Appliction of ozone technology in food industry

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تطبيقات تكنولوجيا الأوزون فى التصنيع الغذائى

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تعريف غاز الاوزون

- الأوزون عبارة عن أكسجين ثلاثي يتكون من ثلاث ذرات أكسجين O_3، نتيجة الأشعة فوق البنفسجية أو التفريغ الكهربائي يحدث تتحلّل جزيئات الأكسجين العادي الثنائي O_2 ثم تتحد ذرة منه O مع ذرة الأكسجين العادي O_2 فتكون الأكسجين الثلاثي O_3، ويوجد الأوزون في الحالات الثلاث، ففي الحالة الغازية يكون على هيئة غاز أزرق، وفي الحالة السائلة يكون على هيئة سائل أزرق غامق، وفي الحالة الصلبة يكون على هيئة بلورات لونها بنفسجي غامق، ويذوب الأوزون في الماء وله رائحه مثيرة و مميزة.

INTRODUCTION

- Food industries are currently in need of different innovative technologies to meet the consumer demand.

- There are some of the technologies like HPP, pulsed electric field, high intensity pulsed light etc.

- But attention is now focused on the ozone as a powerful sanitizer.

- Currently – more than 3000 water treatment installations and more than 300 potable water treatment plants all over the world.

تاريخ استخدام الأوزون في التصنيع الغذائى

HISTORY OF OZONE

DATE	EVENT
1840	Discovered by Schonbein
1909	Firstly used as a food preservative for cold storage of meats
1939	Was found to prevent the growth of yeast & mold during the storage of fruits
1982	O3 declared GRAS for treatment of bottled water
1997	Industrial expert panel reported that use of ozone is safe in foods processing for human consumption
2000	FDA and USDA approval granted for direct contact food additive.

- و في يونيو عام ٢٠٠١ اعتمدت إدارة الرقابة الفيدرالية لخدمات فحص الحبوب الأمريكية الـ FGIS الأوزون كغاز آمن والموافقة عليه كغاز مضاد للبكتريا Antibacterial ويمكن استخدامه في الحبوب والمنتجات الغذائية في الأغراض الصحية وأغراض التعقيم. كما اعتبرت وكالة حماية البيئة EPA الأوزون بأنه غير كيميائي وبديل صديق للبينة في تصنيع الغذاء.ووافق المجلس الوطني للقياسات العضوية NOSB على اعتماد الأوزون للاستخدام في المحاصيل العضوية كمادة مضادة للميكروبات.وعلية قامت العديد من الهيئات الدولية مثل وكالة حماية البيئة (EPA) الامريكية وهيئة الاغذية والادوية الامريكية ادارة السلامة والصحة المهنية الامريكية (OSHA)

Occupational Safety and Health Administration

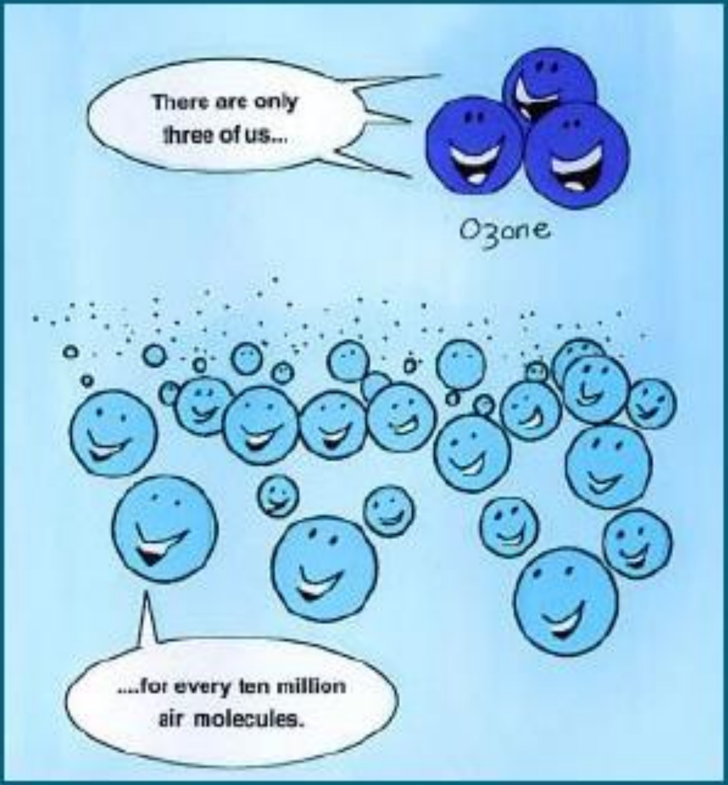
بوضع حدود تنظيمية للمتعاملين مع غاز الاوزون

WHAT IS OZONE?

- OZONE: **highly reactive form** of oxygen consisting of three oxygen atoms i.e. O3

- It is a colorless and **extremely unstable** gas.

- It is **an effective antimicrobial** agent used directly into the food.



- Ozone is naturally found in the stratosphere and protects us from **harmful radiations.**

HOW OZONE IS PRODUCED?

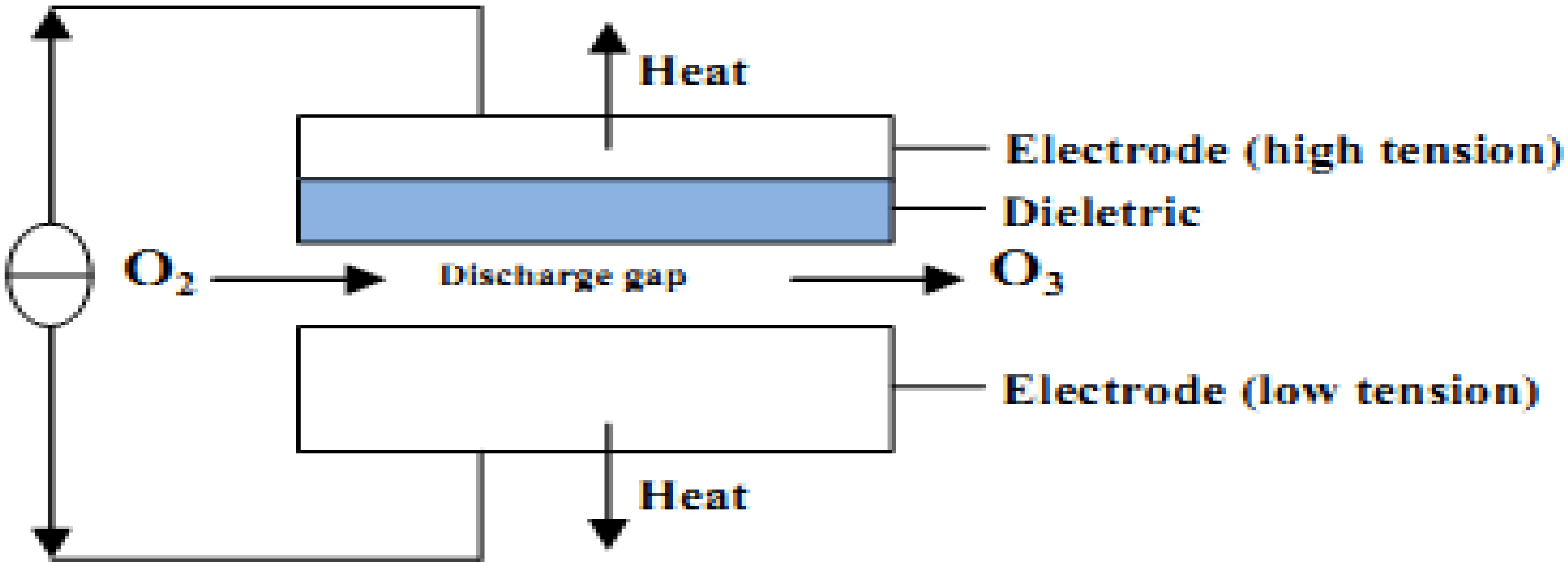
- There are **two methods** of producing ozone

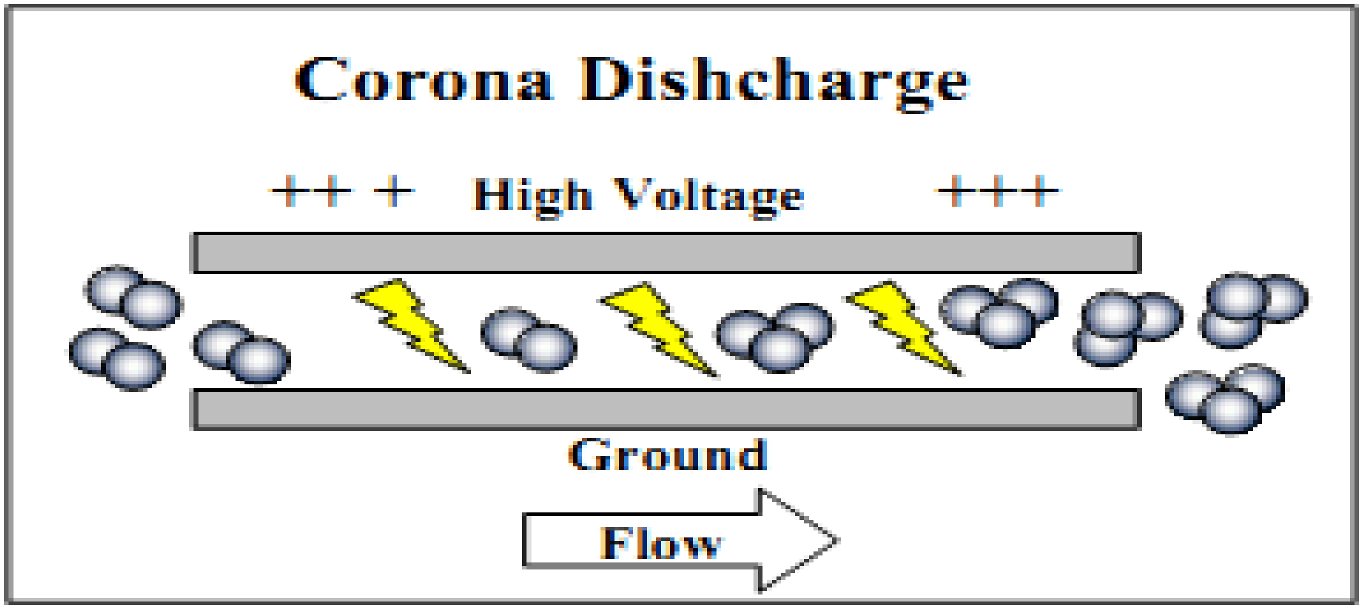
1) **Ultra-violet method**
2) **Corona discharge method**

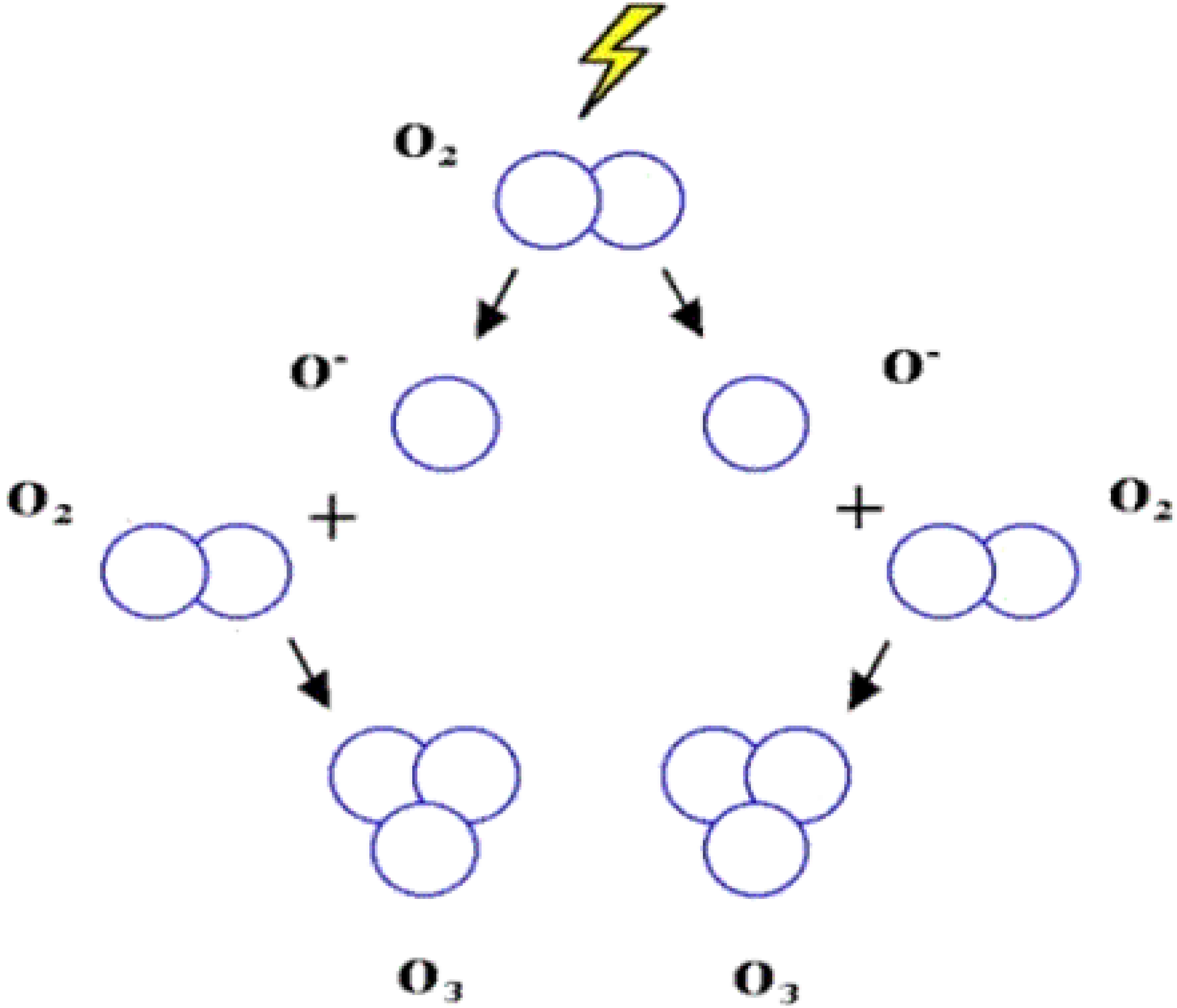
- Ultra-violet light creates ozone when a **wavelength > 254 nm** hits an oxygen atom.
- Corona discharge creates ozone **by applying high voltage** to a metallic grid sandwiched between two dielectrics.
- Splitting of oxygen molecules into single oxygen atom & combine with another oxygen molecule (O2) to form ozone (O3).

- **إنتاج غاز الاوزون بواسطة اسطوانة التفريغ Corona Discharge (CD) ozone generation**
- **وهى من اكثر الطرق شيوعا واستخداما لإنتاج غاز الاوزون صناعيا وفيها ينتج الاوزون كالتالى:**

أحداث فرق جهد عالى بين قطبين او طرفين الاسطوانة مع مرور الهواء الجاف او الاكسيجين بها كما هو الشكل رقم (١) حيث يقوم الجهد العالى من التيار المار فى الأسطوانة بأثارة الكترونات الاكسجين مما يؤدي الى تقسيم جزيئات الأكسجين إلى ذرات، والتي تتحد مع جزيئات الأكسجين الأخرى لإنتاج الأوزون. تركيز غاز الأوزون الناتج بواسطة هذه الطريقة يعتمد على مقدار فرق الجهد، درجة الحرارة ، نسبة الرطوبة فى الهواء المستخدم، سمك الاسطوانة ، فجوة التفريغ، الضغط المطلق في غضون فجوة التفريغ، وطبيعة الغاز التي تمر عبر الأقطاب حيث تختلف كمية الاوزون الناتج ايضا حسب مصدر الاكسجين المستخدم حيث اذا كان يتم استخدام الهواء كمصدر للأكسجين فأن ١-٤٪ من غاز الأوزون يمكن أن تنتج. ، واستخدام الأكسجين النقي تصل كمية غاز الاوزون المنتج إلى ٦-١٤٪ من حجم الاكسجين المار فى الأسطوانة.


Heat
Electrode (high tension)
Dieletric
O_2
Discharge gap
O_3
Electrode (low tension)
Heat


Corona Dishcharge
+++ High Voltage +++
Ground
Flow

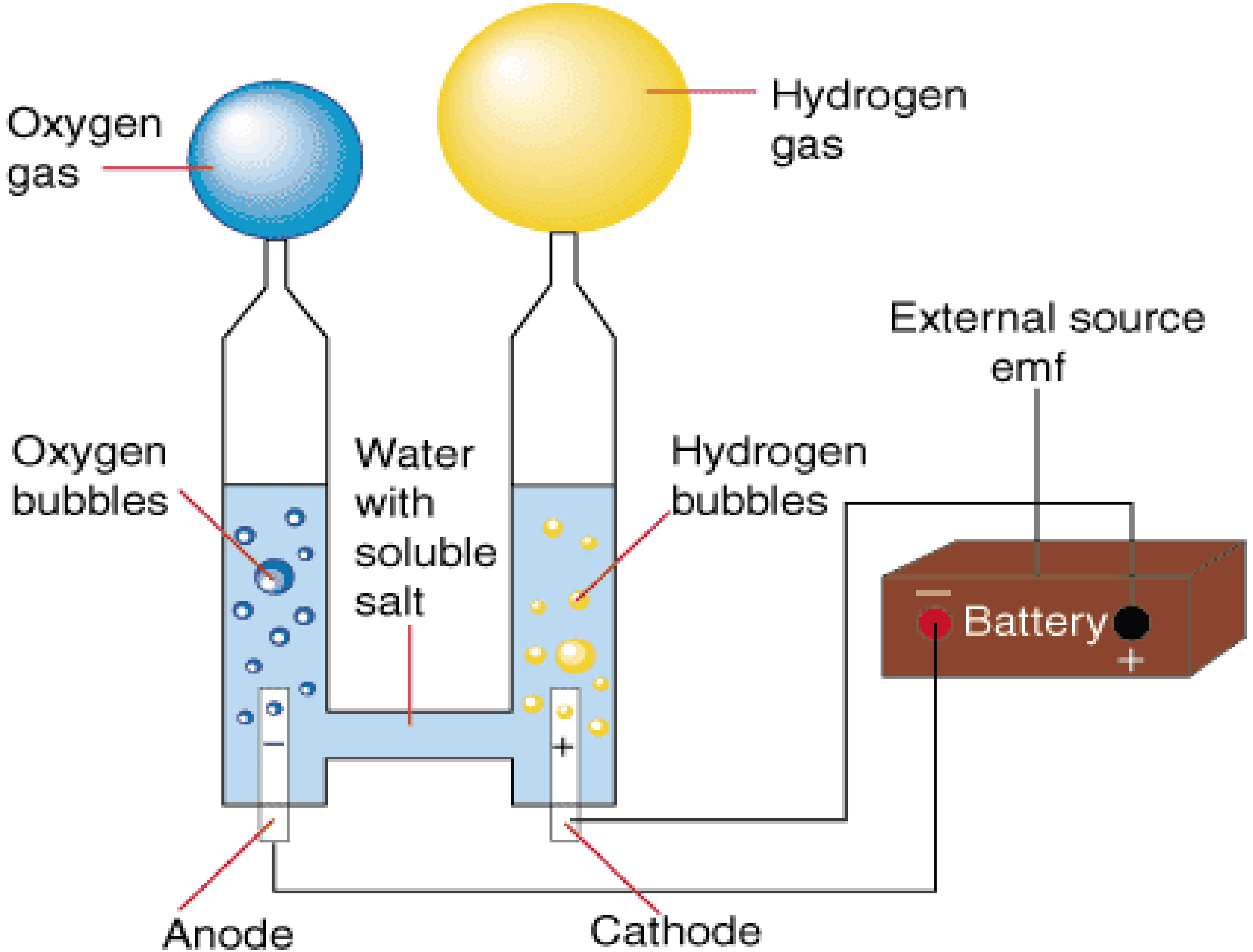


O_2
O^-
O^-
O_2
+
+
O_2
O_3
O_3

إنتاج غاز الاوزون بواسطة لمبات الاشعة فوق البنفسيجية Ultraviolet (UV) ozone generation:

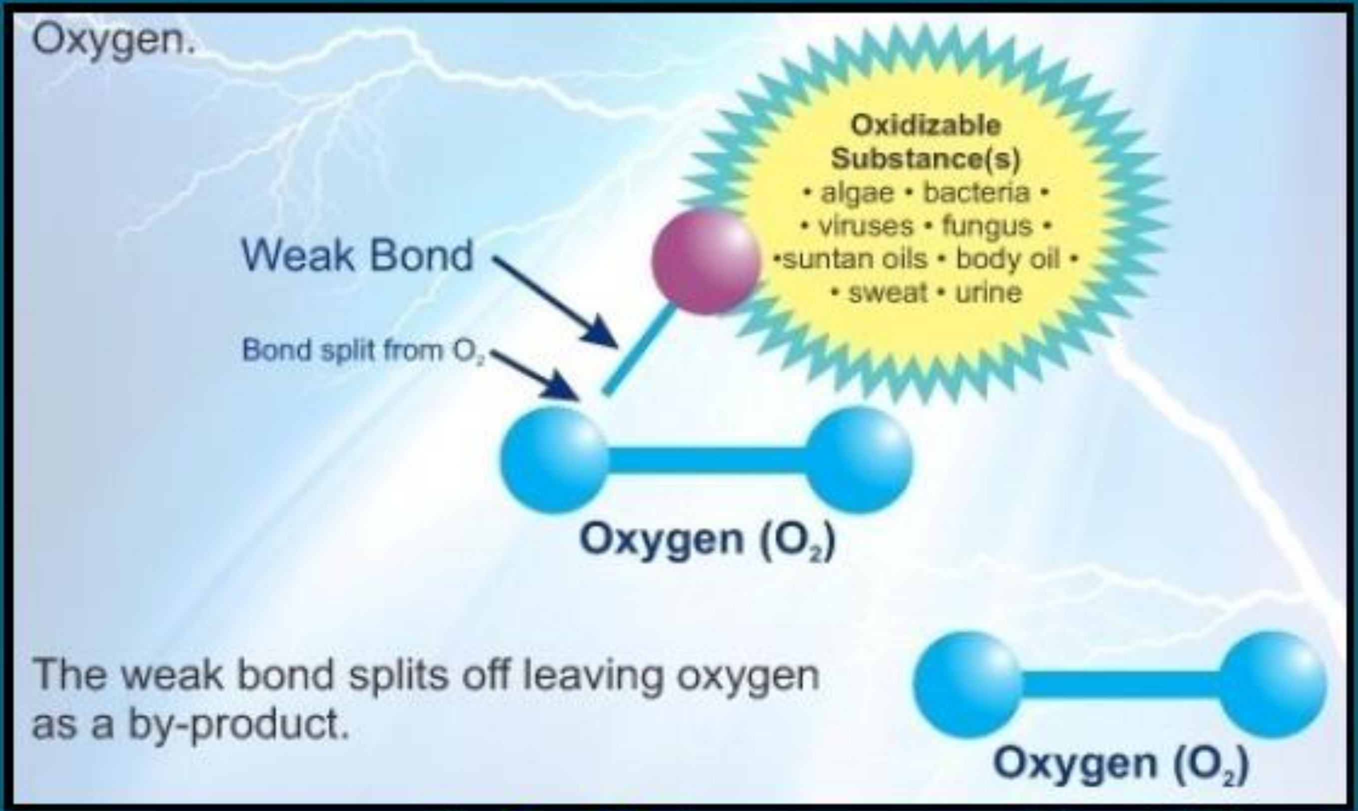
- انتاج الاوزون بهذه الطريقة هى مماثلة لتلك التى تحدث فى طبقة الستراتوسفيرز. وهنا يتم استخدام لمبة اشعة فوق بنفسيجية تبعث اشعة فى مدى ١٨٥ نانوميتر وبالتالى عند مرور هواء حولها فيحدث تكسير للاكسيجين (O_2) ونحصل على اكسجين نشط (O) يقوم بالارتباط مع الأكسجين ويعطى O_3

- **إنتاج غاز الاوزون بطريقة التحليل الكهربى Electrolytic ozone generation:**
- التحليل الكهربائي هو العملية التي يتم فيها تمرير تيار كهربائي خلال سائل، مما تسبب في تفاعل كيميائي. هنا يتم استخدام الماء (H2O) مع تمرير تيار كهربائى بها لكى يحدث تحلل كهربى ونحصل على اكسجين حر (O) الذى يستطيع الارتباط بعد ذلك مع الاكسجين معطيا الاوزون.


Oxygen gas
Hydrogen gas
External source emf
Oxygen bubbles
Water with soluble salt
Hydrogen bubbles
Battery
−
+
Anode
Cathode

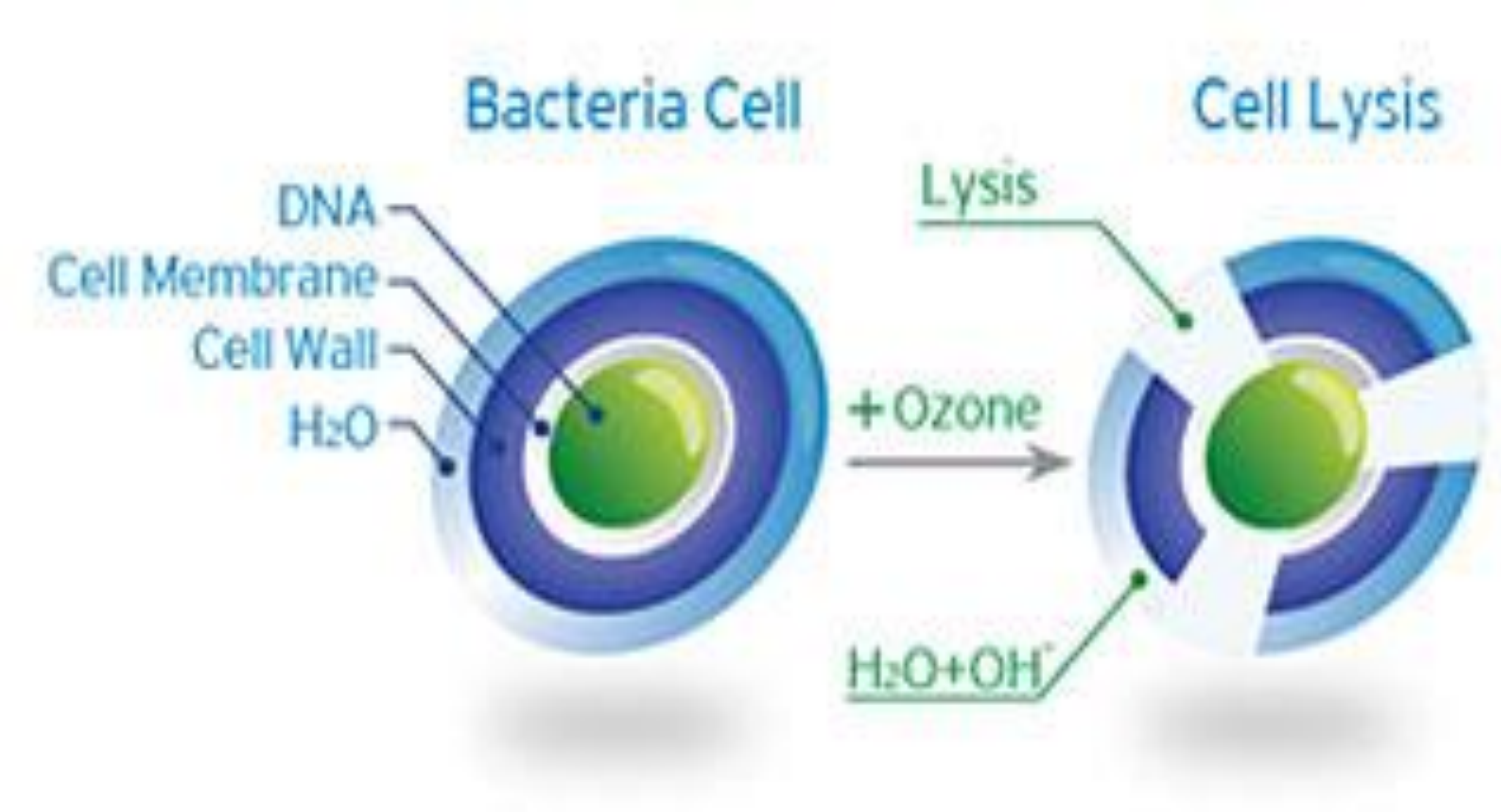
HOW DOES OZONE WORK?

- The unstable third oxygen atom of ozone can **combine with organic and inorganic molecules** to destroy them through oxidation.



- Finally ozone **reverts back to oxygen** after it is used. This makes it a very environmentally friendly oxidant.

شكل يوضح تأثير غاز الاوزون على الخلية البكتيرية



COMPARATIVE STUDY

Comparison between ozone and chlorine shows that-

Action in water	Chlorine	Ozone
Oxidation Potential (Volts)	1.36	2.07
Disinfection: Bacteria, Viruses	Moderate	Excellent
Carcinogen Formation	Likely	Unlikely
Operation Hazards : Skin Toxicity	High	Moderate
Capital investment	Low	High
Operational cost	High	Low

أمثلة على بعض تطبيقات الأوزون في التصنيع الغذائى

- **يطلق على المعاملة بالاوزون Ozonation**
- يستخدم غاز الاوزون مع العديد من المواد الغذائية بتركيزات مختلفة و زمن تعرض مختلفة حسب طبيعة المادة الغذائية وحسب درجة الحرارة والحموضة المتواجد حيث يكون الاوزون اكثر كفاءه و فاعلية فى الاوساط المعتدلة فى رقم الحموضة وكذا فى درجات الحرارة المنخفضة و فى الاونة الاخيرة زاد استخدام غاز الاوزون بشكل تجارى على حساب المركبات الكيماوية المختلفة التى تستخدم فى التطهير او القضاء على البكتريا او الفطريات نظرا الى انة لا يوجد اى اثار متبقية منة بعد المعاملة على عكس المواد الكيماوية الاخرى.

DIFFERENT APPLICATIONS

Some applications of ozone in food processing –

- ❑ Ozone in seafood processing
- ❑ Ozonated fruits and vegetables
- ❑ Ozone and milling
- ❑ Ozone in wine industries
- ❑ Bottled water production
- ❑ Surface sanitation
- ❑ Storage and packaging of food

OZONE IN SEAFOOD PROCESSING

- Effective at lowering water consumption by increasing water re-use.
- Lowers diseases and maintains clean water in fish growing ponds.
- Odor control in offal rooms and other processing area.
- Storage of live seafood.
- Improved antimicrobial intervention in processing of whole fish.

اللحوم والدواجن والاسماك

يستخدم الاوزون الذائب فى الماء فى عمليات الغسيل التى تجرى للحوم او منتجاتها بغرض القضاء على العديد من البكتريا الممرضة مثل السالمونيلا و بكتريا القولون وغيرها حيث يستطيع غاز الاوزون القضاء على جميع انواع البكتريا الموجبة او السالبة لجرام وكذا الخمائر نتيجة أكسدة المكونات الخلوية مثل مجموعات السلفهيدريل والأحماض الأمينية من الأنزيمات، والببتيدات والبروتينات والأحماض الدهنية غير المشبعة، و أكسدة مكونات الغلاف الخلوى للخلية البكتيرية مما يتسبب في موت الخلايا السريع. لذا يستخدم غاز الاوزون فى عمليات التطهير والتعقيم للأسطح المختلفة وكذا يمكن استخدامه مع مواد التعبئة والتغليف و فى مخازن التخزين. ومن ناحية اخرى يمكن استخدام غاز الاوزون فى تطهير البيض قبل إجراء عمليات التعبئة والتغليف.

ومن الامور الشائعة استخدام غاز الاوزون فى مبردات اللحوم لمنع نمو أي ميكروبات من شأنها احداث الفساد او التلف مما يزيد من مدة حفظ تخزين اللحوم والاسماك. وهذا افضل من استخدام لمبات الاشعة فوق البنفسجية التى يترتب عليها احداث تزنخ فى الدهون المتواجدة فى اللحوم و الاسماك.

جدول يوضح استخدام الاوزون لاطالة مدة الحفظ مع بعض منتجات الاسماك واللحوم وشروط التخزين

Food	Extension	Storage Conditions
Fish	50-80%	ozone sterilized ice
Salmon	50%	ozone sterilized ice
Jack Mackerel and Shimaaji (fish)	1.2 - 1.6 days	Soak in 30% NaCl cont. 0.6 mg/L O_3 30-60 min. every 2 days.
Beef (frozen)	30-40%	0.4°C; 85-90% RH; 10-20 mg/m^3 O_3, provided original microbial count is below 10^3/cm^2
Poultry	2.4 days	Soak in ice water while passing in O_3 (3.88 mg/L) 20 min.
Bananas	substantial	A few ppm O_3 @ 12°C, if fruit is not within a few days of its period of rapid ripening.
Strawberries, Raspberries, Currents, Grapes	100%	2-3 ppm O_3, continuously or several hours each day.
Apples	several	1.95 cm^3 O_3/m^3
Potatoes	6 months	3 mg/L O_3; 6-14°C; 93-97% RH
Eggs	8 months	0.6 ppm O_3; 31°F; 90% RH
Cheeses	63 days	0.2 - 0.3 ppm O_3

Use of Ozone to Improve the Safety of Fresh Fruits and Vegetables

Ozone can replace traditional sanitizing agents such as chlorine and provide other benefits in the washing, sanitizing, and storage of produce

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In recent years, increasing attention has been focused on the safety of fruits and vegetables, and in particular on the intervention methods to reduce and eliminate human pathogens from fresh produce.

Traditional technology utilizes water with or without a sanitizing agent to wash fresh fruits and vegetables. Chlorine is the most widely used sanitizing agent available for fresh produce, but it has a limited effect in killing bacteria on fruit and vegetable surfaces. The most that can be expected at permitted concentrations is a 1- to 2-log population reduction (Sapers, 1998). Furthermore, the environmental and health communities have expressed concerns about the residual by-products of chlorine.

An alternative treatment is being sought to improve food safety. Research and commercial applications have verified that ozone can replace traditional sanitizing agents and provide other benefits (Bott, 1991; Cena, 1998; Graham, 1997). Many research and industrial trials are underway to validate the use of ozone in the produce industry. Several meetings on this topic have been sponsored by the Electric Power Research Institute (EPRI), including a "Conference on Ozone for Processing Fresh-Cut Fruit and Vegetables" in April 1998 and an "Ozone Workshop" in May 1998. The produce industry is very interested in this technology. However, many questions still have not been resolved, since experience in commercial application in the United States is lacking (Graham, 1997).

Seeking an Alternative to Traditional Sanitizers

In the past two decades, the consumption of fresh fruits and vegetables in the U.S. has dramatically increased. In the meantime, the incidence of foodborne illness due to food pathogens, chemicals, and wastewater has greatly increased. This has been drawing significant public and government attention.

The number of produce-associated foodborne disease outbreaks and the number of cases of illness due to food pathogens have significantly increased in recent years (Tauxe et al., 1997). Moreover, losses in the fresh produce industry that are attributable to microbial spoilage between the time of harvest and consumption are estimated to be as high as 30% (Beuchat, 1991).

Chlorine is commonly used in the fresh fruit and vegetable industry to improve microbiological quality and control pathogens. However, many research studies have indicated that it is limited in its ability to kill bacteria on fruit and vegetable surfaces (Bott, 1991; Cena, 1998; Graham, 1997; Rice et al., 1982; Sapers, 1998). Environmental and health organizations have expressed concerns with traditional sanitizing agents with respect to the formation of by-products, such as trihalomethanes (THMs) and other chemical residues formed in the wastewater returned to the environment (Anonymous, 1998; Cena, 1998; EPRI, 1997; Graham, 1997). The produce industry is concerned about the possibility of future regulatory constraints on the use of chlorine as a sanitation agent.

الخضروات والفاكهة:

- يستخدم غاز الاوزون خاصة الذائب فى الماء فى عمليات غسيل الخضروات و الفاكهة كبديل للهيبوكلوريد ويعتبر الاوزون اكثر كفاءة وفاعلية حيث يستطيع التخلل بسرعة الى الانسجة المختلفة للخضروات والفاكهة . وتم استخدام هذا مع البطاطس والفراولة ويمكن استخدام غاز الاوزون فى تعديل جو المخازن بغرض القضاء على البكتريا او فطريات المخازن والتى تظهر اثناء التخزين مما يمكنا من اطالة مدة الحفظ والحفاظ على الخضروات والفاكهة من الفساد الناتج عن الكائنات الحية الدقيقة.
- من الامور الهامة عند استخدام غاز الاوزون فى معاملة الخضروات و الفاكهة مراعاة الجرعة المستخدمة& زمن التعرض حيث تعتبر الخضروات والفاكهة من الاغذية الحساسة للمعاملة بالأوزون نتيجة المحتوى العالى من الفيتامينات والعناصر الغذائية الهامة لذا معظم المعاملات تكون بجرعات بسيطة ولفترات تعرض لا تتعدى الدقائق للحفاظ حتى لا يحدث تكسير فى العناصر الغذائية المتواجدة فى الخضروات والفاكهة وكذا المحافظة على اللون الطبيعى المميز لكل نوع منها على حدة.

- **الخضروات والفاكهة:**
- يمكن استخدام غاز الاوزون فى اثناء تخزين الخضروات و الفاكهة على النحو التالى:
- اثناء تخزين البطاطس والبصل ويستطيع ايقاف عمليات التنبيت & تخزين الموالح& تخزين الفراولة والتوت البرى
- ومن الامور الشائعة استخدام الاوزون الذائب فى الماء فى عمليات التبريد الاولى التى تجرى لكثير من الخضروات والفاكهة حيث يستطيع غاز الاوزون خفض درجة حرارة الماء ومن ناحية القضاء على أى ميكروبات محتمل تواجدها .

OZONE IN FRUITS & VEGETABLES

- Act as **effective disinfectant** for fresh vegetables and fruits.
- **Greater than 90% reduction** of total bacterial counts in **Chinese cabbages** (Kondo).
- **Suppressed fungal development** for 12 days and didn't show any **observable injury** in blackberries.

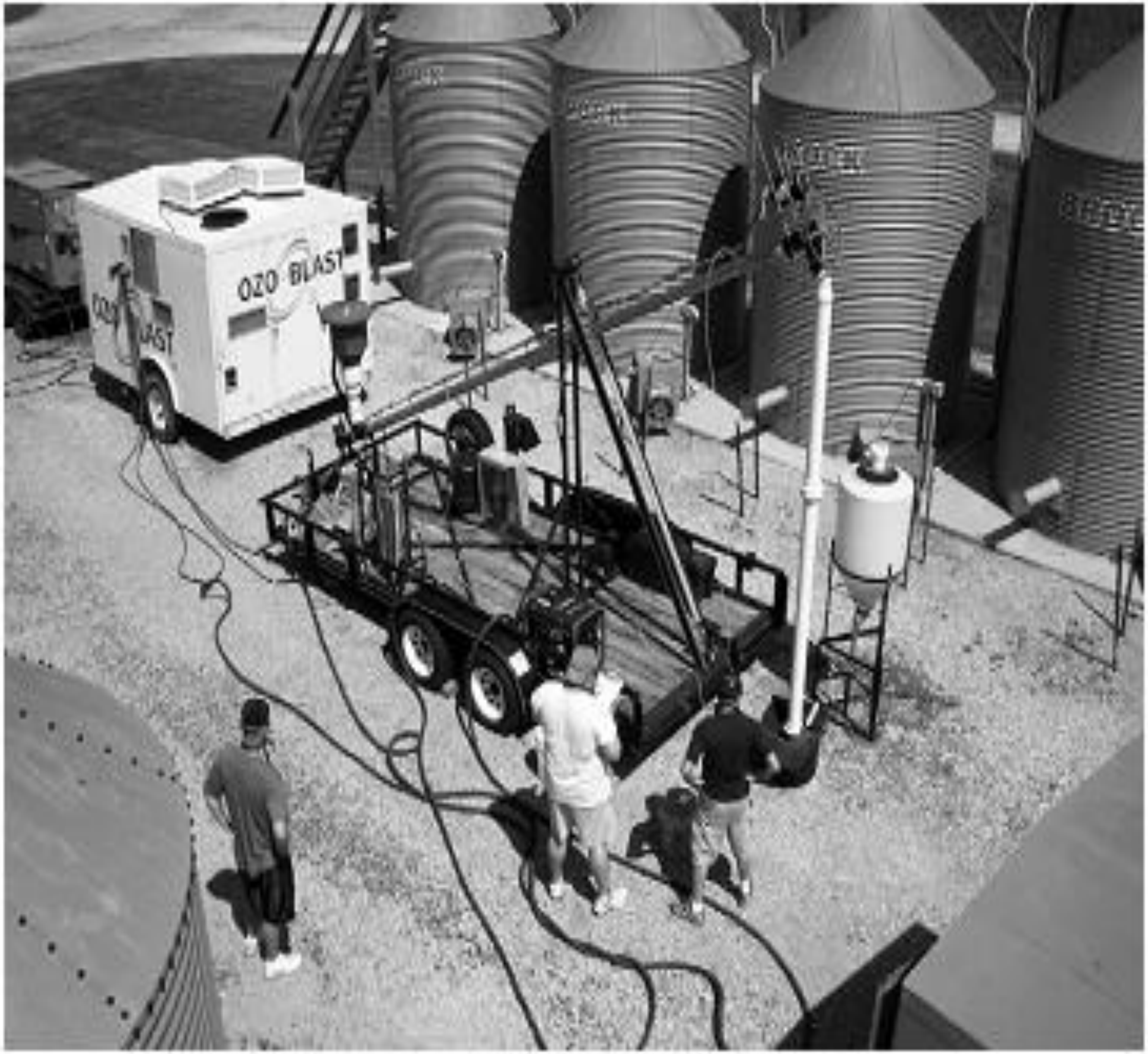
الحبوب:

- تعتبر الحبوب من انسب المواد الغذائية لتطبيقات استخدام غاز الاوزون ويستخدم غاز الاوزون مع الحبوب لعدة اغراض منها
- القضاء على السموم الفطرية و الفطريات المتواجدة
- ايقاف عمليات التنبيت اثناء التخزين
- القضاء على الحشرات
- تعقيم المخازن و الصوامع
- استخدام الاوزون الذائب فى الماء اثناء عمليات تعديل الرطوبة (التنميش) التى تجرى للحبوب قبل الطحن
- استبدال المبيدات الحشرية أو المضادات الفطرية الشائعة الاستخدام أثناء تخزين حبوب القمح بالمعاملة بالأوزون نظرا لأن المعاملة بالأوزون لا ينتج عنها وجود أي متبقيات فى حبوب القمح عكس استخدام المبيدات الحشرية أو المضادات الفطرية.
- ويستطيع غاز الاوزون القضاء تماما على الافلاتوكسينات المتواجدة فى

الحبوب:

- وتعتبر المعاملة بالأوزون (Ozonation) أحد هذه الطرق التى تم مؤخرا وضعها كطريقه مؤكسدة للتخلص من الافلاتوكسين فى الأغذية والتى تعتمد على أن الأوزون يتفاعل مع الرابطة المزدوجة بين ذرتي الكربون رقم ٨ و ٩ فى حلقه الفيوران مكونا مركب ابتدائي سريعا ما يتحول الى مركب **ozonide** والذى بدورة يتحول فى النهاية الى الدهيدات وكيتونات وأحماض عضوية وثانى اكسيد الكربون.

وبالاضافة الى ما سبق أظهرت نتائج الابحاث التى تمت فى هذا الصدد زيادة كفاءة وفاعلية المعاملة بالأوزون مع زيادة المحتوى الرطوبى لحبوب القمح بعد عملية التنميش. حيث أن المحتوى العالى من الرطوبة يزيد من نفاذ غاز الاوزون للحبة كما ان الشقوق أو الشوارد الحرة مثل مجموعه الهيدروكسيل (OH) الناتجة بعد تفاعل الماء الموجودة فى الحبه مع الأوزون تزيد من فاعليه غاز الأوزون.


OZO BLAST

OZONE AND MILLING

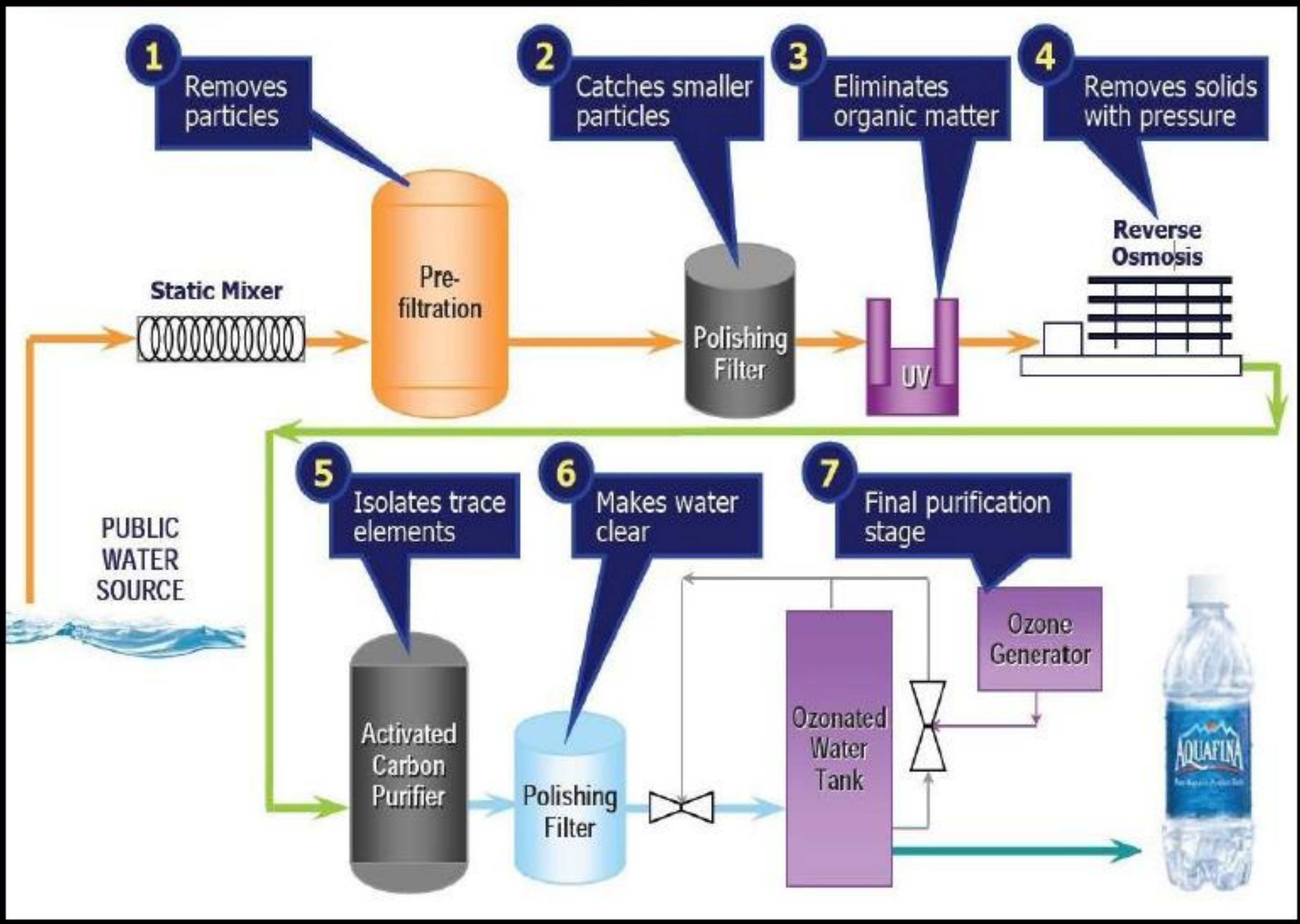
- Aqueous ozone - **grain tempering process to inactivate mold and bacteria.**
- Ozone gas - **surface sanitation of enclosed equipment and conveyors and transport equipment** as an antimicrobial intervention.
- **About 45-55% reduction in bacteria and mold** using ozone in tempering process as compared to normal water use.
- Resulting into **increase in shelf life** of milled product.

Effect of ozone on bacterial attack in wheat tempering process

Particular	Reduction in bacteria (%)	Reduction in mold (%)
Soft wheat water tempered	13	6.2
Soft wheat ozone tempered	45.65	49.42
Hard wheat water tempered	7.27	5.4
Hard wheat ozone tempered	42.27	47.39

BOTTLED WATER PRODUCTION

- Ozone - Initially used in the US for water treatment.
- In 1982 – approval (GRAS) for use in bottled water.
- Today, the majority of bottled water companies use ozone to ensure pure water.
- Removes most of the bacteria from water without any residue.
- Oxidizes iron, manganese and sulpher based compounds in water.
- Removes color and odor of water.
- The International Bottled Water Association (IBWA) suggests that, a residual ozone level of 0.2 to 0.4 ppm which also disinfects both the water and the bottle.



Bottled water production with ozonation

SURFACE SANITIZATION

- In food processing - **important to provide pathogen free food.**
- So, for that, reducing the contamination of potentially deadly pathogens is very crucial.
- **Equipment, walls, floors, drains, tanks, tubes, racks, knives and tables** can all be sprayed with aqueous ozone.
- **Enclosed piping** – sanitized **with ozone** spraying.

- Sanitization is two step process-
- In first step - **bio-films are removed with a hot water** cleaning.
- Then **aqueous ozone is used to sanitize the surface** destroying all bacteria, viruses, fungi, and spores.

Effectiveness of ozone with different materials

Surface material	Percent reduction in plate count
Stainless Steel Tabletop	98 - 99
Plastic Shipping Containers	95 - 97
Stainless Steel Kettle	89 - 96

STORAGE AND PACKAGING

- Storage rooms are treated with ozone gas in order to remove the micro-organisms.
- Ozonization - refrigerated meat destroys micro-organisms of pseudomonas family.
- Ozone prevents ethylene formation, retards ripening of fruits and spoilage by micro organisms.
- Sometimes, raw food products are stored in an atmosphere containing gaseous ozone.

  e.g. onions and potatoes

- Packaging materials are also **disinfected using ozone and UV radiation.**
- In air-tight sealed plastic bags, **ozone treatment is followed by carbon dioxide or nitrogen.**
- Since ozone will not leave any toxic residue, ozone surface disinfection can apply for products and **packing materials like container, bottles, pouches etc.**

Extension of Storage Life With Ozone

Food type	Extension of life	Storage Conditions
Fish	50-80%	With ozone sterilized ice
Poultry	2.5 days	When soak in ozonated water (3.88 mg/L) 20 min.
Strawberries and Grapes	Double	Spraying of (2-3 ppm) aqueous ozone for 5 - 6 hours per day.
Potatoes	6 months	3 ppm O_3 at 6-14°C and 93-97% RH
Eggs	8 months	0.6 ppm O_3 at -0.5°C and 90% RH

استخدامات اخرى لغاز الاوزون

- يستخدم غاز الاوزون فى العديد من العمليات الصناعية التى تتطلب عمليات أكسدة . كما يستخدم كمادة مبيضة تستخدم لتبيض المركبات العضوية مثل الشموع والزيوت. يستخدم غاز الاوزون فى ازالة الروائح الكرية من بعض المواد الغذائية.
- يستخدم الأوزون في تعقيم وتكرير المياه ومعالجة مياه الشرب؛ حيث وجد أنه أسرع من الكلور ٣٢٠٠ مرة في قتل البكتريا والفيروسات، فضلا عنٍ الفطريات والطفيليات، وبدون أي آثار جانبية. والأوزون يعد عاملا منظفًا للبيئة، لكن زيادة نسبته عن الحد المسموح به تحوله إلى عنصر ضار ومتلف ومدمر لها.

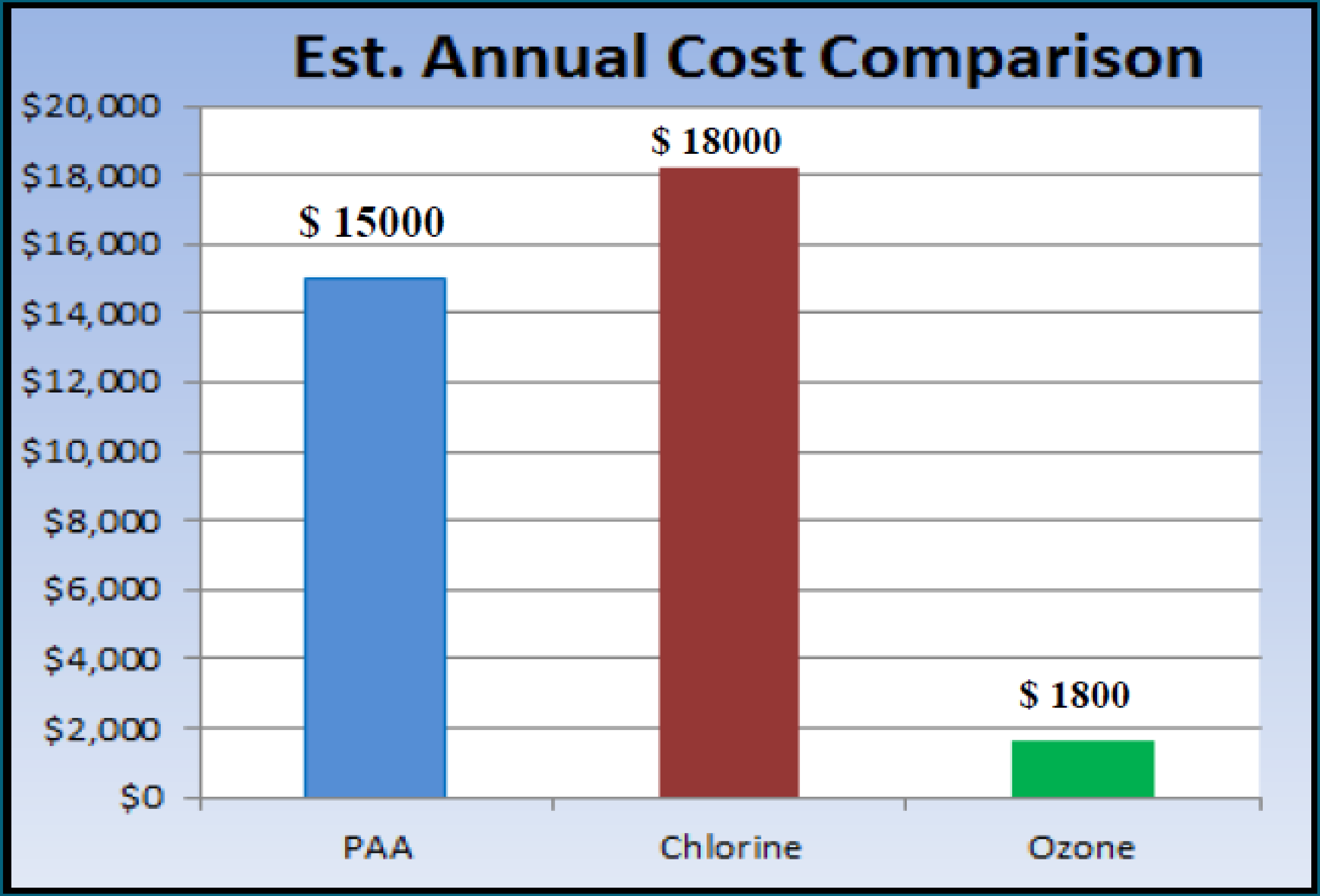
ADVANTAGES OF OZONE

- Ozone oxidation is nature friendly process .
- Highly effective in cold water, which saves energy & cost.
- Significant reduction in processing time.
- Effective against wide range of harmful microorganisms.
- Dissolves metallic substances like iron and manganese.
- Removes unwanted color, taste, and odor of product.

DISADVANTAGES OF OZONE

- Installation of ozone technology unit is complicated.
- May produce undesirable aldehydes and ketones by reacting with certain organics.
- Less soluble in water than chlorine; thus special mixing devices are necessary.
- In light, it may degrades the material.
- The initial investment is more as compared to other chemicals.

COST ANALYSIS OF OZONE TECHNOLOGY



CONCLUSIONS

From the above study it is concluded that,

- Ozone is a potent sanitizer with promising applications in the modern food industry

- It is environment-friendly disinfectant.

- There is a considerable difference (about 10 times less) in operational cost with the use of ozone as compared to other chemical sanitizers.

- It is the most likely alternative to chlorine and hydrogen peroxide etc. in food applications.

Environmental & Analytical
Toxicology

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http://dx.doi.org/10.4172/2161-0525.1000128

Research Article Open Access

Effect of Ozone Gas on Degradation of Aflatoxin B1 and *Aspergillus Flavus* Fungal

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Abstract

Wheat (*Triticum aestivum*) is one of the most important agricultural crops. Requirements, wheat must be produced free of hazardous contaminants. However, previous investigations showed that wheat could be contaminated by aflatoxins above the limits that may be critical for health. In this study, use of the high oxidising power of ozone achieved degradation of aflatoxin B_1. Samples were subjected to ozonation at various ozone concentrations (20, 40ppm) and exposure time (5, 10, 15, 20 min). The reduction percentages of aflatoxin B_1 in artificially contaminated wheat 10 µg/kg were 84.1 and 86.75% after exposures to 20 and 40 ppm ozone for 20min respectively, and 86.7 and 96.66 % with 20 µg/kg after exposures 20 and 40 ppm ozone for 20 min, respectively. The percentage of inhibition zone from *Aspergillus flavus* was 46.4 % to 87.8 % after ozonation using concentration 20 ppm for 5 and 20 min respectively. While with 40 ppm ozone inhibition zone was 65.6% and 95.6 % with 5 and 20 min respectively. Exposure of strain to 20 ppm ozone inhibition of product of AFB_1 percentage of 40.94%, 52.5%, 59.32% and 60.4 % for exposure time 5, 10, 15 and 20 min, respectively. However, observed increase on inhibition of AFB_1 with topping up ozone dose to 40 ppm attained 55.2%, 64%, 74.5% and 77.2% with 5, 10, 15 and 20 min exposure time respectively.

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Evaluation of ozone gas as an anti-aflatoxin B1 in wheat grains during storage

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Abstract

Wheat (*Triticum aestivum*) is one of the most important agricultural crops. Egypt has one of the highest wheat per capita consumption levels in the world (108 kg/person/year).Wheat must be produced free of hazardous contaminants. However, previous investigations showed that wheat could be contaminated by aflatoxins above the limits that may be critical for health. In post-harvest situations, crop spoilage, fungal growth, and mycotoxin formation result from the interaction of several factors in the storage environment.

In this study, use of ozone gas achieved as an anti-aflatoxin B1 in wheat grains during storage. Wheat samples were artificially infected with spores count of *Aspergillus flavus* ATCC 28542 strain (105, 104, 103,102 and 101 spores/kg) and ozonation at 20 and 40 ppm ozone gas for 5, 10, 15 and 20 min at room temperature.

We not observed any amount of AFB1 in wheat samples were ozonation for 10, 15 and 20 min with spores count of *Aspergillus flavus* strain 105 to 101 (spores/kg), But AFB1 was formed in samples treated for 5 min at 20 and 40 ppm ozone gas with artificially infected spores count 105 and 104 (spores/kg) solely. While 5 min of exposure to ozone gas were sufficient inhibition production of AFB1 with the number of spores fewer than 104 spores/kg.

Keywords: Aflatoxin B1; inhibition; Ozone; *Aspergillus flavus*; storage; wheat

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Baratharaj V., Use of ozone in food processing & cold storage:7 -12.

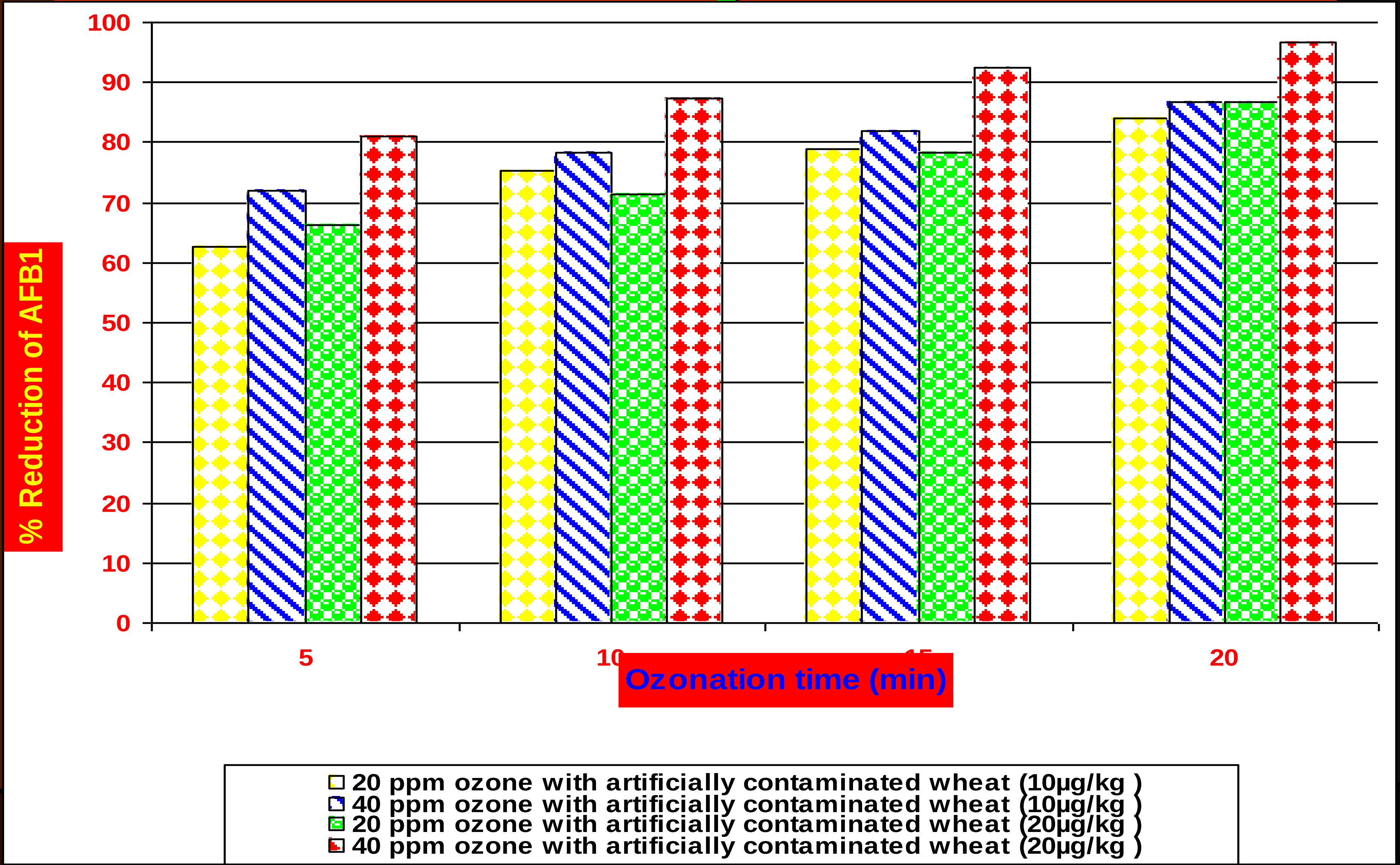
http://www.ozonesolutions.com

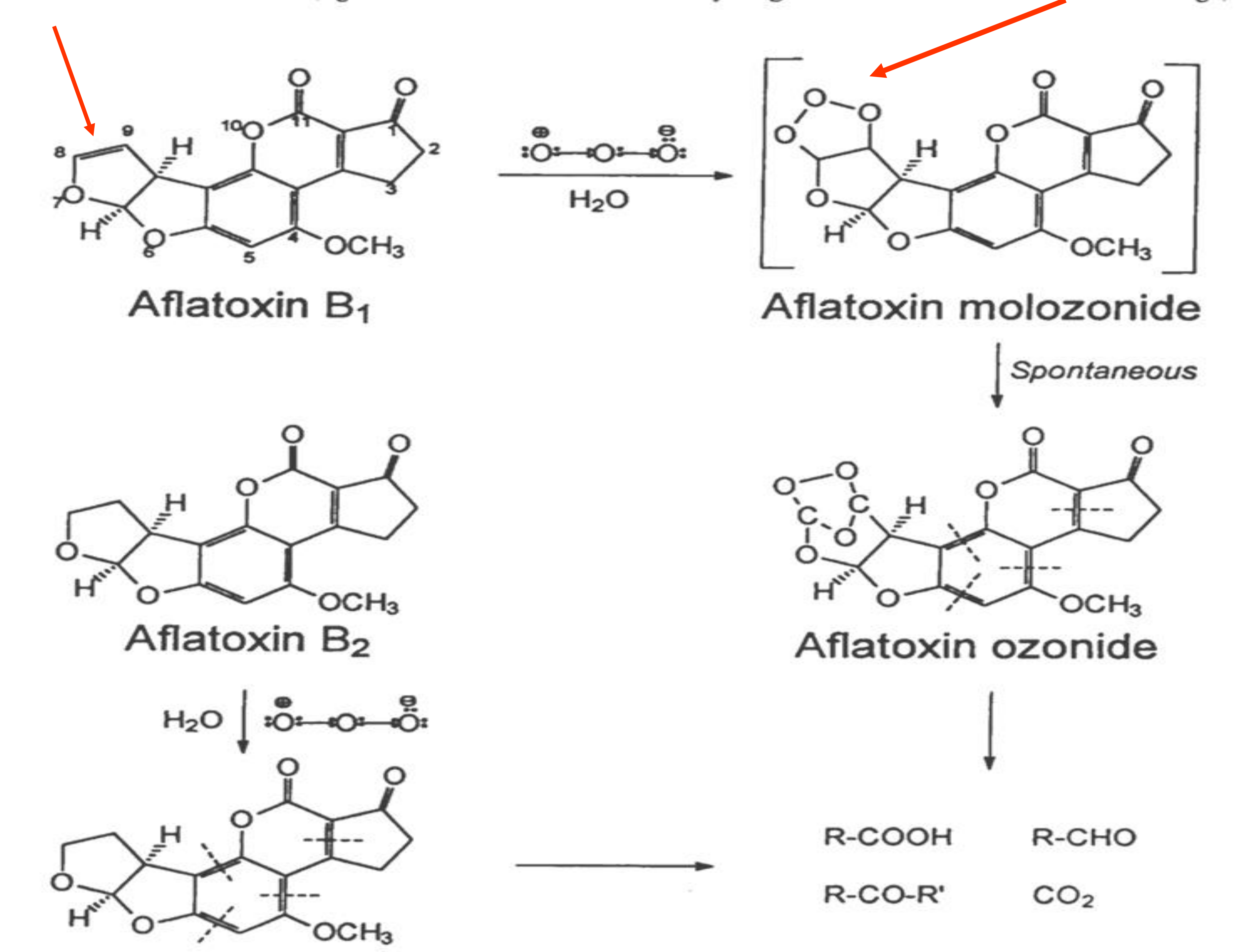
http://www.worldfoodscience.com


كلية الزراعة

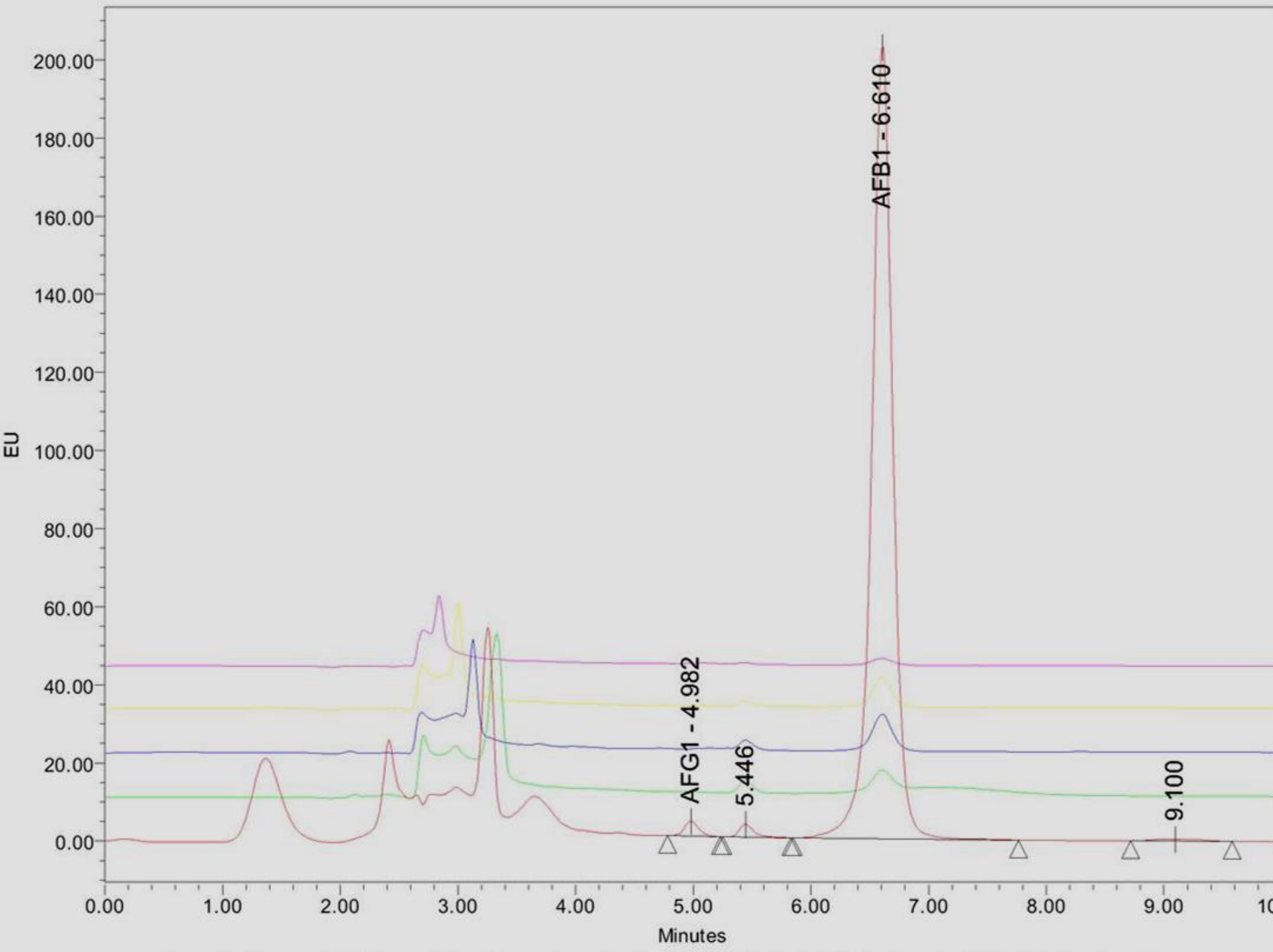
Part I

Study the efficacy of ozone gas in degradation and or decontamination of AFB_1 in artificially contamination wheat grain



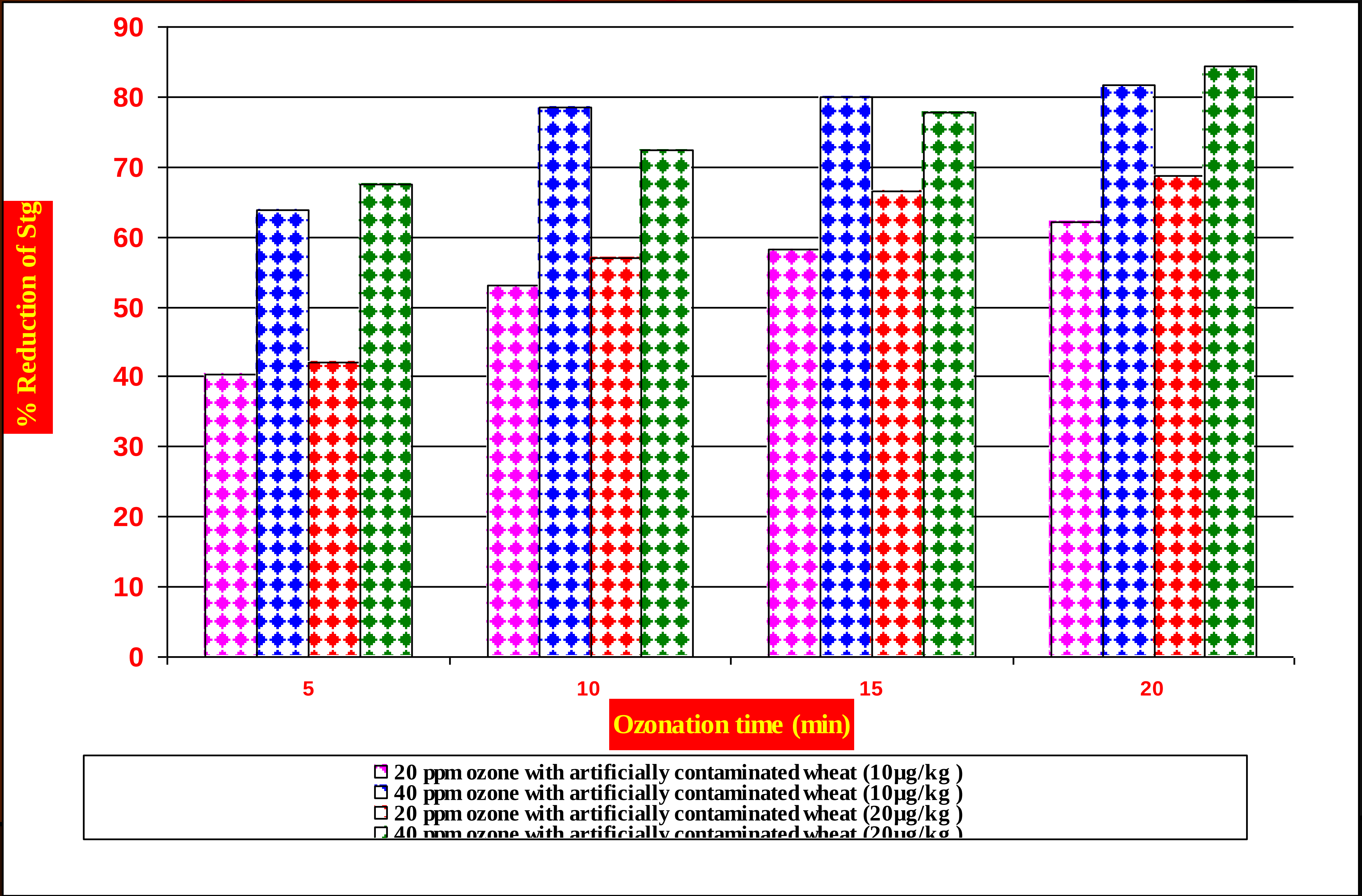


Aflatoxin B1
H2O
Aflatoxin molozonide
Spontaneous
Aflatoxin B2
Aflatoxin ozonide
H2O
OCH3
R-COOH
R-CHO
R-CO-R'
CO2

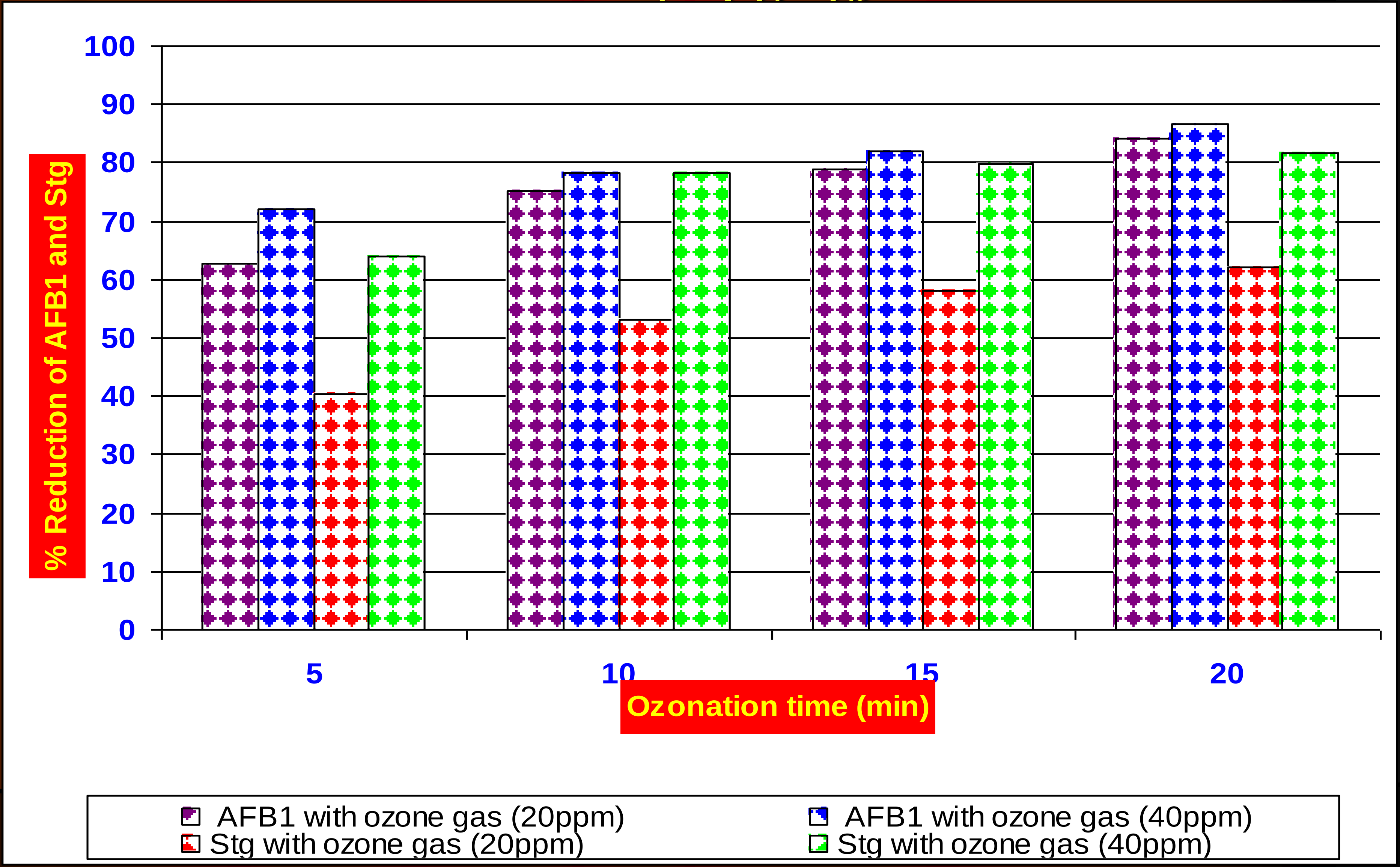


AFB1 - 6.610
AFG1 - 4.982
5.446
9.100
EU
200.00
180.00
160.00
140.00
120.00
100.00
80.00
60.00
40.00
20.00
0.00
0.00
1.00
2.00
3.00
4.00
5.00
6.00
7.00
8.00
9.00
10
Minutes

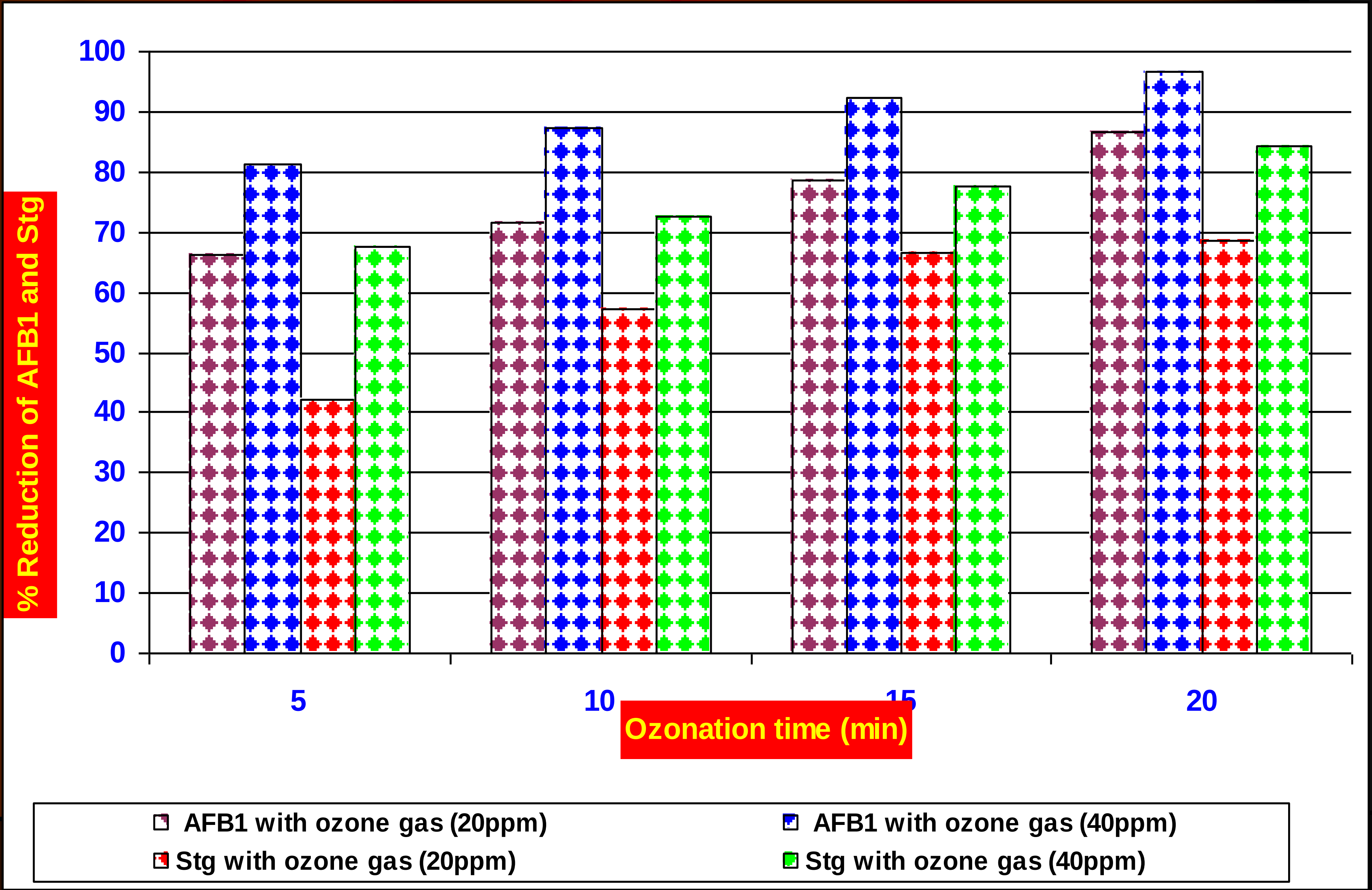
Effect of ozone gas on content of Stg in artificially contaminated wheat grains



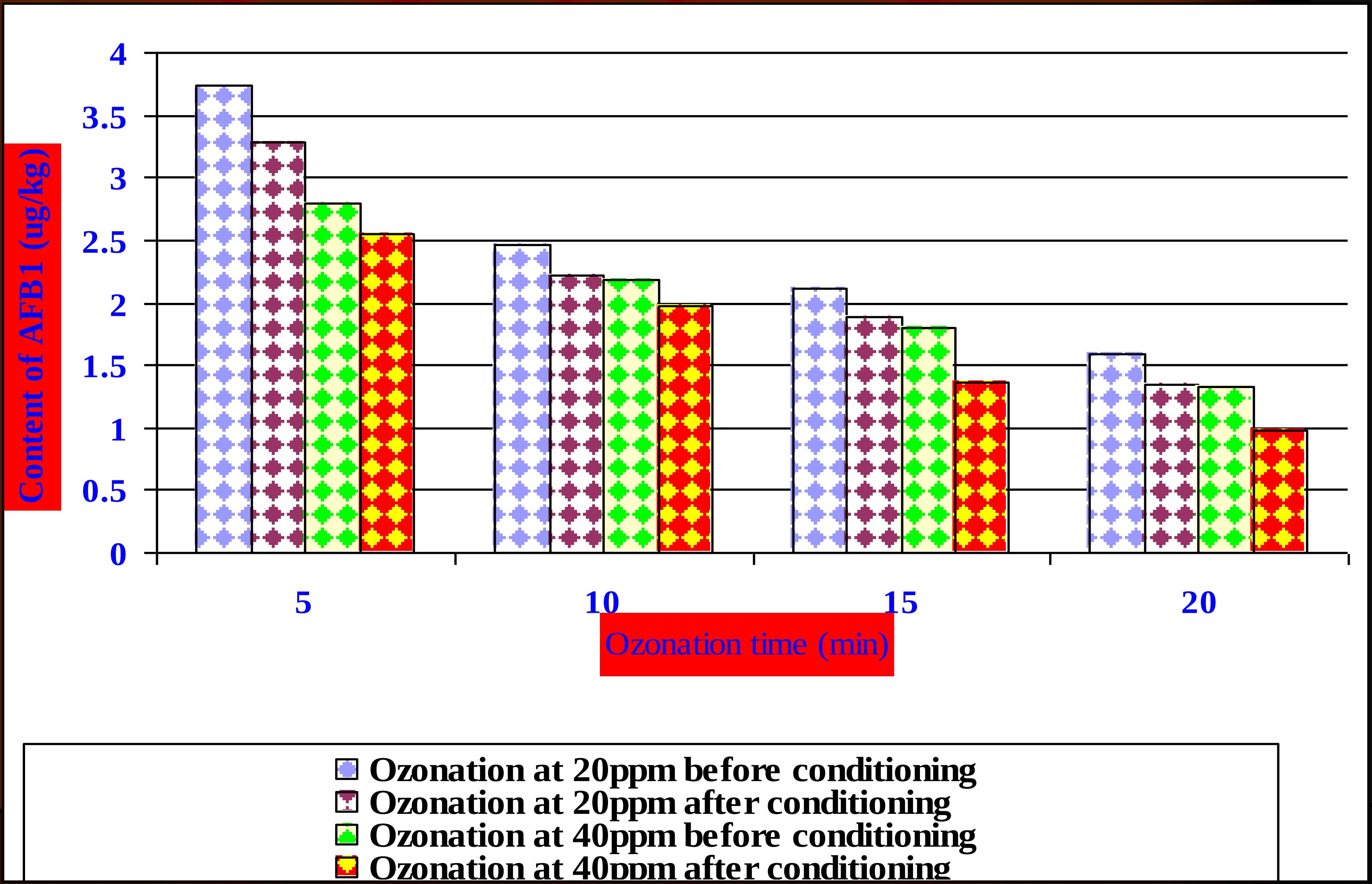
Percentage of reduction AFB_1 and Stg in artificially contaminated wheat (10µg/kg) after ozonation



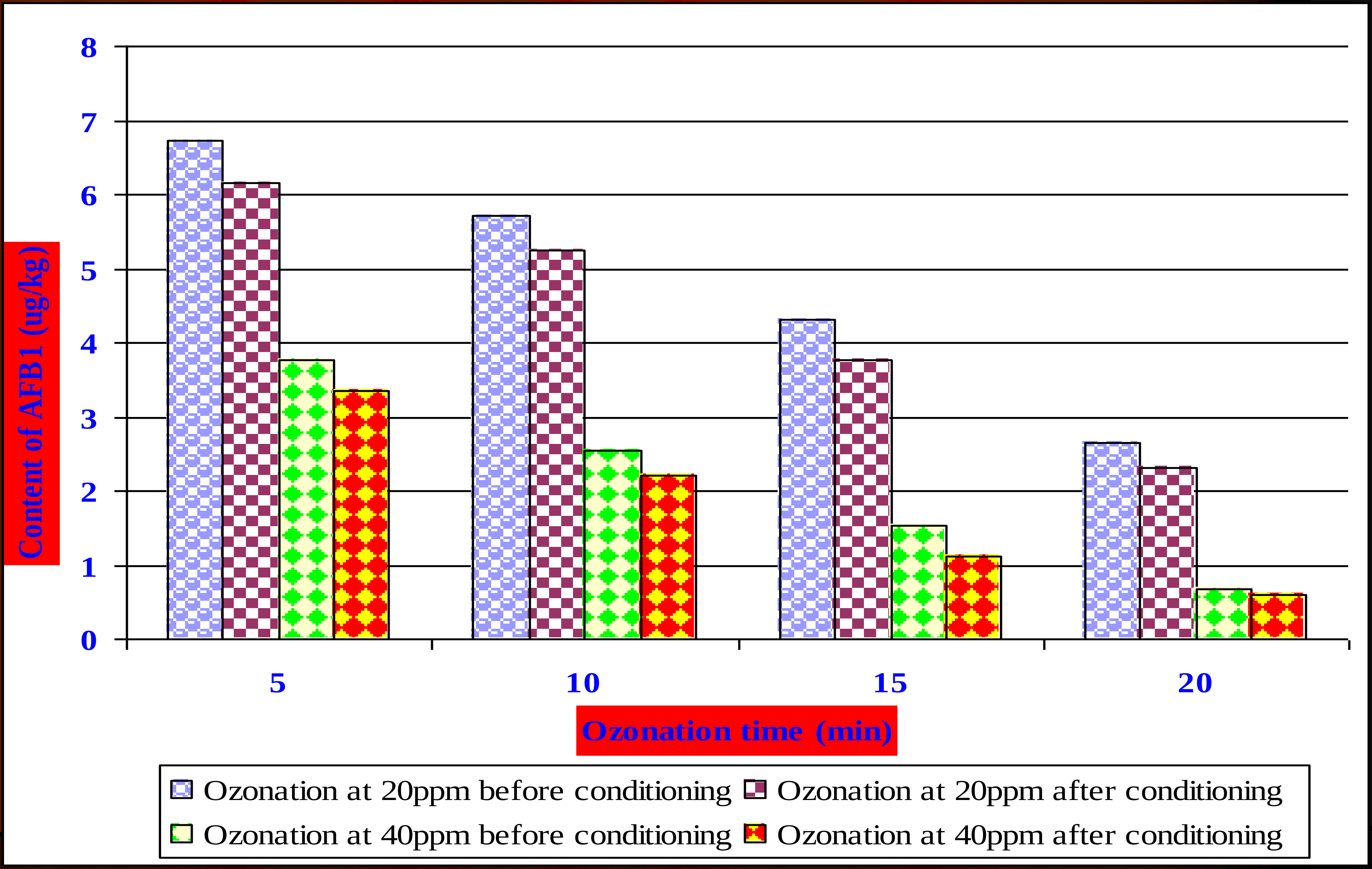
Percentage of reduction AFB1 and Stg in artificially contaminated wheat (20µg/kg) after ozonation.



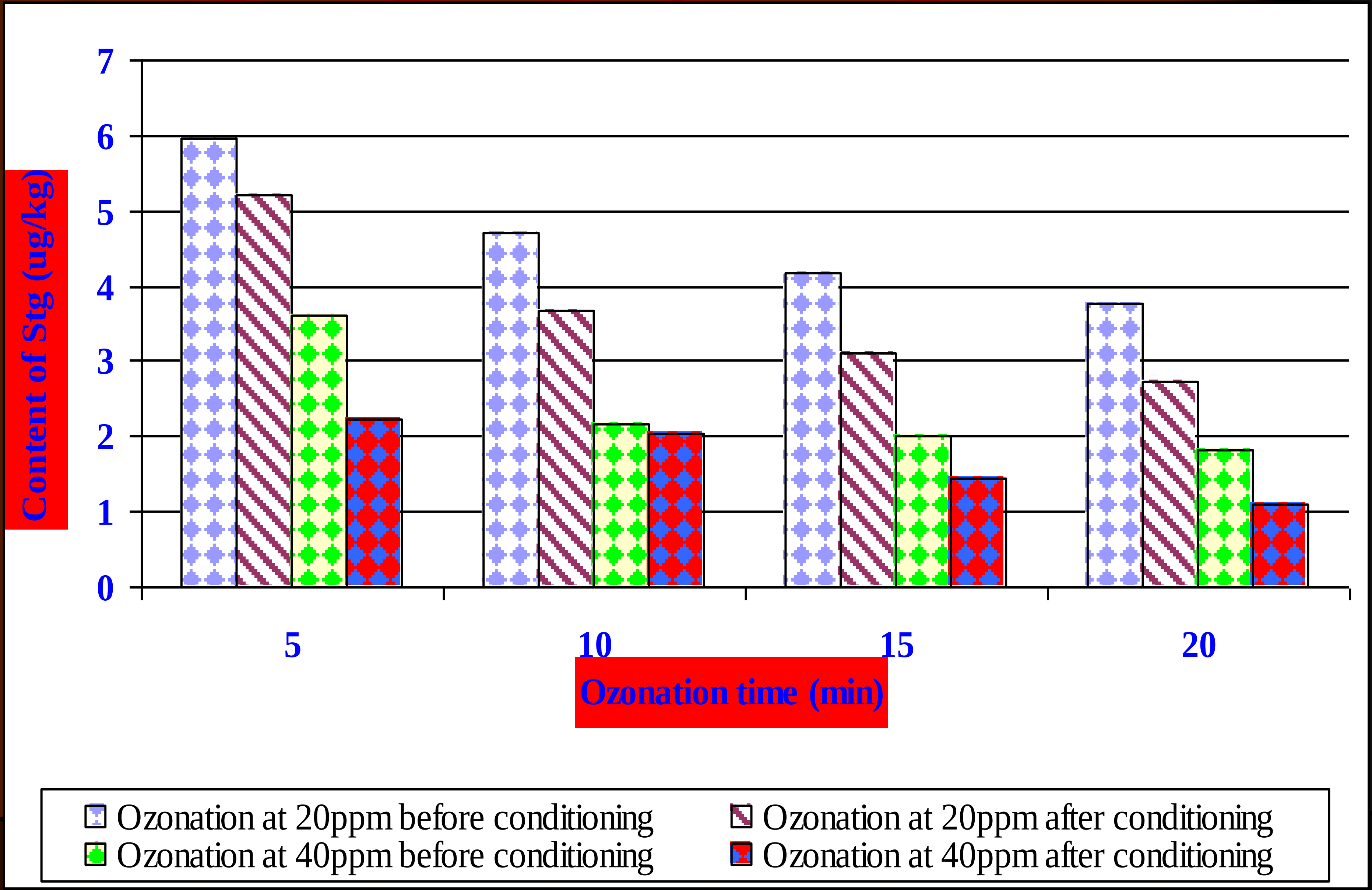
Concentration of AFB_1 in artificially contaminated wheat (10µg/kg) after conditioning and ozonation



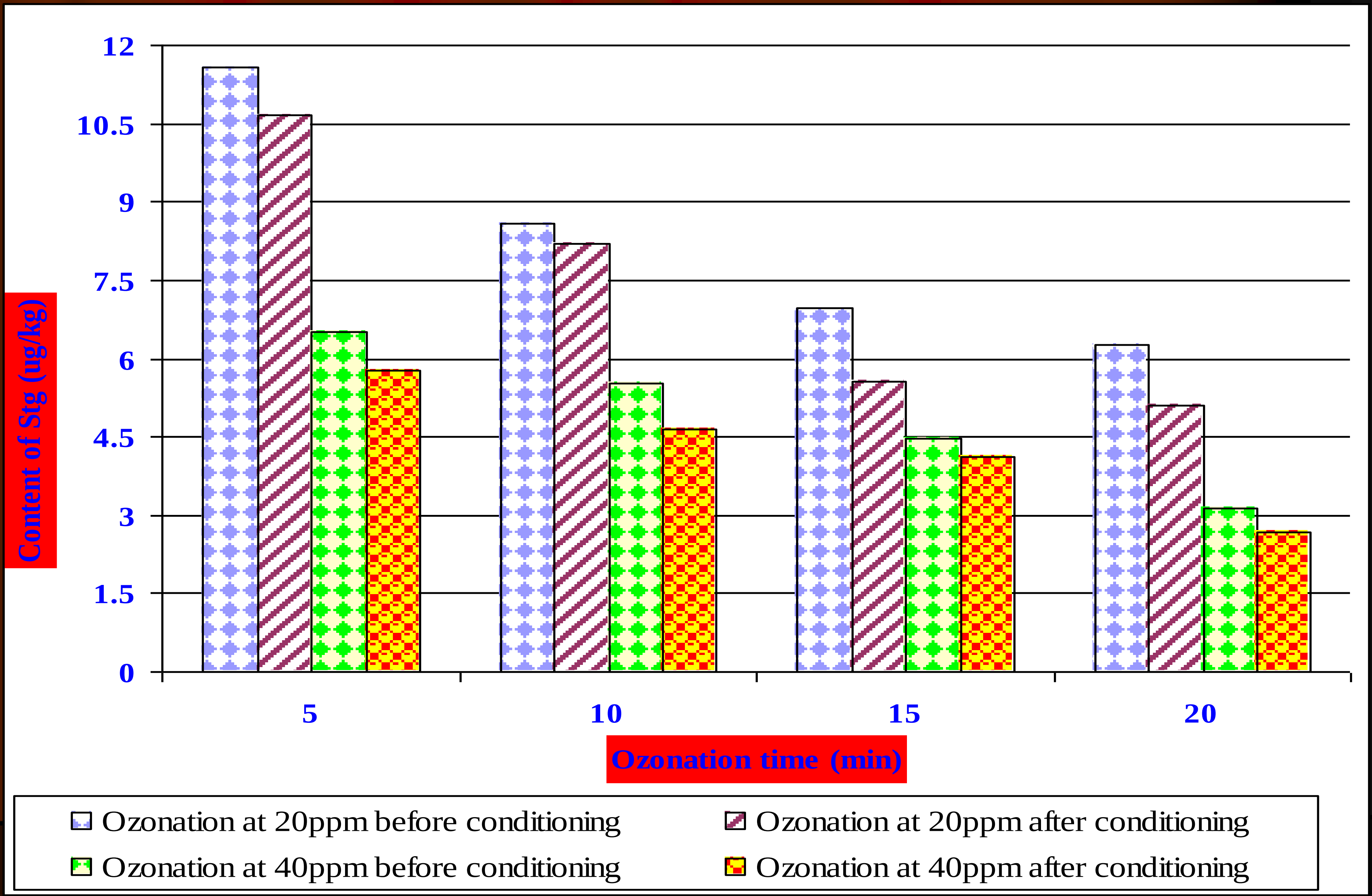
Concentration of AFB1 in artificially contaminated wheat (20µg/kg) after conditioning and ozonation



concentration of Stg in artificially contaminated wheat (10µg/kg) after conditioning and ozonation

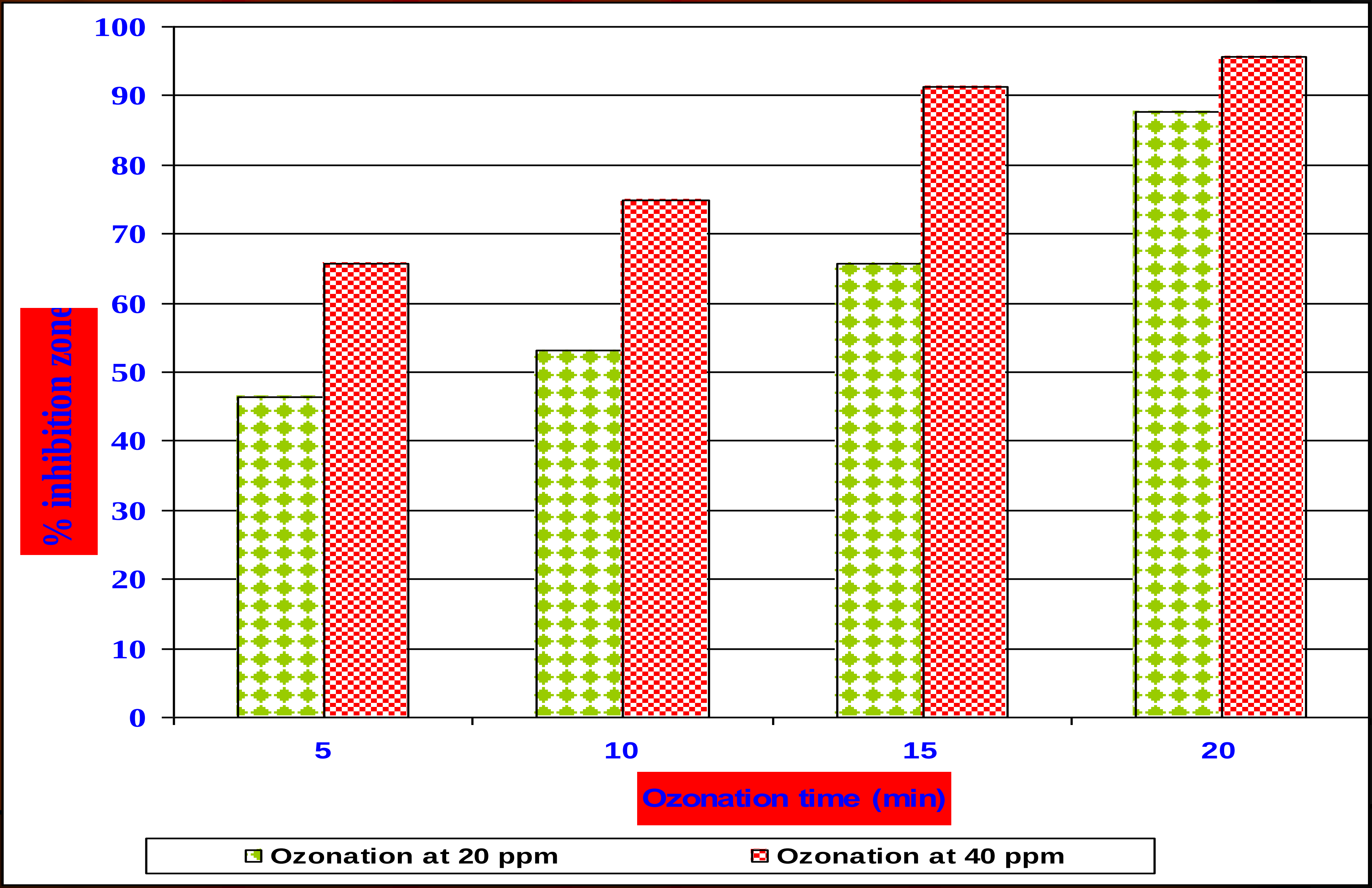


Concentration of Stg in artificially contaminated wheat (20µg/kg) after conditioning and ozonation

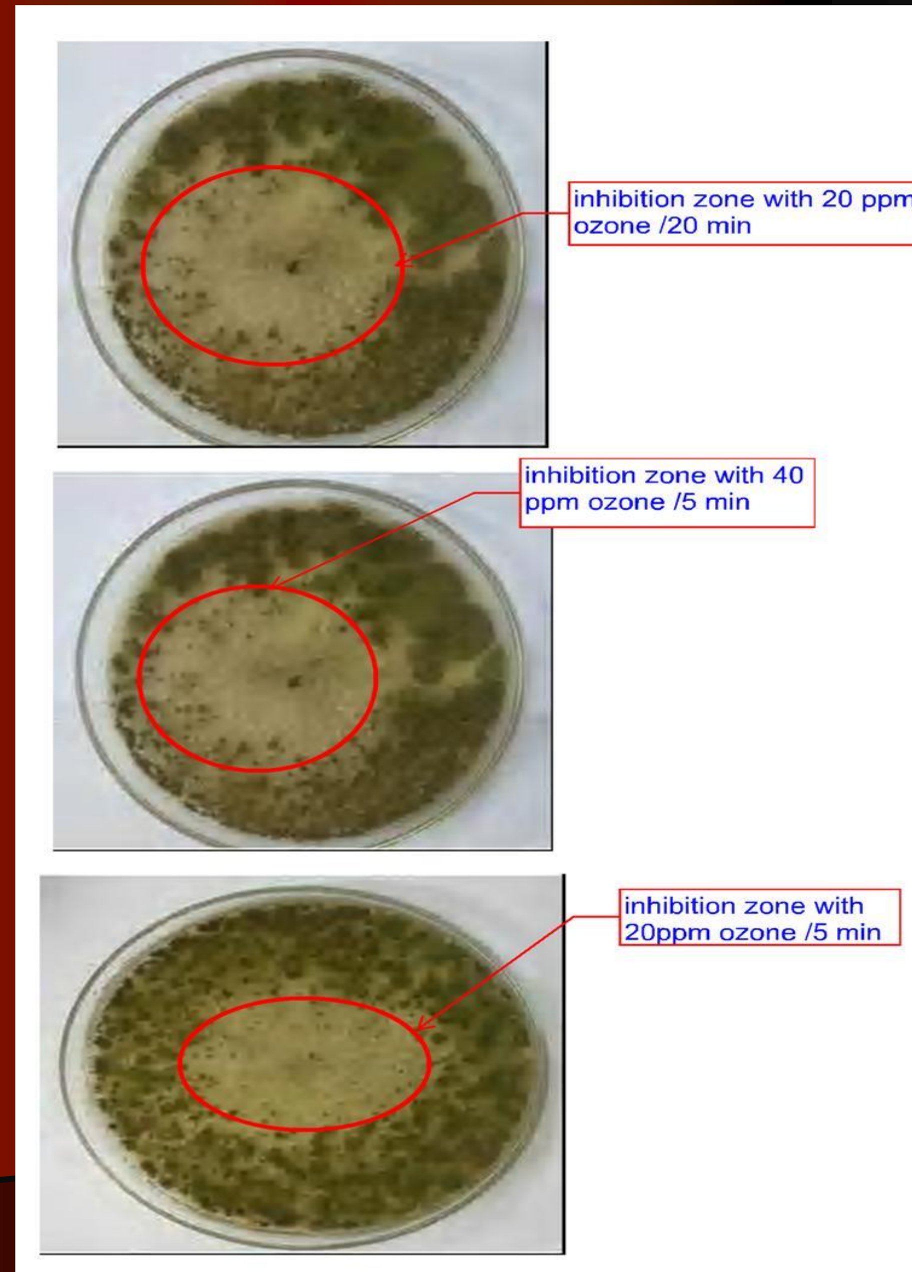


Part II:
Effect of ozonation on growth *Aspergillus flavus* and AFB1 production

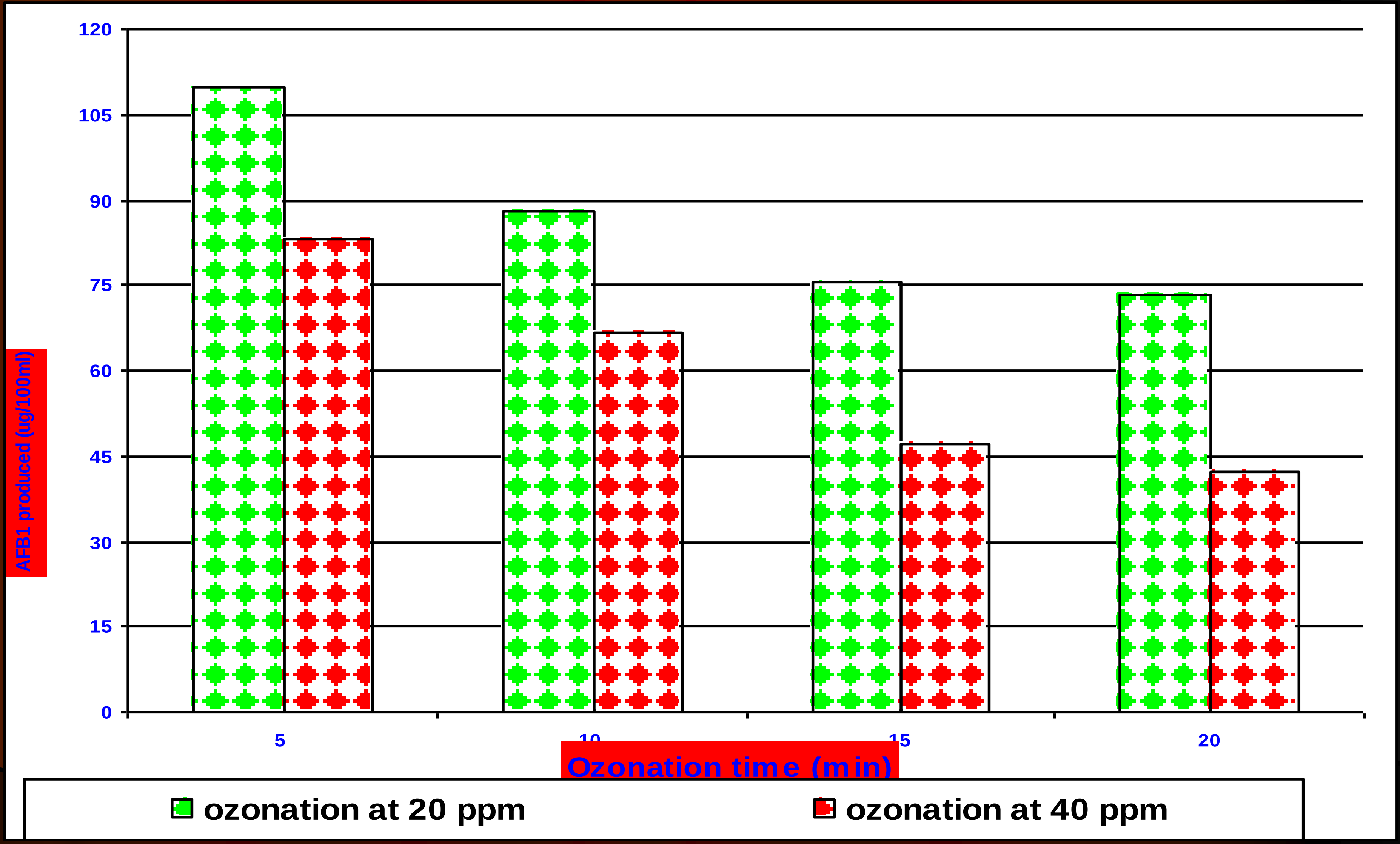
Inhibitory effect of ozone gas against mycelial growth of *Aspergulls flavus*:

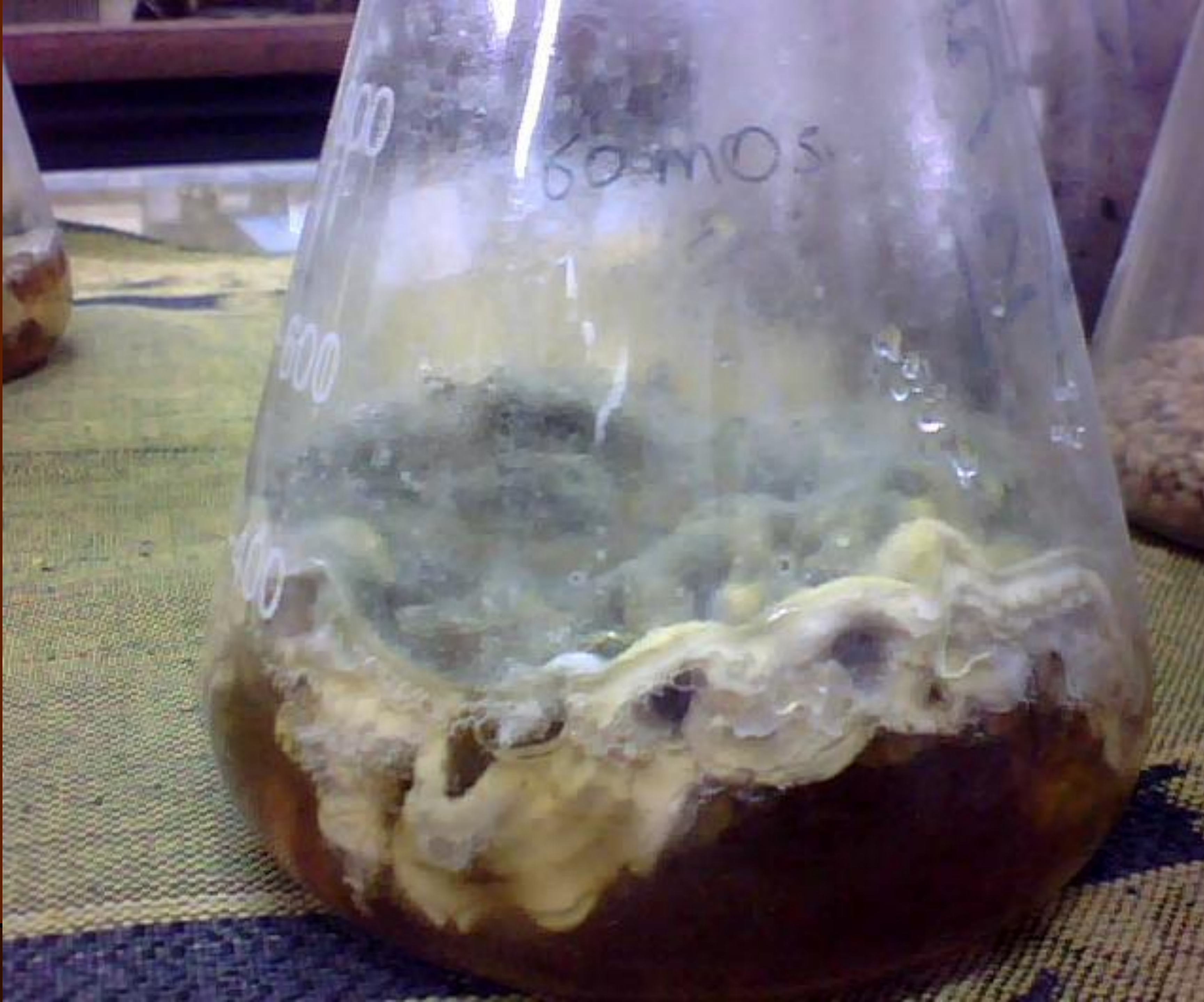


Inhibition of *Aspergillus flavus* growth by ozone gas on PDA media.

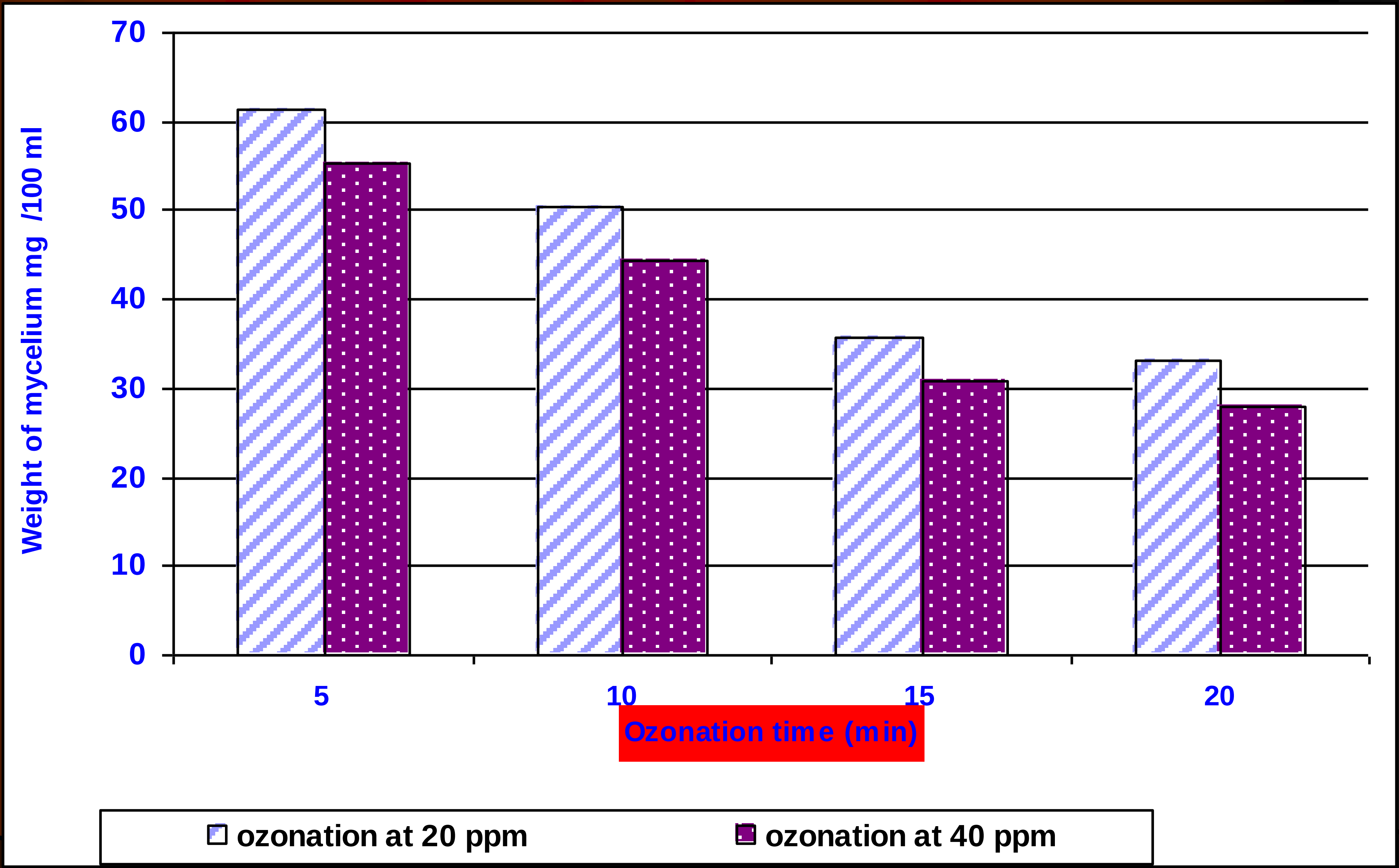


Effect of ozonation on AFB_1 formed by *Aspergillus flavus* in YES media



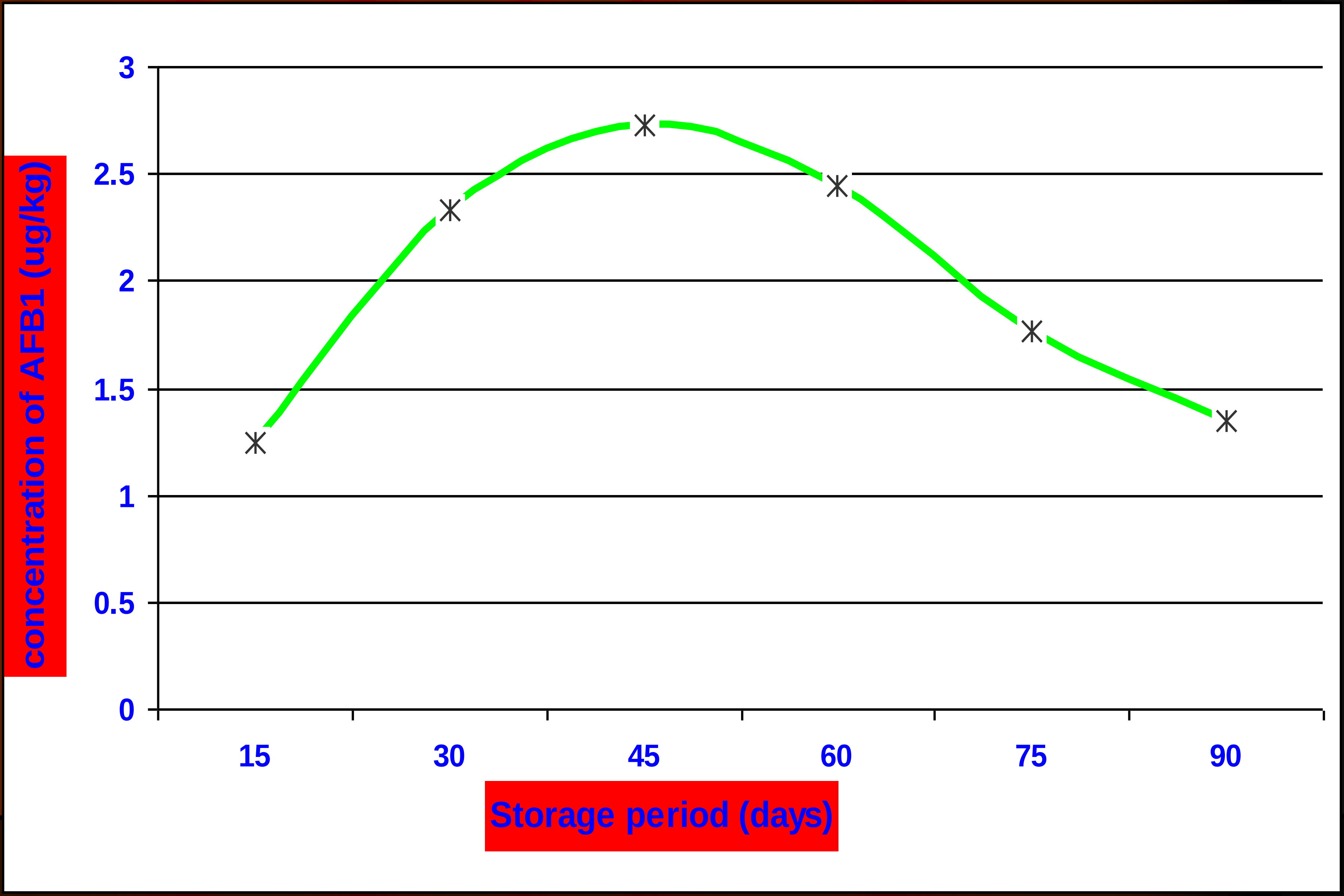


Effect of ozonation on weight of mycelium formed by *Aspergillus flavus* in YES medium.



Effect of ozonation on AFB_1 production in wheat grains during storage

AFB1 in control wheat grain sample during storage.



- **Wheat samples artificially infected with spores of *Aspergillus flavus* ATCC ٢٨٥٤٢ strain (١٠^{5}, 10^{4}, 10^{3},10^{2} and 10^{1} spore/kg) were incubated at 28^{o}C and ozonation at 20 and 40 ppm ozone for 5, 10, 15 and 20 min at room temperature.**
- **We not observed any amount of AFB_1 in wheat samples were ozonation at 20 and 40 ppm for 10, 15 and 20 min with spores of *Aspergillus flavus* count 10^{5} to 10^{1} (spore/kg).**
- **AFB_1 was formed in samples were ozonation for 5 min at 20 and 40 ppm ozone gas with artificially infected spores count 10^{5} and 10^{4} (spore/kg)**


500
APPROX

Thus, fumigation with ozone gas can be a good method for achieving sanitation and decreasing initial microbial load in food storage facilities and aid in curbing spoilage on a long term. However, it is clear from this work that ozone gas exposure time higher than 5 min would be required to achieve complete spore kills. These results were in agreement with other studies.

Part III:

Effect of ozone gas on chemical composition of wheat and rheological properties of dough

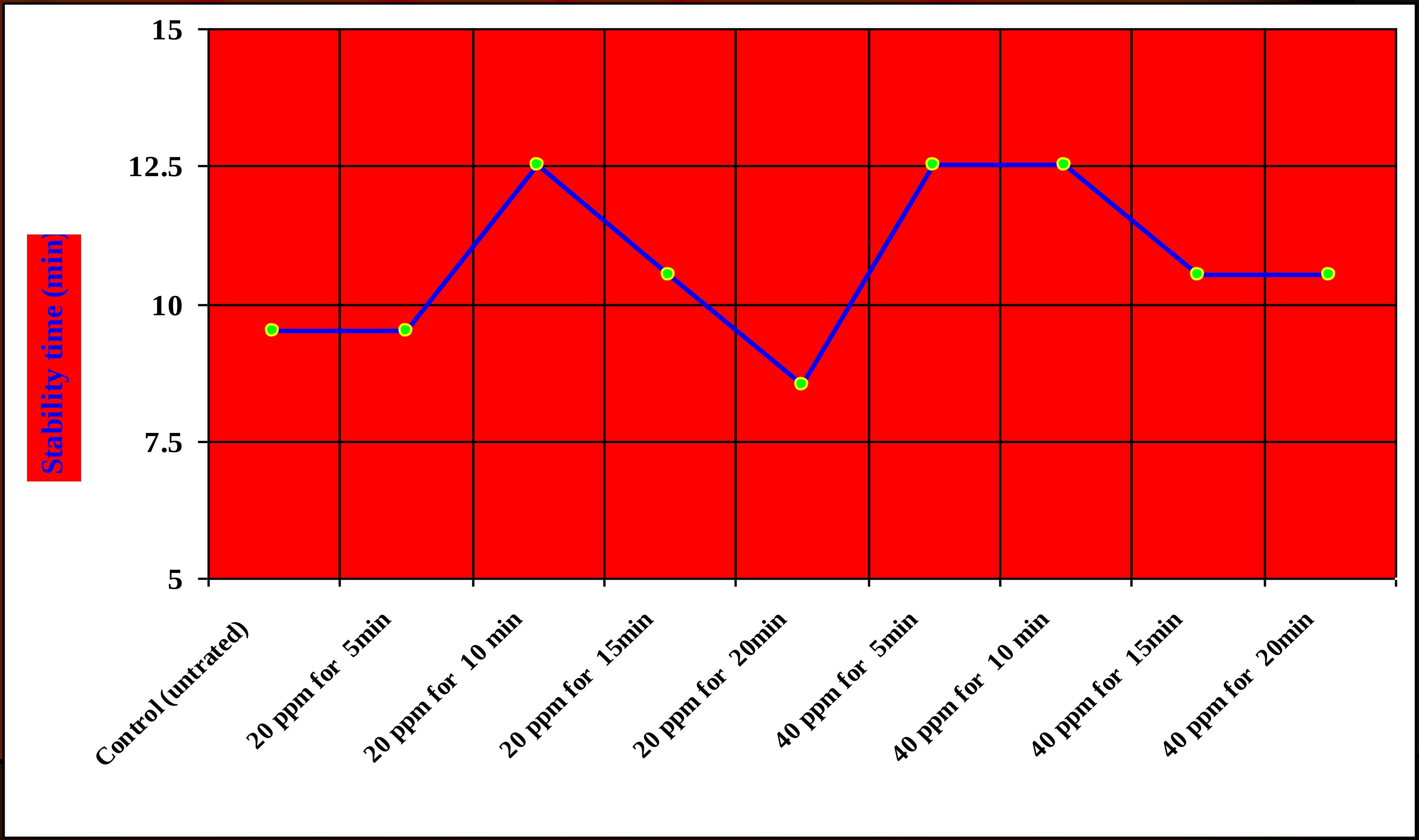
Effect of ozonation on chemical composition of wheat flour

Results indicated that ozonated wheat grain did not significantly alter the chemical of the wheat flour. These results were in agreement with other studies (**Ibanoglu, 2001; Paul *et al.*, 2005 and and Sandhu *et al.*, 2011**).

Effect of ozonation wheat grain on farinogram parameters in wheat flour dough (82% extraction).

Samples	Water absorption %	Arrival time (min	Development time (min)	Stability time (min	Mixing tolerance index (B.U	Degree of weakening (B.U)
Control (untrated)	59.0	1.5	2.0	9.5	30	160
ozonation at 20 ppm for 5min	59.0	1.5	2.0	9.5	30	160
ozonation at 20 ppm for 10 min	59.0	1.5	2.5	12.5	20	160
ozonation at 20 ppm for 15 min	59.0	2.0	3.0	10.5	30	160
ozonation at 20 ppm for 20 min	59.0	2.0	3.0	8.5	20	160
ozonation at 40 ppm for 5min	59.0	1.5	2.5	12.5	20	160
ozonation at 40 ppm for 10min	59.0	1.5	2.5	12.5	20	160
ozonation at 40 ppm for 15min	59.0	2.0	3.0	10.5	20	160
ozonation at 40 ppm for 20min	59.0	2.0	2.5	10.5	30	160

Effect of ozonation wheat grain on stability of dough flour.



Effect of ozone treatment on extensograph parameters

Samples	Extensibility E (m.m)	Resistance to extension (R) (B.U.):	Proportional number (R/E)	Dough energy :(cm2)
Control (untrated)	120	320	2.7	54
ozonation at 20 ppm for 5min	120	320	2.7	45
ozonation at 20 ppm for 10 min	115	320	2.8	45
ozonation at 20 ppm for 15 min	110	320	2.9	42
ozonation at 20 ppm for 20 min	90	320	3.5	40
ozonation at 40 ppm for 5min	110	360	3.3	45
ozonation at 40 ppm for 10min	100	360	3.6	45
ozonation at 40 ppm for 15min	80	360	4.8	39
ozonation at 40 ppm for 20min	75	340	5.2	36

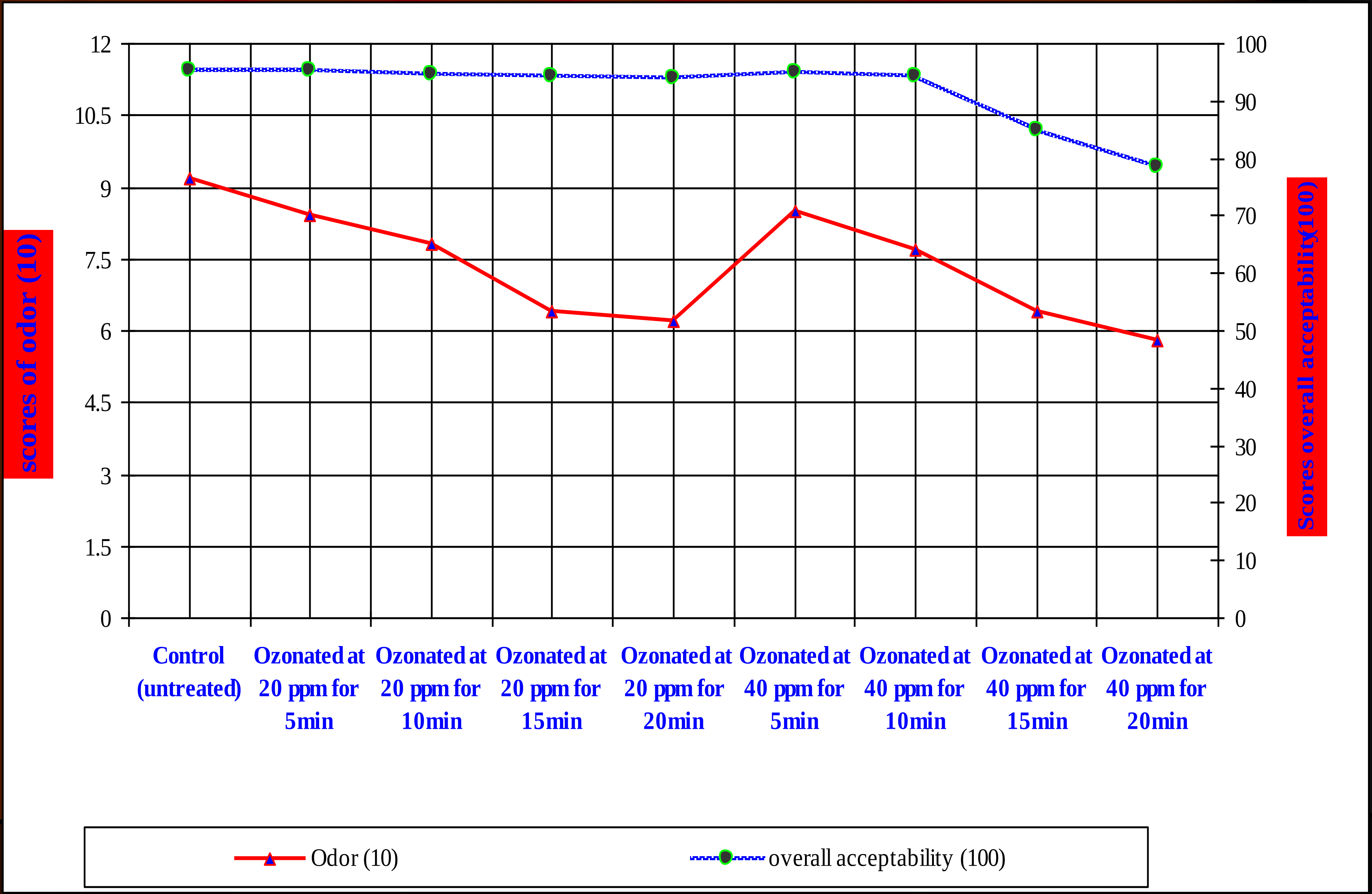
Quality of Balady bread made from ozonated wheat grain

sensory evaluation of balady bread produced from wheat flour 82% extraction which obtained after ozonated wheat grain.

Ozone treatment did not affect crumb distribution, roundness and separation of layers.

The results showed significant differences in taste between the samples as compared with control samples. This is probably due to the odor of ozone gas, which had a significant effect with treatments where there was a decrease score of odor bread, which in turn played an important role in the general acceptance of the bread product

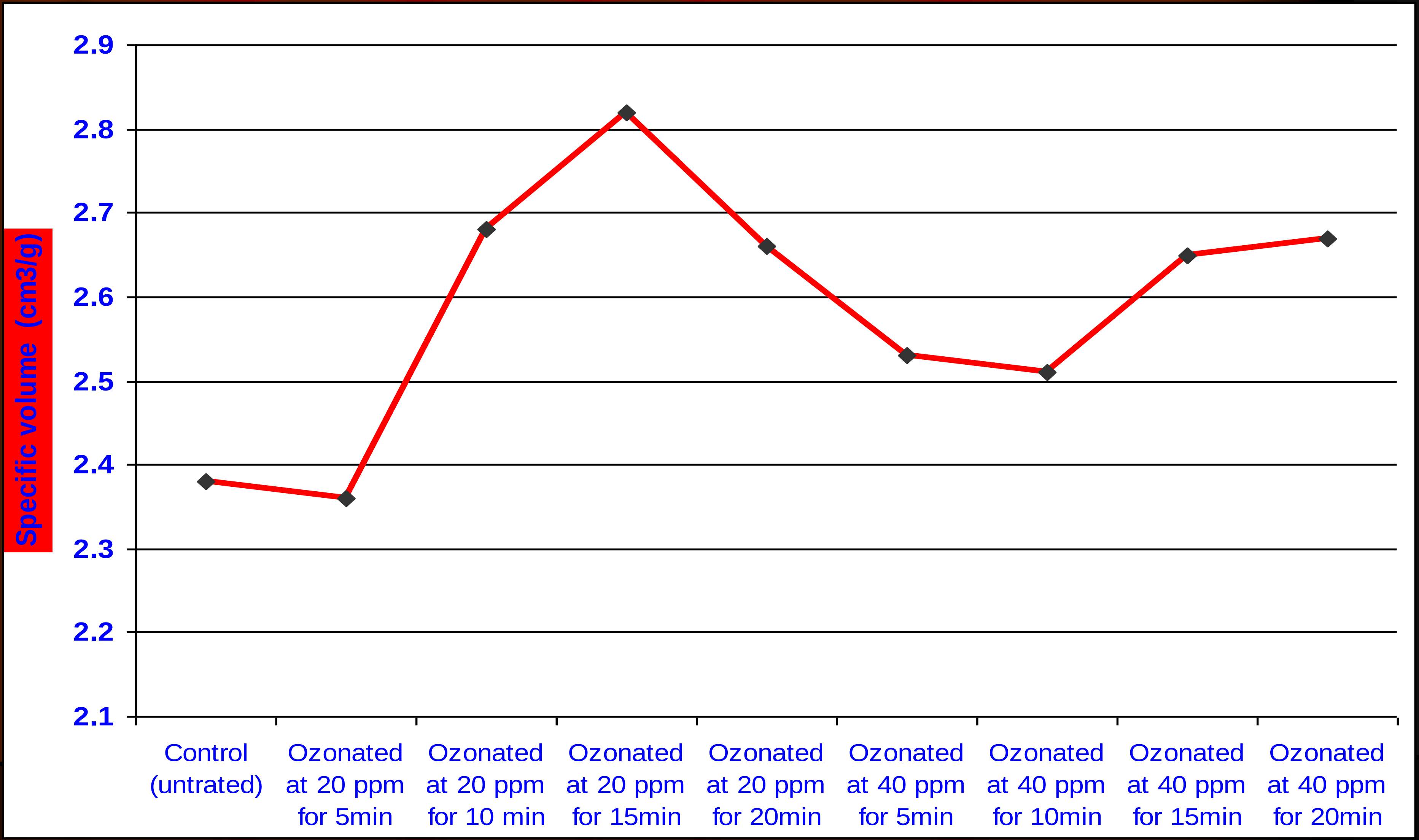
Effect of odor ozone gas on overall acceptability balady bread.



Effect of ozonated wheat grain on physical characteristics (baking quality) of balady bread.

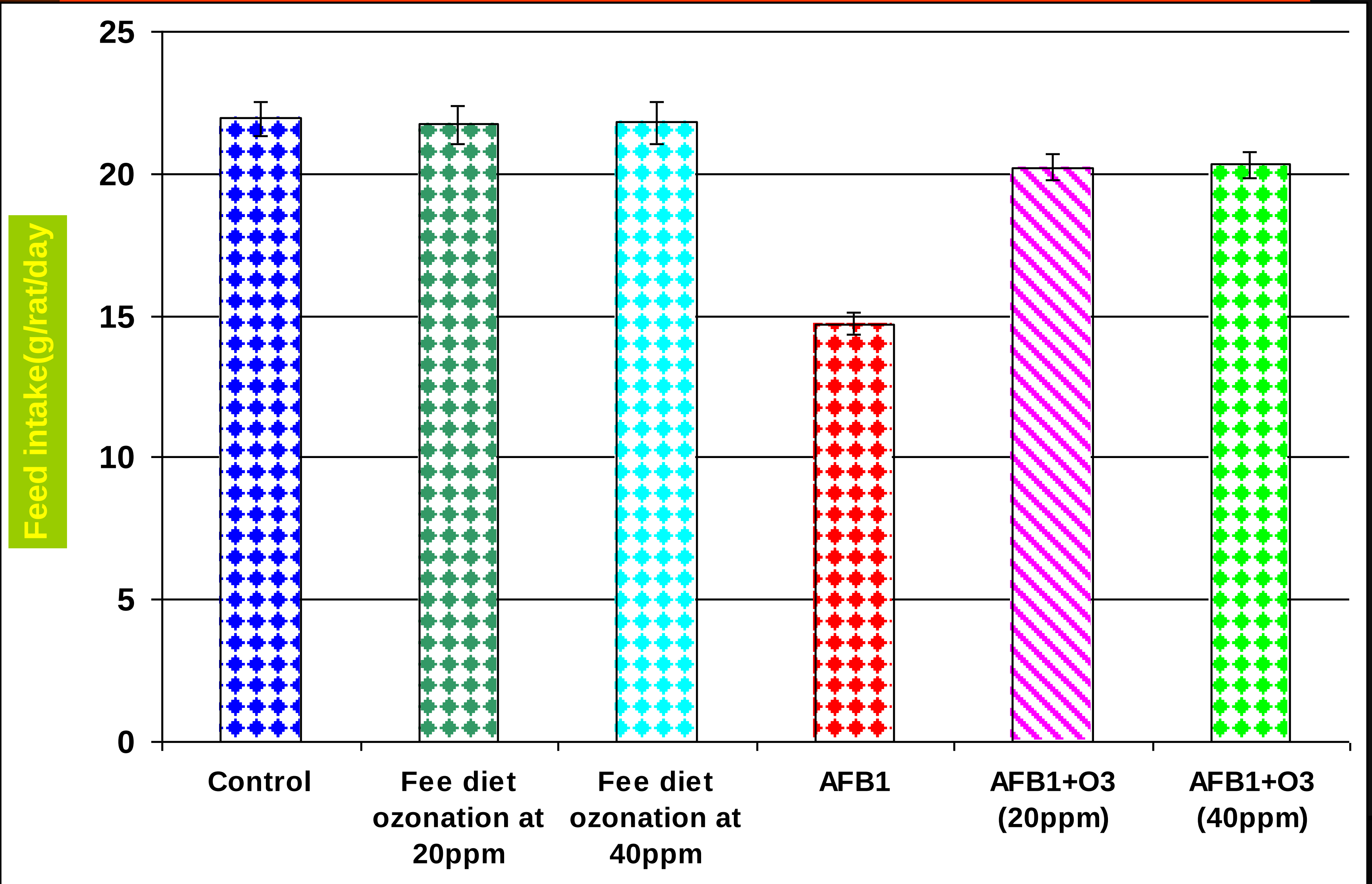
Samples	Laof Weight (g)	Loaf Volume (cm^3)	Loaf Specific volume (cm^3/g)
Control (untrated)	118.57±0.23	282.5±0.28	2.38
ozonation at 20 ppm for 5min	111.0±0.50	262.42±0.22	2.36
ozonation at 20 ppm for 10 min	111.93±0.06	299.87±0.58	2.68
ozonation at 20 ppm for 15 min	113.77±0.14	320.58±0.36	2.82
ozonation at 20 ppm for 20 min	114.33±0.44	303.83±0.60	2.66
ozonation at 40 ppm for 5min	112.76±0.14	285.72±0.63	2.53
ozonation at 40 ppm for 10min	123.56±0.38	310.68±0.51	2.51
ozonation at 40 ppm for 15min	114.83±0.17	304.87±0.58	2.65
ozonation at 40 ppm for 20min	116.56±0.38	311.6±0.086	2.67

Effect of ozonated wheat grain on specific volume of balady bread.

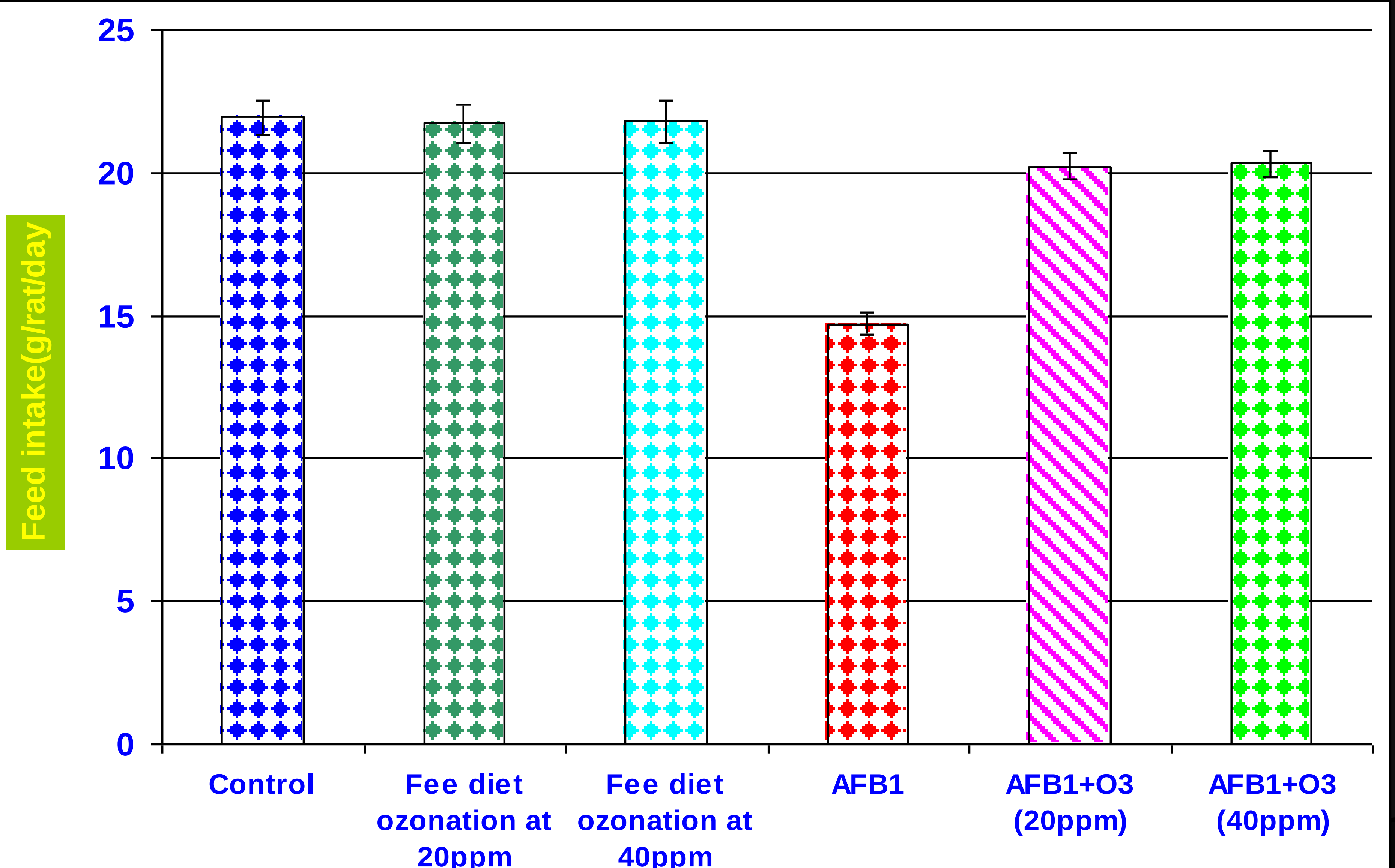


Part IV
Biological and histopathological evaluations of using ozone gas in decontamination of AFB_1 in wheat grains

Effect of ozone treated wheat and AFB_1 on feed intake

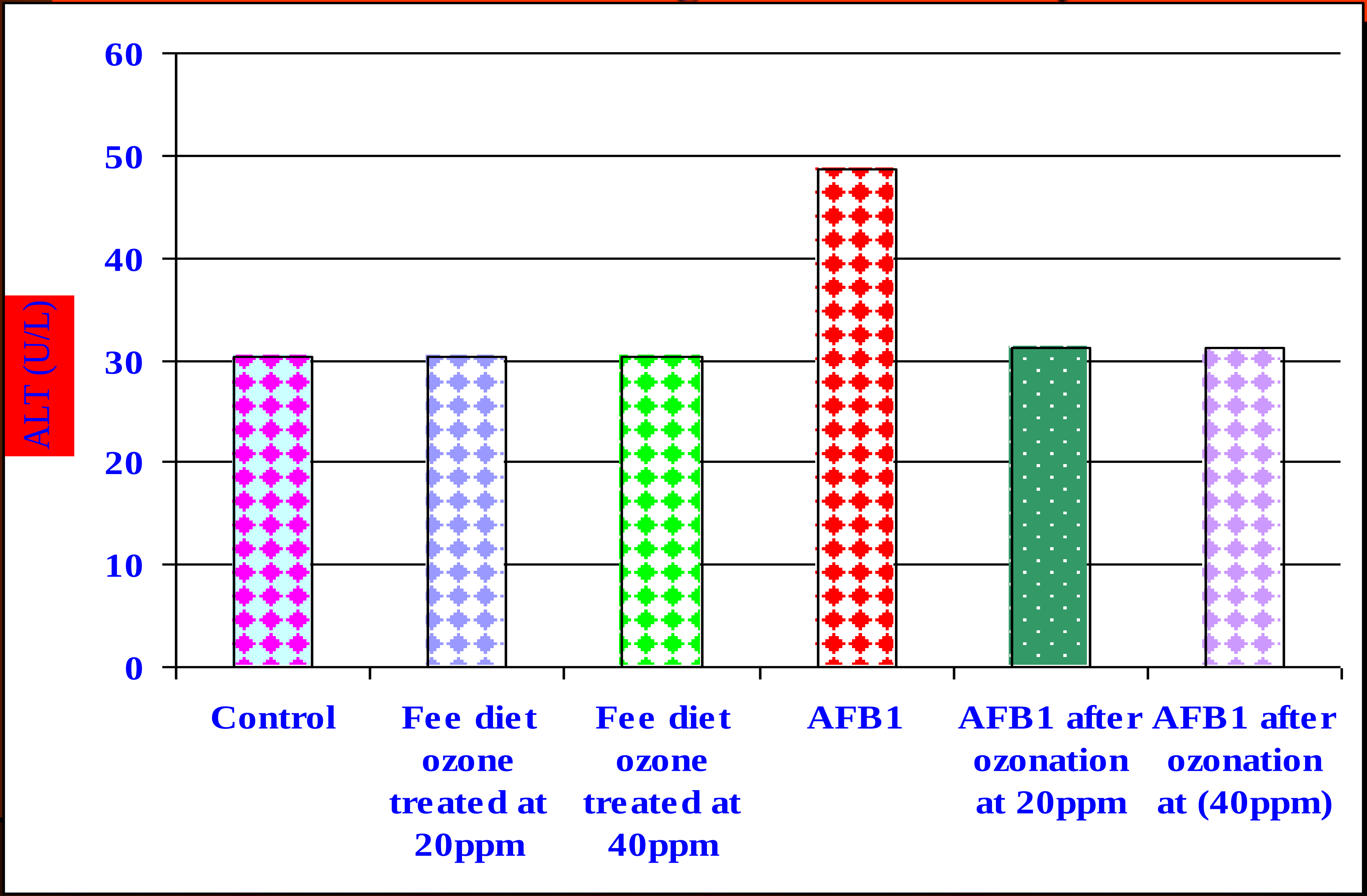


Effect of ozone treated wheat and AFB_1 on body weight

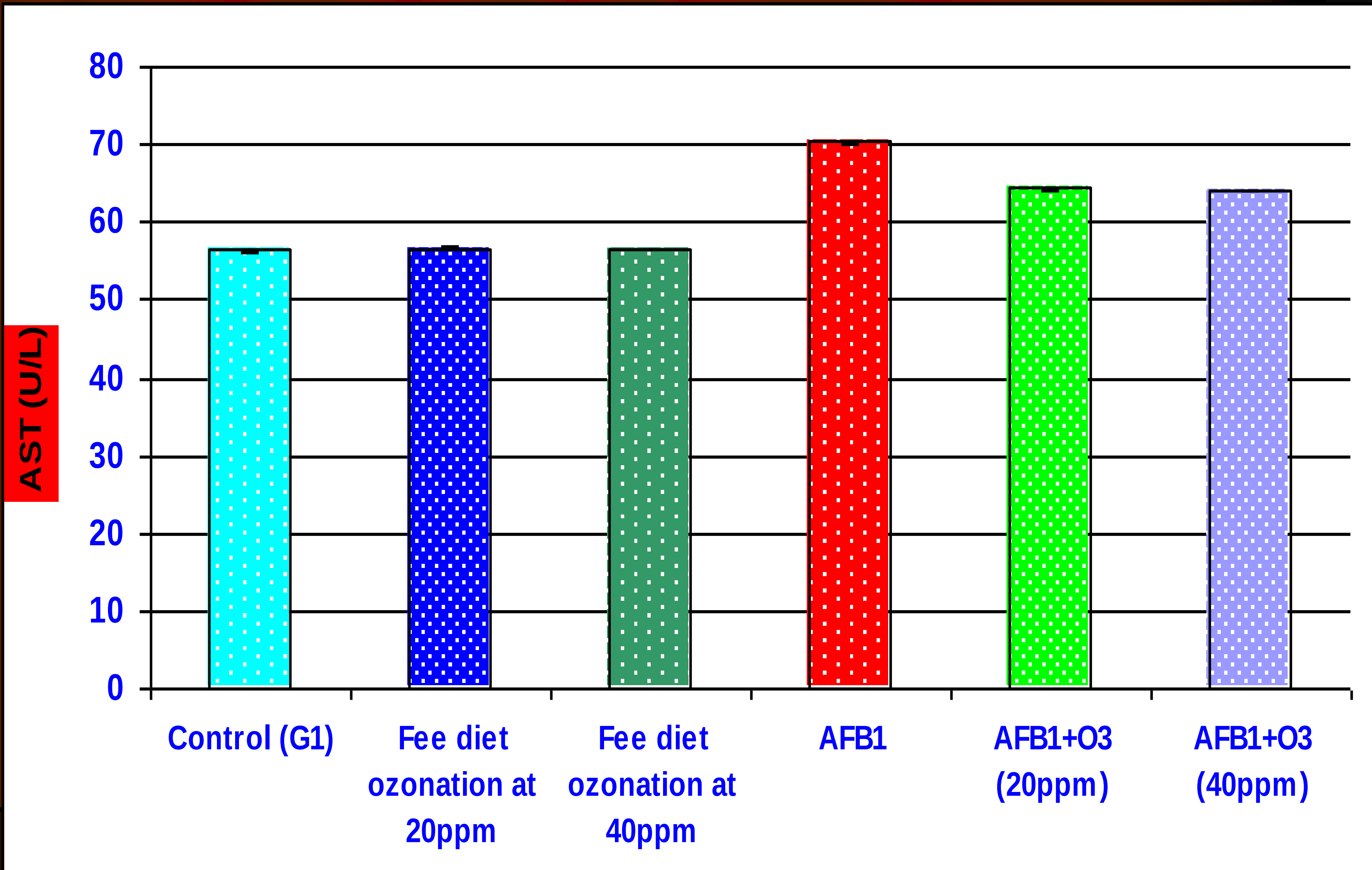


Effect of ozone treatment on liver function parameters in rats fed AFB1-contaminated diet

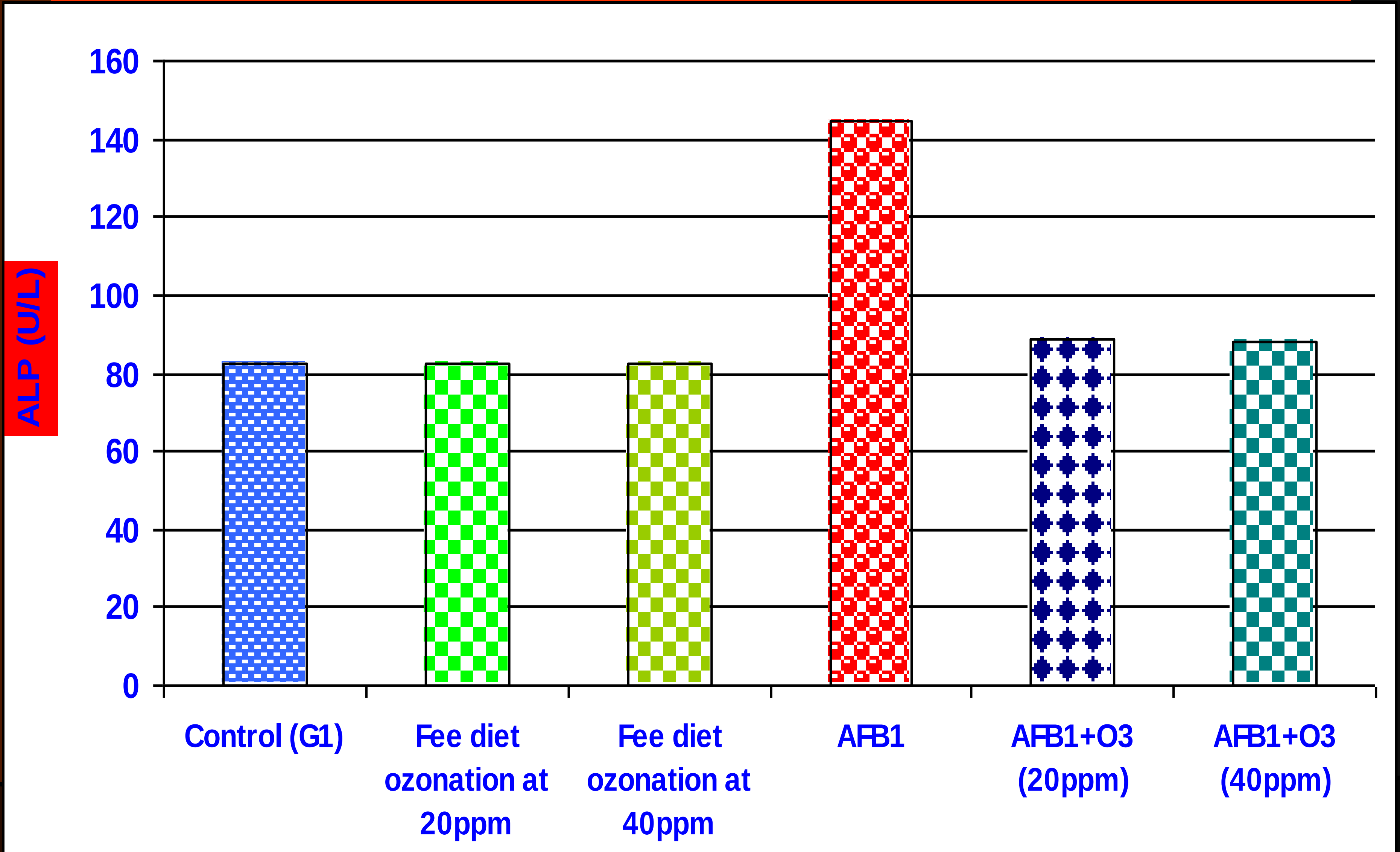
Effect of ozone treated and AFB1-contaminated wheat alone or treated with ozone gas on ALT activity in rats



Effect of ozone treated and AFB1-contaminated wheat alone or treated with ozone gas on AST activity in rats.

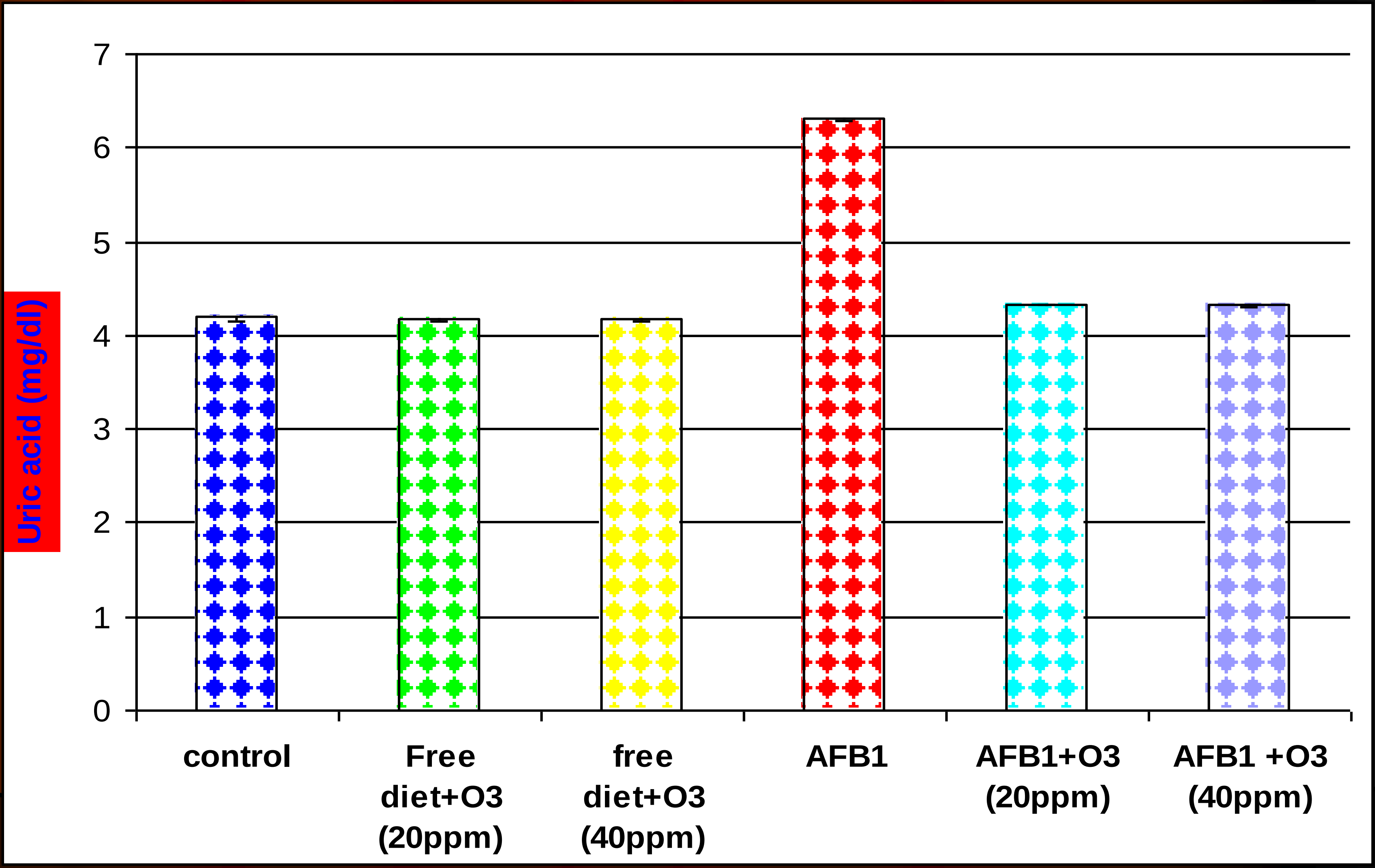


Effect of ozone treated and AFB1-contaminated wheat alone or treated with ozone gas on ALP activity in rats

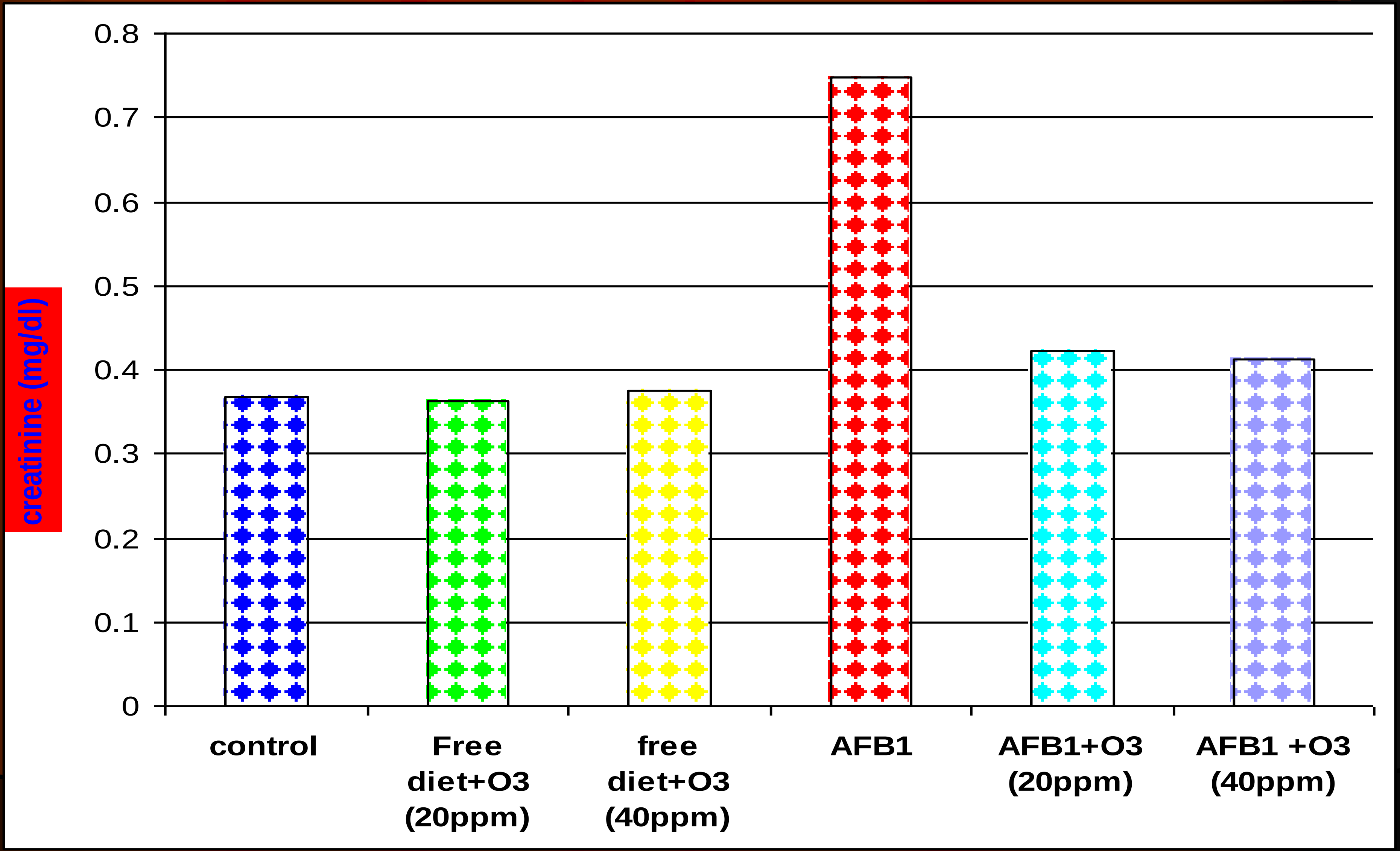


Effect of ozone treated on kidneys function parameters in rats fed AFB_1-contaminated die

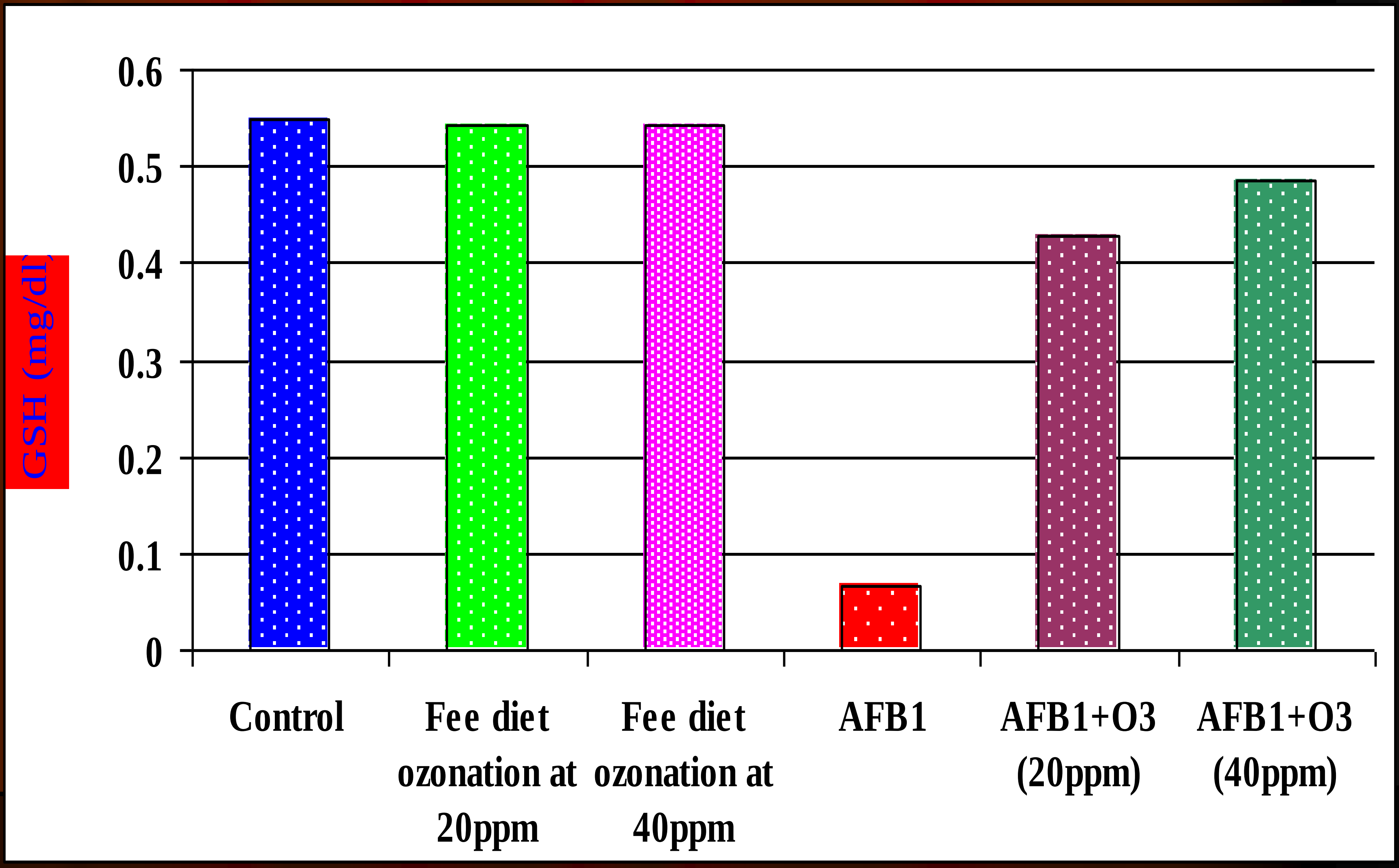
Effect of ozonation on uric acid level in rats fed AFB_1-contaminated wheat



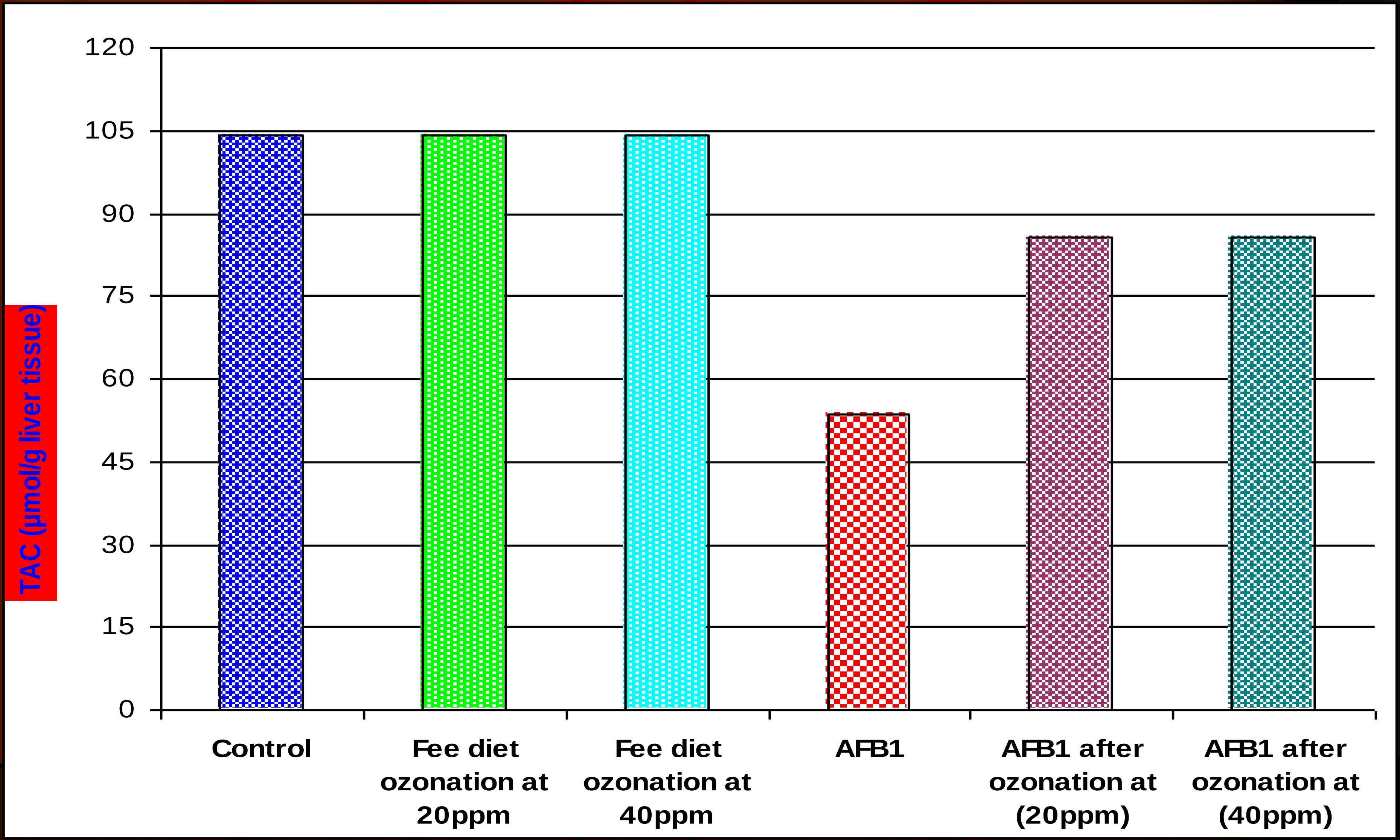
Effect of ozonation on createnine level in rats fed AFB1-contaminated wheat.



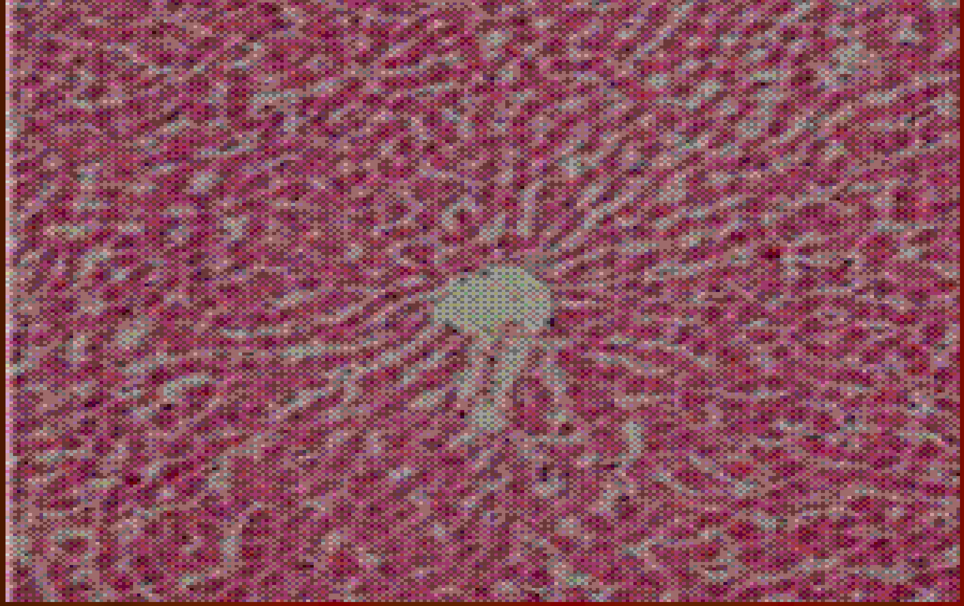
Effect of ozonation on GSH level in rats fed AFB1-contaminated wheat.



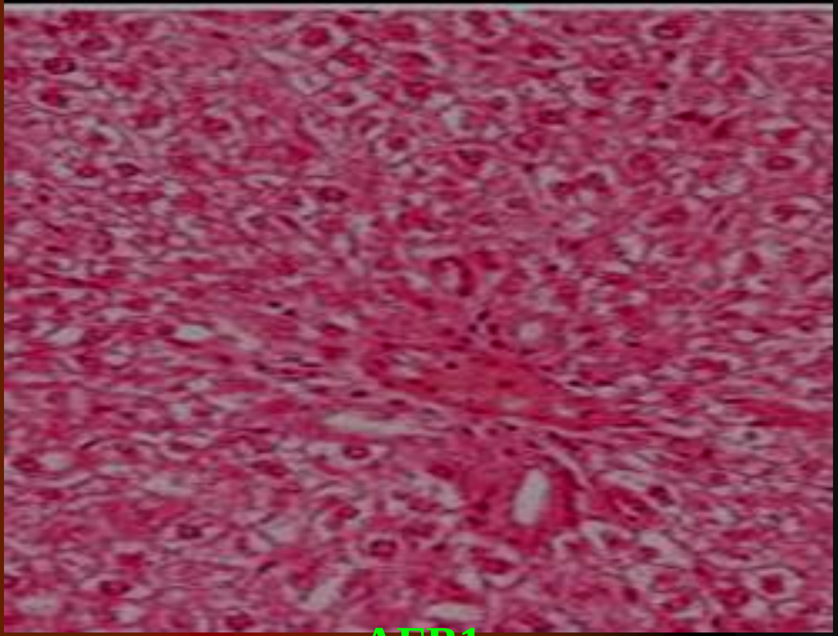
Effect of ozonation on TAC level in rats fed AFB1-contaminated wheat



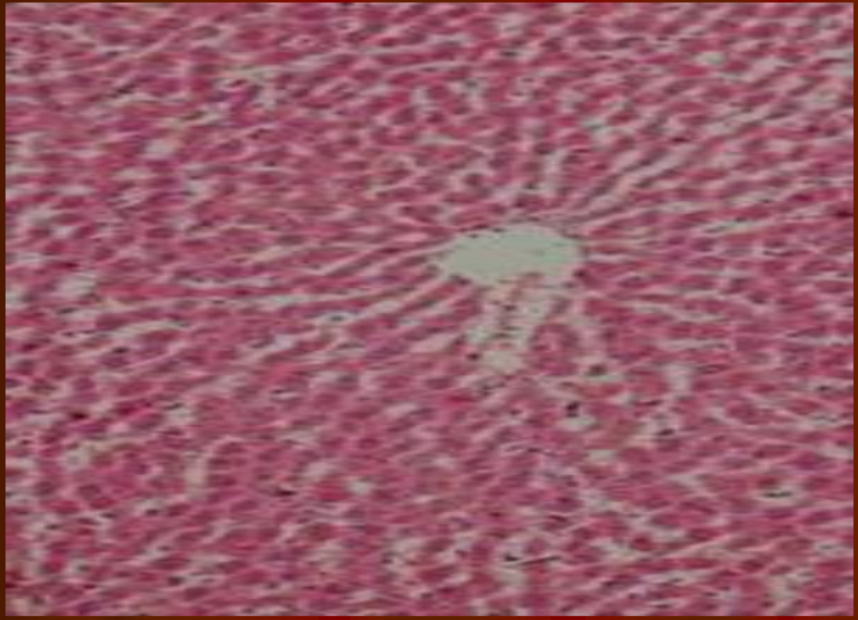
The histopathological study



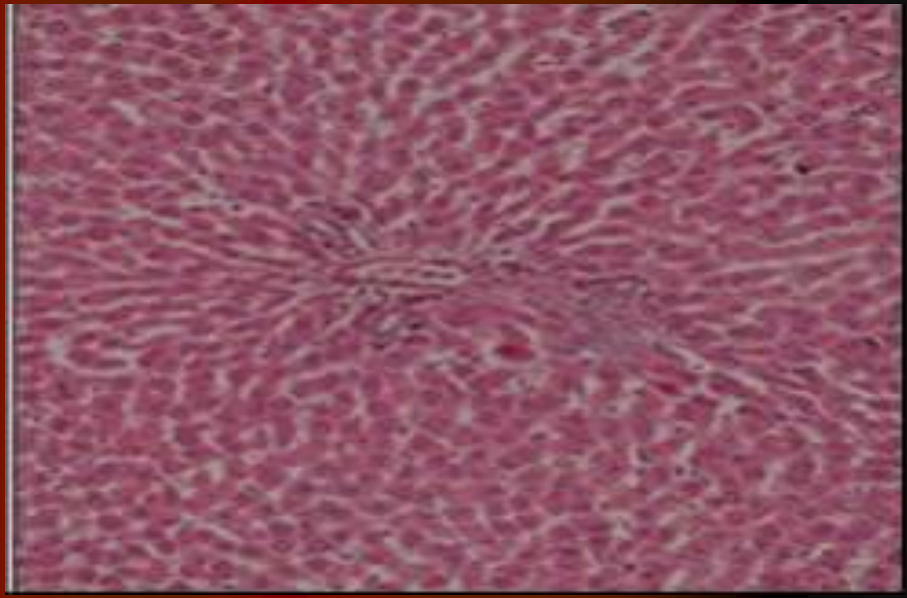
CONTROL



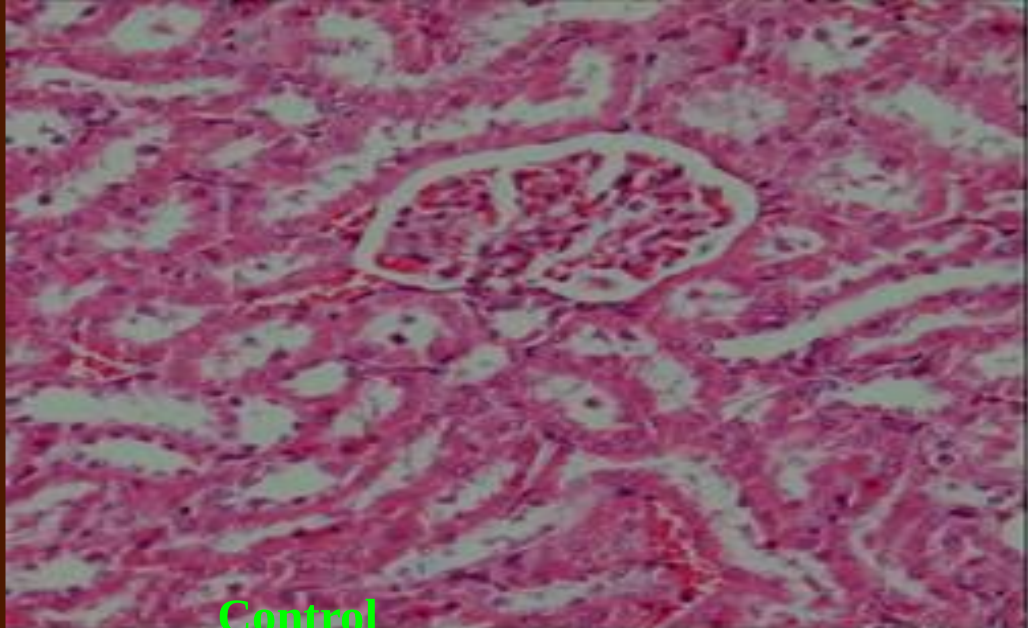
AFB1



section in liver of group fed on AFB1 free diet and treated with ozone gas



section in liver of rat fed AFB1-contaminated diet after ozonation



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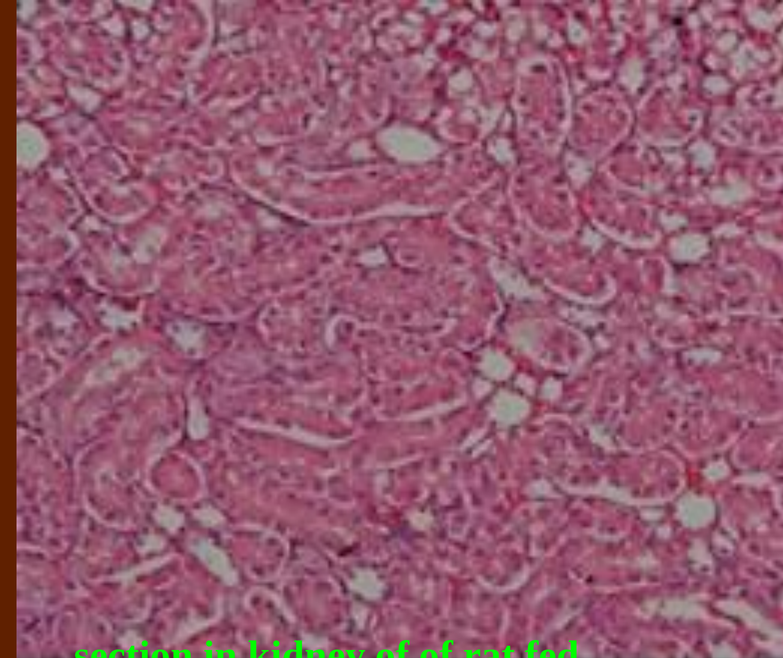
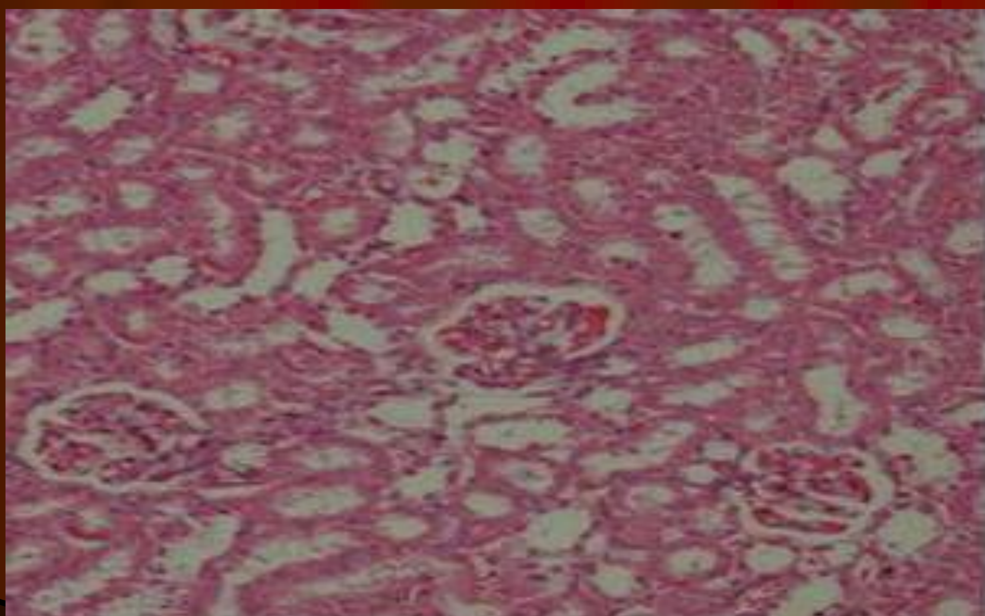
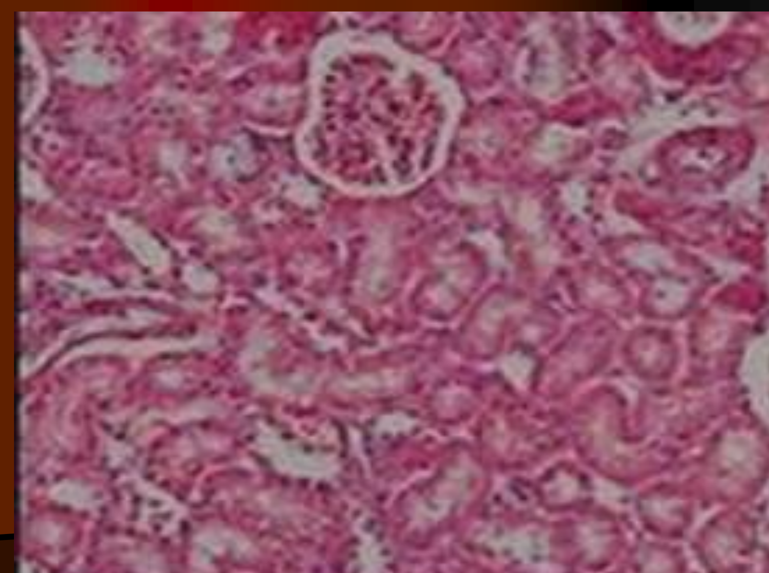


section in kidney of of rat fed AFB1-contaminated diet



section in kidney of group fed on AFB1 free diet and treated with ozone gas



section in kidney of rat fed AFB1-contaminated diet after ozonation

Conclusion

Due to the strong desire to reduce the use of chemicals applied in the food and feed chains, and considering the nonresidual ozone feature as an important advantage, the application of ozone technology in food has been considered safe and effective by the World Health Organization (WHO), the Food and Agriculture Organization (FAO),

and the U.S. Food and Drug administration (WHO, 2007; FDA, 2008).