

Review

# Edible Films and Coatings for Fresh Fish Packaging: Focus on Quality Changes and Shelf-life Extension

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**Abstract:** Fresh fish is extensively consumed and is one of the most-traded food commodities in the world. Conventional preservation technologies include vacuum and modified atmosphere packaging, but they are costly since requires capital investment. In the last decade, research has been directed towards the development of antimicrobial packaging systems, as an economical alternative to these. This paper outlines antimicrobial films and coatings applied so far on fresh fish, their efficacy against targeted microorganism/group and effects on chemical quality of the product. Findings show that edible films/coatings incorporated with different active agents applied to fresh fish are able to inhibit the microbial growth and decrease the rate of fish nutrients degradation, thus preventing the formation of chemical metabolites; a shelf-life extension of 6 to 13 days was obtained for fish fillets, depending on the species on which the active packaging materials were applied. The manufacturing use of these formulations could lead to a significant reduction in fish waste, consequently, a diminution of economic losses for fish traders and retailers. Therefore, their industrial production and commercialization could be an exploitable sector by the packaging industry.

**Keywords:** edible films; edible coatings; antimicrobial agents; fresh fish; spoilage; shelf-life

## 1. Introduction

Fish is one of the most-traded food commodities worldwide [1]. Capture fisheries and aquaculture provide valuable economic and social benefits to those who work in these industries [2]. However, post-harvest handling, processing, and storage of fish lead to food losses and waste [3]. Post-harvest losses occur at all stages in the fish supply chain from capture to consumer [4]. The losses can be physical, economical, or nutritional and are caused by spoilage or poor processing [5]. Spoilage is the process in which fish deteriorates to the point that becomes unacceptable for human consumption (with altered taste, smell, appearance, or texture) [6]. Globally, fish losses that are caused by spoilage account for around 10% (10 to 12 million tons per year) of the total production from capture fisheries and aquaculture [7].

Fresh fish is a highly perishable product due to its high water activity, nutrient availability, nearly neutral-pH (factors that influence microbial growth) and the presence of autolytic enzymes; hence, it is susceptible to post-harvest losses [8,9]. Under normal refrigerated storage conditions, its shelf-life is limited by the development of enzymatic (caused by endogenous or microbial enzymes) and chemical reactions [10]. The main initial causative factor for fish spoilage is microbial growth and invasion, followed by the autolytic enzymes and then by chemical reactions, such as oxidation or hydrolysis [11,12].

Post-harvest losses of fresh fish due to microbial spoilage are a matter of great importance to the fishing industry [13]. So, specific requirements and preservation techniques are needed to minimize the activity of spoilage bacteria. Fresh fish products are presently stored on ice or under refrigeration during their distribution and marketing. In these conditions, their shelf-life is limited to 5–10 days (depending on species, harvest location, and season) and they can result in enormous economic losses to fish traders and retailers [14,15]. Therefore, the fish-process industry is actively seeking alternative methods of shelf-life preservation and marketability of fresh fish [16].

Packaging plays a critical role in the fish supply chain and is part of the solution to tackle food waste [17,18]. Vacuum packaging (VP) and modified atmosphere packaging (MAP) are very commonly used as a supplement to ice or refrigeration to inhibit the normal spoilage flora and extend the shelf-life of fresh fish products [14,19,20]. MAP technology has, however, some disadvantages, such as added costs for packaging equipment, gases, and packaging materials; it also requires special training for food operators [21].

Packaging innovation and new technologies is a necessity for the fishing industry. In recent years, a variety of active packaging systems have been developed to prolong storage life and enhance the safety of fish products. These have a variety of advantages such as biodegradability, edibility, biocompatibility, and aesthetic appearance, respectively, barrier properties against oxygen and physical stress [22]. The purpose of this paper is to provide an overview of published research about edible films and coatings applied to fresh fish. The antimicrobial films and coatings that are used for fish packaging and their effects on chemical quality of fresh fish are reviewed and discussed (Figure 1).

Edible polymers	Active agents	Fresh fish fillets	Microbial indicators	Chemical indicators
<ul style="list-style-type: none"> <li>•Gelatin</li> <li>•Chitosan</li> <li>•Chitosan-gelatin</li> <li>•Gelatin-alginate</li> <li>•Carrageenan</li> <li>•Quince seed mucilage</li> <li>•Whey protein concentrate</li> <li>•Whey protein isolate</li> </ul>	<ul style="list-style-type: none"> <li>•Clove EO</li> <li>•Cinnamon EO</li> <li>•Oregano EO</li> <li>•Thyme EO</li> <li>•Lemon EO</li> <li>•Glycerol monolaurate</li> <li>•<math>\alpha</math>-Tocopherol</li> <li>•Lactoperoxidase</li> <li>•Citric acid</li> <li>•Licorice extract</li> <li>•Grape seed extract</li> <li>•Tea polyphenols</li> </ul>	<ul style="list-style-type: none"> <li>•Rainbow trout</li> <li>•Silver carp</li> <li>•Grass carp</li> <li>•Beluga sturgeon</li> <li>•Salmon</li> <li>•Pike-perch</li> <li>•Japanese sea bass</li> <li>•Red drum</li> <li>•Golden pomfret</li> <li>•Hake</li> </ul>	<ul style="list-style-type: none"> <li>•<i>Listeria innocua</i></li> <li>•<i>Listeria monocytogenes</i></li> <li>•<i>Escherichia coli</i></li> <li>•<i>Salmonella typhimurium</i></li> <li>•<i>Pseudomonas</i> spp.</li> <li>•H<sub>2</sub>S producing bacteria</li> <li>•Lactic acid bacteria</li> <li>•Total viable organisms</li> <li>•Total mesophilic bacteria</li> <li>•Total psychrotrophic bacteria</li> <li>•<i>Enterobacteriaceae</i> (including coliform bacteria)</li> <li>•Total yeasts and moulds</li> </ul>	<ul style="list-style-type: none"> <li>•Thiobarbituric acid reactive substances</li> <li>•Total volatile basic nitrogen</li> <li>•Trimethylamine nitrogen</li> <li>•<i>k</i>-value</li> </ul>

Figure 1. Antimicrobial films and coatings used to extend the shelf-life of fresh fish fillets.

## 2. Microbiological Issues

Fresh fish spoils due to the action of a group of microorganisms, the so-called specific spoilage organisms (SSOs). These organisms have the ability to dominate the fish flora and produce metabolites that directly affect the sensory properties of the product resulting in its rejection by consumers [23]. During storage, the microflora changes owing to different capacities of the microorganisms to tolerate the preservation conditions [24]. Under aerobic iced storage, the flora of fish is composed almost

exclusively of *Pseudomonas* spp. and *Shewanella putrefaciens* (SSOs) regardless of whether it was caught or harvested in temperate or sub-tropical and tropical waters. At ambient temperature (25 °C), microflora is dominated by mesophilic *Vibrionaceae*, and, particularly if the fish is caught in polluted waters, by mesophilic *Enterobacteriaceae* [25].

Microbial spoilage is due to the proliferation of microorganisms after the death of fish as a result of the immune system collapsing, followed by the microbial invasion of the fish body through the skin [12]. Fish have a unique osmoregulatory mechanism to avoid dehydration in marine environments and waterlogging of tissue in freshwater; it contains osmoregulatory compounds, like trimethylamine oxide (TMAO) and urea [26]. Microbial enzymes that are present in fish can break down TMAO to trimethylamine (TMA) and urea to ammonia, volatile organic compounds associated with microbial spoilage [12]. Many other volatile compounds can be formed by microbial enzymatic degradation of other substrates, such as hydrogen sulphide (from cysteine), methanethiol and methyl sulphide (from methionine), histamine (from histidine), acetate, carbon dioxide and water (from carbohydrates and lactate), hypoxanthine (from inosine and inosine-5'-monophosphate), esters, ketones, aldehydes (from amino acids, like glycine, serine, and leucine), as well as ammonia (from amino acids and urea) [12,26]. These molecules are responsible for sweet, fruity, ammonia-like, putrid, and sulphuric off-flavours in spoiled fish [27].

### 3. Antimicrobial Films and Coatings Applied on Fresh Fish

This chapter provides an overview of previous research on the antimicrobial packaging of fresh fish. Table 1 lists active edible films and coatings applied to fresh fish fillets (of rainbow trout, silver carp, grass carp, beluga sturgeon, salmon, pike-perch, Japanese sea bass, red drum, golden pomfret, and hake) to extend its shelf-life. These films and coatings were produced from edible polymers like gelatin, chitosan, chitosan-gelatin, gelatin-alginate, carrageenan, quince seed mucilage, whey protein concentrate, and whey protein isolate incorporated with various active agents (essential oils (EOs) of clove, cinnamon, oregano, thyme, and lemon, glycerol monolaurate,  $\alpha$ -tocopherol, lactoperoxidase, citric acid, licorice extract, grape seed extract, and tea polyphenols). Their antimicrobial efficacy was investigated in situ against spoilage and pathogenic microorganisms. Different levels of effectiveness were noticed, depending on the active agent used, its concentration, storage temperature, atmosphere composition (normal or modified), and targeted microorganism/group.

#### 3.1. Efficacy against Tested Microorganism/Group at the End of Monitoring Time

##### 3.1.1. Efficacy against Spoilage Microorganisms

Several authors have investigated the potential of edible films/coatings in extending the shelf-life of fresh fish fillets by retarding the growth of spoilage bacteria. Jouki et al. (2014) [28] have tested the efficacy of films based on 1% quince seed mucilage incorporated with different concentrations of oregano and thyme EOs (1%, 1.5%, and 2%) against *Pseudomonas* spp., H<sub>2</sub>S producing bacteria, and lactic acid bacteria in rainbow trout fillets; Kazemi & Rezaei (2015) [29] of films based on 3% gelatin and 1.5% alginate containing 1.5% oregano EO against *Pseudomonas* spp. and lactic acid bacteria; Volpe et al. (2015) [30] of the coating based on 1% carrageenan incorporated with 1% lemon EO against H<sub>2</sub>S producing bacteria and lactic acid bacteria; Yıldız & Yangilar (2016) [31] of coatings based on 8% whey protein concentrate/glycerol in ratios of 1:1 and 2:1 against lactic acid bacteria. On grass carp fillets, Yu et al. (2017) [32] have evaluated the efficacy of coatings based on 2% chitosan incorporated with different concentrations of glycerol monolaurate (0.1% and 0.3%) against *Pseudomonas* spp. and H<sub>2</sub>S producing bacteria. In a study on pike-perch fillets, Shokri & Ehsani (2017) [33] have tested the efficacy of coatings based on 10% whey protein isolate incorporated with 2.5% lactoperoxidase, 1.5% and 3.0%  $\alpha$ -tocopherol, respectively, combinations of lactoperoxidase and  $\alpha$ -tocopherol (2.5%/1.5% and 2.5%/3.0%) against *Pseudomonas* spp. and H<sub>2</sub>S producing bacteria.

Edible films/coatings incorporated with 2% thyme EO [28], 1.5% oregano EO [29], respectively 1% lemon EO [30] applied on rainbow trout fillets, 0.3% glycerol monolaurate [32] on grass carp fillets, and 2.5% lactoperoxidase [33] on pike-perch fillets have been proven to be the most effective against *Pseudomonas* spp. The most effective against H<sub>2</sub>S producing bacteria were edible films/coatings incorporated with 2% thyme EO [28] applied on rainbow trout fillets, 0.3% glycerol monolaurate [32] on grass carp fillets, and 2.5% lactoperoxidase [33] on pike-perch fillets, but against lactic acid bacteria, the ones incorporated with 2% thyme EO [28], 1.5% oregano EO [29], 1% lemon EO [30], and 8% whey protein concentrate/glycerol, 2:1 [31] applied on rainbow trout fillets.

In a recent study, Carrión-Granda et al. (2018) [34] have examined the efficacy of coatings based on 10% whey protein isolate incorporated with different concentrations of oregano and thyme EOs (1% and 3%) under air and MAP conditions against *Pseudomonas* spp., H<sub>2</sub>S producing bacteria, and lactic acid bacteria in hake fillets. The application of coating with 1% thyme EO under MAP has shown the best results against *Pseudomonas* spp. but against H<sub>2</sub>S producing bacteria and lactic acid bacteria, the one with 3% oregano EO under the MAP. Different inhibitory effects displayed by an essential oil against various bacteria are most probably due to its chemical composition [35]. The antimicrobial mechanism of action of plant EOs is related to the hydrophobicity of their components [36], which enables them to migrate in the lipids of the bacterial cell membrane and mitochondria, disturbing their structures and rendering them more permeable [37]; leakage of ions and intracellular constituents can thus occur [38].

### 3.1.2. Efficacy against Pathogenic Microorganisms

According to current literature, few studies on the efficacy of active packaging materials against pathogenic microorganisms in fresh fish have been published. Findings of such in situ investigations are presented in Table 1. Gómez-Estaca et al. (2009) [39] have tested the efficacy of edible films based on 8% gelatin and 8% gelatin/chitosan, both incorporated with 7.5% clove EO on salmon fillets, in vitro against *Listeria innocua* and *Escherichia coli*, then in situ against total viable organisms. The film based on gelatin was more effective against both bacteria than the one based on gelatin/chitosan; the ionic and hydrogen bonds that were formed between gelatin and chitosan diminished the solubility of the resulting film, thus reducing the amount of clove EO released. However, in the in situ experiment, they used the film based on gelatin/chitosan for storage trials. Their previous studies revealed that the low water solubility of the gelatin/chitosan matrix gives the film stability under fish contact conditions during chilled storage.

There are also some studies on fish fillets challenged with pathogenic bacteria. Han et al. (2013) [40] have investigated the efficacy of films based on 6.75% (*w/w*) gelatin, with and without nisin-incorporated, against *Listeria monocytogenes* in rainbow trout fillets that were challenged with 2 log CFU/g inoculum before and after coating. The edible film incorporated with 18 µg/cm<sup>2</sup> nisin, applied before inoculation, showed the highest inhibitory effect on *Listeria monocytogenes*.

The efficacy of gelatin coatings containing different concentrations of oregano EO (0.5%, 1.0%, and 2.0% *v/v*) was also investigated by Min and Oh (2009) [41], in catfish fillets that were inoculated with *Salmonella typhimurium* and *Escherichia coli* O157:H7. The coating based on 3% (*w/v*) gelatin containing 2% oregano EO exhibited the best inhibitory effect on both bacteria.

### 3.1.3. Efficacy against Spoilage and/or Pathogenic Microorganisms

The following groups of microorganisms we have included into this category: total viable organisms, total mesophilic bacteria, total psychrotrophic bacteria, *Enterobacteriaceae* (including coliform bacteria), respectively total yeasts and moulds. Against total viable organisms, the most effective edible films/coatings were those that were incorporated with 2% thyme EO [28], 1.5% oregano EO [29], 1% lemon EO [30], and 1.5% cinnamon EO [42] applied on rainbow trout fillets, 0.3% glycerol monolaurate [32] on grass carp fillets, 2.5% lactoperoxidase [33] on pike-perch fillets, 0.2% tea polyphenols [43] on red drum fillets, 0.5% citric acid on Japanese sea bass fillets [44] and beluga sturgeon fillets [45], and 3% oregano EO under MAP conditions [34] on hake fillets. Edible coatings

based on chitosan [46] applied to salmon fillets, respectively chitosan-gelatin [47] to golden pomfret fillets exhibited an antimicrobial effect compared to uncoated controls.

Regarding total psychrotrophic bacteria, the most effective were edible films/coatings incorporated with 2% thyme EO [28], 1.5% oregano EO [29], and 1.5% cinnamon EO [42] applied on rainbow trout fillets, 0.3% glycerol monolaurate [32] on grass carp fillets, 1.5% cinnamon EO on beluga sturgeon fillets [45], and 2.5% lactoperoxidase [33] on pike-perch fillets.

Edible coatings with 8% whey protein concentrate/glycerol, 2:1 applied on rainbow trout fillets [31], 1% chitosan [48] on salmon fillets, and 2% nanochitosan on silver carp fillets [49] have shown to be effective against both total psychrotrophic bacteria and total mesophilic bacteria.

The most effective edible films/coatings against *Enterobacteriaceae* (including coliform bacteria) were those incorporated with 2% thyme EO [28], 1.5% oregano EO [29], and 1% lemon EO [30] that were applied on rainbow trout fillets. Edible coating with 8% whey protein concentrate/glycerol, 2:1 has also shown to be effective against *Enterobacteriaceae* in rainbow trout fillets as compared with the other formulations tested in the study [31].

When tested against total yeasts and moulds, the edible coating based on 0.4% chitosan and 3.6% gelatin applied to golden pomfret fillets was the most effective among all formulations [47].

In the work of Carrión-Granda et al. (2018) [34], the edible coating incorporated with 3% oregano EO was the most effective against total viable organisms, total psychrotrophic bacteria, as well as *Enterobacteriaceae* when applied under the MAP conditions.

The results of these investigations are not comparable, since, on the same fish species, were applied edible films/coatings with different polymer matrices, respectively active agents and evaluated in different storage conditions (temperature, atmosphere composition, and storage time). We noticed, however, some tendencies that allow us to affirm that:

- edible films/coatings with the highest concentration of active agent tested have shown the greatest antimicrobial efficacy;
- antimicrobial films/coatings were more effective at lower temperatures when tested in different storage temperature conditions; and,
- under modified atmosphere packaging conditions, antimicrobial films/coatings were more effective than under air conditions.

Other authors have noticed that the effectiveness of antimicrobial packaging material depends also on the initial microbial load [40], chemical composition, and pH of tested food products [37]. Generally, the susceptibility of bacteria to the antimicrobial effect of EOs is increased in products with low-fat content and low pH, respectively.

**Table 1.** Antimicrobial films and coatings used for packaging fish.

Tested Fish Product	Antimicrobial Packaging Materials		Storage Conditions	Targeted Microorganism/ Group	Type of Microorganism	Level of Effectiveness against Targeted Microorganisms/Group at the End of Monitoring Time	MAL for Targeted Microorganism/ Group	Shelf-life of Fish Product		Ref.
	Film/Coating	Active Agent/ Concentration						Uncoated	Treated	
Rainbow trout fillets	Coating based on 2% (w/v) chitosan, acetic acid, and glycerol	Cinnamon EO/1.5% (v/v)	4 °C/16 days	Total viable organisms	Pathogenic and/or spoilage	1.5% (v/v) cinnamon EO > control	7.0 log CFU/g for TVC	Uncoated control-up to 8 days	Control-up to 16 days	[42]
									1.5% (v/v) cinnamon EO-up to 16 days	
				Total psychrotrophic bacteria	Pathogenic and/or spoilage	Idem section TVC	7.0 log CFU/g for TPC	See section TVC	See section TVC	
	Film based on 1% (w/w) quince seed mucilage, glycerol, and Tween 80	Oregano EO/1%, 1.5%, and 2% (v/v) Thyme EO/1%, 1.5%, and 2% (v/v)	4 °C/18 days	<i>Pseudomonas</i> spp.	Spoilage	2% (v/v) thyme EO > 2% (v/v) oregano EO > 1.5 (v/v) thyme EO > 1.5% (v/v) oregano EO > 1% (v/v) thyme EO > 1% (v/v) oregano EO > control	7.0 log CFU/g for <i>Pseudomonas</i> spp.	See section TVC	See section TVC	
				H <sub>2</sub> S producing bacteria	Spoilage	2% (v/v) thyme EO > 2% (v/v) oregano EO > 1.5 (v/v) thyme EO > 1% (v/v) thyme EO > 1.5% (v/v) oregano EO > 1% (v/v) oregano EO > control	7.0 log CFU/g for H <sub>2</sub> S producing bacteria	See section TVC	See section TVC	[28]
				Lactic acid bacteria	Spoilage	2% (v/v) thyme EO > 1.5% (v/v) thyme EO > 1% (v/v) thyme EO > 2% (v/v) oregano EO > 1.5% (v/v) oregano EO > 1% (v/v) oregano EO > control	6.0 log CFU/g for LAB	See section TVC	See section TVC	
				Total viable organisms	Pathogenic and/or spoilage	2% (v/v) thyme EO > 1.5 (v/v) thyme EO > 2% (v/v) oregano EO > 1% (v/v) thyme EO > 1.5% (v/v) oregano EO > 1% (v/v) oregano EO > control	7.0 log CFU/g for TVC	Uncoated control-up to 6 days	Control-up to 9 days	
									1% (v/v) Oregano EO-up to 9 days	
									1.5% (v/v) Oregano EO-up to 12 days	
					2% (v/v) Oregano EO-up to 15 days					
							1% (v/v) Thyme EO-up to 12 days			
							1.5% (v/v) Thyme EO-up to 15 days			
						2% (v/v) Thyme EO-up to 18 days				
			Total psychrotrophic bacteria	Pathogenic and/or spoilage	2% (v/v) thyme EO > 1.5% (v/v) thyme EO > 2% (v/v) oregano EO > 1.5% (v/v) oregano EO > 1% (v/v) thyme EO > 1% (v/v) oregano EO > control	7.0 log CFU/g for TPC	See section TVC	See section TVC		
			<i>Enterobacteriaceae</i>	Pathogenic and/or spoilage	Idem section <i>Pseudomonas</i> spp.	5.0 log CFU/g for <i>Enterobacteriaceae</i>	See section TVC	See section TVC		
Film based on 3% (w/v) gelatin and 1.5% (w/v) alginate, glycerol, and Tween 80	Oregano EO/1.5% (w/v)	4 °C/15 days	<i>Pseudomonas</i> spp.	Spoilage	1.5% (w/v) oregano EO > control	–	See section TVC	See section TVC	[29]	
			Lactic acid bacteria	Spoilage	Idem section <i>Pseudomonas</i> spp.	–	See section TVC	See section TVC		
			Total viable organisms	Pathogenic and/or spoilage	Idem section <i>Pseudomonas</i> spp.	7.0 log CFU/g for TVC	Uncoated control-up to 3 days	Control-up to 3 days		
								1.5% (w/v) oregano EO-up to 9 days		
			Total psychrotrophic bacteria	Pathogenic and/or spoilage	Idem section <i>Pseudomonas</i> spp.	–	See section TVC	See section TVC		
<i>Enterobacteriaceae</i>	Pathogenic and/or spoilage	Idem section <i>Pseudomonas</i> spp.	–	See section TVC	See section TVC					

Table 1. Cont.

Tested Fish Product	Antimicrobial Packaging Materials		Storage Conditions	Targeted Microorganism/ Group	Type of Microorganism	Level of Effectiveness against Targeted Microorganisms/Group at the End of Monitoring Time	MAL for Targeted Microorganism/ Group	Shelf-life of Fish Product		Ref.
	Film/Coating	Active Agent/ Concentration						Uncoated	Treated	
	Coating based on 1% (w/w) carrageenan	Lemon EO/1% (w/w)	4 °C/15 days	H <sub>2</sub> S producing bacteria	Spoilage	1% (w/w) lemon EO > control	–	See section TVC	See section TVC	[30]
				Lactic acid bacteria	Spoilage	Idem section H <sub>2</sub> S producing bacteria	–	See section TVC	See section TVC	
				Total viable organisms	Pathogenic and/or spoilage	Idem section H <sub>2</sub> S producing bacteria	7.0 log CFU/g for TVC	Uncoated control-up to 3 days	Control-up to 12 days 1% (w/w) lemon EO-up to 15 days	
				<i>Enterobacteriaceae</i>	Pathogenic and/or spoilage	Idem section H <sub>2</sub> S producing bacteria	–	See section TVC	See section TVC	
	Coating based on 8% (w/w) whey protein concentrate	–	4 °C/15 days	Lactic acid bacteria	Spoilage	8% (w/w) whey protein concentrate/glycerol, 2:1 > 8% (w/w) whey protein concentrate/glycerol, 1:1 > 8% (w/w) whey protein concentrate	–	See section TMC	See section TMC	[31]
				Total mesophilic bacteria	Pathogenic and/or spoilage	Idem section LAB	–	Uncoated control-up to 9 days	8% (w/w) whey protein concentrate-up to 12 days 8% (w/w) whey protein concentrate/glycerol, 1:1-up to 15 days 8% (w/w) whey protein concentrate/glycerol, 2:1-up to 15 days	
	Coating based on 8% (w/w) whey protein concentrate/glycerol, 1:1 and 2:1			Total psychrotrophic bacteria	Pathogenic and/or spoilage	Idem section LAB	–	See section TMC	See section TMC	
	<i>Enterobacteriaceae</i>			Pathogenic and/or spoilage	8% (w/w) whey protein concentrate/glycerol, 2:1 > 8% (w/w) whey protein concentrate > 8% (w/w) whey protein concentrate/glycerol, 1:1	–	See section TMC	See section TMC		
Silver carp fillets	Coating based on 2% (w/v) chitosan and glycerol	–	4 °C/12 days	Total mesophilic bacteria	Pathogenic and/or spoilage	2% (w/v) nanochitosan > 2% (w/v) chitosan	7.0 log CFU/g for TMC	See section TPC	See section TPC	[49]
	Coating based on 2% (w/v) nanochitosan and glycerol			Total psychrotrophic bacteria	Pathogenic and/or spoilage	Idem section TMC	7.0 log CFU/g for TPC	Uncoated control-up to 6 days 1% glacial acetic acid-up to 6 days	2% (w/v) chitosan-up to 9 days 2% (w/v) nanochitosan-up to 12 days	

Table 1. Cont.

Tested Fish Product	Antimicrobial Packaging Materials		Storage Conditions	Targeted Microorganism/Group	Type of Microorganism	Level of Effectiveness against Targeted Microorganisms/Group at the End of Monitoring Time	MAL for Targeted Microorganism/Group	Shelf-life of Fish Product		Ref.
	Film/Coating	Active Agent/Concentration						Uncoated	Treated	
Grass carp fillets	Coating based on 2% (w/v) chitosan, acetic acid, and glycerol	Glycerol monolaurate/0.1% and 0.3%	4 °C/20 days	<i>Pseudomonas</i> spp.	Spoilage	0.3% glycerol monolaurate > 0.1% glycerol monolaurate > control	–	See section TVC	See section TVC	[32]
				H <sub>2</sub> S producing bacteria	Spoilage	Idem section <i>Pseudomonas</i> spp.	–	See section TVC	See section TVC	
				Total viable organisms	Pathogenic and/or spoilage	Idem section <i>Pseudomonas</i> spp.	7.0 log CFU/g for TVC	Uncoated control-up to 7 days	Control-up to 15 days 0.1% glycerol monolaurate-up to 15 days 0.3% glycerol monolaurate-up to 20 days	
				Total psychrotrophic bacteria	Pathogenic and/or spoilage	Idem section <i>Pseudomonas</i> spp.	–	See section TVC	See section TVC	
Beluga sturgeon fillets	Coating based on 8% (w/v) whey protein concentrate, glycerol, and Tween 80	Cinnamon EO/1.5% (v/v)	4 °C/20 days	Total viable organisms	Pathogenic and/or spoilage	1.5% (v/v) cinnamon EO > control	7.0 log CFU/g for TVC	Uncoated control-up to 4 days	Control-up to 4 days 1.5% (v/v) cinnamon EO-up to 16 days	[45]
				Total psychrotrophic bacteria	Pathogenic and/or spoilage	Idem section TVC	–	See section TVC	See section TVC	
Salmon fillets	Coating based on 1% (w/w) chitosan, acetic acid, and glycerol	–	2 °C/6 days	Total mesophilic bacteria	Pathogenic and/or spoilage	1% (w/w) chitosan > 1% (w/w) chitosan and 2% (w/w) tapioca starch	–	Not specified	All treated samples-up to 6 days	[48]
	Coating based on 1% (w/w) chitosan, acetic acid, glycerol, and 2% (w/w) tapioca starch			Total psychrotrophic bacteria	Pathogenic and/or spoilage	Idem section TMC	–	See section TMC	See section TMC	
	Film based on 8% (w/v) gelatin/chitosan, 3:1, sorbitol and glycerol	Clove EO/7.5% (v/w)	2 °C/11 days	Total viable organisms	Pathogenic and/or spoilage	7.5% (v/w) clove EO	–	Uncoated control-up to 9 days	7.5% (v/w) clove EO-up to 11 days	[39]
	Coating based on 1.0, 1.5, and 2% (w/v) chitosan, lactic acid solution, and Tween 80	–	0 °C/18 days	Total viable organisms	Pathogenic and/or spoilage	1%, 1.5%, and 2% (w/v) chitosan	7.0 log CFU/g for TVC	Uncoated control-up to 9 days	All treated samples-up to 15 days	[46]

Table 1. Cont.

Tested Fish Product	Antimicrobial Packaging Materials		Storage Conditions	Targeted Microorganism/ Group	Type of Microorganism	Level of Effectiveness against Targeted Microorganisms/Group at the End of Monitoring Time	MAL for Targeted Microorganism/ Group	Shelf-life of Fish Product		Ref.
	Film/Coating	Active Agent/ Concentration						Uncoated	Treated	
Pike-perch fillets	Coating based on 10% (w/v) whey protein isolate, glycerol, and Tween 80	Lactoperoxidase/2.5% (v/v)	4 °C/16 days	<i>Pseudomonas fluorescens</i>	Spoilage	2.5% (v/v) lactoperoxidase > 2.5% (v/v) lactoperoxidase and 1.5% (v/v) α-tocopherol > 2.5% (v/v) lactoperoxidase and 3% (v/v) α-tocopherol > 3% (v/v) α-tocopherol > 1.5% (v/v) α-tocopherol > 10% (w/v) whey protein isolate and 3% (v/v) ethanol	-	See section TVC	See section TVC	[33]
				H <sub>2</sub> S producing bacteria	Spoilage	Idem section <i>Pseudomonas fluorescens</i>	-	See section TVC	See section TVC	
	Coating based on 10% (w/v) whey protein isolate, glycerol, ethanol, and Tween 80	α-Tocopherol/1.5% (v/v) α-Tocopherol/3% (v/v) Lactoperoxidase and α-tocopherol/2.5% (v/v) and 1.5% (v/v) Lactoperoxidase and α-tocopherol/2.5% (v/v) and 3% (v/v)		Total viable organisms	Pathogenic and/or spoilage	2.5% (v/v) lactoperoxidase > 2.5% (v/v) lactoperoxidase and 1.5% (v/v) α-tocopherol > 2.5% (v/v) lactoperoxidase and 3% (v/v) α-tocopherol > 3% (v/v) α-tocopherol > 1.5% (v/v) α-tocopherol > control for coating with lactoperoxidase >control for other coatings	7.0 log CFU/g for TVC	-	Control-up to 4 days 2.5% (v/v) lactoperoxidase-up to 12 days	
								-	Control-up to 4 days 1.5% (v/v) α-tocopherol-up to 4 days 3% (v/v) α-tocopherol-up to 4 days 2.5% (v/v) lactoperoxidase and 1.5% (v/v) α-tocopherol-up to 12 days 2.5% (v/v) lactoperoxidase and 3% (v/v) α-tocopherol-up to 8 days	
Total psychrotrophic bacteria	Pathogenic and/or spoilage	Idem section TVC	7.0 log CFU/g for TPC	See section TVC	See section TVC					
Japanese sea bass fillets	Coating based on 1.5% (w/v) chitosan and acetic acid	Citric acid/0.5% (w/v) Licorice extract/1% (w/v)	4 °C/12 days	Total viable organisms	Pathogenic and/or spoilage	0.5% (w/v) citric acid > 1% (w/v) licorice extract > control	6.0 log CFU/g for TVC	Uncoated control-up to 8 days	Control-up to 8 days 0.5% (w/v) citric acid-up to 12 days 1% licorice extract-up to 12 days	[44]
Red drum fillets	Coating based on 1.5% chitosan, acetic acid, and glycerol	Grape seed extract/0.2% (w/v) Tea polyphenols/0.2% (w/v)	4 °C/20 days	Total viable organisms	Pathogenic and/or spoilage	0.2% (w/v) tea polyphenols > 0.2% (w/v) grape seed extract	7.0 log CFU/g for TVC	Uncoated control-up to 8 days	0.2% (w/v) grape seed extract-up to 16 days 0.2% (w/v) tea polyphenols-up to 16 days	[43]

Table 1. Cont.

Tested Fish Product	Antimicrobial Packaging Materials		Storage Conditions	Targeted Microorganism/ Group	Type of Microorganism	Level of Effectiveness against Targeted Microorganisms/Group at the End of Monitoring Time	MAL for Targeted Microorganism/ Group	Shelf-life of Fish Product		Ref.	
	Film/Coating	Active Agent/ Concentration						Uncoated	Treated		
Golden pomfret fillets	Coating based on 0.4% (w/w) chitosan Coating based on 0.4% (w/w) chitosan and gelatin	-	4 °C/17 days	Total viable organisms	Pathogenic and/or spoilage	0.4% (w/w) chitosan = 0.4% (w/w) chitosan and 3.6% (w/w) gelatin = 0.4% (w/w) chitosan and 5.4% (w/w) gelatin = 0.4% (w/w) chitosan and 7.2% (w/w) gelatin	6.0 log CFU/g for TVC	Deionized water-up to 17 days	All treated samples-up to 17 days	[47]	
				Total yeasts and moulds	Pathogenic and/or spoilage	0.4% (w/w) chitosan and 3.6% (w/w) gelatin > 0.4% (w/w) chitosan and 5.4% (w/w) gelatin > 0.4% (w/w) chitosan and 7.2% (w/w) gelatin > 0.4% (w/w) chitosan	-	See section TVC	See section TVC		
Hake fillets	Coating based on 10% (w/w) whey protein isolate and glycerol	Oregano EO/1% and 3% (w/w) Thyme EO/1% and 3% (w/w)	4 °C/8 days	Total viable organisms	Pathogenic and/or spoilage	3% (w/w) thyme EO > 1% (w/w) thyme EO > 3% (w/w) oregano EO > 1% (w/w) oregano EO > control	7.0 log CFU/g for TVC	Uncoated control-up to 4 days	All treated samples-up to 4 days	[34]	
				Total psychrotrophic bacteria	Pathogenic and/or spoilage	Idem section TVC	7.0 log CFU/g for TPC	See section TVC	See section TVC		
				<i>Enterobacteriaceae</i>	Pathogenic and/or spoilage	3% (w/w) oregano EO > 3% (w/w) thyme EO > 1% (w/w) thyme EO > 1% (w/w) oregano EO > control	4.0 log CFU/g for <i>Enterobacteriaceae</i>	See section TVC	See section TVC		
				Lactic acid bacteria	Spoilage	Idem section <i>Enterobacteriaceae</i>	-	See section TVC	See section TVC		
				H <sub>2</sub> S producing bacteria	Spoilage	Idem section TVC	-	See section TVC	See section TVC		
				<i>Pseudomonas</i> spp.	Spoilage	Idem section TVC	-	See section TVC	See section TVC		
			4 °C under MAP conditions/ 16 days	Total viable organisms	Pathogenic and/or spoilage	3% (w/w) thyme EO > 3% (w/w) oregano EO > 1% (w/w) thyme EO > 1% (w/w) oregano EO > control	7.0 log CFU/g for TVC	Uncoated control-up to 8 days	Control-up to 8 days 3% (w/w) oregano EO-up to 16 days 1% (w/w) oregano EO-up to 8 days 3% (w/w) thyme EO-up to 16 days 1% (w/w) thyme EO-up to 16 days		
				Total psychrotrophic bacteria	Pathogenic and/or spoilage	3% (w/w) oregano EO > 1% (w/w) thyme EO > 3% (w/w) thyme EO > 1% (w/w) oregano EO > control	7.0 log CFU/g for TPC	See section TVC	See section TVC		
				<i>Enterobacteriaceae</i>	Pathogenic and/or spoilage	3% (w/w) oregano EO > 3% (w/w) thyme EO > 1% (w/w) thyme EO > 1% (w/w) oregano EO > control	4.0 log CFU/g for <i>Enterobacteriaceae</i>	See section TVC	See section TVC		
				Lactic acid bacteria	Spoilage	3% (w/w) oregano EO > 3% (w/w) thyme EO > 1% (w/w) oregano EO > 1% (w/w) thyme EO > control	-	See section TVC	See section TVC		
				H <sub>2</sub> S producing bacteria	Spoilage	Idem section LAB	-	See section TVC	See section TVC		
				<i>Pseudomonas</i> spp.	Spoilage	1% (w/w) thyme EO > 3% (w/w) oregano EO > 1% (w/w) oregano EO > 3% (w/w) thyme EO > control	-	See section TVC	See section TVC		
				4 °C under air and MAP conditions/ 12 days	Total viable organisms	Pathogenic and/or spoilage	3% (w/w) oregano EO (MAP) > 3% (w/w) oregano EO (air)	7.0 log CFU/g for TVC	Uncoated control (air)-up to 4 days Uncoated (MAP)-up to 4 days		3% (w/w) oregano EO (MAP)-up to 12 days 3% (w/w) oregano EO (air)-up to 4 days
					Total psychrotrophic bacteria	Pathogenic and/or spoilage	Idem section TVC	7.0 log CFU/g for TVC	See section TVC		See section TVC
					<i>Enterobacteriaceae</i>	Pathogenic and/or spoilage	3% (w/w) oregano EO (MAP) > 3% (w/w) oregano EO (air)	4.0 log CFU/g for <i>Enterobacteriaceae</i>	See section TVC		See section TVC
Lactic acid bacteria	Spoilage	Idem section <i>Enterobacteriaceae</i>	-		See section TVC	See section TVC					
H <sub>2</sub> S producing bacteria	Spoilage	Idem section <i>Enterobacteriaceae</i>	-		See section TVC	See section TVC					
<i>Pseudomonas</i> spp.	Spoilage	Idem section <i>Enterobacteriaceae</i>	-		See section TVC	See section TVC					

EO, essential oil; CFU, colony-forming units; TVC, total viable count; TMC, total mesophilic bacteria; TPC, total psychrotrophic bacteria; LAB, Lactic Acid Bacteria; MAL, maximum acceptable level.

### 3.2. Efficacy of Edible Films/Coatings on Enhancing the Shelf-Life of Fresh Fish

The application of above-mentioned edible films and coatings to fish fillets resulted in an extension of their shelf-life as compared to uncoated controls. The film based on 1% quince seed mucilage incorporated with 2% thyme EO prolonged the shelf-life of rainbow trout fillets by 12 days [28] and the one based on 3% gelatin and 1.5% alginate incorporating 1.5% oregano EO by 6 days [29]; the coating based on 2% chitosan incorporated with 1.5% cinnamon EO by 8 days [42], the one based on 1% carrageenan incorporated with 1% lemon EO by 12 days [30], and the one based on 8% whey protein concentrate/glycerol, 2:1 by 6 days [31]. In these cases, the shelf-life was stated considering a maximum acceptable level of 7.0 log CFU/g for the total viable count.

Shelf-lives of silver carp and grass carp fillets were extended by 6 and 13 days, respectively, when coatings based on 2% nanochitosan [49] and 2% chitosan incorporated with 0.3% glycerol monolaurate [32] was used.

When applied to salmon fillets, the film based on 8% gelatin/chitosan, 3:1 incorporated with 7.5% clove EO [39] and coatings based on 1%, 1.5%, and 2% chitosan [46] enhanced the shelf-lives by 6 days.

On beluga sturgeon fillets, the coating based on 8% whey protein concentrate incorporated with 1.5% cinnamon EO [45] extended the shelf-life by 12 days.

The study of Shokri & Ehsani (2017) [33] on pike-perch fillets show a shelf-life prolongation by 8 days when a packaging material based on 10% whey protein isolate incorporated with 2.5% lactoperoxidase was used for coating.

Another study, carried out by Qiu et al. (2014) [44], has shown an increased storage stability (from 8 to 12 days) of Japanese sea bass fillets coated with a solution containing 1.5% chitosan and 0.5% citric acid [44].

The coating formulation of Li et al. (2013) [43], also based on 1.5% chitosan but incorporated with 0.2% tea polyphenols, prolonged the microbiological shelf-life of red drum fillets by 8 days.

In a study on hake fillets, Carrión-Granda et al. (2018) [34] reported a shelf-life prolongation by 8 days when a coating based on 10% whey protein isolate incorporated with 3% oregano EO was used under MAP conditions.

Our review also revealed some studies in the existing literature focused on the application of synthetic films to fresh fish fillets. Cardoso et al. (2017) [50] have tested the efficiency of films based on poly(butylene adipate-co-terephthalate) incorporated with different levels of oregano EO (2.5%, 5.0%, 7.5%, and 10%) in lessening coliform bacteria, *Staphylococcus aureus*, and total psychrotrophic bacteria in fish fillets. The film incorporated with 10% (*w/w*) oregano EO showed the highest inhibitory effect on all bacteria leading to a shelf-life extension of 6 days for wrapped samples. The shelf-life was established considering a maximum acceptable level of 5.0 log CFU/g for *Staphylococcus aureus*.

In another study, Rollini et al. (2016) [51] have evaluated the efficacy of film based on polyethylene terephthalate coated with 3% (*w/v*) lysozyme and lactoferrin water solution, respectively, coextruded multilayer film based on polypropylene incorporated with 4.8% carvacrol against total mesophilic bacteria, total psychrotrophic bacteria, *Enterobacteriaceae* (including coliform bacteria), lactic acid bacteria, *Pseudomonas* spp., and H<sub>2</sub>S producing bacteria. The film that was coated with 3% lysozyme-lactoferrin has shown the best antibacterial results on total mesophilic bacteria, total psychrotrophic bacteria, lactic acid bacteria, and H<sub>2</sub>S producing bacteria, but the one incorporated with 4.8% carvacrol on *Enterobacteriaceae* (including coliform bacteria) and *Pseudomonas* spp. All of the samples were stored for up to four days; therefore, no extension of shelf-life was possible to notice for treated samples in such a short period of storage.

At high levels of incorporation with EOs, active films/coatings may impart foreign flavours to the products on which are applied. Of all the studies that are mentioned in Table 1, only two mentioned their effects on the sensory attributes of fresh fish. The study of Jouki et al. (2014) [28] revealed no significant negative effect of films based on 1% quince seed mucilage incorporated with oregano and thyme EOs in concentrations of up to 2% on the organoleptic acceptability of rainbow trout fillets.

Similar observations were also reported by Ojagh et al. (2010) [42] when a coating based on 2% chitosan incorporated with 1.5% cinnamon EO treatment was applied.

### 3.3. Effects of Edible Films/Coatings on the Chemical Quality of Fresh Fish

Table 2 summarizes the effects of the above-mentioned edible films and coatings on the chemical quality of fresh fish. Chemical indicators of lipid oxidation (TBARS—thiobarbituric acid reactive substances), degradation of nitrogen-containing compounds (TVB-N—total volatile basic nitrogen and TMA-N—trimethylamine nitrogen), and adenosine triphosphate breakdown (*k*-value) were measured during storage of fish fillets.

The thiobarbituric acid reactive substances (TBARS) assay is commonly used to evaluate malondialdehyde (MDA) content. MDA is one of the most significant products of lipid damage [52]. Several researchers [28,33,42] have proposed maximum permitted levels for TBARS although the threshold criteria have not yet received regulatory approval; values  $<3$  mg MDA/kg for perfect quality material,  $3 \leq$  MDA/kg  $< 5$  for good quality material, and  $5 \leq$  MDA/kg  $< 8$  for suitable for human consumption. In the published data reviewed in the current paper, TBARS values ranged from 0.2 to 0.9 mg MDA/kg for rainbow trout fillets, 3.0 to 4.0 mg MDA/kg for silver carp fillets, 0.9 to 1.2 mg MDA/kg for grass carp fillets, 0.06 to 0.12 mg MDA/kg for beluga sturgeon fillets, 1.1 to 1.8 mg MDA/kg for salmon fillets, 1.0 to 2.5 mg MDA/kg for pike-perch fillets, 0.2 to 2.0 mg MDA/kg for Japanese sea bass fillets, and 0.8 to 1.8 mg MDA/kg for red drum fillets; samples meeting the requirements for good quality material, respectively perfect quality material.

Total volatile base nitrogen (TVB-N) is one of the most widely used fish spoilage indicator [53]. It represents the sum of ammonia, methylamine, dimethylamine, trimethylamine, and other basic nitrogenous volatile compounds resulted from fish degradation [54,55]. Commission Regulation (EC) 2074/2005 [56] set limits for TVB-N only for redfish, flatfish, Atlantic salmon, hake, and gadoids; values  $\leq 25$  mg N/100 g for *Sebastes* spp., *Helicolenus dactylopterus*, and *Sebastichthys capensis*,  $\leq 30$  mg N/100 g for species belonging to the *Pleuronectidae* family (with the exception of halibut: *Hippoglossus* spp.), and  $\leq 35$  mg N/100 g for *Salmo salar*, species belonging to the *Merlucciidae* family, and species belonging to the *Gadidae* family. Since no limits of acceptability for rainbow trout, grass carp, beluga sturgeon, pike-perch, Japanese sea bass, and red drum have been established by EC Regulation 2074/2005 [56], the values that were reported previously in the literature were taken as threshold limits by Ojagh et al. (2010) [42], Jouki et al. (2014) [28], Kazemi & Rezaei (2015) [29], Volpe et al. (2015) [30], Yıldız & Yangilar (2016) [31], Yu et al. (2017) [32], Bahram et al. (2016) [45], Shokri and Ehsani (2017) [33], Qiu et al. (2014) [44], and Li et al. (2013) [43]; levels of 25–35 mg N/100 g for rainbow trout,  $\leq 15$  mg N/100 for grass carp, levels of 35–40 mg N/100 g for beluga sturgeon,  $\leq 35$  mg N/100 for pike-perch, levels of 30–35 mg N/100 g for Japanese sea bass, and  $\leq 25$  mg N/100 for red drum. TVB-N values reported in the reviewed studies ranged from 10 to 65 mg N/100 g for rainbow trout fillets, 44 to 60 mg N/100 g for silver carp fillets, 15 to 28 mg N/100 g for grass carp fillets, 50 to 70 mg N/100 g for beluga sturgeon fillets, 28 to 33 mg N/100 g for salmon fillets, 35 to 45 mg N/100 g for pike-perch fillets, 30 to 100 mg N/100 g for Japanese sea bass fillets, 34 to 51 mg N/100 g for red drum fillets, and 11 to 94 mg N/100 g for golden pomfret fillets.

Most marine fish contain TMAO [57]. TMAO is also found, with few exceptions, in freshwater fish, but only in small concentrations [58]. Certain bacteria that occur naturally on the skin, in the guts of fish, and in water can break down TMAO to TMA. The amount of trimethylamine nitrogen (TMA-N) produced is a measure of the activity of spoilage bacteria in the flesh and so is an indicator of the degree of spoilage [57]. There are no regulatory limits available for TMA level in fish. The rejection limit proposed by Jouki et al. (2014) [28] was  $<5$  mg N/100 g and by Souza et al. (2010) [46]  $\leq 5$  mg N/100 g.

**Table 2.** Effects of antimicrobial packaging on chemical quality of fresh fish.

Tested Fish Product	Antimicrobial Packaging Materials		Storage Conditions	ML Obtained for TBARS during Storage	TLV for TBA	ML Obtained for TVB-N during Storage	TLV for TVB-N	ML Obtained for TMA-N during Storage	TLV for TMA-N	ML Obtained for K-Value during Storage	TL for K-Value	Ref.
	Film/Coating	Active Agent/Concentration										
Rainbow trout fillets	Coating based on 2% (w/v) chitosan, acetic acid, and glycerol	Cinnamon EO/1.5% (v/v)	4 °C/16 days	1.5% (v/v) cinnamon EO (~0.2 mg MDA/kg) < uncoated control (below 0.25 mg MDA/kg) < control (below 0.25 mg MDA/kg)	5 mg MDA/kg-good quality; 8 mg MDA/kg-suitable for human consumption	1.5% (v/v) cinnamon EO (~10 mg N/100 g) < control (~20 mg N/100 g) < uncoated control (~40 mg N/100 g)	25 mg N/100 g	-	-	-	-	[42]
	Film based on 1% (w/w) quince seed mucilage, glycerol, and Tween 80	Oregano EO/1%, 1.5%, and 2% (v/v) Thyme EO/1%, 1.5%, and 2% (v/v)	4 °C/18 days	2% (v/v) oregano EO (~0.4 mg MDA/kg) < 1.5% (v/v) oregano EO (~0.4 mg MDA/kg) < % (v/v) thyme EO (below 0.5 mg MDA/kg) < 1% (v/v) oregano EO (below 0.5 mg MDA/kg) < 1.5% (v/v) thyme EO (below 0.6 mg MDA/kg) < 1% (v/v) thyme EO (below 0.6 mg MDA/kg) < control (~0.8 mg MDA/kg) < uncoated control (~0.9 mg MDA/kg)	below 5 mg MDA/kg	2% (v/v) thyme EO (below 20 mg N/100 g) < 1.5% (v/v) thyme EO (below 25 mg N/100 g) < 2% (v/v) oregano EO (below 25 mg N/100 g) < 1% (v/v) thyme EO (below 30 mg N/100 g) < 1.5% (v/v) oregano EO (below 30 mg N/100 g) < 1% (v/v) oregano EO (below 35 mg N/100 g) < control (below 35 mg N/100 g) < uncoated control (below 45 mg N/100 g)	25 mg N/100 g	2% (v/v) thyme EO (~5 mg N/100 g) < 1.5% (v/v) thyme EO (~6 mg N/100 g) < 2% (v/v) oregano EO (~6 mg N/100 g) < 1% (v/v) thyme EO (below 8 mg N/100 g) < 1.5% (v/v) oregano EO (below 8 mg N/100 g) < 1% (v/v) oregano EO (~8 mg N/100 g) < uncoated control (~12 mg N/100 g) < control (~12 mg N/100 g)	below 5 mg N/100 g	-	-	[28]
	Film based on 3% (w/v) gelatin and 1.5% (w/v) alginate, glycerol, and Tween 80	Oregano EO/1.5% (w/v)	4 °C/15 days	-	-	1.5% (w/v) oregano EO (~60 mg N/100 g) < control (~65 mg N/100 g) < uncoated control (~65 mg N/100 g)	35 mg N/100 g	-	-	-	-	[29]
	Coating based on 1% (w/w) carrageenan	Lemon EO/1% (w/w)	4 °C/15 days	-	-	1% (w/w) lemon EO (20 mg N/100 g) < control (below 35 mg N/100 g) < uncoated control (40 mg N/100 g)	25 mg N/100 g	-	-	-	-	[30]
	Coating based on 8% (w/w) whey protein concentrate Coating based on 8% (w/w) whey protein concentrate/glycerol, 1:1 and 2:1	-	4 °C/15 days	8% (w/w) whey protein concentrate/glycerol, 2:1 (0.4 mg MDA/kg) < 8% (w/w) whey protein concentrate/glycerol, 1:1 (0.5 mg MDA/kg) < 8% (w/w) whey protein concentrate (0.6 mg MDA/kg) < uncoated control (0.7 mg MDA/kg)	-	8% (w/w) whey protein concentrate/glycerol, 2:1 (21.1 mg N/100 g) < 8% (w/w) whey protein concentrate/glycerol, 1:1 (24.6 mg N/100 g) < 8% (w/w) whey protein concentrate (27.4 mg N/100 g) < uncoated control (32.5 mg N/100 g)	25 mg N/100 g	-	-	-	-	-

Table 2. Cont.

Tested Fish Product	Antimicrobial Packaging Materials		Storage Conditions	ML Obtained for TBARS during Storage	TLV for TBA	ML Obtained for TVB-N during Storage	TLV for TVB-N	ML Obtained for TMA-N during Storage	TLV for TMA-N	ML Obtained for K-Value during Storage	TL for K-Value	Ref.
	Film/Coating	Active Agent/Concentration										
Silver carp fillets	Coating based on 2% (w/v) chitosan and glycerol/Coating based on 2% (w/v) nanochitosan and glycerol	-	4 °C/12 days	2% (w/v) chitosan (below 3 mg MDA/kg) < 2% (w/v) nanochitosan (below 3 mg MDA/kg) < uncoated control (below 4 mg MDA/kg) < 1% glacial acetic acid (~4 mg MDA/kg)	-	2% (w/v) nanochitosan (44.4 mg N/100 g) < 2% (w/v) chitosan (30.8 mg N/100 g) < 1% glacial acetic acid (below 60 mg N/100 g) < uncoated control (~60 mg N/100 g)	-	-	-	-	-	[49]
Grass carp fillets	Coating based on 2% (w/v) chitosan, acetic acid, and glycerol	Glycerol monolaurate/ 0.1% and 0.3%	4 °C/ 20 days	0.3% glycerol monolaurate (~0.9 mg MDA/kg) < 0.1% glycerol monolaurate (~0.9 mg MDA/kg) < control (~0.9 mg MDA/kg) < uncoated control (below 1.2 mg MDA/kg)	-	0.3% glycerol monolaurate (15 mg N/100 g) < 0.1% glycerol monolaurate (below 20 mg N/100 g) < control (~22.5 mg N/100 g) < uncoated control (below 1.2 mg MDA/kg) (~27.5 mg N/100 g)	15 mg N/100 g	-	-	0.3% glycerol monolaurate (~69%) < 0.1% glycerol monolaurate (77.7%) < control (78.2%) < uncoated control (90.5%)	<20%-vf; <60%-mf; >60%-rp	[32]
Beluga sturgeon fillets	Coating based on 8% (w/v) whey protein concentrate, glycerol, and Tween 80	Cinnamon EO/1.5% (v/v)	4°C/ 20 days	1.5% (v/v) cinnamon EO (below 0.06 mg MDA/kg) < control (below 0.1 mg MDA/kg) < uncoated control (below 0.12 mg MDA/kg)	-	1.5% (v/v) cinnamon EO (~50 mg N/100 g) < control (below 70 mg N/100 g) < uncoated control (~70 mg N/100 g)	35–40 mg N/100 g	-	-	-	-	[45]
Salmon fillets	Coating based on 1.0%, 1.5%, and 2% (w/v) chitosan, lactic acid solution, and Tween 80	-	0 °C/ 18 days	All treated samples (1.1 mg MDA/kg) < uncoated control (1.8 mg MDA/kg)	1 mg MDA/kg	All treated samples (28 mg N/100 g) < uncoated control (33 mg N/100 g)	30 mg TVB-N/100 g	All treated samples (5 mg N/100 g) < uncoated control (6 mg N/100 g)	5 mg N/100 g	All treated samples (46%) < uncoated control (50%)	40%	[46]
Pike-perch fillets	Coating based on 10% (w/v) whey protein isolate, glycerol, and Tween 80	Lactoperoxidase/ 2.5% (v/v)	4 °C/ 16 days	3% (v/v) α-tocopherol (below 1 mg MDA/kg) < 2.5% (v/v) lactoperoxidase and 3% (v/v) α-tocopherol (below 1 mg MDA/kg) < 1.5% (v/v) α-tocopherol (~1 mg MDA/kg) < 2.5% (v/v) lactoperoxidase and 1.5% (v/v) α-tocopherol (~1 mg MDA/kg) < control for other coatings (below 2.5 mg MDA/kg) < control for coating with lactoperoxidase (below 2.5 mg MDA/kg) < 2.5% (v/v) lactoperoxidase (~2.5 mg MDA/kg)	below 3 mg MDA/kg-perfect quality material; below 5 mg MDA/kg-good quality material	2.5% (v/v) lactoperoxidase (below 35 mg N/100 g) < 2.5% (v/v) lactoperoxidase and 1.5% (v/v) α-tocopherol (below 40 mg N/100 g) < control for coating with lactoperoxidase (~40 mg N/100 g) < control for other coating! (~40 mg N/100 g) < 3% (v/v) α-tocopherol (below 45 mg N/100 g) < 1.5% (v/v) α-tocopherol (below 45 mg N/100 g) < 2.5% (v/v) lactoperoxidase and 3% (v/v) α-tocopherol (below 45 mg N/100 g)	35 mg N/100 g	-	-	-	-	[33]
	Coating based on 10% (w/v) whey protein isolate, glycerol, ethanol, and Tween 80	α-Tocopherol/ 1.5% (v/v) α-Tocopherol/ 3% (v/v) Lactoperoxidase and α-tocopherol/ 2.5% (v/v) and 1.5% (v/v) Lactoperoxidase and α-tocopherol/ 2.5% (v/v) and 3% (v/v)										

Table 2. Cont.

Tested Fish Product	Antimicrobial Packaging Materials		Storage Conditions	ML Obtained for TBARS during Storage	TLV for TBA	ML Obtained for TVB-N during Storage	TLV for TVB-N	ML Obtained for TMA-N during Storage	TLV for TMA-N	ML Obtained for K-Value during Storage	TL for K-Value	Ref.
	Film/Coating	Active Agent/Concentration										
Japanese sea bass fillets	Coating based on 1.5% (w/v) chitosan and acetic acid	Citric acid/0.5% (w/v) Licorice extract/1% (w/v)	4 °C/ 12 days	0.5% (w/v) citric acid (~0.2 mg MDA/kg) < 0.2% (w/v) licorice extract (~0.2 mg MDA/kg) < control (below 1.5 mg MDA/kg) < uncoated control (below 2.0 mg MDA/kg)	–	0.5% (w/v) citric acid (29.7 mg N/100 g) < 1% (w/v) licorice extract (48.0 mg N/100 g) < control (60.5 mg N/100 g) < uncoated control (100.2 mg N/100 g)	30–35 mg N/100 g	–	–	–	–	[44]
Red drum fillets	Coating based on 1.5% chitosan, acetic acid, and glycerol	Grape seed extract/0.2% (w/v) Tea polyphenols/0.2% (w/v)	4 °C/20 days	0.2% (w/v) tea polyphenols (~0.8 mg MDA/kg) < 0.2% (w/v) grape seed extract (~1.0 mg MDA/kg) < uncoated control (~1.8 mg MDA/kg)	–	0.2% (w/v) tea polyphenols (33.69 mg N/100 g) < 0.2% (w/v) grape seed extract (38.17 mg N/100 g) < uncoated control (51.25 mg N/100 g)	25 mg N/100 g	–	–	0.2% (w/v) tea polyphenols (~40%) < 0.2% (w/v) grape seed extract (~45%) < uncoated control (62.57%)	60%	[43]
Golden pomfret fillets	Coating based on 0.4% (w/w) chitosan	–	4 °C/ 17 days	–	–	0.4% (w/w) chitosan and 7.2% (w/w) gelatin (10.51 mg N/100 g) < 0.4% (w/w) chitosan and 5.4% (w/w) gelatin (12.31 mg N/100 g) < 0.4% (w/w) chitosan and 3.6% (w/w) gelatin (13.48 mg N/100 g) < deionized water (93.52 mg N/100 g)	–	–	–	–	–	[47]
	Coating based on 0.4% (w/w) chitosan and gelatin											

TBARS, thiobarbituric acid reactive substances; TLV, threshold limit value; ML, maximum levels; TVB-N, total volatile basic nitrogen; TMA-N, trimethylamine nitrogen; MDA, malondialdehyde; vf, very fresh; mf, moderately fresh; rp, rejection point.

K-value is an important chemical index widely used for fish freshness [59]. During post-mortem storage of fish, autolytic changes take place in the muscle that determines adenosine triphosphate (ATP) degradation with the formation of adenosine-5'-diphosphate (ADP), adenosine-5'-monophosphate (AMP), inosine-5'-monophosphate (IMP), inosine (HxR), and hypoxanthine (Hx). K-value is calculated as the percentage of the sum of HxR and Hx, divided by the sum of ATP, ADP, AMP, IMP, HxR, and Hx [12,59]. Since there are no legally enforceable limits for k-value in fish, Yu et al. (2017) [32] proposed the following freshness criteria: very fresh fish ( $k$ -value < 20%), moderately fresh ( $k$ -value < 60%), and spoiled ( $k$ -value > 60%). K-values reported in the discussed studies ranged from 68.7% to 90.5% for grass carp fillets, 46% to 50% for salmon fillets and 40% to 62.6% for red drum fillets; samples meeting freshness criteria for moderately fresh, respectively spoiled.

#### 4. Conclusions

The active packaging of fish represents an economic alternative to conventional preservation technologies (vacuum and modified atmosphere packaging) due to the limited capital investment as compared to those. Besides being biodegradable, edible films and coatings improve the microbiological stability of fish and reduce waste; moreover, retard lipid oxidation. For the past 10 years, research on the use of antimicrobial packaging materials for fresh fish applications has undergone considerable evolution; nevertheless, as far as we know, there is not yet an edible film or coating commercially available on the market.

Fish represent one of the most-traded segments of the world food sector. Therefore, there is a great demand for the packaging of this good. Industrial production and commercialization of antimicrobial packaging materials for fresh fish could be an exploitable sector by the packaging industry. Suppliers of active packaging materials on the European market need to make sure that their products comply with the requirements of Regulations (EC) 1935/2004 [60] and (EC) 450/2009 [61] regarding active and intelligent materials that are intended to come into contact with food, respectively, Regulation (EC) 1333/2008 [62] that lays down specifications for food additives. Additional studies are however needed to further validate these findings, especially on the stability of antimicrobial films/coatings during shipment, storage, and handling.

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