



وزارت بهداشت
و درمان



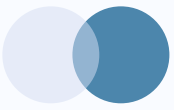
گروه علوم و صنایع غذایی (کنترل کیفی و بهداشتی)
ژورنال کلاب دوره کارشناسی علوم و صنایع غذایی



تأثیر بسته‌بندی فعال بر پایه پوشش بیوپلیمری کیتوزان غنی‌شده با اسانس آویشن شیرازی بر ماندگاری ماهی کپور نقره‌ای در طول نگهداری در یخچال

استاد راهنما: دکتر پرنیان پزشکی
ارائه دهندگان: محمدمهدی تیموری - محمدمهدی فهمیده
آبان ماه ۱۴۰۴





مقدمه

ماهی کپور نقره‌ای (*Hypophthalmichthys molitrix*) در سیستم پرورش چندگانه کشورهای شرقی از اهمیت ویژه‌ای برخوردار است. گوشت ماهی به دلیل ارزش غذایی بالا، فعالیت آبی و pH مناسب، از نظر میکروبیولوژیکی و شیمیایی بسیار فسادپذیر می‌باشد.

مهمترین چالش صنعت غذاهای دریایی، افزایش مدت ماندگاری و بهبود قابلیت پذیرش گوشت ماهی از طریق مهار اکسیداسیون لیپیدی و جلوگیری از تکثیر میکروبی است. پوشش‌های خوراکی در بسته‌بندی فعال، فناوری نوینی است که غذا، بسته‌بندی و حفظ کیفیت را در یک مفهوم واحد تلفیق می‌نماید.

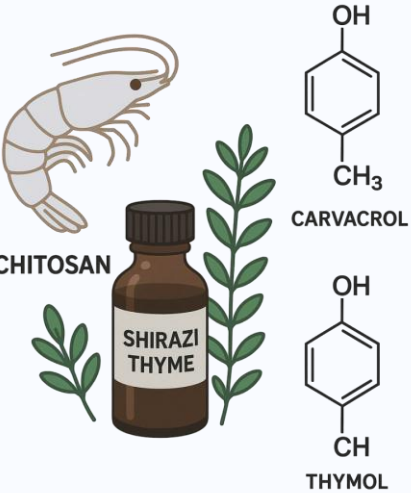


پوشش‌های خوراکی لایه‌های نازک و خوراکی هستند که مستقیماً روی سطح مواد غذایی اعمال می‌شوند. این پوشش‌ها با کنترل انتشار گازها، رطوبت و انتقال ترکیبات معطر، به حفظ کیفیت و مدت ماندگاری محصول کمک می‌کنند.



مقدمه ادامه...

پوشش خوراکی فعال پیشنهادی متشکل از کیتوزان به عنوان بیوپلیمر پایه و اسانس *Zataria multiflora* (آویشن شیرازی) به عنوان عامل فعال می‌باشد.



کیتوزان یک بیوپلیمر طبیعی است که از پوسته سخت‌پوستان استخراج می‌شود. اسانس آویشن شیرازی حاوی ترکیبات فنولی اصلی شامل کارواکرول و تیمول است که دارای خواص ضد میکروبی و آنتی‌اکسیدانی قوی می‌باشند.

سازمان FDA آمریکا ترکیبات فنولی از جمله کارواکرول و تیمول را در گروه GRAS (Generally Recognized as Safe) طبقه‌بندی کرده است. این ترکیب طبیعی به عنوان یک جایگزین ایمن برای مواد شیمیایی مصنوعی در بسته‌بندی فعال مواد غذایی دریایی پیشنهاد می‌شود.

هدف

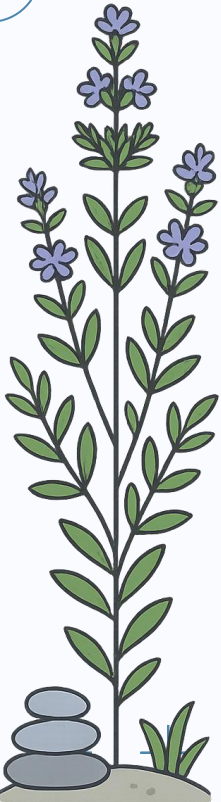
هدف از انجام این پژوهش، بررسی اثر ترکیب کیتوزان با اسانس روغنی آویشن (ZMEO) بر افزایش ماندگاری و کیفیت فیله‌های تازه ماهی نقره‌ای در طی دوره نگهداری در دمای ۴ درجه سانتی‌گراد بود. در این مطالعه تلاش شد تا با به‌کارگیری پوشش‌های زیست‌تخریب‌پذیر و ترکیبات طبیعی ضد میکروبی، از فساد میکروبی و شیمیایی محصول جلوگیری شده و زمان ماندگاری آن افزایش یابد.



مواد و روش‌ها - تهیه اسانس آویشن شیرازی

منبع و شناسایی گیاه:

- گیاه آویشن شیرازی (*Zataria multiflora*) از استان فارس جمع‌آوری شد.
- شناسایی و تأیید هویت گیاه توسط مؤسسه گیاهان دارویی دانشگاه علوم پزشکی تهران انجام گرفت.



مواد و روش‌ها - تهیه اسانس آویشن شیرازی

روش استخراج اسانس:

- اندام‌های هوایی گیاه پس از خشک‌شدن در محیط طبیعی
- استخراج به روش تقطیر با بخار آب به مدت ۳ ساعت
- استفاده از دستگاه کلونجر (Clevenger) برای استخراج
- جمع‌آوری اسانس استخراج شده برای آنالیزهای بعدی

تجزیه و تحلیل ترکیبات اسانس:

- شناسایی ترکیبات موجود در اسانس با دستگاه گاز کروماتوگرافی متصل به طیف‌سنج جرمی (GC-MS)
- انجام آنالیز مطابق با روش منتشر شده توسط موسوی و همکاران (۲۰۰۸)



مواد و روش‌ها - تهیه پوشش کیتوزان

تهیه محلول پایه کیتوزان:

- ۲۰ گرم کیتوزان با وزن مولکولی بالا در ۱۰۰ میلی‌لیتر اسید استیک ۱٪ حل شد.
- محلول به مدت ۳ ساعت در دمای اتاق همزده شد تا کاملاً همگن شود.



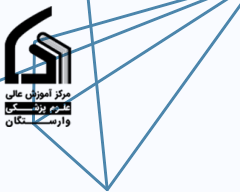
افزودن نرم‌کننده:

- گلیسرول به میزان ۰٫۷۵ میلی‌لیتر به ازای هر گرم کیتوزان اضافه شد.
- مخلوط به مدت ۱۰ دقیقه دیگر همزده شد.



صاف کردن محلول:

- محلول کیتوزان از فیلتر کاغذی واتمن شماره ۳ عبور داده شد تا ذرات جامد حل‌نشده جدا شود.



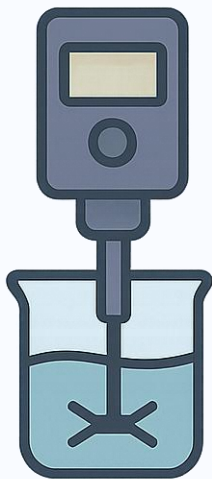
مواد و روش‌ها - تهیه پوشش کیتوزان

افزودن ترکیبات فعال:

- Tween 80 با غلظت ۰٫۲۵٪ حجمی/حجمی به محلول اضافه شد
- اسانس آویشن شیرازی در چهار غلظت مختلف به محلول افزوده شد:
 - ۰٪ (شاهد)
 - ۰٫۳٪
 - ۰٫۴۵٪
 - ۰٫۶٪

همگن‌سازی نهایی:

- مخلوط در شرایط استریل با سرعت ۲۱۶۰۰ دور بر دقیقه به مدت ۱ دقیقه همگن شد.



مواد و روش‌ها - آماده‌سازی و فرآوری ماهی

منبع و مشخصات ماهی:

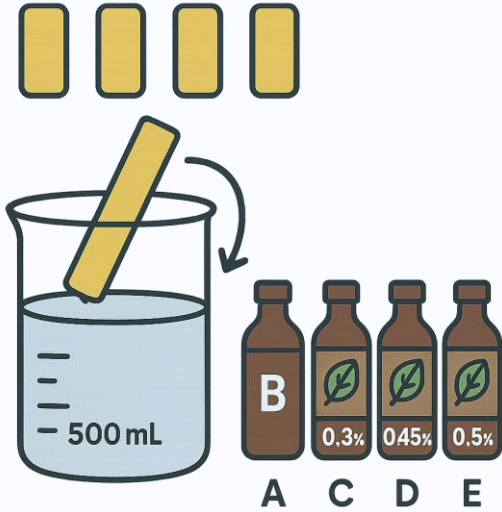
- ماهی‌های کپور نقره‌ای (*Hypophthalmichthys molitrix*) از مزرعه پرورش ماهی در استان تهران صید شدند.
- میانگین وزن: ۱۰۰۰ گرم
- میانگین طول: ۳۵۰ میلی‌متر

انتقال و آماده‌سازی اولیه:

- ماهی‌ها به آزمایشگاه علوم گوشت گروه بهداشت مواد غذایی دانشکده دامپزشکی دانشگاه تهران منتقل شدند.
- ماهی‌ها به قطعات مربعی با وزن تقریبی ۶۰ گرم تقسیم شدند.
- ترکیبات پایه نمونه‌ها شامل درصد پروتئین و چربی سه فیله با روش انجمن شیمی‌دانان تحلیلی (AOAC) اندازه‌گیری شد



مواد و روش‌ها - آماده‌سازی و فرآوری ماهی



Coating treatment process

فرآیند تیمار با پوشش:

· نمونه‌ها به مدت ۳۰ ثانیه در ۵۰۰ میلی‌لیتر از محلول‌های پوشش مختلف غوطه‌ور شدند.

تیمارهای مورد بررسی:

- A: نمونه‌های شاهد (آب مقطر)
- B: پوشش کیتوزان
- C: پوشش کیتوزان غنی‌شده با ۰٫۳٪ اسانس
- D: پوشش کیتوزان غنی‌شده با ۰٫۴۵٪ اسانس
- E: پوشش کیتوزان غنی‌شده با ۰٫۶٪ اسانس

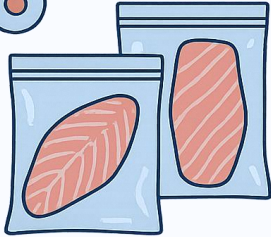
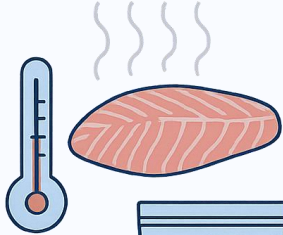
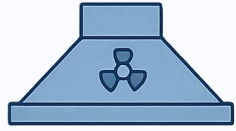
مواد و روش‌ها - آماده‌سازی و فرآوری ماهی

خشک‌کردن و بسته‌بندی:

- فیله‌های ماهی از محلول خارج شده و به مدت ۳۰ دقیقه زیر هود میکروبیولوژی در دمای ۲۵ درجه سانتی‌گراد خشک شدند.
- پوشش بر روی سطح فیله‌ها تشکیل شد.
- نمونه‌ها در دمای 4 ± 1 درجه سانتی‌گراد نگهداری شدند.

دوره نگهداری و ارزیابی:

- مدت زمان مطالعه: ۱۵ روز
- ارزیابی‌ها در روزهای ۰، ۱، ۳، ۵، ۷، ۹، ۱۱، ۱۳ و ۱۵ انجام شد.
- پارامترهای میکروبی، شیمیایی و حسی در هر بازه زمانی مورد سنجش قرار گرفت.





مواد و روش‌ها - بررسی‌های میکروبیولوژیکی

آماده‌سازی نمونه برای آنالیز میکروبی:

- ۱۰۰ گرم از هر نمونه با ۹۰ میلی‌لیتر آب پپتون ۰/۱٪ در کیسه‌های استریل مخلوط شد.
- به مدت ۲ دقیقه با دستگاه استومیگر همگن شد.
- رقت‌های متوالی اعشاری تهیه شد.



شمارش کل باکتری‌ها (TVC):

- محیط کشت: پلیت کانت آگار (PCA)
- روش کشت: روش سطحی
- شرایط انکوباسیون: ۳۰ درجه سانتی‌گراد به مدت ۳ روز
- گزارش نتیجه: log CFU/g

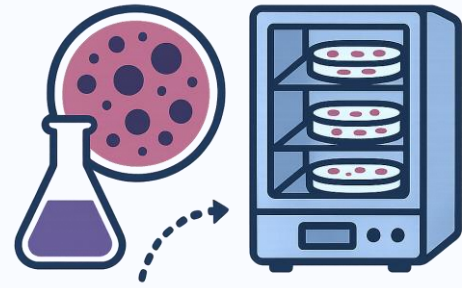
مواد و روش‌ها - بررسی‌های میکروبیولوژیکی

شمارش انتروباکتریاسه (EBC):

• محیط کشت: وایلت رد بایل گلوکز آگار (VRBGA)

• روش کشت: روش پور پلیت

• شرایط انکوباسیون: ۲۵ درجه سانتی‌گراد به مدت ۴۸ ساعت



شمارش باکتری‌های اسید لاکتیک (LAB):

• محیط کشت: دمان روگوزا شارپ آگار (MRS)

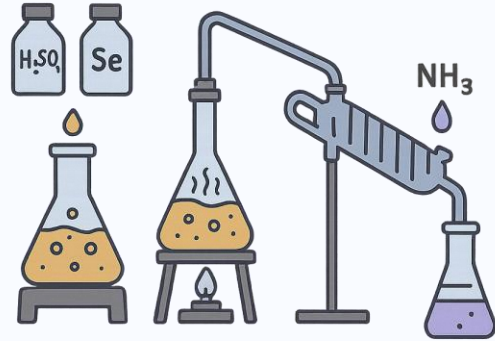
• شرایط انکوباسیون: ۳۰ درجه سانتی‌گراد به مدت ۲-۳ روز



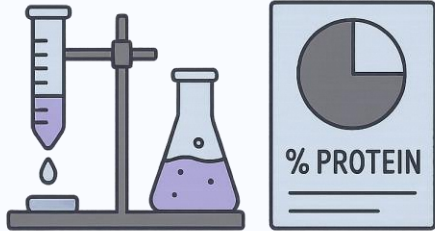
تجزیه و تحلیل داده‌ها:

• تمام داده‌های میکروبیولوژیکی به لگاریتم واحد تشکیل کلونی در گرم تبدیل شد.

مواد و روش‌ها - آنالیزهای شیمیایی (پروتئین و چربی)



Digestion Distillation Titration



NITROGEN TO
PROTEIN
CONVERSION FACTOR: 6,25

REPORTING

اندازه‌گیری پروتئین کل:

· روش مورد استفاده: کجلدال

· مراحل انجام آزمایش:

· هضم: استفاده از اسید سولفوریک غلیظ، سولفات پتاسیم

و کاتالیزور سلنیوم

· تقطیر: جداسازی آمونیاک

· تیتراسیون: سنجش نهایی مقدار نیتروژن

· ضریب تبدیل نیتروژن به پروتئین: ۶٫۲۵

· گزارش نتیجه: درصد پروتئین خام

مواد و روش‌ها - آنالیزهای شیمیایی (پروتئین و چربی)



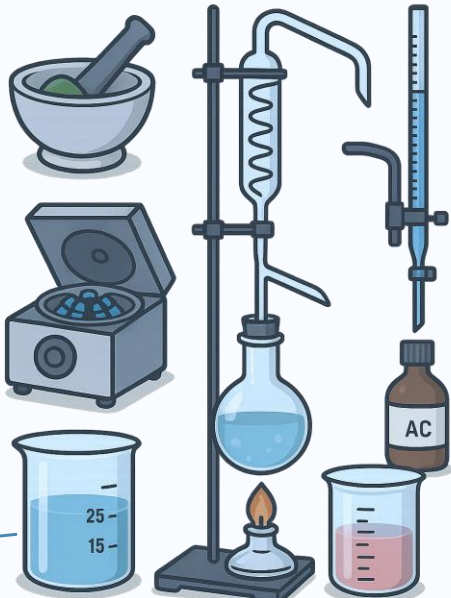
اندازه‌گیری چربی کل:

- روش مورد استفاده: استخراج با مخلوط کلروفرم-متانول
- نسبت حلال: ۲ به ۱ (حجم به حجم)
- مراحل انجام:
 - هموژنیزاسیون نمونه در مخلوط حلال
 - سانتریفیوژ به مدت ۵ دقیقه با نیروی ۴۰۰g
 - جداسازی فاز چربی
 - تبخیر حلال با روتاری اوپراتور در دمای ۴۰ درجه سانتی‌گراد
 - توزین و محاسبه درصد چربی
 - تکرار آزمایش: سه بار برای هر نمونه

مواد و روش‌ها - آنالیزهای شیمیایی (شاخص‌های فساد)

اندازه‌گیری TVB-N (نیتروژن بازهای فرار کل):

- روش: بر اساس روش جوکی و همکاران (۲۰۱۴)
- ۲۰۰ میلی‌لیتر از محلول ۷/۵٪ تری‌کلرواستیک اسید (TCA) به نمونه ماهی اضافه شد.
- همگن‌سازی و سانتریفیوژ با نیروی ۴۰۰ g به مدت ۵ دقیقه
- فیلتراسیون با کاغذ صافی واتمن شماره ۳
- ۲۵ میلی‌لیتر از فیلترات با ۵ میلی‌لیتر سود ۱۰٪ مخلوط شد.
- تقطیر با دستگاه تقطیر کج‌لدا
- جذب در ۱۰ میلی‌لیتر اسید بوریک ۴٪ حاوی شناساگر
- تیتراسیون با اسید سولفوریک ۰/۱ نرمال



مواد و روش‌ها - آنالیزهای شیمیایی (شاخص‌های فساد)



اندازه‌گیری عدد پراکسید (PV):

روش: پیرسون

- ۰/۳ گرم نمونه حاوی چربی استخراج شده
- حلال‌زدایی با روتاری اوپراتور
- اضافه کردن ۱۰ میلی‌لیتر مخلوط اسید استیک-کلروفرم
- افزودن یدید پتاسیم و نگهداری در تاریکی
- تیتراسیون با تیوسولفات سدیم ۰/۰۱ نرمال

اندازه‌گیری TBA (اسید تیوباربتوریک):

روش: کالریتری

- هضم اسیدی نمونه و تقطیر هدفمند
- واکنش با ۲-تیوباربتوریک اسید
- اندازه‌گیری جذب در طول موج ۵۳۸ نانومتر



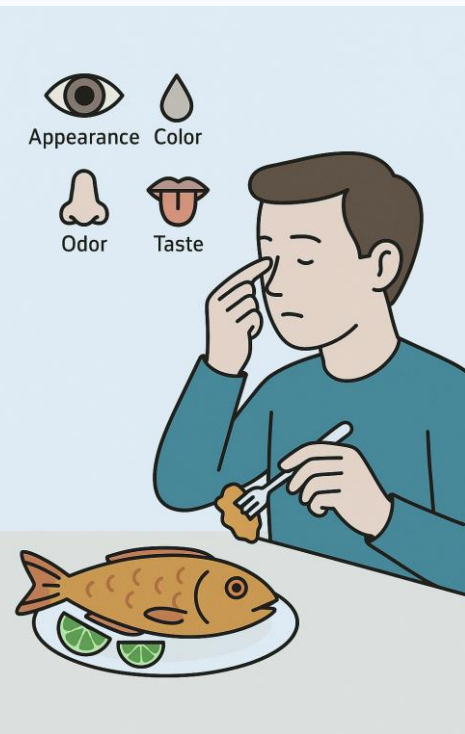
مواد و روش‌ها - ارزیابی حسی

روش انجام ارزیابی حسی:

- نمونه‌های ماهی سرخ شده مورد ارزیابی قرار گرفتند.
- تعداد ارزیاب‌ها: ۵۰ نفر از اعضای دانشکده دامپزشکی دانشگاه تهران
- ارزیاب‌ها آموزش‌دیده نبودند (Untrained panelists)

معیارهای ارزیابی:

- ظاهر
- رنگ
- بو
- طعم



مواد و روش‌ها - ارزیابی حسی

مقیاس امتیازدهی:

از مقیاس ۹ نقطه‌ای هدونیک استفاده شد:

۹۰: علاقه بسیار زیاد

۸۰: علاقه زیاد

۷۰: علاقه متوسط

۶۰: علاقه کم

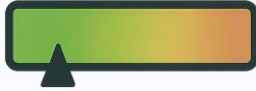
۵۰: بدون علاقه یا بی‌علاقگی

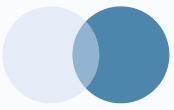
۴۰: بی‌علاقگی کم

۳۰: بی‌علاقگی متوسط

۲۰: بی‌علاقگی زیاد

۱۰: بی‌علاقگی بسیار زیاد

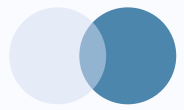




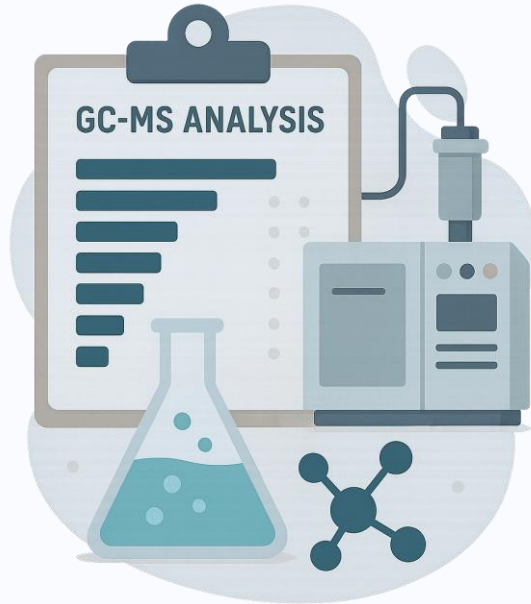
تحلیل آماری



- تحلیل داده‌ها با نرم‌افزار SPSS نسخه ۱۶ انجام شد.
- از آزمون ANOVA یک طرفه و Tukey برای مقایسه میانگین‌ها استفاده گردید.
- سطح معنی‌داری ۰/۰۵ در نظر گرفته شد.
- داده‌ها به صورت میانگین \pm انحراف معیار گزارش شدند.



نتایج - ترکیبات شیمیایی اسانس آویشن شیرازی



نتایج آنالیز GC-MS نشان داد:

- کارواکرول: ۷۳٫۶۵٪
- تیمول: ۴٫۳۱٪
- ترانس-کاریوفیلین: ۲٫۹۳٪
- گاما-ترپینن: ۲٫۲۷٪
- لینالول: ۱٫۲۸٪
- ترپینن-۴-اول: ۱٫۷۰٪
- متیل اتر تیمول: ۰٫۶۲٪
- ۱ و ۸-سینئول: ۰٫۵۲٪
- سایر ترکیبات: ۱۴٫۷۲٪

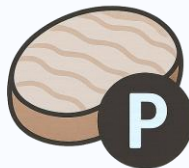
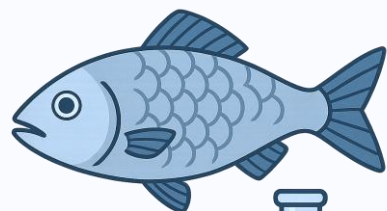


نتایج - ترکیبات پروتئین و چربی نمونه‌های ماهی

مقادیر پایه نمونه‌های ماهی:

· محتوای پروتئین: $13,97 \pm 0,59\%$

· محتوای چربی: $5,12 \pm 0,48\%$



PROTEIN



FAT



Table 1. Median rating for sensory properties of fish fillets as affected by different concentrations of ZEMO

| combination | Odor | Taste | Overall acceptability |
|------------------------|------|-------|-----------------------|
| control | 7 | 7 | 7 |
| Chitosan | 7 | 8 | 7 |
| Chitosan + 0.3 % ZEMO | 8 | 8 | 8 |
| Chitosan + 0.45 % ZEMO | 8 | 6 | 6 |
| Chitosan + 0.6 % ZEMO | 6 | 4 | 4 |

Means followed by the same letters are not significantly ($P < 0.05$) different.

SD: standard deviation.

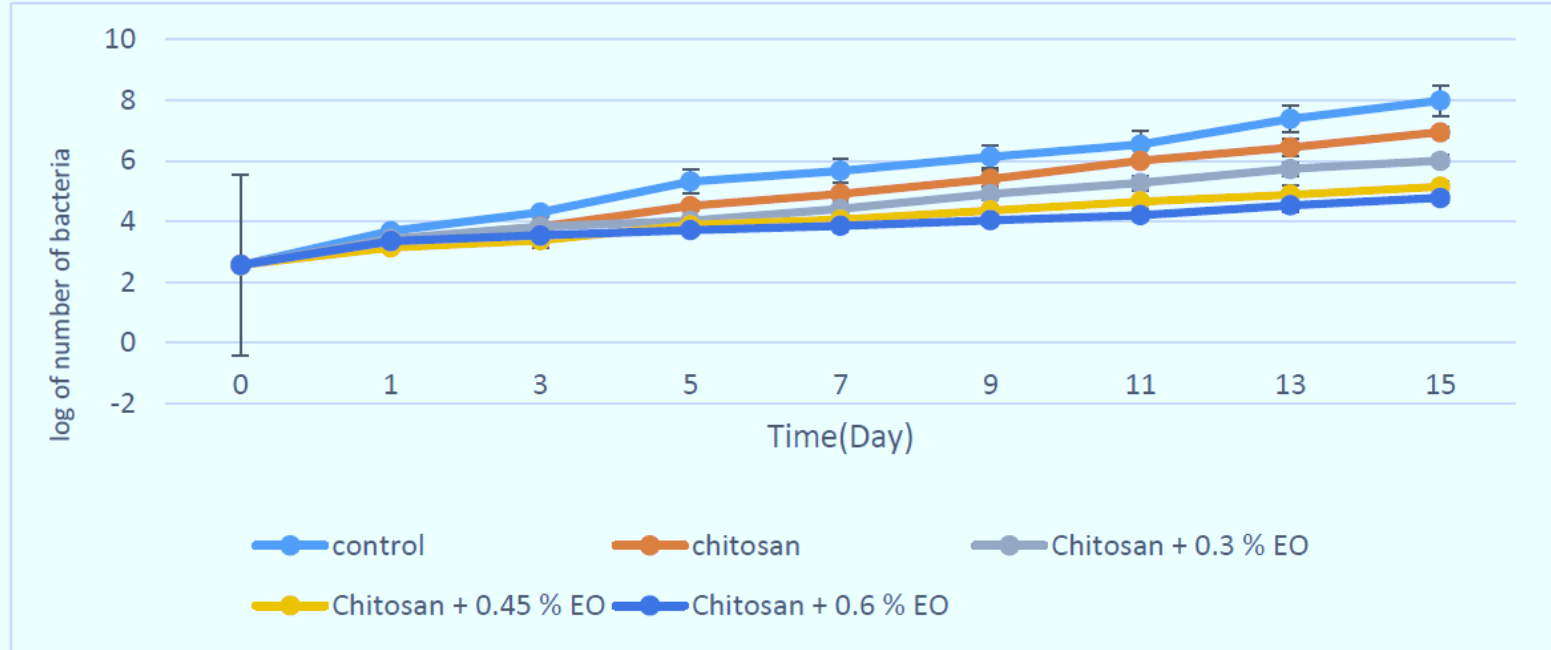


Figure 5. Changes in total viable counts (TVC) of Silver carp during chilled storage

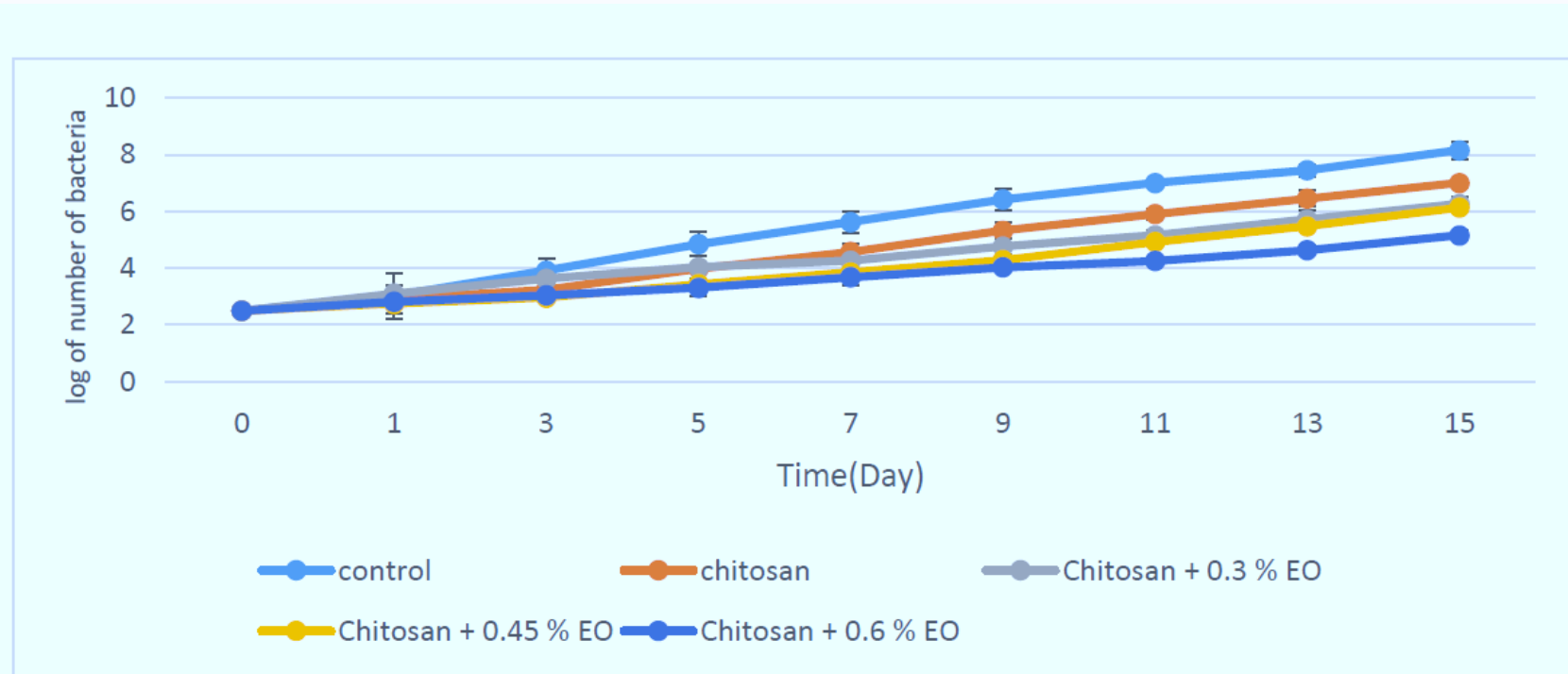


Figure 6. Changes in Enterobacteriaceae count (EBC) of silver carp during chilled storage

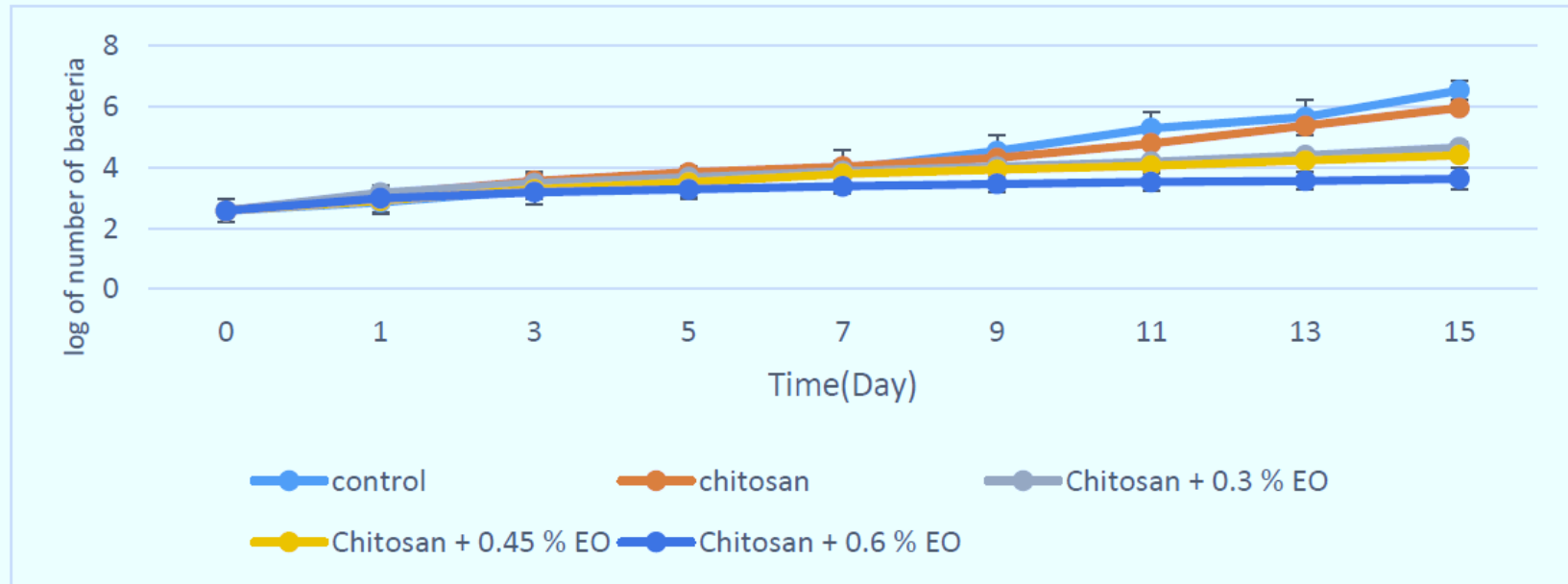


Figure 7. Changes in lactic acid bacteria count (LAB) of silver carp during chilled storage

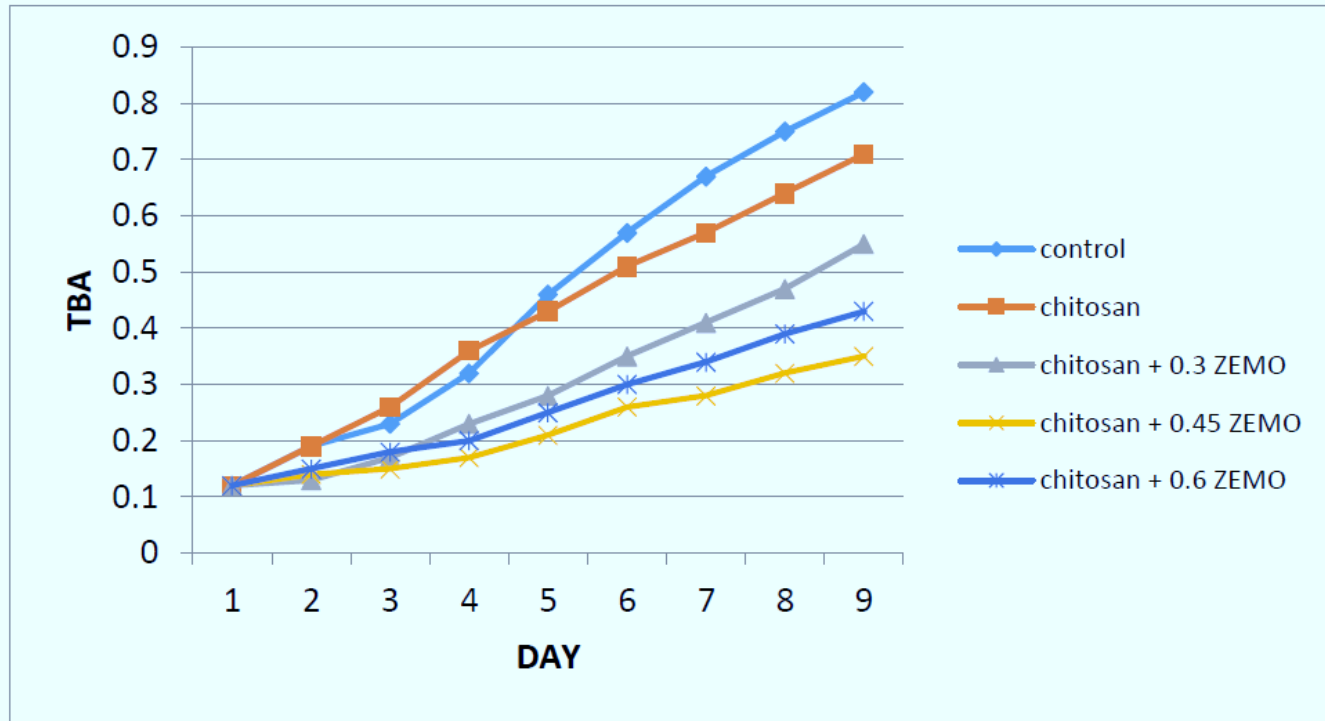


Figure 1. Changes in TBA values of the fish sample during chilled

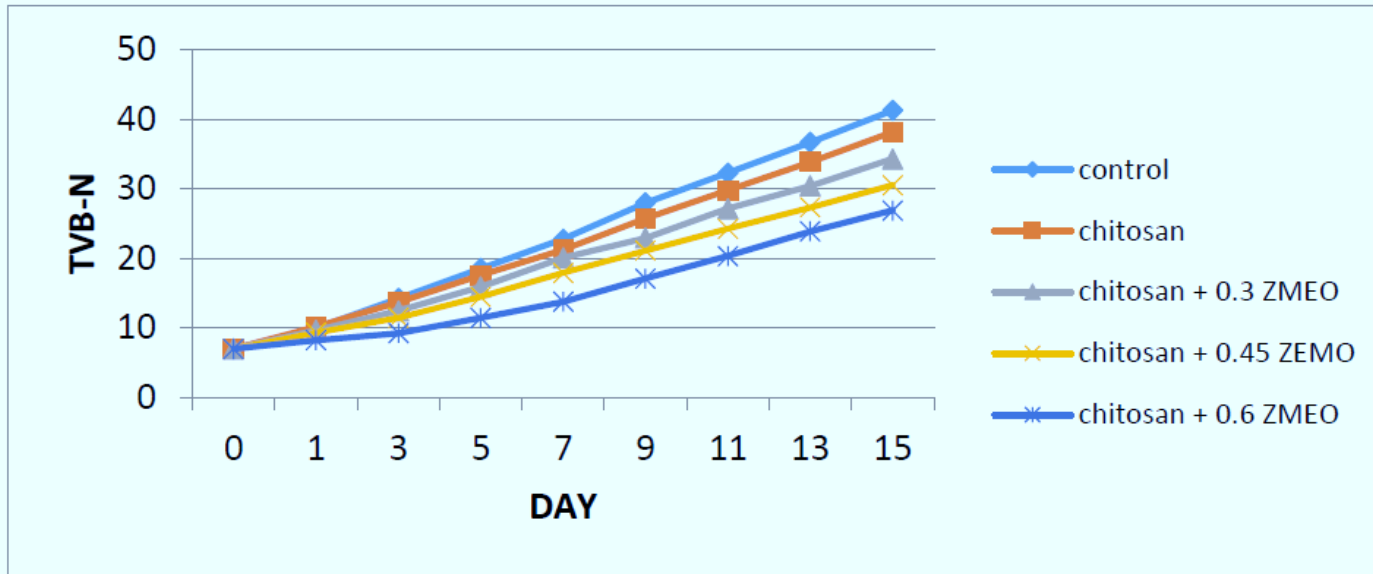


Figure 2. Changes in TVB-N values of fish samples during chilled storage.

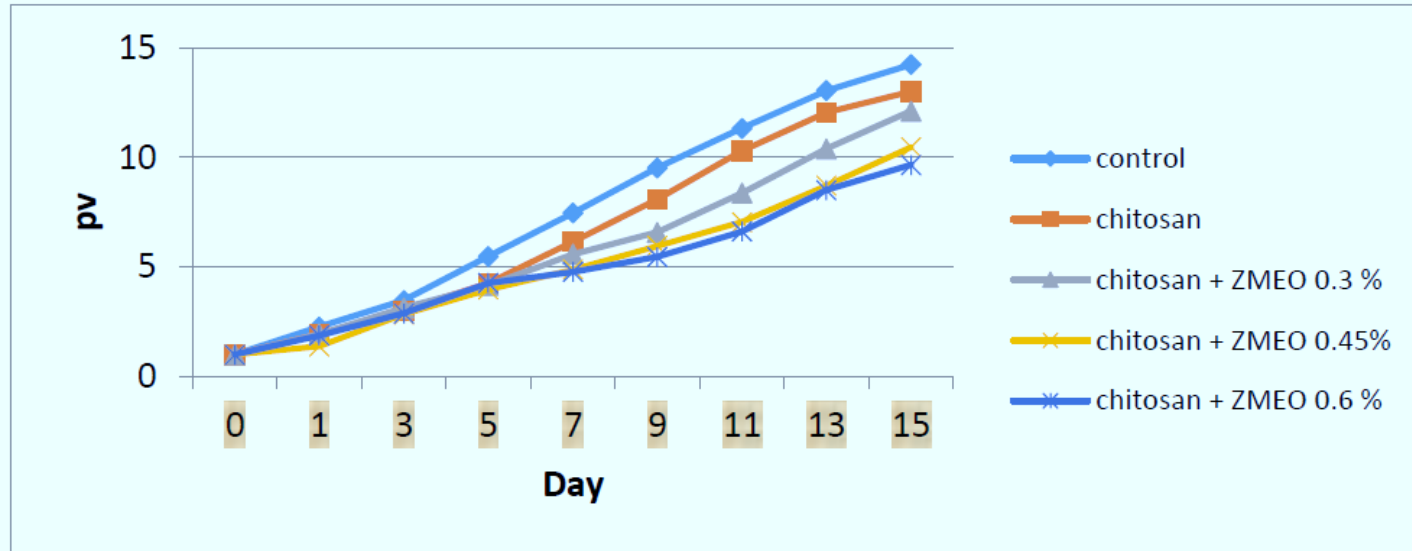


Figure 3. Changes in PV values of the fish sample during chilled storage

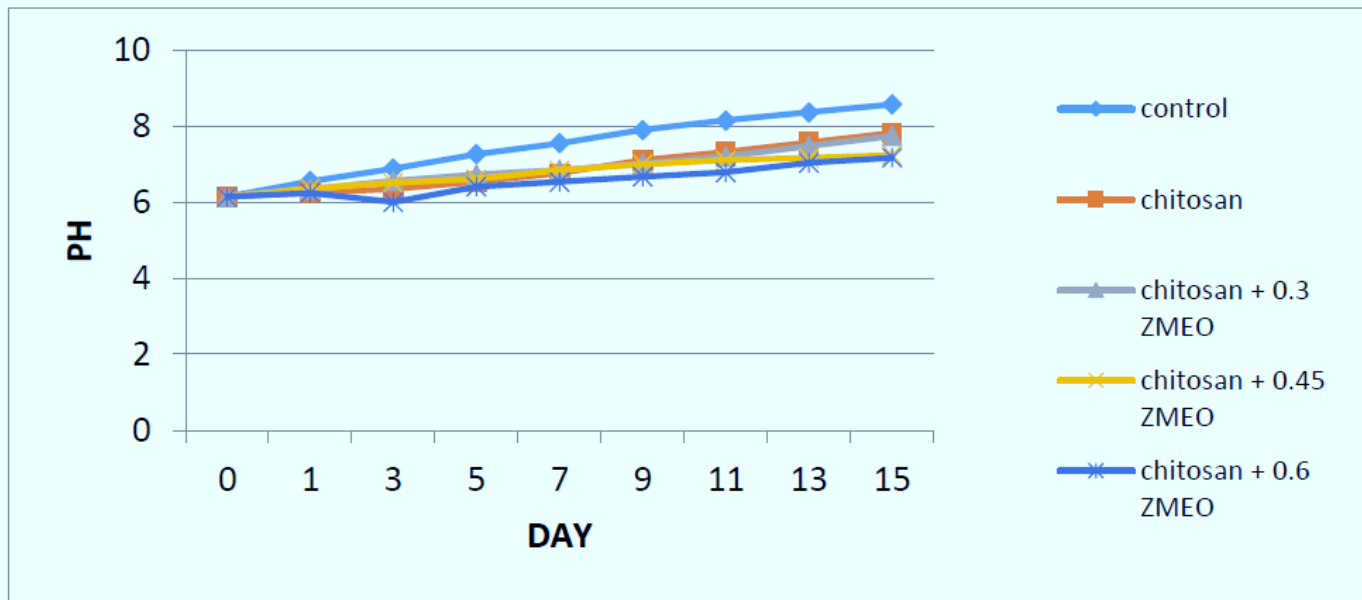


Figure 4. Changes in pH values of fish samples during chilled storage

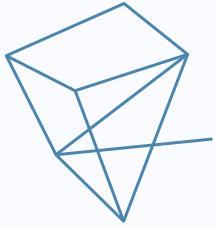


بحث

ترکیب شیمیایی اسانس آویشن (ZMEO) تحت تأثیر عوامل متعددی مانند فصل برداشت، خاک و روش استخراج قرار دارد. نتایج ترکیب تقریبی با تحقیق اصغرزاده و همکاران مطابقت داشت، اگرچه پروتئین در نمونه‌های کنترل این مطالعه کمتر بود.

در این پژوهش، از ترکیب کیتوزان و ZMEO برای تیمار فیله‌های کپور نقره‌ای استفاده شد. این ترکیب خواص ضد باکتریایی نشان داد و ماندگاری فیله‌ها را در یخچال تا ۱۵ روز افزایش داد. این یافته با تحقیقات گذشته درباره پوشش‌های حاوی روغن‌های ضروری مطابقت دارد.

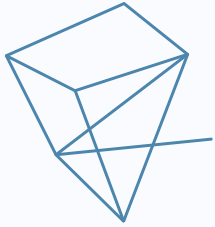
بحث



فعالیت آنتی‌اکسیدانی پوشش نیز به دلیل وجود ترکیباتی مانند تیمول و کارواکرول تأیید شد. که با یافته‌های خنجری و همکاران مشابهت دارد.

مطابق استاندارد ICMSF(22)، حد مجاز باکتری‌ها $7 \log \text{CFU/g}$ است. تفاوت‌های مشاهده شده از نظر آماری معنادار بود ($P < 0.05$).

جمعیت اولیه انتروباکتریاسه (EBC) در نمونه‌ها $2.49 \log \text{CFU/g}$ بود. تیمار با کیتوزان و ZMEO باعث کاهش چشمگیر ۱ تا ۳ واحدی لگاریتمی (\log) این جمعیت در مقایسه با نمونه کنترل شد. در روز پانزدهم، جمعیت در نمونه کنترل به $8.15 \log \text{CFU/g}$ رسید، در حالی که در نمونه تیمار شده با کیتوزان حاوی ۰/۶٪ ZMEO، این مقدار $5.15 \log \text{CFU/g}$ بود (شکل ۶). این کاهش را می‌توان به خاصیت ضد باکتریایی این ترکیبات نسبت داد.



نتیجه‌گیری

کاربرد پوشش کیتوزان حاوی اسانس آویشن (ZMEO) بر روی فیله‌های کپور نقره‌ای:

- منجر به کند شدن رشد میکروارگانیسم‌ها می‌شود
- واکنش‌های اکسیداتیو را در بافت گوشت مهار می‌کند
- مدت زمان نگهداری در یخچال را افزایش می‌دهد

این اثرات ناشی از وجود ترکیبات طبیعی ضد میکروب و آنتی‌اکسیدان در پوشش است. برای استفاده تجاری، مطالعات آینده باید بر بهبود پایداری و ویژگی‌های فیزیکی پوشش تمرکز کنند.

به طور خلاصه، این پوشش پتانسیل بالایی برای استفاده به عنوان یک عامل ضد میکروبی مؤثر در محصولات غذایی، به ویژه فرآورده‌های شیلاتی دارد.



آشنایی با ژورنال





JOURNAL OF NUTRITION FASTING AND HEALTH



JOURNAL OF NUTRITION
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JOURNAL OF NUTRITION
FASTING AND HEALTH

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The **Journal of Nutrition, Fasting and Health** is a peer-reviewed open access journal.

Journal of Nutrition, Fasting and Health, Abbreviated as **J Nutr Fast Health**, provides publication of articles quarterly in a different areas of nutrition such as clinical nutrition, community nutrition, public health nutrition, nutrigenomics, molecular nutrition, nutritional biochemistry, food health and safety, nutrition physiology, sport nutrition, health sciences, ERAS and also all aspects of fasting in health and diseases such as intermittent fasting, fasting health benefits.

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دارای رتبه علمی-پژوهشی از وزارت بهداشت درمان و آموزش پزشکی

Current Issue: Volume 13, Issue 4, October 2025, Pages 216-292

Case Report

Improvement of Bone Mineral Density with Mena7 360 Mcg: a Case Report

Pages 216-220

Area: Health Sciences, Pub: Med Sciences

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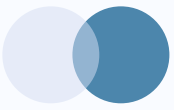
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Journal of Nutrition, Fasting and Health

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| View Per Article | 3942.39 |
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Indexing and Abstracting

ISC

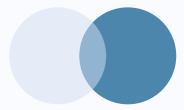
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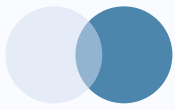


Index Copernicus

HINARI

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Aims and scope of the Journal of Nutrition, Fasting and Health

The Journal of Nutrition, Fasting and Health (JNFH) is a peer review English language journal that provides publication of articles in a different areas of nutrition and such as:

- Clinical nutrition
- Community nutrition
- Public health nutrition
- Nutrigenomics
- Molecular nutrition
- Nutritional biochemistry
- Food health and safety
- Sport nutrition
- Nutrition physiology
- ERAS (Enhanced Recovery After Surgery)
- Food Security
- Health Sciences

and also all aspects of fasting in health and diseases such as:

- prolonged fasting, fasting and weight loss, fasting therapy, intermittent fasting, preoperative fasting, postprandial fasting and water fasting



Preparation of Manuscript

Manuscripts are returned to the authors in case any of the following instructions are not followed by the authors.

Solid English language should be used in the entire manuscript. Manuscripts should be typewritten and in DOUBLE-SPACE format with 1-inch (2.5 cm) margin. Manuscript pages should be consecutively numbers starting from the title page. Manuscripts should be organized into sections with following headings; Title page, Abstract, Keywords, Introduction, Materials and Methods, Results, Discussion, Conclusion, Acknowledgements, references, Figure legends, and Tables.

Discussion

The chemical composition of ZMEO is influenced by various elements, including the season of harvest, the age of the plant, the soil it grows in, the geographical location, the climate conditions, the species, the method of herb drying, and the extraction method (19).

References

1. Fan W, Sun J, Chen Y, Qiu J, Zhang Y, Chi Y. Effects of chitosan coating on quality and shelf life of silver carp during frozen storage. Food Chemistry. 2009; 115(1):66-70.

Conclusion

According to the findings of this research, applying a layer of chitosan infused with ZMEO onto silver carp fillets. Slows down the growth of microorganisms and unwanted chemical reactions in meat, hence increasing the amount of time silver carp fillets can be stored in refrigerated conditions. The capabilities of ZMEO and chitosan can be attributed to the presence of natural components that function as antioxidants and antimicrobials. Future studies should focus on enhancing the stability and hardness of the coating's physical qualities to make it suitable for commercial application. In summary, this coating has the potential to serve as an effective antimicrobial agent that can be utilized to prevent the growth of undesirable microbes in food products, particularly those derived from fish and shellfish.



Abstract:

Abstracts should be self-explanatory and follow the Medline limitation. Abstracts should not exceed 250 words. Title and scope of the manuscript, significant statistical findings, major findings and conclusions should be stated in the abstract. Abstracts should be prepared using complete sentences in the past tense and by using third person is sentences. Abstracts use standard nomenclature and should not include abbreviations and reference citations. Original papers should have structures abstracts with following sections:

Introduction: Introduction should be brief and include the aim or research question or study hypothesis that is tested in the study.

Methods: Methods should include the basic design, study duration, and statistical methods of the study.

Results: Results should be prepared in narrative form and include the main findings of the study. Measurements and other information should be defined in the results section. Findings should be presented using statistical significance level and reporting other statistical parameters.

Conclusion(s): Conclusions that are directly derived from study findings should be stated in the conclusions section. This section should include the implications of the study findings.

Keywords: Abstracts should be immediately followed by 3 to 6 keywords based on the Medical Subject Headings (MeSH) list. Keywords should not be duplicated title words. Keywords should be chosen from the text and sorted alphabetically.

ARTICLE INFO

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Zataria multiflor
Boiss.
Silver carp
Shelf life
Essential oil

ABSTRACT

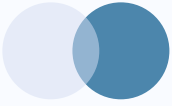
Introduction: This study examined the impact of a chitosan coating with *Zataria multiflora* Boiss essential oil (ZMEO) as an antioxidant and antibacterial agent on the quality and shelf life of silver carp fillets when stored in the refrigerator.

Methods: The ZMEO-enriched chitosan coating was prepared at concentrations of 0%, 0.3%, 0.45% and 0.6%. After packaging the filet sample, the antioxidant and antibacterial properties were evaluated and analyzed after 0, 1, 3, 5, 7, 9, 11, 13, and 15 days at 4 °C temperature.

Results: At day 15, the chitosan coating + 0.6% ZMEO resulted in considerably reduced numbers of TVC (total viable counts), EBC (Enterobacteriaceae), and LAB (lactic acid bacteria) compared to other treatments ($p < 0.05$). The log CFU/g counts for TVC, EBC, and LAB were 4.78, 5.15, and 4.78, respectively. The application of chitosan coating + 0.6% ZMEO effectively suppressed the increase in thiobarbituric acid (TBA), total volatile elemental nitrogen (TVB-N), peroxide value (PV), and pH in the silver carp fillets. The findings showed that applying a chitosan coating with ZMEO can prolong the nutritional value of silver carp fillets when stored in refrigerated circumstances for a maximum of 15 days, without causing any negative changes in taste, smell, or texture.

Conclusions: In summary, our work has demonstrated that a chitosan coating enhanced with ZMEO has the potential to effectively prolong the shelf life of silver carp fillets when stored in refrigerated conditions.





Structure of Text:

Introduction: Should include study context.

Materials and Methods: Should include detailed information so that the reader could reproduce the study. Authors should cite the original manuscript in case the study methodology was previously published and only indicate modifications they made in the study methodology. Ethical approval should be mentioned in the Materials and Methods section.

Results: The findings of the study should be clearly and concisely stated. Reported results in text should not be repeated in tables and vice versa.

Discussion: Discussions should be prepared in relation to the study findings using current literature. It is also appropriate if authors combine the results and discussion. Discussions should be kept brief with the minimum possible citations.

Conclusion: Should include the summary of the study findings.

Measurement of the Total Amount of Nitrogen in a Sample That Exists in a Volatile Form (TVB_N)

The fish samples were analyzed for their total volatile basic nitrogen (TVB-N) content using the method previously outlined by Jouki et al (13).

Materials and Methods

Zataria Multiflora Boiss. Extraction of the Essential Oil

ZMEO was obtained from Fars Province, Iran, and authenticated by the Institute of Medicinal Plants, Tehran University of Medical Sciences, Iran. The plant portions that had been dried in the air were subjected to steam distillation for a duration of three hours using a Clevenger-type equipment (7). The components of the ZMEO were analyzed using gas chromatography-mass spectrometry [GC-MS], following the method published by Moosavy et al (8).



References:

References should be consecutively numbered in tests in order of first appearance. Reference numbers should be in square brackets. All cited references in the manuscript text should appear in the end of the manuscript in a separate page. Conversely all references presented in the reference list should be cited in manuscript text. References should be formatted using the **Vancouver style**. In the reference list, the first 6 authors should be cited and for references with more than 6 authors, the first 6 authors should be cited followed by et al. The page range should be cited for references of more than one page.

References

1. Fan W, Sun J, Chen Y, Qiu J, Zhang Y, Chi Y. Effects of chitosan coating on quality and shelf life of silver carp during frozen storage. Food Chemistry. 2009; 115(1):66-70.

Figures:

Figure legends: All figures should have a figure legend. Figure legends should be prepared to be brief and should not be duplicates of manuscript text. Figures should have a title and an explanatory legend. The explanatory legend should encompass the titles and figure title should not appear in the figure itself. Figure legends should be placed in a separate page after references at the end of the manuscript. The number of the figure to which the figure legend is attributed should be presented at the beginning of the figure legend. Figure legend should explain symbols, abbreviations and statistical information used in the figure. Electron micrographs should be accompanied with scale markers with the type of staining indicated in figure legend.

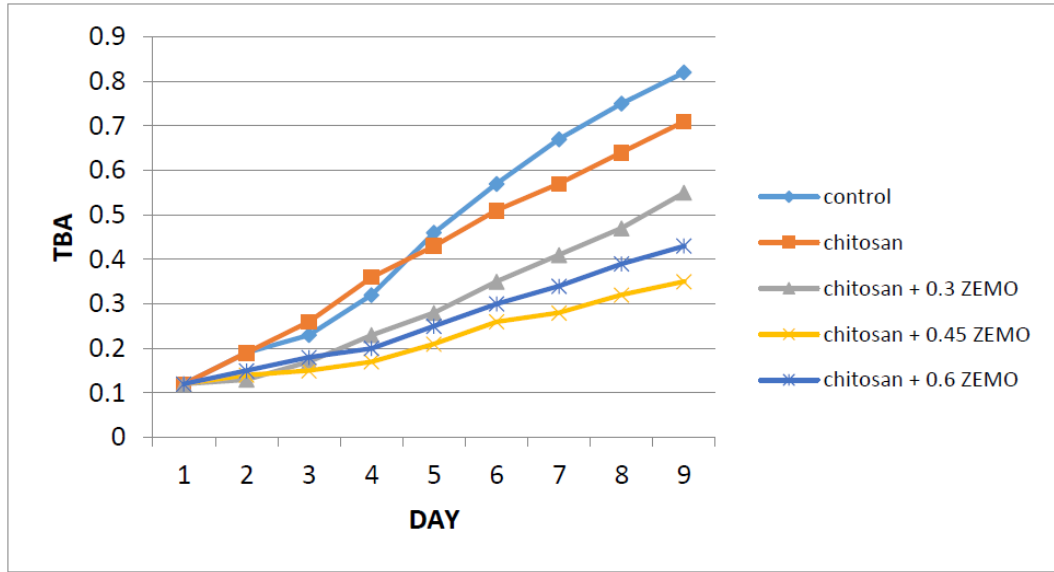
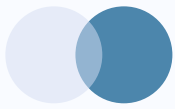


Figure 1. Changes in TBA values of the fish sample during chilled





Tables:

Tables should be consecutively cited and numbered in text. Each table should be prepared on a separate page and should be accompanied with a title, appropriate column heads, and legends. Definition of the abbreviations in the table should be explained in the table legend. Tables should not be placed in the manuscript text body. Tables should be self-explanatory and should not be a duplicate of manuscript text.

*For short communication, follow the instructions for original articles, except that the total word number of the main text (excluding references, tables and figure legends) is limited to 2000 with no more than 2 figures and/or tables and no more than 15 references. An abstract, not exceeding 150 words, should be presented at the beginning of the article.

Table 1. Median rating for sensory properties of fish fillets as affected by different concentrations of ZEMO

| combination | Odor | Taste | Overall acceptability |
|------------------------|------|-------|-----------------------|
| control | 7 | 7 | 7 |
| Chitosan | 7 | 8 | 7 |
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| Chitosan + 0.6 % ZEMO | 6 | 4 | 4 |

Means followed by the same letters are not significantly ($P < 0.05$) different.

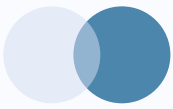
SD: standard deviation.





نقد ساختاری مقاله





Effects of Active Packaging Based on Chitosan Biopolymer Coating Enriched with *Zataria multiflora* Essential Oil on Shelf Life of Silver Carp during Chilled Storage

ایرادات ساختاری:

- عدم درج نام کامل در عنوان (Boiss)
- ایتالیک بودن Essential





Introduction: This study examined the impact of a chitosan coating with *Zataria multiflora* Boiss essential oil (ZMEO) as an antioxidant and antibacterial agent on the quality and shelf life of silver carp fillets when stored in the refrigerator.

Methods: The ZMEO-enriched chitosan coating was prepared at concentrations of 0%, 0.3%, 0.45% and 0.6%. After packaging the filet sample, the antioxidant and antibacterial properties were evaluated and analyzed after 0, 1, 3, 5, 7, 9, 11, 13, and 15 days at 4 °C temperature.

ایرادات ساختاری:

- نام خاص باید ایتالیک باشد.
- قبل and باید ویرگول بزاره.
- باید s جمع بگیرد: Samples
- وقتی C آورده دیگه نیازی به temperature نیست، باید حذف شود.



Keywords:

Chitosan coating
Zataria multiflora

Boiss.

Silver carp

Shelf life

Essential oil

Results: At day 15, the chitosan coating + 0.6% ZMEO resulted in considerably reduced numbers of TVC (total viable counts), EBC (Enterobacteriaceae), and LAB (lactic acid bacteria) compared to other treatments ($p < 0.05$). The log CFU/g counts for TVC, EBC, and LAB were 4.78, 5.15, and 4.78, respectively. The application of chitosan coating + 0.6% ZMEO effectively suppressed the increase in thiobarbituric acid (TBA), total volatile elemental nitrogen (TVB-N), peroxide value (PV), and pH in the silver carp fillets. The findings showed that applying a chitosan coating with ZMEO can prolong the nutritional value of silver carp fillets when stored in refrigerated circumstances for a maximum of 15 days, without causing any negative changes in taste, smell, or texture.

Conclusions: In summary, our work has demonstrated that a chitosan coating enhanced with ZMEO has the potential to effectively prolong the shelf life of silver carp fillets when stored in refrigerated conditions.

ایرادات ساختاری:

- on day 15
- اصطلاح با مخفف همخوانی ندارد: total volatile basic nitrogen
- بهتر بود بنویسه condition
- for up to 15 days
- بهتر بود study یا research نوشته میشد
- باید در ادامه نام گیاه می امد و ایتالیک می شد، آخرش نقطه ندارد!

Introduction

The production and consumption of silver carp (*Hypophthalmichthys molitrix*) has increased in many countries in recent decades. The silver carp holds significant importance in the polyculture system of eastern countries.

Whole fresh fish and fillets are the main forms sold in the markets (1). Fish meat is highly perishable both microbiologically and chemically due to its high nutritional value, aw and pH (2, 3). The primary obstacle faced by the seafood sector is to prolong the shelf life and enhance the overall acceptability of **fish meat** by inhibiting lipid oxidation and avoiding microbial proliferation (4). Edible coatings in active packaging are a potential technology that integrates food, packaging, and preservation into a unified idea.

ایرادات ساختاری:

- اسم علمی اشتباه تایپی دارد:
- استفاده از fish meat دقیق و علمی نیست؛ بهتر است از fish products یا fish fillets استفاده شود.

by-products of the food industry or underutilized sources of lipids, polysaccharides, or proteins to develop an efficient system that maintains food quality during its storage period. Edible coatings are thin films, either in solid or liquid form that are applied to food. They serve to prolong the shelf life and enhance the quality of food by influencing characteristics such as diffusibility, permeability, and trait retention (5).

Numerous studies have demonstrated the antibacterial, antioxidant, antiviral and antifungal properties of essential oils extracted from various parts of plants. The properties of an essential oil are determined by the number of phenolic chemicals it contains, including eugenol, thymol, and carvacrol (6).

Zataria multiflora, sometimes known as Shirazi thyme, belongs to the Lamiaceae family.

ایرادات ساختاری:

- قبل از and باید ویرگول بیاید طبق فرمت آکادمیک Oxford comma
- نام علمی باید ایتالیک باشد: *Zataria multiflora*

It is widely used in Iran as a flavoring ingredient in many types of food and for some medicinal purposes. The essential oil of this plant is rich in carvacrol and thymol. Carvacrol and thymol are the primary phenolic molecules that contribute to the antibacterial and antioxidant properties of ZMEO (7). The US FDA classifies phenolic chemicals, including carvacrol and thymol, as GRAS (generally regarded as safe) (6).

Some studies have shown that edible coatings of seafood with chitosan infused with antimicrobial or antioxidant agents help prolong the duration of freshness for these products (1).

The purpose of this study was to investigate the impact of combining chitosan with ZMEO on the longevity of fresh silver carp fillets while stored at a temperature of 4°C.

ایرادات ساختاری:

- عبارت phenolic chemicals بهتر است با واژه تخصصی تر phenolic compounds جایگزین شود.
- مخفف ZMEO باید در اولین کاربرد تعریف شود، اما اینجا قبلاً تعریف نشده بود.
- "US FDA" بهتر است به صورت کامل در اولین اشاره نوش_____ته ش_____ود:
United States Food and Drug Administration (FDA)
- longevity بهتر است با اصطلاح علمی تر مثل shelf life جایگزین شود.

Materials and Methods

Zataria Multiflora Boiss. Extraction of the Essential Oil

ZMEO was obtained from Fars Province, Iran, and authenticated by the Institute of Medicinal Plants, Tehran University of Medical Sciences, Iran. The plant portions that had been dried in the air were subjected to steam distillation for a duration of three hours using a Clevenger-type equipment (7). The components of the ZMEO were analyzed using gas chromatography-mass spectrometry [GC-MS], following the method published by Moosavy et al (8).

ایرادات ساختاری:

- نام علمی باید ایتالیک باشد: *Zataria multiflora* Boiss
- Boiss مخفف نام گیاهشناس است و معمولاً در ایتالیک قرار نمی‌گیرد.

Preparation of the Chitosan Coating Solution

The chitosan solution with a high molecular weight was made in the following manner: A solution was prepared by dissolving 2 grams of chitosan in 100 cubic centimeters of 1% acetic acid (volume/volume) and stirring it for three hours at room temperature. A plasticizer, glycerol, was introduced to the solution at a rate of 0.75 ml per gram of chitosan weight. The mixture was then agitated for a duration of 10 minutes. In order to exclude solid particles that had not dissolved, the chitosan coating solution was passed through a Whatman No.3 filter paper. The chitosan solution was supplemented with Tween 80 at a concentration of 0.25% v/v, along with different concentrations of ZMEO (0%, 0.3%, 0.45%, and 0.6% v/v). The solution was uniformly mixed under sterile conditions at a speed of 21600 revolutions per minute for a duration of 1 minute (9).

ایرادات ساختاری:

- cubic centimeters نباید نوشته شود؛ باید mL باشد.
- ml باید به صورت mL نوشته شود.
- Ratio تغییر به



Preparation and Processing of the Fish Samples

Silver carp (*Hypophthalmichthys molitrix*) were captured in a fish farm in Tehran Province, with an average weight of 1000 grams and an average length of 350 millimeters. The fish were transported to the meat science laboratory of the Food Hygiene Department at Tehran University and subsequently divided into square pieces weighing approximately 60 grams each. The fat and protein content percentages of three fillets were analyzed using the Association of Analytical Communities (10) method.

The fillets were immersed for 30 seconds in 500 cc of different coating solutions, including A (control samples, distilled water), B (chitosan), C (chitosan enriched with 0.3% ZMEO), D (chitosan enriched with 0.45% ZMEO) and E (chitosan enriched with 0.6% ZMEO). The fish fillets were extracted and left to drain for 30 minutes at a temperature of 25°C in a microbiological hood to create the coatings. Subsequently, they were stored at a temperature of 4±1°C (9).

The shelf life of the fish was determined by conducting microbiological, chemical, and sensory investigations at eight specific time intervals (0, 1, 3, 5, 7, 9, 11, 13, and 15 days).

ایرادات ساختاری:

- نام علمی باید ایتالیک باشد.
- واحدها باید خلاصه شوند g، mm
- 500cc باید به صورت 500mL نوشته شود.
- ویرگول قبل از E and باید اضافه شود.
- in 15 days (every other days)

Bacteriological Examination

Bacteriological values were assessed by combining a 10 g sample of each treatment with 90 cc of 0.1% peptone water in sterile bags and homogenizing the mixture using a stomacher for 2 minutes. Subsequent decimal dilutions were made and subsequently plated onto the suitable medium. The TVC was calculated using the surface plate method and the plate counting agar PCA. The plates that were treated with a little amount of a substance to prevent the growth of microorganisms were thereafter placed in a controlled environment at a temperature of 30°C for a duration of three days (11, 12).

The enumeration of Enterobacteriaceae (EBC) was conducted on violet red bile glucose agar (VRBGA) using the pour plate method. The plates were subsequently infected at a temperature of 25 °C for a duration of 48 hours. The enumeration of lactic acid bacteria (LAB) was performed on de Man Rogosa Sharpe Agar (MRS) through incubation at a temperature of 30 °C for a period of 2-3 days (12). The microbiological data were transformed into logarithmic values representing the number of colony-forming units (CFU/g).

ایرادات ساختاری:

- cc باید به mL اصلاح شود.
- نام محیط باید دقیق نوشته شود: plate count agar (PCA)
- عبارت " "plate counting agar" اشتباه است
- بهتر است TVC یکبار کامل تعریف شود
- EBC :entrobacteriaceae count
- واژه infected کاملاً اشتباه است (باید incubated باشد).
- "representing the number of" تکراری است چون CFU/g خودش مفهوم را می‌رساند.

Chemical Analyses

Protein and Lipid Content

The Kjeldahl technique was used to determine the total crude protein. This technique employs a three-step process to measure the amount of protein: digestion, distillation, and titration. The organic substance is digested by employing concentrated H₂SO₄, heat, K₂SO₄ (to elevate the boiling point), and a catalyst (such as selenium) to expedite the reaction. The lipid content of the fillets was evaluated using the method developed by the Fraunhofer Institute for Molecular Biology and Applied Ecology (10). The lipids were extracted from the fillets by homogenizing them in a mixture of chloroform and methanol (at a ratio of 2:1, volume to volume). The amount of lipids was then determined by measuring the weight of the extracted lipids, and this process was repeated three times for accuracy.

ایرادات ساختاری:

- بهتر است گفته شود Kjeldahl method نه technique
- اضافه گویی در بند اول
- منبع ۱۰ به این محل ارجاع نخورده



Measurement of the Total Amount of Nitrogen in a Sample That Exists in a Volatile Form (TVB_N)

The fish samples were analyzed for their total volatile basic nitrogen (TVB-N) content using the method previously outlined by Jouki et al (13). The fish muscles were blended in 200 cc of a 7.5% water-based solution of TCA (trichloroacetic acid). The homogenate underwent centrifugation at a force of 400 times the acceleration due to gravity for a duration of 5 minutes. The resulting liquid above the sediment was then passed through a Buchner funnel equipped with a Whatman filter paper No. 3. The process of steam distillation was conducted using a distiller of the Kjeldahl type. Twenty-five milliliters of the filtrate were introduced into the distillation tube, and then 5 ml of a 10% NaOH solution were added. A 10 ml beaker containing a 4% aqueous solution of boric acid and 0.04 ml of methyl red and bromocresol green indicator was positioned near the condenser's endpoint for the ammonia titration. The distillation process was carried out until the volume in the beaker reached a final measurement of 50 ml, with 40 ml of distillate collected. The boric acid solution underwent a color change to green when it was made alkaline by the addition of distilled TVBN. The titration was performed using a 0.01 ml microburette filled with a 0.1 N aqueous solution of sulphuric acid.

ایرادات ساختاری:

- اندازه گیری TVBN مینوشت کافی بود.
- نقطه گذاری قبل از رفرنس رعایت نشده.
- fillets

Determination of the Peroxide Value (PV)

The peroxide content was quantified in the whole lipid extracts using Pearson's technique (14). A total of 0.3 grams of the sample was carefully deposited into a stoppered flask with a capacity of 250 milliliters. The solvent was eliminated using a rotary evaporator under reduced pressure at a temperature of 40°C (equivalent to the temperature of a water bath). A volume of 10 ml of the acetic acid/chloroform combination with a ratio of 3/2 was added, and the lipids were dissolved through agitation. Next, 1 milliliter of a concentrated chlorine solution was added and the mixture was left undisturbed in a dark environment for a duration of 5 minutes. Dispense 20 milliliters of distilled water and agitate. Perform a titration of the liberated iodine using a 0.01 N sodium thiosulfate solution until the solution turns a pale yellow color. Introduce 1 milliliter of a 1.5% starch solution into the mixture as an indicator, and continue the titration process until the solution becomes colorless. Conduct blank tests in a same manner, excluding any fats. Utilize a conventional formula to denote the peroxide value.

ایرادات ساختاری:

- کلا متن اضافات هست و از نظر نگارشی ایراد دارد.
- بایستی فرمول نوشته نشود.



Quantification of Thiobarbituric Acid Reactive Substances (TBARS)

The thiobarbituric acid value was measured using calorimetry, following the method described by Porkony and Dieffenbacher, as outlined by Kirk and Sawyer (15). The biological sample underwent acid digestion and was subsequently separated using targeted reflux distillation. The chemical substances present in the distillate were determined by spectrophotometry measurements at a wavelength of 538 nm, following their interaction with 2-thiobarbituric acid.

Sensory Evaluation

An acceptance test was conducted on fried fish fillet samples to analyze the sensory effects obtained from them. A panel test was conducted to evaluate the fish fillets from each treatment of chitosan enriched with varying concentrations of ZMEO (0, 0.3, 0.45, and 0.6% v/v) and a control group. The panel consisted of 50 untrained judges who were members of the Faculty of Veterinary Medicine at the University of Tehran. The panelists assessed the samples by assigning ratings on a 9-point scale, where 9 indicates a remarkable liking, 8 indicates a strong liking, 7

ایرادات ساختاری:

- به جای judges بهتر است panelists
- به جای 9-point scale بهتر است hedonic



indicates a moderate liking, 6 indicates a slight liking, 5 indicates neither liking nor disliking, 4 indicates a slight disliking, 3 indicates a moderate disliking, 2 indicates a strong disliking, and 1 indicates an intense disliking. These ratings were given for different attributes such as appearance, color, odor, and flavor.

Statistical Analysis

The statistical analysis was conducted using SPSS 16. The data underwent analysis using one-way ANOVA, followed by **Turkey's** multiple comparison test. The values are presented as the mean together with their corresponding standard deviation for all outcomes. Statistical significance was determined at a p-value of less than 0.05.

ایرادات ساختاری:

- میتونست به صورت جدولی بیاره یا اصلا استفاده نکند
- نام آزمون اشتباه است؛ باید Tukey's باشد، نه Turkey's.

ZMEO's Chemical Components

The gas chromatography-mass spectrometry examination revealed that the major constituents of the ZMEO were carvacrol (73.65%), thymol (4.31%), Trans-caryophyllene (2.93%), and Gamma-terpinene (2.27%). The remaining chemicals included linalool (1.28%), thymol methyl ether (0.62%), carvacrol terpinene-4-ol (1.7%), and 1, 8-cineole (0.52%).

Protein and Lipid Content

Protein and lipid contents were measured $13.97 \pm 0.59 \%$ and $5.12 \pm 0.48 \%$, respectively.

Bacteriological Changes

The alterations in bacterial composition (total viable count, Enterobacteriaceae count, and lactic acid bacteria count) of silver carp fillets treated with various concentrations of ZMEO (0, 0.3, 0.45, and 0.6 % v/v) throughout a 15-day period of refrigerated storage are depicted in Figure 5-7.

ایرادات ساختاری:

- ترتیب نتایج با ترتیب جداول و نمودارها یکسان نیست!
- بهتر است در پایان هر نوشتار نتیجه شکل آن گذاشته شود نه اینکه تمام نتایج را بنویسد و بعد همه اشکال پشت سر هم آورده شود.
- نباید از ۵ شروع بشه. باید ترتیب نتایج و نمودارها یکسان و از ۱ شروع شود.



Figure 5 displays the variations in TVC (Total Viable Count) of silver carp fillets for each treatment. The initial total viable counts of fresh silver carp fillets were 2.97 ± 0.02 log cfu/g. The total volatile compounds (TVC) exhibited an increase in all treatments when stored at a temperature of 4°C. The count of mesophilic bacteria in the treated samples was considerably lower ($P < 0.05$) compared to the control group. The total viable count (TVC) of microorganisms in the control sample of fresh fish reached a value of 7 log cfu/g, which is the upper limit set by Salam (12), between days 13 to 15. The use of chitosan enriched with ZMEO (at concentrations of 0, 0.3, 0.45, and 0.6 %v/v) as a coating for silver carp fillets led to an increase in the fillet's shelf-life.

According to Figure 6, the initial count of Enterobacteriaceae (EBC) was 2.49 log cfu/gr. The treated samples had a significantly lower EBC count compared to the control sample, with a p-value of less than 0.5. The initial count of lactic acid bacteria (LAB) in the silver carp fillets was 2.58 log cfu/g on the first day of the investigation (Figure 7). The application of chitosan with varying quantities of essential oils resulted in a significant reduction in the count of lactic acid bacteria ($P < 0.05$).

ایرادات ساختاری:

- غلط نوشتاری
- دوباره گویی غیر ضروری: TVC و Total Viable Count هر دو ذکر شده
- log cfu/g باید به صورت استاندارد و یک دست نوشته شود:
log CFU/g حروف بزرگ برای CFU
- Significantly
- ۰/۰۵

Thiobarbituric Acid Value

As shown in Figure 1, the initial TBA value of silver carp fillets were 0.12 ± 0.20 and increased to 0.82 for control samples by the end of the storage period. By contrast, the value reached 0.43 for samples that were treated with chitosan containing 0.6% ZMEO.

Total Volatile Elemental Nitrogen (TVB-N)

Figure 2 displays the variations in TVB-N levels in silver carp fillets. The initial TVB-N value of the samples was 7 ± 0.39 mg of nitrogen per 100 grams. The TVB-N values exhibited a progressive increase throughout the investigation for all treatments, eventually reaching 41.25 and 26.87 mg nitrogen/100 g for the control group and the samples coated with chitosan integrated by 0.6% ZMEO, respectively.

The total volatile base nitrogen (TVB-N) is utilized to ascertain the existence of nitrogenous substances produced by proteolytic bacteria (16).

The TVN (Total Volatile Nitrogen) of the treated samples exhibited a decreased value compared to the control samples. The antimicrobial activity of essential oil and chitosan may be the cause. This finding is consistent with the results of prior research studies (1, 9, 13).

ایرادات ساختاری:

- واحد درصد یا mg MDA/kg ذکر نشده
- لحن توضیحی است نه گزارش نتایج < این جمله باید در Discussion یا Introduction باشد، نه Results
- بر عکس اینجا معنی نداره! باید مینوشت while یا however

Peroxide Value (PV)

The PV value index is a useful tool for quantifying the extent of primary lipid oxidation in seafood. Figure 3 displays the PV values data. The peroxide value (PV) of the samples was 0.97 milliequivalents of oxygen per kilogram (meq O₂/kg) on day 0. The study period revealed a decrease in PV values in samples that were coated with chitosan containing ZMEO, as shown in **figure 3**. The PV values exhibited a substantial rise ($P < 0.05$) as the storage time at 4°C progressed for all treatments. There was a statistically significant decrease ($P < 0.05$) in the PV value of the treated samples compared to the untreated samples during the storage period at 4°C.

The study shown that the use of chitosan supplemented with ZMEO effectively delays the occurrence of primary lipid oxidation. This finding is consistent with the findings of Ojagh et al (9) and Mexis et al (18). ZMEO's substantial protective impact can be attributed to its carvacrol and thymol concentration, as well as its antioxidant properties in scavenging free radicals (18).

ایرادات ساختاری:

- "figure" باید با حرف بزرگ باشد. (Figure 3)
- جاش توی بحث مقاله هست

pH

The average initial pH of the fish samples was determined to be 6.14. Figure 4 displays the alterations in pH levels of fish fillets over the period of refrigerated storage.

The pH values of the control samples and the samples coated with chitosan integrated by 0.6 ZMEO were 8.57 and 7.17, respectively, at the end of the investigation.

ایرادات ساختاری:
توی متد اصلا در مورد pH حرف نزده!
علامت % یادش رفته

Sensory Analysis

Table 1 displays the median scores for odor, taste, and acceptability of the ZEMO fish fillet samples. The fillet treated with chitosan mixed with 0.3% EO for 15 days of storage was the most favored sample. The fillet treated with chitosan combined with 0.3% EO during 15 days of storage was the most preferred sample. The evaluation of odor, color, and general acceptability of silver carp fillet samples were found in Table 1. According to the findings of this investigation, applying a coating of chitosan with 0.3% ZMEO resulted in the required sensory qualities.

ایرادات ساختاری:

- اختصار به اشتباه ZEMO نوشته شده
- اختصارها باید یکسان باشند. به جای ZMEO از EO استفاده شده.

Table 1. Median rating for sensory properties of fish fillets as affected by different concentrations of ZEMO

| combination | Odor | Taste | Overall acceptability |
|------------------------|------|-------|-----------------------|
| control | 7 | 7 | 7 |
| Chitosan | 7 | 8 | 7 |
| Chitosan + 0.3 % ZEMO | 8 | 8 | 8 |
| Chitosan + 0.45 % ZEMO | 8 | 6 | 6 |
| Chitosan + 0.6 % ZEMO | 6 | 4 | 4 |

Means followed by the same letters are not significantly ($P < 0.05$) different.

SD: standard deviation.

ایرادات ساختاری:

- عدم یکنواختی مخفف: ZEMO نوشته شده، باید ZMEO باشد.
- "properties" غلط املائی است. شکل صحیح properties است.
- در Methods گفته شده که ویژگی‌ها شامل: appearance, color, odor, flavor, overall acceptability هستند.
- اما در جدول فقط از موارد odor, taste, overall acceptability نام برده شده است.
- در عنوان جدول Median نوشته شده در حالی که در پانویس از Means استفاده شده است.

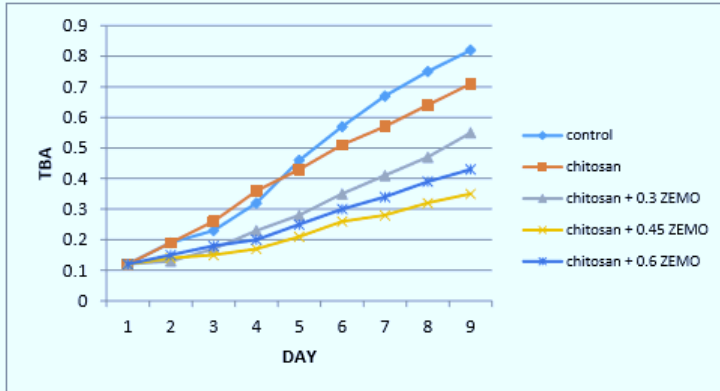


Figure 1. Changes in TBA values of the fish sample during chilled

نقد ساختاری:

- نمودارها معمولا ساده و بدون خط باید باشند.
- عدم یکنواختی مخفف: ZEMO نوشته شده، باید ZMEO باشد.
- مقیاس بندی نامناسب محور X
- برای غلظت های ZEMO باید % گذاشته شود.
- باید از عبارت samples استفاده شود.
- استفاده از chilled storage به جای chilled

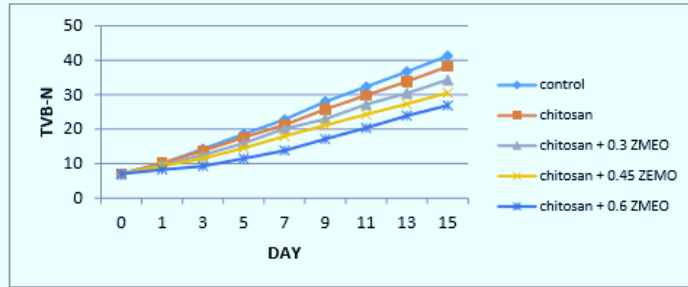


Figure 2. Changes in TVB-N values of fish samples during chilled storage.

نقد ساختاری:

- نمودارها معمولا ساده و بدون خط باید باشند.
- درجه بندی نامناسب محور Y
- برای غلظت های ZME0 باید % گذاشته شود.

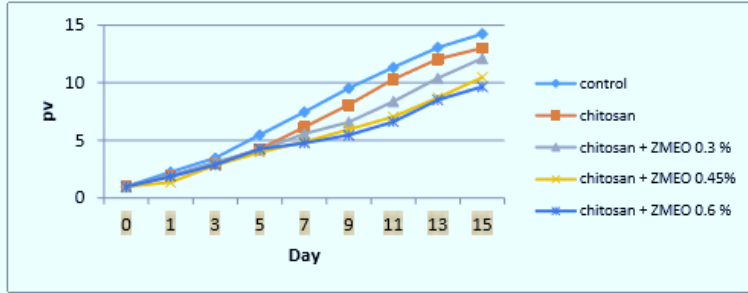


Figure 3. Changes in PV values of the fish sample during chilled storage

نقد ساختاری:

- نمودارها معمولا ساده و بدون خط باید باشند.
- درجه بندی نامناسب محور Y
- باید از عبارت samples استفاده شود.

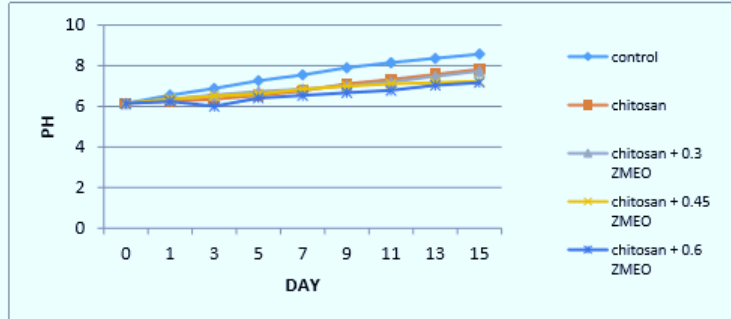


Figure 4. Changes in pH values of fish samples during chilled storage

- ایرادات ساختاری:
- نمودارها معمولاً ساده و بدون خط باید باشند.
 - درجه بندی نامناسب محور Y



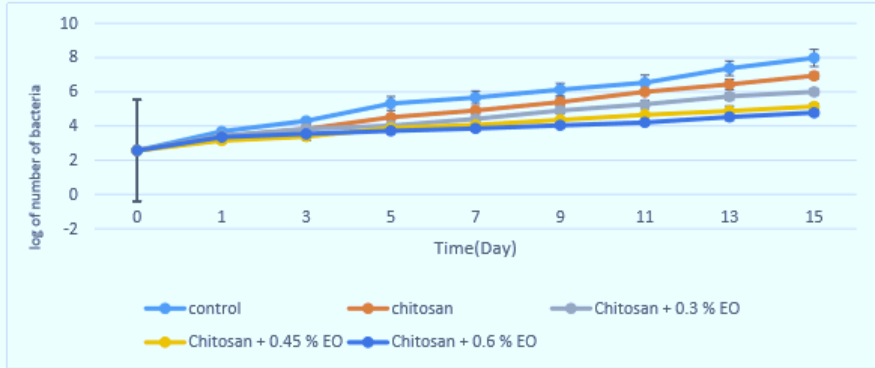


Figure 5. Changes in total viable counts (TVC) of Silver carp during chilled storage

نقد ساختاری:

- نمودارها معمولا ساده و بدون خط باید باشند.
- درجه بندی نامناسب محور Y
- عدم یکنواختی مخفف: EO استفاده شده.
- عدم تمایز نمودارها از یکدیگر
- یا باید برای تمام نمودارها از عبارت carps استفاده می شد یا مانند سایر نمودارها از samples استفاده می کرد.
- باید محور X برای تمام نمودارها یکسان باشد یا از DAY استفاده شود یا Time (Day)

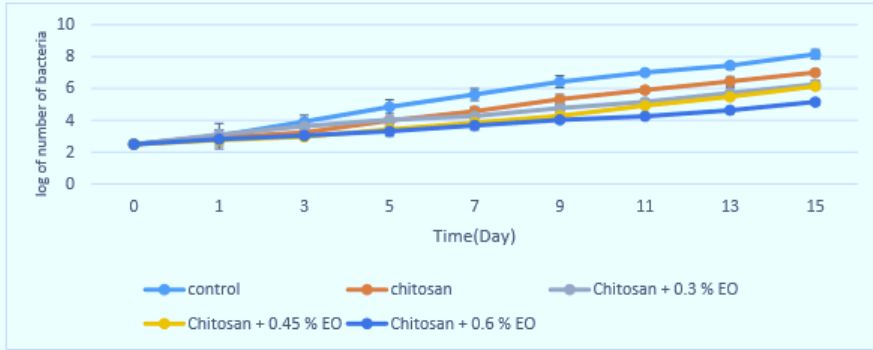


Figure 6. Changes in Enterobacteriaceae count (EBC) of silver carp during chilled storage

نقد ساختاری:

- نمودارها معمولا ساده و بدون خط باید باشند.
- درجه بندی نامناسب محور Y
- عدم تمایز نمودارها از یکدیگر
- عدم یکنواختی مخفف: EO استفاده شده.
- باید محور X برای تمام نمودارها یکسان باشد یا از DAY استفاده شود یا Time (Day)
- یا باید برای تمام نمودارها از عبارت carps استفاده می شد یا مانند سایر نمودارها از samples استفاده می کرد.

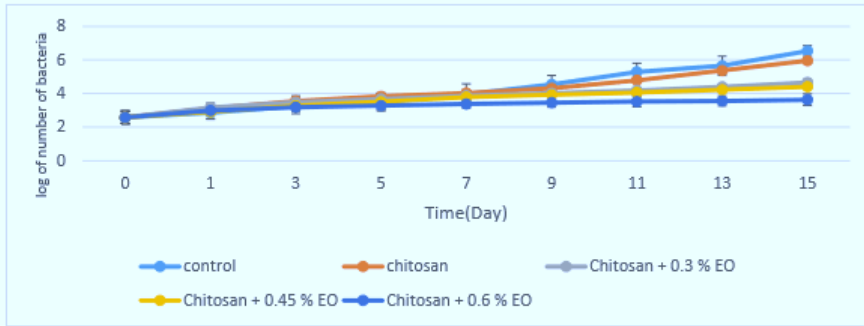


Figure 7. Changes in lactic acid bacteria count (LAB) of silver carp during chilled storage

نقد ساختاری:

- نمودارها معمولا ساده و بدون خط باید باشند.
- درجه بندی نامناسب محور Y
- عدم تمایز نمودارها از یکدیگر
- عدم یکنواختی مخفف: EO استفاده شده
- یا باید برای تمام نمودارها از عبارت carps استفاده می شد یا مانند سایر نمودارها از samples استفاده می کرد

count of the sample was 2.49 log CFU/g. After treatment, the final population of the treated samples reduced dramatically by roughly 1-3 log CFU/g compared to the control sample (Figure 6). Specifically, on the 15th day, the population of Enterobacteriaceae in the control sample was measured to be 8.15 log CFU/g. However, when Chitosan + 0.6% ZMEO was present, their population was seen to be 5.15 log CFU/g (Figure 6). Prior studies have indicated that several essential oils and extracts have the ability to hinder the growth of the Enterobacteriaceae population in fish products (18).

- ایرادات ساختاری:
حرف بزرگ Chitosan در وسط جمله اشتباه است
(نباید بزرگ باشد).

Conclusion

According to the findings of this research, applying a layer of chitosan infused with ZMEO onto silver carp fillets. Slows down the growth of microorganisms and unwanted chemical reactions in meat, hence increasing the amount of time silver carp fillets can be stored in refrigerated conditions. The capabilities of ZMEO and chitosan can be attributed to the presence of natural components that function as antioxidants and antimicrobials. Future studies should focus on enhancing the stability and hardness of the coating's physical qualities to make it suitable for commercial application. In summary, this coating has the potential to serve as an effective antimicrobial agent that can be utilized to prevent the growth of undesirable microbes in food products, particularly those derived from fish and shellfish.

ایرادات ساختاری:

- جمله اول ناقص است. به نقطه ختم شده اما فعل ندارد
- جمله دوم فاعل ندارد



نقد علمی مقاله



by-products of the food industry or underutilized sources of lipids, polysaccharides, or proteins to develop an efficient system that maintains food quality during its storage period. Edible coatings are thin films, either in solid or liquid form that are applied to food. They serve to prolong the shelf life and enhance the quality of food by influencing characteristics such as diffusibility, permeability, and trait retention (5).

Numerous studies have demonstrated the antibacterial, antioxidant, antiviral and antifungal properties of essential oils extracted from various parts of plants. The properties of an essential oil are determined by the number of phenolic chemicals it contains, including eugenol, thymol, and carvacrol (6).

Zataria multiflora, sometimes known as Shirazi thyme, belongs to the Lamiaceae family.

تمام روغن های ضروری حاوی این ترکیبات نیستند!



It is widely used in Iran as a flavoring ingredient in many types of food and for some medicinal purposes. The essential oil of this plant is rich in carvacrol and thymol. Carvacrol and thymol are the primary phenolic molecules that contribute to the antibacterial and antioxidant properties of ZMEO (7). The US FDA classifies phenolic chemicals, including carvacrol and thymol, as GRAS (generally regarded as safe) (6). Some studies have shown that edible coatings of seafood with chitosan infused with antimicrobial or antioxidant agents help prolong the duration of freshness for these products (1). The **purpose** of this study was to investigate the impact of combining chitosan with ZMEO on the longevity of fresh silver carp fillets while stored at a temperature of 4°C.

نقطه قوت: در آخرین پاراگراف مقدمه هدف از مطالعه بیان شده



Materials and Methods

Zataria Multiflora Boiss. Extraction of the Essential Oil

ZMEO was obtained from Fars Province, Iran, and authenticated by the Institute of Medicinal Plants, Tehran University of Medical Sciences, Iran. The plant portions that had been dried in the air were subjected to steam distillation for a duration of three hours using a Clevenger-type equipment (7). The components of the ZMEO were analyzed using gas chromatography-mass spectrometry [GC-MS], following the method published by Moosavy et al (8).

مشخصات GC باید ذکر شود

Bacteriological Examination

Bacteriological values were assessed by combining a 10 g sample of each treatment with 90 cc of 0.1% peptone water in sterile bags and homogenizing the mixture using a stomacher for 2 minutes. Subsequent decimal dilutions were made and subsequently plated onto the suitable medium. The TVC was calculated using the surface plate method and the plate counting agar PCA. The plates that were treated with a little amount of a substance to prevent the growth of microorganisms were thereafter placed in a controlled environment at a temperature of 30°C for a duration of three days (11, 12).

مشخصات دستگاه استومیکر باید ذکر شود



Chemical Analyses

Protein and Lipid Content

The Kjeldahl technique was used to determine the total crude protein. This technique employs a three-step process to measure the amount of protein: digestion, distillation, and titration. The organic substance is digested by employing concentrated H₂SO₄, heat, K₂SO₄ (to elevate the boiling point), and a catalyst (such as selenium) to expedite the reaction. The lipid content of the fillets was evaluated using the method developed by the Fraunhofer Institute for Molecular Biology and Applied Ecology (10). The lipids were extracted from the fillets by homogenizing them in a mixture of chloroform and methanol (at a ratio of 2:1, volume to volume). The amount of lipids was then determined by measuring the weight of the extracted lipids, and this process was repeated three times for accuracy.

این اسم پژوهشکده است
باید نام روش را بیان می کرد.
روشی که در ادامه گفته شده برای مواد غذایی اصلاً معتبر و دقیق نیست.



Determination of the Peroxide Value (PV)

The peroxide content was quantified in the whole lipid extracts using Pearson's technique (14). A total of 0.3 grams of the sample was carefully deposited into a stoppered flask with a capacity of 250 milliliters. The solvent was eliminated using a rotary evaporator under reduced pressure at a temperature of 40°C (equivalent to the temperature of a water bath). A volume of 10 ml of the acetic acid/chloroform combination with a ratio of 3/2 was added, and the lipids were dissolved through agitation. Next, 1 milliliter of a concentrated chlorine solution was added and the mixture was left undisturbed in a dark environment for a duration of 5 minutes. Dispense 20 milliliters of distilled water and agitate. Perform a titration of the liberated iodine using a 0.01 N sodium thiosulfate solution until the solution turns a pale yellow color. Introduce 1 milliliter of a 1.5% starch solution into the mixture as an indicator, and continue the titration process until the solution becomes colorless. Conduct blank tests in a same manner, excluding any fats. Utilize a conventional formula to denote the peroxide value.

بایستی فرمول نوشته شود.

Sensory Evaluation

An acceptance test was conducted on **fried** fish fillet samples to analyze the sensory effects obtained from them. A panel test was conducted to evaluate the fish fillets from each treatment of chitosan enriched with varying concentrations of ZMEO (0, 0.3, 0.45, and 0.6% v/v) and a control group. The panel consisted of 50 untrained judges who were members of the Faculty of Veterinary Medicine at the University of Tehran. The panelists assessed the samples by assigning ratings on a 9-point scale, where 9 indicates a remarkable liking, 8 indicates a strong liking, 7

بایستی در مورد چگونگی سرخ کردن و اینکه آیا مزه دار شده یا نه را توضیح می داد!

indicates a moderate liking, 6 indicates a slight liking, 5 indicates neither liking nor disliking, 4 indicates a slight disliking, 3 indicates a moderate disliking, 2 indicates a strong disliking, and 1 indicates an intense disliking. These ratings were given for different attributes such as appearance, color, odor, and flavor.

مغایرت با جدول ۱ دارد.



Protein and Lipid Content

Protein and lipid contents were measured $13.97 \pm 0.59 \%$ and $5.12 \pm 0.48 \%$, respectively.

چرا رطوبت اندازه گیری نشد؟
یکی از اثرات پوشش ها، جلوگیری از تغییرات رطوبت است که در فساد هم می باشد!

Bacteriological Changes

The alterations in bacterial composition (total viable count, Enterobacteriaceae count, and lactic acid bacteria count) of silver carp fillets treated with various concentrations of ZMEO (0, 0.3, 0.45, and 0.6 % v/v) throughout a 15-day period of refrigerated storage are depicted in Figure 5-7.

Figure 5 displays the variations in TVC (Total Viable Count) of silver carp fillets for each treatment. The initial total viable counts of fresh silver carp fillets were $2.97 \pm 0.02 \log \text{cfu/g}$. The total volatile compounds (TVC) exhibited an increase in all treatments when stored at a temperature of 4°C . The count of mesophilic bacteria in the treated samples was considerably **lower** ($P < 0.05$) compared to the control group. The total viable count (TVC) of microorganisms in the control sample of fresh fish reached a value of 7 log cfu/g, which is the upper limit set by Salam (12), between days 13 to 15. The use of chitosan enriched with ZMEO (at concentrations

نه اینجا و نه در نمودار به تفاوت بین گروه ها اشاره نکرده
است. یعنی تفاوت تیمارها باهم معنادار نبوده؟

of 0, 0.3, 0.45, and 0.6 %v/v) as a coating for silver carp fillets led to an increase in the fillet's shelf-life.

According to Figure 6, the initial count of Enterobacteriaceae (EBC) was 2.49 log cfu/gr. The treated samples had a significantly lower EBC count compared to the control sample, with a p-value of less than 0.5. The initial count of lactic acid bacteria (LAB) in the silver carp fillets was 2.58 log cfu/g on the first day of the investigation (Figure 7). The application of chitosan with varying quantities of essential oils resulted in a significant reduction in the count of lactic acid bacteria ($P < 0.05$).

این نتیجه رو بر چه مبنایی آورده؟
اولا مدت ماندگاری یک واژه کلی است که از مجموع پارامترها قابل ارائه است.
اینجا فقط در مورد باکتریها داره توضیح میده . پس باید میگفت فساد میکروبی رو به تاخیر میندازه



Thiobarbituric Acid Value

As shown in Figure 1, the initial TBA value of silver carp fillets were 0.12 ± 0.20 and increased to 0.82 for control samples by the end of the storage period. By contrast, the value reached 0.43 for samples that were treated with chitosan containing 0.6% ZMEO.

بقیه تیمارها چی؟ و اینکه آیا این تفاوت ها معنادار بوده؟

Peroxide Value (PV)

The PV value index is a useful tool for quantifying the extent of primary lipid oxidation in seafood. Figure 3 displays the PV values data. The peroxide value (PV) of the samples was 0.97 milliequivalents of oxygen per kilogram (meq O₂/kg) on day 0. The study period revealed a decrease in PV values in samples that were coated with chitosan containing ZMEO, as shown in figure 3. The PV values exhibited a substantial rise ($P < 0.05$) as the storage time at 4°C

جمله بالا را نقض میکند

طبق نمودار همه تیمارها روند افزایشی داشته اند!



چرا میانه؟ ما در آمار با میانگی سروکار داریم نه میانه!
با این حال ظاهراً اعداد صحیح در جدول ۱ نشانه مد است که بیشترین
عدد تکرار شده در بین نظرات پنلیست ها می باشد.

Sensory Analysis

Table 1 displays the **median** scores for odor, taste, and acceptability of the ZEMO fish fillet samples. The fillet treated with chitosan mixed with 0.3% EO for 15 days of storage was the most favored sample. The fillet treated with chitosan combined with 0.3% EO during 15 days of storage was the most preferred sample. The evaluation of **odor**, **color**, and general acceptability of silver carp fillet samples were found in Table 1. According to the findings of this investigation, applying a coating of chitosan with 0.3% ZMEO resulted in the required sensory qualities.

اینجا میگه بو و رنگ
بالا گفته بو و مزه.
تو جدول هم بو و مزه هست.

Table 1. Median rating for sensory properties of fish fillets as affected by different concentrations of ZEMO

| combination | Odor | Taste | Overall acceptability |
|------------------------|------|-------|-----------------------|
| control | 7 | 7 | 7 |
| Chitosan | 7 | 8 | 7 |
| Chitosan + 0.3 % ZEMO | 8 | 8 | 8 |
| Chitosan + 0.45 % ZEMO | 8 | 6 | 6 |
| Chitosan + 0.6 % ZEMO | 6 | 4 | 4 |

Means followed by the same letters are not significantly ($P < 0.05$) different.

SD: standard deviation.

از میانگین هایی با حروف یکسان نام برده شده اما وجود ندارند. انحراف استاندارد وجود ندارد.

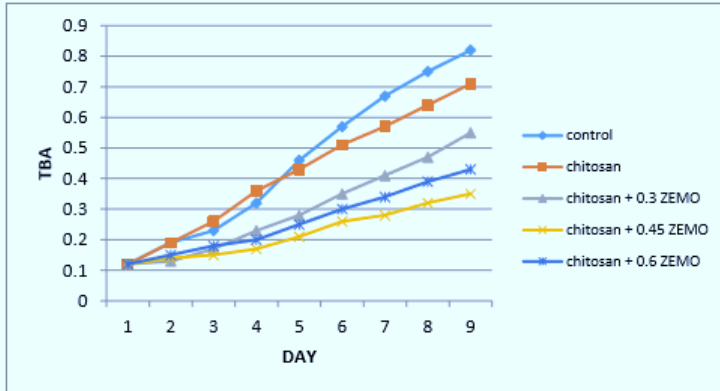


Figure 1. Changes in TBA values of the fish sample during chilled

نقد علمی:

- محور γ فاقد واحد است
- عدم نمایش انحراف معیار و نشانگر اختلافات معنی دار
- عدم تطابق کامل با مقادیر گزارش شده در متن
- چرا تا روز نهم بررسی شده؟

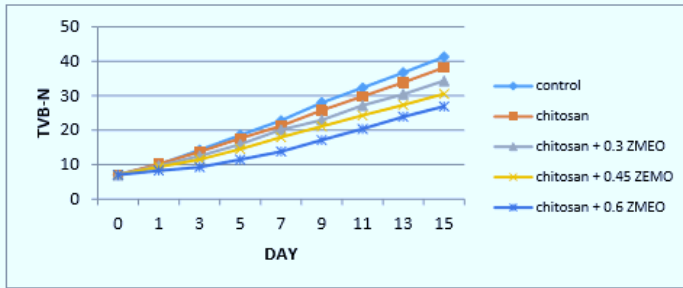


Figure 2. Changes in TVB-N values of fish samples during chilled storage.

- نقد علمی:
- محور یافقدواحد است.
- عدم نمایش انحراف معیار و نشانگر اختلافات معنی دار

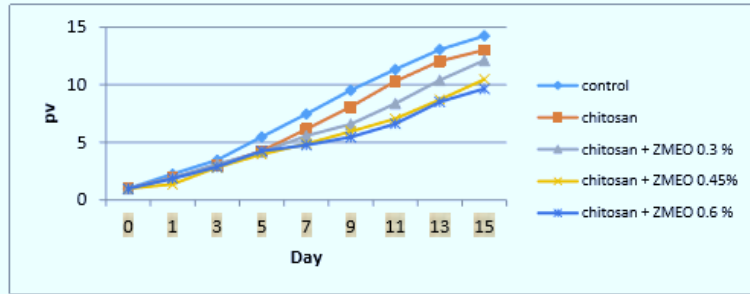


Figure 3. Changes in PV values of the fish sample during chilled storage

نقد علمی:

- محور γ فاقد واحد است.
- عدم نمایش انحراف معیار و نشانگر اختلافات معنی دار
- عدم تطابق کامل با مقادیر گزارش شده در متن

Discussion

The chemical composition of ZMEO is influenced by various elements, including the season of harvest, the age of the plant, the soil it grows in, the geographical location, the climate conditions, the species, the method of herb drying, and the extraction method (19).

The proximate composition results are consistent with the findings reported by Asgharzadeh et al (20). In this investigation, the protein content of the control samples was found to be lower than the protein content of silver carp fillets in the Asgharzadeh et al (20) study. The presence of both report and wild silver carp can be ascribed to variations in diet.

به طور کلی :
بحث ناقص است و به دلایل مشاهده نتایج اشاره نکرده.
نقطه قوت: با مطالعات دیگر مقایسه شده است.

Conclusion

According to the findings of this research, applying a layer of chitosan infused with ZMEO onto silver carp fillets. Slows down the growth of microorganisms and unwanted chemical reactions in meat, hence increasing the amount of time silver carp fillets can be stored in refrigerated conditions. The capabilities of ZMEO and chitosan can be attributed to the presence of natural components that function as antioxidants and antimicrobials. Future studies should focus on enhancing the stability and hardness of the coating's physical qualities to make it suitable for commercial application. In summary, this coating has the potential to serve as an effective antimicrobial agent that can be utilized to prevent the growth of undesirable microbes in food products, particularly those derived from fish and shellfish.

نقطه قوت: پیشنهاد برای مطالعات بعدی



پایان

