modify the intestinal flora through competition and adhesion to mucous epithelial membranes and the ability to absorb fats through the breakdown of bile salts and inhibition of the enzyme lipoprotein lipase which controls the deposition of triglycerides.

In adipocytes, the compound (LPS) is reduced, which leads to regulating the work of hormones, including those that control appetite, the most important of which is leptin, as well as insulin, improving their sensitivity. It also improves the function of the intestinal barrier, which leads to controlling body weight.

Table (2-4) shows that the mean  $\pm$  standard deviation of liver weight in the positive control group was (1.33 $\pm$ 6.53) grams, while the negative control group was (0.91 $\pm$ 7.18) grams. As for the third and treatment group, the liver weight was (0.6 $\pm$ 7.65).) gloom.

Arithmetic mean	Member weights		
Kidney (gm)	Spleen (gm)	Liver (gm)	Study totals
0.28±1.39	0.03±0.58	1.33±6.53	control
0.05±1.69	0.12±0.62	0.91±7.18	Induction
0.07±1.69	0.01±0.6	0.6±7.65	Induction and treatment
0.174*	0.076	1.031	LSD
0.002	0.724	0.101	p-value

Table- 2- Mean ± standard deviation of organ weights in the albino rats under study

The different letters have significant differences below the probability level less than P<0.05. ANOVA test with least significant difference (LSD.(

The results of the current study showed that there was a very slight, non-significant increase in liver weight in the group induced with formaldehyde (negative control) compared to the non-induced group.

While the results also showed a slight increase in liver weight in the group treated with bacteria compared to the positive control group, with no significant differences between the treated group and the induced group in liver weight, as in Figure 2.

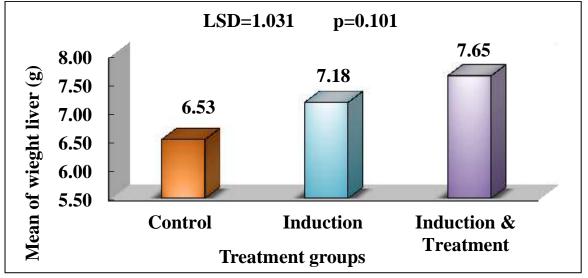


Figure - 2: Liver weight in the study groups

The results of the current study agreed with the results of (Mahmoud . 2016), which indicated an increase in liver weight in the group induced with formaldehydethe results agreed with the results of (Ucmakli, 2013), which indicated a slight increase in the group treated with bacteria.

The reason for the increase in the group treated with formaldehyde, whether through contact inhalation, is skin or that formaldehyde stimulates reactions within tissues and increases the decomposition of some components of the cytoplasm, as well as affecting the genetic material, whether RNA or DNA. The increase in weight can be attributed to saturation of the cells with liquid as a result. Because there are some elements that the immune system uses to interact with the substance and stop the reactions

The reason for the increase in the group treated with formaldehyde, whether through skin contact or inhalation, is that formaldehyde stimulates reactions within tissues and increases the decomposition of some components of the cytoplasm, as well as affecting the genetic material, whether RNA or DNA. The increase in weight can be attributed to The of the cells with liquid as a result. Because there are some elements that the immune system uses to interact with the substance and stop the reactions

Therefore, when amino acids are introduced into the body, the dehydrogenase enzymes need glutathione as one of the auxiliary factors during the reaction. Therefore, increasing the concentration of formaldehyde reduces the level of glutathione in the blood and thus depletes it. As an antioxidant, its decrease leads to an increase in formaldehyde toxicity and thus leads to injuries. Extremely large in the body, and this is what the rats were exposed to

The current study may therefore suggest that the depletion of antioxidants as a result of formaldehyde toxicity may lead to skin infections, including eczema, which affects white rats.

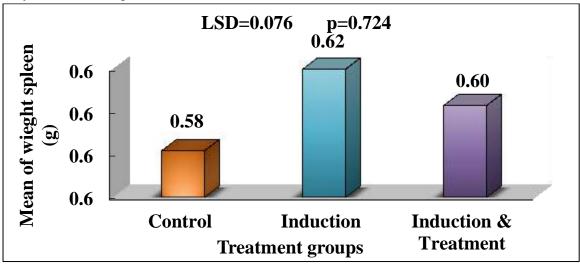
On the other hand, lactic acid bacteria caused a slight increase in liver weight, as the literature did not indicate the role of bacteria on liver weight in rats induced with formaldehyde.

However, a study (Jameel. 2021) indicated that mice induced with melamine led to a decrease in the weight of the liver, but when treated with lactic acid bacteria, it led to an increase in the weight of the liver, spleen, and kidneys.

However, according to the results of the study, probiotics, including L. acidophilus bacteria, may not have a negative effect on the health of animals and liver activity, and the probiotic may work to reduce the levels of fats and cholesterol manufactured in the liver and excrete quantities of them through the feces.

As for the weight of the spleen, the weight of the spleen in the positive control group was  $(0.03\pm0.58)$  grams, while the weight of the negative control group was  $(0.12\pm0.62)$  grams. As for the third and treatment group, the weight of the spleen was  $(0.01\pm0.6)$  grams.

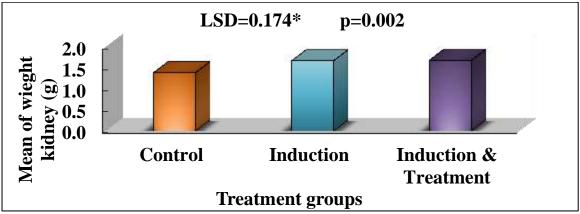
The results of the current study showed that there was a very slight, non-significant increase in the weight of the spleen in the group induced with formaldehyde (negative control) compared to the non-induced group (positive control), while the results also showed a slight, non-significant increase in the group treated with bacteria compared to the positive control group, with There were no significant differences between the treated and induced groups for spleen weight



)Figure-3): Spleen weight in the study groups

The results of the current study agreed with the results of (Cheng, 2003), who indicated in his study an increase in the weight of the spleen. The results also agreed with the results of (Gurel., 2005) and (Sandikci., 2007.( The increase in the weight of the spleen may be attributed to the nature of the tissue and cells of the spleen, as its importance lies in resisting infection and purifying the blood from non-essential substances. Therefore, treatment with formaldehyde, which is a substance It is toxic, so the spleen must purify the body of it. Since formaldehyde is present in the human body as a metabolic intermediate, exposure to it leads to its metabolism into other chemicals, including formic acid. Therefore, the presence of formaldehyde in the body leads to its transfer to the body's organs, including the liver. The spleen and lung, thus can lead to increased oxygen activity, leading to oxidationFats accumulate in the body, which negatively affects the health of the body, and thus leads to changes in the body's cells, including skin cells (Du., 2014.( As for the kidney weight, it was  $(0.28\pm1.39)$  grams in the positive control group, while the negative control group was  $(0.05\pm1.69)$ ) grams. As for the third and treatment group, the kidney weight was  $(0.07\pm1.69)$  grams.

The results of the current study showed that there was a very slight, non-significant increase in kidney weight in the formaldehyde-induced group (negative control) compared to the non-induced group. There were no significant differences in kidney weight between the treated group and the induced group, as in (Figure - 4).



)Figure-4): Kidney weight in the study groups

The results of the current study agreed with the results of (Abd Ellah. 2007), and the results also agreed with the results of (Raja. 2012) and the results of (Amina. 2016). The reason for the increase in kidney weight may be attributed to the release of formaldehyde when it is metabolized into formic acid in the liver and red blood cells. It is secreted

Either through urine or feces, so the kidneys do not retain the liquid, but rather purify it and excrete it outside the body, thus transferring the chemicals or their metabolic types from the blood plasma to the tubes of the urinary organs and thus leading to an increase in the concentration of formaldehyde toxicity (İnci. 2013). Formaldehyde can react with various cell components such as lipids, proteins, and nucleic acids (Dixit. 2008(,

Therefore, chronic inhalation of formaldehyde may deplete the activities of antioxidant enzymes and stimulate oxidative stress, thus causing toxicity to vital organs, including the kidneys (Akwiwu 2015). We also note from the results of the study that the group treated with L.acidophilus bacteria did not

Significant differences were recorded with the induced group, so the reason may be that the probiotic may have the ability to keep the kidney weight within normal limits or the dose concentrations were not high to cause some changes.

A study of the effect of L.acidophilus + L.plantarum bacteria on liver functions in male rats with eczema experimentally induced with formaldehyde is under study.

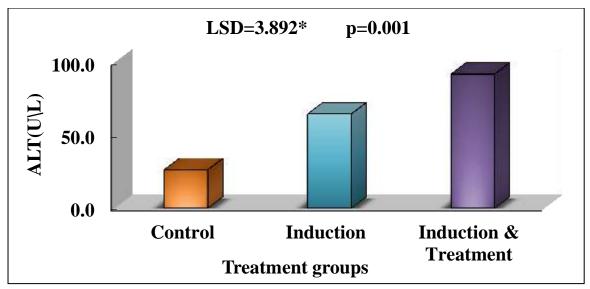
Efficacy of the enzyme N-Aminotransferase in blood serum

Table (3-4) shows that the mean  $\pm$  standard deviation of ALT enzyme activity in the

positive control group was  $(0.96 \pm 26.48)$  IU/L, while the negative control group was  $(5.93 \pm 64.77)$  IU/L, and as for the treated group it was  $(2.44\pm92.2)$  IU/L.

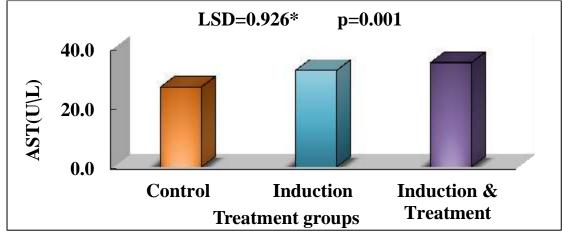
Arithmetic mean ± st	Member weights	
AST(U\L)	ALT(U\L)	Study treatment
1.07±27.02 C	0.96±26.48 C	Control
0.75±32.58 B	5.93±64.77 B	Induction
0.82±35.22 A	2.44±92.2 A	Induction and treatment
0.926*	3.892*	LSD
0.001	0.001	p-value

Different letters have significant differences below the probability level less than P<0.05. ANOVA test with least significant difference (LSD).The results of the current study showed a significantly significant increase in the activity of the ALT enzyme in the blood serum of the group induced with formaldehyde compared to the control group that was not induced (control).positive), while an increase in enzyme activity was observed in the group treated with bacteria compared to the negative and positive control groups, as in Figure (5-4.(



)Figure-5): The activity of the ALT enzyme in the blood serum of the study groups

Efficacy of aspartate aminotransferase enzyme in blood serum 3(3-4) shows that the mean  $\pm$ standard deviation of AST enzyme activity in the positive control group was (1.07 $\pm$ 27.02) international units/liter, while the negative control group was (0.75 $\pm$ 58.32) international units/liter, and as for the treated group, it was (2.44 $\pm$ 92.2) IU/L The results of the current study showed a significant increase in the activity of the AST enzyme in the blood serum of the group induced with formaldehyde compared to the non-induced control group (positive control), while an increase in the activity of the enzyme was observed in the group treated with bacteria compared to the negative and positive control groups, as in Figure 6.





The results of the current study agreed with the results of (Cikmaz. 2010), the results of (Hernán. 2018), and the results of (Wei. 2017), and (Samah. 2020), which indicated an increase in liver enzymes in the groups treated with formaldehyde. Formaldehyde is a colorless gas with a pungent odor that is widely used as a preservative.

Its use in different concentrations on the skin can cause allergies or skin infections (N. Latorre., 2011), so atopic dermatitis is a chronic relapsing disease that prevails all over the world, especially in developed countries. Therefore, in studies, formaldehyde was tested on laboratory animals to determine its effect on the development of skin inflammation (eczema) (Rafael., 2017.(

The reason for the increase in liver enzymes may be attributed to the fact that the Formal glutathione - FGSH using NADP+ as a cofactor (Cikmaz., 2010). Therefore, deletion of the enzyme in mice leads to an increase in the levels of induced DNA addition in the liver and kidneys, which leads to higher levels of liver enzymes (Zemba. 2019.(

Exposure to formaldehyde can cause increased oxidative stress and increased formation of free radicals due to cell breakdown and necrosis. (Table-4) Arithmetic mean standard deviation Study totals.

,Hepatitire results in damage to the liver tissue and loss or destruction of enzyme receptors located on the epithelial cells lining the bile duct and around the central vessel, which leads to increased release of liver enzymes outside the hepatic cells (Zhang et al. 2020). An increase in liver enzymes also indicates

the occurrence of inflammation that contributes to cell damage Clearly, it is found that the damaged liver cells are surrounded by inflammatory cells as a result of the leakage of some hydrolytic enzymes from the damaged liver cells, which may lead to an increase in liver enzymes as a result of the destruction of the liver cells. In addition, free radicals cause significant damage to the liver cell through their effect on Cellular molecules, especially the phosphorylated lipids present In the plasma membrane, which leads to the release of these enzymes from the cells into the serum and an increase in lipid peroxidation, causing the breakdown of the plasma membrane. The damage may also reach the cellular nuclei,

formaldehyde metabolism process begins with the spontaneous reaction between aldehyde and glutathione to form 5-hydroxymethyl glutathione -HMGSH, as the alcohol dehydrogenase enzyme 5-ADH5 oxidizes 5 hydroxymethyl glutathione to 5-formyl.

causing changes in the shape and composition of these nuclei through the occurrence of a breakdown of the genetic material (DNA), which in turn stimulates Programmed cell death occurs (Prahalathan et al. 2005).On the other hand, it was found from the results of the current study that treatment with bacteria L.acidophilus and Lactobacillus plantarum in rats induced with formaldehyde led to an increase in the effectiveness of liver enzymes. The literature did not indicate the role of bacteria in treating liver enzymes in rats induced with formaldehyde, but there are other studies that indicated the role of bacteria, a including study L.acidophilus and Lactobacillus plantarum in rats induced with formaldehyde led to an increase in the effectiveness of liver enzymes. The literature did not indicate the role of bacteria in treating liver enzymes in rats induced with formaldehyde, but there are other studies that indicated the role of bacteria, including a study

)Hoda., 2019), which indicated that treatment with ethephon, which causes oxidative stress, and then treatment with bacteria led to a decrease in the effectiveness of liver enzymes, through its role as an antioxidant nutritional supplement that plays a role in reducing the levels of liver enzymes, and this is not consistent with the results of the current study. L.acidophilus bacteria is supposed to work to return liver enzymes to their normal level through its antioxidant and antioxidant capacity. infections Nonetheless, the larger influence might result from formaldehyde's toxicity hindering the bacterial treatment of liver tissue, or the levels may not be adequate for the bacteria to restore the enzymes to their typical ranges. (Peran, 2007..(

The study (Yemisi, 2019) also indicated that treatment with bacteria in mice induced to develop diabetes led to a decrease in the effectiveness of liver enzymes. As the results of the study (Al-Sultani, 2022) indicated,

Rats treated withAlloxan showed elevated liver enzymes, but when given L.acidophilus bacteria, there was a reduction in liver enzymes. Change to This suggests that bacteria play an effective role. which is an effective probiotic in maintaining the activity and integrity of the epithelial cells lining the bile duct, indicates a direct effect of the probiotic in improving liver function. This is not consistent with the results of the current study, (Mahjida., 2014.(

Study of the effect of L.acidophilus and Lactobacillus plantarum on the level of immunoglobulin in male rats with experimentally induced eczema.

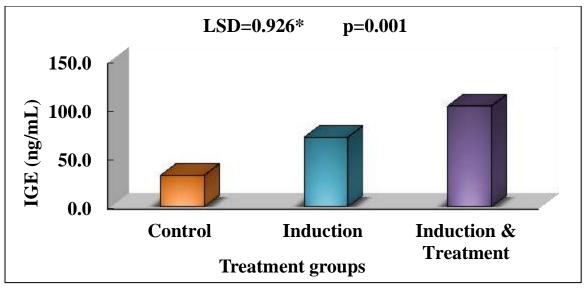
IgE level in blood serum Table (4-4) shows the mean  $\pm$  standard deviation of the Ige level in the control group The positive test group  $(4.94\pm32.5)$  ng/ml, while the negative control group was  $(5.23\pm71.56)$  ng/ml, and the treated group was  $(6.38\pm103.62)$  ng/ml. induced, with an increase in the level of IgE in the treated group, as in (7). (4): Mean  $\pm$  standard deviation of immunoglobulin level in albino rats for the current study The different letters have significant differences below the probability

(4): Mean ± standard deviation of the immunoglobulin level in the albino rats For the current study.

Arithmetic mean ± st	Study is peeking	
IGE (ng/mL)		
	4.94±32.5 C	Control
LSD=5.775 *	5.23±71.56 <b>B</b>	Do you have an idea?
p-value=0.001	6.38±103.62 <b>A</b>	Induction and treatment

\*The different letters have significant differences below the probability level less than P<0.05. ANOVA

test with least significant difference (LSD).



:(7)IGE level in the blood serum of the study groups

The results of the current study agreed with the results of (Huaxiao., 2016) and the results of (Sayed., 2024). so the World Health Organization reported The results of the **References** 

[1]Zahraa Raysan Karim Shati. The effect of some probiotics on a number of nutritional and biological indicators that cause obesity in rats. Doctoral thesis. University of Baghdad, College of Agriculture. (2017..(

[2]Al-Sultani. Zahraa Hussein Malik Al-Sultani. Study of the effect of Lactobacillus acidophilus bacteria on the nutritional and health indicators of male rats with experimentally induced diabetes. 2022..

[3]]Al-Khazraji, Aseel Adnan Hussein. 2005. The use of Lactobacillus GG bacteria in the production and prolonging the shelf life of some medicinal soft cheeses. Master's thesis. College of Agriculture, University of Baghdad.

[4]A.H. Jameel , M.J. Mohammed , M.S. Mahdi and K.M. Thalj. Physiological effects of lactic acid bacteria against melamine induced toxicity in female albino rats. current study agreed with the results of (Huaxiao., 2016) and the results of (Sayed., 2024). The reason for the increase in the level of IGE may explain that the immunotoxicity occurring is due to formaldehyde, (7-4.(

[5]Abd Ellah, M.R., Okada, K and Yasuda, J. (2007). Oxidative stress and bovine liver diseases: Role of glutathione peroxidase and glucose 6-phosphate dehydrogenase. Japanese Journal of Veterinary Research. 54(4):163-173.

[6[Abdel Mohsen, A.H.; Abdel Wahab, H. and Emad, A. (2011). New markers of disease activity in children with atopic dermatitis. J. Am.Sci.; 7 (10): 404-408.

[7]Akan, Aysegul, et al. "Single center experience: clinical features of children with atopic dermatitis." Clinical and Translational Allergy 1.Suppl 1 (2011): P23.

[8]Akwiwu, E. C., Usoro, C. A.O., Akpotuzor, J. O., Etukudo, M. H. (2015) Occupational Health and the Impact of Long Term Formaldehyde Exposure on Health Professionals in Calabar, Nigeria. Journal of Natural Sciences Research: 5, 119-126. [9]Amina Mohamed Tolbaa and Abeer A. A. Salamab\*. Adverse effects of melamine formaldehyde on the liver, kidney and brain in rats. Der Pharma Chemica, 2016, 8(2):398-409.

[10]Amouri, Meriem, et al. "Atopic dermatitis in Tunisian schoolchildren." Pan African Medical Journal 9.1 (2011.(

[11]Chen, G, Shi, Y, Sturla, SJ, Jalas, JR, McIntee, EJ, Villalta, PW, Wang, M & Hecht, SS 2003, 'Reactions of formaldehyde plus ac etaldehyde with deoxyguanosine and DNA: Formation of cyclic deoxyguanosine adducts and formaldehyde cross-links', Chemical research in toxicology, vol. 16, no. 2, pp. 145-152. https://doi.org/10.1021/tx025614r.

[12]Cheng, G, Shi, Y, Sturla, SJ, Jalas, JR, McIntee, EJ, Villalta, PW, Wang, M & Hecht, SS 2003, 'Reactions of formaldehyde plus ac etaldehyde with deoxyguanosine and DNA: Formation of cyclic deoxyguanosine adducts and formaldehyde cross-links', Chemical research in toxicology, vol. 16, no. 2, pp. 145-152.

[13]Cheng, G, Shi, Y, Sturla, SJ, Jalas, JR, McIntee, EJ, Villalta, PW, Wang, M & Hecht, SS 2003, 'Reactions of formaldehyde plus ac etaldehyde with deoxyguanosine and DNA: Formation of cyclic deoxyguanosine adducts and formaldehyde cross-links', Chemical research in toxicology, vol. 16, no. 2, pp. 145-152. https://doi.org/10.1021/tx025614r.

[14]Cikmaz S, Kutoglu T, Kanter M, Mesut R. Effect of formaldehyde inhalation on rat livers: A light and electron microscopic study. Toxicology and Industrial Health. 2010;26(2):113-119.

# doi:10.1177/0748233710362384

[15]Clark, I.A. and Vissel, B. The meteorology of cytokine storms, and the clinical usefulness of this knowledge. InSeminars in immunopathology 2017 (Vol. 39, No. 5, pp. 505-516). Springer Berlin Heidelberg.

[16]Dixit, D. (2008). Role of standardized embalming fluid in reducing the toxic effects of formaldehyde. Indian Journal of Forensic Medical Toxicology. 2:1006-1012.

[17]Duong et al. Reproductive and developmental toxicity of formaldehyde: a systematic review. Mutat. Res. (2011.(

[18]El-Bayoumy, Karam. "The protective role of selenium on genetic damage and on cancer." Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis 475.1-2 (2001): 123-139.

[19]Evrard B, Coudeyras S, Dosgilbert A, Charbonnel N, Alamé J, et al.- 183 (2011). Dose-dependent immunomodulation of human dendritic.110.extract. Journal of Morphology and Embryology, 51(1):91–95.

[20]Fischer MH. THE TOXIC EFFECTS OF FORMALDEHYDE AND FORMALIN. J Exp Med. 1905 Feb 1;6(4-6):487-518. doi: 10.1084/jem.6.4-6.487. PMID: 19866982; PMCID: PMC2124506.

[22]Gurel A, Coskun O, Armutcu F, Kanter M, Ozen OA. Vitamin E against oxidative damage caused by formaldehyde in frontal cortex and hippocampus: biochemical and histological studies. J Chem Neuroanat. 2005 May;29(3):173-8. doi: 10.1016/j.jchemneu.2005.01.001. PMID: 15820619.

[23]Hassan, Hassan pyar Ali; Pazilah, I. and Liong, Min-Tze (2011). Isolation of probiotics Lactobacillus acidophilus from commericial yoghurt Alandalus for social and Applied Sciences 4 (7):5-15.

[24]Hernán Reingruber, Lucas Blas Pontel, Formaldehyde metabolism and its impact on human health, Current Opinion in Toxicology, Volume 9, 2018, Pages 28-34, [25]Hernán Reingruber, Lucas Blas Pontel, Formaldehyde metabolism and its impact on human health, Current Opinion in Toxicology, Volume 9, 2018, Pages 28-34, ISSN 2468-2020,

https://doi.org/10.1016/j.cotox.2018.07.001.

[26]Hoda I Bahr,1,\* Rania Hamad,2 and Shimaa AA Ismail3. The impact of Lactobacillus acidophilus on hepatic and colonic fibrosis induced by ethephon in a rat model. Iran J Basic Med Sci. 2019 Aug; 22(8): 956–962.

[27]Huaxiao Wen, Langyue Yuan, Chenxi Wei, Yun Zhao, Yan Qian, Ping Ma, Shumao Ding, Xu Yang, Xianliang Wang, Effects of combined exposure to formaldehyde and benzene on immune cells in the blood and spleen in Balb/c mice, Environmental Toxicology and Pharmacology, Volume 45, 2016, Pages 265-273, https://doi.org/10.1016/j.etap.2016.05.007.

[28]İnci M, Zararsız İ, Davarcı M, Görür S. Toxic effects of formaldehyde on the urinary system. Turk J Urol. 2013 Mar;39(1):48-52. doi: 10.5152/tud.2013.010. PMID: 26328078; PMCID: PMC4548585.

[29]Kim, Ki-Hyun, Shamin Ara Jahan, and Jong-Tae Lee. "Exposure to formaldehyde and its potential human health hazards." Journal of Environmental Science and Health, Part C 29.4 (2011): 277-299.

[30]Kim, Woon-ki, et al. "Lactobacillus acidophilus KBL409 Ameliorates Atopic Dermatitis in a Mouse Model." Journal of Microbiology 62.2 (2024): 91-99.

[31]Lee, J. H., Son, S. W., & Cho, S. H. (2016). A comprehensive review of the treatment of atopic eczema. Allergy, asthma & immunology research, 8(3), 181-190.

[32]Mahmoud S.M., Hegab A. S., Ibrahim I.H. and Farag A. THE EFFECT OF FORMALDEHYDE ON THE LIVER OF ADULT MALE ALBINO RATS AND POSSIBLE PROTECTIVE ROLE OF VITAMIN C ESCTJ;Vol. (4) N0. (1(

[33]Majida AJ, Abass DE. Effects of probiotics (Lactobacillus acidophilus) on liver functions in experimental colitis in rats. The Iraqi Journal of Veterinary Medicine. 2014;38(2):48-54.

[34]Mamoru Tanida Jiao Shen, Keiko Maeda, Yuko Horii, Toshihiko Yamano, Yoichi Fukushima, Katsuya Nagai. (2008). High-fat diet- induced obesity is attenuated by probiotic strain Lactobacillus paracasei ST11 (NCC2461) in rats. Obesity research & clinical practice, Volume 2, Issue 3, Pages 159-169.

[35]N. Latorre,\* J.F. Silvestre, A.F. Monteagudo. Allergic Contact Dermatitis Caused by Formaldehyde and Formaldehyde Releasers. Actas Dermosifiliogr. 2011;102(2):86–97.

[36]Omar M.E. Abdel-Salam, Eman R. Youness , Yasser Ashry Khadrawy and Amany A. Sleem. Brain and liver oxidative stress after sertraline and haloperidol treatment in mice. J Basic Clin Physiol Pharmacol 2013; 24 (2), pp. 115-123.

[37]Peran L, Camuesco D, Comalada M, Bailon E, Henriksson A, Xaus J, et al. A comparative study of the preventative effects exerted by three probiotics, Bifidobacterium lactis, Lactobacillus casei and Lactobacillus acidophilus, in the TNBS model of rat colitis. J Appl Microbiol. 2007;103:836–844.

[38]Rafael Taeho Han, 1 Seung Keun Back, 2 JaeHee Lee, 1 Hye young Kim, 1 Hee Jin Kim, 3 and Heung Sik Na . Effects of Exposure of Formaldehyde to a Rat Model of Atopic Dermatitis Induced by Neonatal Capsaicin Treatment. J Vis Exp. 2017; (127): 55987. [39]Rafael Taeho Han, 1 Seung Keun Back, 2 JaeHee Lee, 1 Hye young Kim, 1 Hee Jin Kim, 3 and Heung Sik Na . Effects of Exposure of Formaldehyde to a Rat Model of Atopic Dermatitis Induced by Neonatal Capsaicin Treatment. J Vis Exp. 2017; (127): 55987.

[40]Raja, D.S. (2012). Potential health hazards for students exposed to formaldehyde in the gross anatomy laboratory. Journal Environmental Health. 74: 36-40.

[41]Remus N, Reichenbach J, Picard C, Rietschel C, Wood P, Lammas D, Kumararatne DS, Casanova JL. Impaired interferon gamma-mediated immunity and susceptibility to mycobacterial infection in childhood. Pediatr Res. 2001 Jul;50(1):8-13. doi: 10.1203/00006450-200107000-00005. PMID: 11420412.

[42]Samah, A. El-Nagdy, Ebtesam Khalid Ali Nasif. TOXIC Elgezawy, **EFFECTS** OF FORMALDEHYDE ON CELLULAR IMMUNITY AND POSSIBLE RELATIONSHIP TO ITS RISK OF CARCINOGENESIS: AN EXPERIMENTAL STUDY. J. Zagazig Forensic Med.& Toxicology. Vol.(18) No. (1) Jan 2020.

[43]Sayed M. El-Sayed, Eyad M.T. Ali, and Hesham I. Abdallah. Formaldehyde Inhalation Induces Histological and Immunohistochemical Aberrations in the Spleen of the Male Albino Rats. Egypt. Acad. J. Biolog. Sci., 16(2): 1-26 (2024(

[44]Schulien, I., Hockenjos, B., van Marck, V., Ayata, C. K., Follo, M., Thimme, R., & Hasselblatt, P. (2020). Extracellular ATP and Purinergic P2Y2 Receptor Signaling Promote Liver Tumorigenesis in Mice by Exacerbating DNA DamagePurinergic Receptor Signaling in HCC. Cancer Research, 80(4), 699-708.

[45]Teijaro, J.R. Cytokine storms in infectious diseases. In Seminars in

immunopathology. Springer Berlin Heidelberg 2017; 39 (5) : 501-503.

[46]Ucmakli, E.,Armutcu, F.,Özturk, A. The effects of formaldehyde intoxication on the inducible nitric oxide synthase expression and nitric oxide level in the liver tissue of rats. J.Med. Sci. 43: 52-56. (2013.(

[47]Wei, C., Wen, H., Yuan, L. et al. Formaldehyde induces toxicity in mouse bone marrow and hematopoietic stem/progenitor cells and enhances benzene-induced adverse effects. Arch Toxicol 91, 921–933 (2017)WHO. (2016). "Obesity and overweight Fact sheet N°311". WHO.

[48]Woo, J. Y.; Gu, W.; Kim, K. A.; Jang, S. E.; Han, M.J. and Kim, D. H., (2014). Lactobacillus pentosus var. plantarum C29 ameliorates memory impairment and inflammaging in a D-galactose-induced accelerated aging mouse model. Anaerobe 27, 22–26.

[49]Yemisi Adesiji1, Samsudeen Owolabi2, Olubunmi Gloria Ayelagbe3, Adekunle Olowe. Effects of Lactobacillus acidophilus on Biochemical Indices and Liver Histology in Streptozotocin-induced Diabetic Rats. Journal of Clinical and Diagnostic Research. 2019 Jul, Vol-13(7): OC11-OC15.

[51]Zemba, S., Damiano, L., Little, H., Doris, J., and Estabrooks M. (2019). Formaldehyde A Leading Air Toxic The Magazine for Environmental Managers. Pp: 5-12.

[52] Zhang, C., Chen, Y., Zhang, M., Xu, C., Gong, G., Veeraraghavan, V. P., ... & Li, Y. (2020). Vicenin-2 treatment attenuated the Diethylnitrosamine-induced liver carcinoma and oxidative stress through increased apoptotic protein expression in experimental rats. Journal of Environmental Pathology, Toxicology and Oncology