



VICTORIA UNIVERSITY
MELBOURNE AUSTRALIA

Essential Oils and Their Principal Constituents as Antimicrobial Agents for Synthetic Packaging Films

This is the Accepted version of the following publication

Kuorwel, Kuorwel Kuai, Cran, Marlene, Sonneveld, Kees, Miltz, Joseph and Bigger, Stephen W (2011) Essential Oils and Their Principal Constituents as Antimicrobial Agents for Synthetic Packaging Films. *Journal of Food Science*, 76 (9). R164-R177. ISSN 0022-1147 (print) 1750-3841 (online)

The publisher's official version can be found at
<http://onlinelibrary.wiley.com/doi/10.1111/j.1750-3841.2011.02384.x/abstract>
Note that access to this version may require subscription.

Downloaded from VU Research Repository <https://vuir.vu.edu.au/7673/>

1 **Essential Oils and Their Principal Constituents as Antimicrobial Agents for Synthetic**
2 **Packaging Films**

3
4 Kuorwel K. Kuorwel¹, Marlene J. Cran², Kees Sonneveld³,
5 Joseph Miltz⁴ and Stephen W. Bigger^{1*}

6
7 1. School of Engineering and Science, 2. Institute for Sustainability and Innovation
8 Victoria University, PO Box 14428, Melbourne, 8001, Australia.

9 3. KS PackExpert & Associates, PO Box 399, Mansfield, 3724, Australia

10 4. Department of Biotechnology and Food Engineering, Technion-Israel Institute of
11 Technology, Haifa, 3200, Israel

12
13
14 ***Corresponding Author**

15 Prof. Stephen W. Bigger

16 School of Engineering and Science, Victoria University,

17 PO Box 14428, Melbourne, 8001, Australia

18 Tel: +61 3 9919 2959

19 Fax: +61 3 9919 2005

20 E-mail: stephen.bigger@vu.edu.au

21
22
23 **Short version of title:** AM Agents Used in Synthetic Films (. . .)

24 **Choice of journal section:** Concise Reviews and Hypotheses in Food Science

25 **Word Count:** 12606

27 **Abstract**

28 Spices and herbal plant species have been recognized to possess a broad spectrum of active
29 constituents that exhibit antimicrobial (AM) activity. These active compounds are produced
30 as secondary metabolites associated with the volatile essential oil (EO) fraction of these
31 plants. A wide range of AM agents derived from EOs have the potential to be used in AM
32 packaging systems which is one of the promising forms of active packaging systems aimed at
33 protecting food products from microbial contamination. Many studies have evaluated the AM
34 activity of synthetic AM and/or natural AM agents incorporated into packaging materials and
35 have demonstrated effective AM activity by controlling the growth of microorganisms. This
36 review examines the more common synthetic and natural AM agents incorporated into or
37 coated onto synthetic packaging films for AM packaging applications. The focus is on the
38 widely studied herb varieties including basil, oregano and thyme and their essential oils.

39

40 **Keywords:** Essential oils, natural AM agents, active packaging, antimicrobial packaging,
41 microbial contamination

42

43 **1 Introduction**

44 Food products can be subjected to microbial contamination that is mainly caused by bacteria,
45 yeasts and fungi. Many of these microorganisms can cause undesirable reactions that
46 deteriorate the flavour, odour, colour, sensory, and textual properties of foods (Appendini and
47 Hotchkiss 1997; Vermeiren and others 1999; Weng and others 1999; Appendini and
48 Hotchkiss 2002; Vermeiren and others 2002; Devlieghere and others 2004a; Han 2005;
49 Rupika and others 2005; Davidson and Taylor 2007; Gutierrez and others 2008). Microbial
50 growth in food products is a major concern because some microorganisms can potentially
51 cause food-borne illness (Padgett and others 1998; Natrajan and Sheldon 2000; Cha and
52 Chinnan 2004; Davidson and others 2005; de Oliveira and others 2007). In packaged foods,
53 the growth and survival of common spoilage and pathogenic microorganisms such as *Listeria*
54 *monocytogenes*, *Escherichia coli* O157, *Salmonella*, *Staphylococcus aureus*, *Bacillus cereus*,
55 *Campylobacter*, *Clostridium perfringens*, *Aspergillus niger* and *Saccharomyce cerevisiae* are
56 affected by a variety of intrinsic factors such as pH, water activity, and the presence of
57 oxygen or by extrinsic factors associated with storage conditions including temperature, time
58 and relative humidity (Singh and others 2003; López-Malo and others 2005; Rydlo and others
59 2006). Many food products including various types of cheeses, meats, poultry and baked
60 products are highly susceptible to microbial spoilage (Weng and Hotchkiss 1993; Suppakul
61 2004; Limjaroen and others 2005; Schelz and others 2006; Silveira and others 2007).

62

63 To prevent the growth of spoilage and pathogenic microorganisms on foods, various
64 traditional preservation techniques such as heat treatment, salting, acidification and drying
65 are used in the food industry (Quintavalla and Vicini 2002; Ozdemir and Floros 2004;
66 Davidson and Taylor 2007; Farkas 2007). In recent years, a rise in consumer demand for safe,
67 fresh and minimally-processed foods, has led to the development of new preservation

68 techniques. Active packaging (AP) technologies, for example, can provide safe food products
69 with longer shelf lives (Rooney 1995; Lau and Wong 2000; Vermeiren and others 2002;
70 Fitzgerald and others 2003; Ozdemir and Floros 2004; Gutierrez and others 2008). In the
71 food industry, spoilage of food products, including spoilage that is caused by microorganisms
72 is a major concern. The AP technologies designed primarily to protect food products from
73 deterioration and from the growth of microorganisms can involve the use of synthetic or
74 natural antimicrobial (AM) agents (Juneja and Sofos 2005). To diminish food spoilage by
75 microorganisms, different AM agents (primarily synthetic) are commonly incorporated
76 directly into the food. This method has many disadvantages: (i) consumers prefer foods with
77 no or minimal synthetic additives because of concerns about side-effects; (ii) since food
78 spoilage occurs primarily on the surface, incorporation of relatively large quantities of the
79 agents in the bulk of the food is not justified; (iii) some of the synthetic agents possess a
80 distinct flavour that may rendered the food flavour, and (iv) synthetic additives have to be
81 declared on the package. Therefore, packaging materials that incorporate in them the AM
82 agent as an additional protective barrier are emerging as the preferred preservation method.
83 Several authors have reported AP technologies that involve the use of films produced from
84 synthetic polymers (Miltz and others 1995; Rooney 1995; Smith and others 1995). These
85 materials can act as carriers for active agents, including AM compounds, in order to maintain
86 high concentrations of the agent on or near the food surface to control or prevent the growth
87 of spoilage and pathogenic microorganisms (Krochta and De Mulder-Johnston 1997; Joerger
88 2007; Raybaudi-Massilia and others 2009; Rojas-Graü and others 2009; Suppakul and others
89 2011a). Thus a packaging film impregnated or coated with an AM agent could potentially
90 extend the shelf life and improve the microbial safety of food products (Appendini and
91 Hotchkiss 2002; Suppakul and others 2003b; Burt 2004; Kuorwel and others 2011b).

92 Although AM agents such as essential oils (EOs) and/or their principal components may
93 exhibit AM activity against various microorganisms when incorporated into packaging
94 materials, the organoleptic properties of the packaged food products are one of the important
95 factors that must also be taken into consideration. According to Davison and Zivanovic
96 (2003), the concentration of AM agents required to demonstrate AM activity against various
97 microorganisms on food products might be higher than the concentration applied for
98 flavouring purposes. As a result, this might cause food tainting and/or adverse sensorial
99 effects to food products (Smith-Palmer and others 2001; Bagamboula and others 2004). The
100 adverse sensorial effects of AM agents to food products can be overcome by masking the
101 odor of AM agents with other approved aroma compound as suggested by Gutiérrez and
102 others (2009). An understanding of the relationship between minimum inhibitory
103 concentration and acceptable organoleptic properties of AM Agents such as EOs and/or their
104 constituents is also important (Lambert and others 2001). In some cases, the replacement of
105 EOs with one or a number of their principal constituents may provide equal AM effectiveness
106 but with milder flavouring attributes (Lambert and others 2001; Smith-Palmer and others
107 2001).

108

109 In a recent review, the current authors presented an evaluation of the AM activity of
110 biodegradable polysaccharide and protein based films containing natural agents (Kuorwel and
111 others 2011b). These films showed the potential for wide-range of applications in food
112 packaging where undesirable microbial growth is a concern. Moreover, these films degrade
113 readily in the environment but the acquisition of this attribute may harm the processability
114 and mechanical stability of the film. Thus, in spite of the increasing concern in recent years
115 about the use of synthetic polymers due to their poor biodegradability, these materials have
116 several advantages including low cost, good processability and sound mechanical and

117 physical properties. Therefore, development of AM packaging materials manufactured from
118 synthetic polymers such as low-density polyethylene (LDPE), high-density polyethylene
119 (HDPE), polystyrene (PS), polyethylene terephthalate (PET) and polypropylene (PP) is still
120 important in offering commercial benefits for packaging food products. In the current review,
121 a detailed summary of synthetic films utilising common synthetic and natural AM agents is
122 presented with an emphasis on the principal components of basil, oregano, and thyme
123 essential oils (EOs) namely, linalool, carvacrol and thymol respectively. This is followed by a
124 list of other natural AM agents that have the potential for controlling microbial growth on
125 foods.

126

127 **2 Antimicrobial Packaging Systems**

128 Studies have shown that AM packaging systems can increase the shelf life of packaged foods
129 by extending the lag phase and reducing the growth rate of spoilage microorganisms (Han
130 2000; Cooksey 2001; Appendini and Hotchkiss 2002; Rydlo and others 2006; Coma 2007; de
131 Oliveira and others 2007; Gutierrez and others 2008; Rardniyom and others 2008; Rupika
132 and others 2008). In the past, preservatives were directly added into food products to protect
133 them from microbial contamination. This process of direct addition of preservatives into
134 foods may result in levels of additives in excess to those required for an efficient AM effect.
135 New AM packaging systems have attracted much attention in the food industry with the aim
136 of replacing the conventional food preservation systems (Weng and Hotchkiss 1993; An and
137 others 1998; Quintavalla and Vicini 2002; Bagamboula and others 2004; Devlieghere and
138 others 2004b; Miltz and others 2006).

139

140 An AM packaging system can be produced by: directly incorporating AM agents into
141 packaging films; coating of the packaging films with AM agents; developing packaging

142 materials from polymers that have inherent AM properties (Vermeiren and others 2002;
143 Suppakul and others 2003b). Typically, AM packaging systems can be regarded as migrating
144 or non-migrating with the distinction depending on the specific AM agent used and on its
145 interactions with the packaging and food matrix (Cooksey 2000). In a migrating system, the
146 AM agent is released from the packaging film into the package headspace and onto the food
147 surface and such systems are most useful when direct contact between the packaging film and
148 food product is not required for efficient AM activity (Weng and Hotchkiss 1993; Cooksey
149 2000). The non-migrating systems involve packaging materials in which the AM agent is
150 immobilised within the material (Brody and others 2001) and these systems can be applied
151 where direct contact between the food and the material can be achieved or is required for
152 effective AM activity (Vermeiren and others 2002; Suppakul and others 2003b). In either AM
153 packaging system, both synthetic and natural AM agents can be incorporated into or coated
154 onto the packaging material. The mode of action of AM agents incorporated in a packaging
155 material is influenced by the controlled and slow release of the agent onto the food surfaces.
156 This is required in order to maintain an adequate concentration of the agent on the food and
157 effectively inhibit microbial growth throughout the product shelf life (Cooksey 2005; Salleh
158 and others 2007; Cran and others 2010; Tunç and Duman 2011). Han (2005) suggested that
159 the mass transfer rate of an AM agent should not be faster than the growth rate of the target
160 microorganism, otherwise the AM agent might be diluted on the surface of the packaged food
161 product, thus limiting the AM activity.

162

163 During extrusion or compression moulding of AM films, the temperature and mechanical
164 energy input, such as shearing forces, must be carefully considered (Han 2003). High-
165 processing temperatures, for example, may result in considerable losses of volatile AM
166 agents (Han and Floros 1997; Han 2000; Rupika and others 2005; Suppakul and others

167 2011b). Moreover, Cooksey (2005) suggested that an AM agent might partly or completely
168 lose its AM activity if incorporated into a film under harsh processing conditions. Therefore,
169 to minimise the loss of AM agent during processing, temperatures that are as low as possible
170 should be applied (Han and Floros 1998; Suppakul and others 2011b). The storage
171 temperature may also influence the activity of AM agents that are incorporated into
172 packaging films (Vojdani and Torres 1989; Han 2005). The concentration of AM agents
173 retained in the film may decrease during long-term storage. However, the amount of AM
174 agent retained in the film after a long storage period may be sufficient to demonstrate AM
175 activity as shown by Suppakul and others (2011b). Du and others (2008) reported AM
176 activity against *E. coli* (using an agar disc diffusion method) of carvacrol incorporated into
177 films for edible apples that were stored for 7 weeks.

178

179 **2.1 Synthetic AM Agents**

180 In the past few decades, various synthetic AM agents have been investigated and developed
181 into food packaging materials (Weng and Hotchkiss 1992; Weng and Hotchkiss 1993). Many
182 of these agents including various organic acids and salts have been approved by regulatory
183 agencies and have since been used for the preservation of food products (Davidson and
184 Taylor 2007). Synthetic AM agents that have demonstrated inhibitory activity against
185 different microorganisms include sodium benzoates and propionates, potassium sorbates,
186 sulfites, chlorides, nitrites, triclosan, fungicides (e.g. benomyl, imazalil) and various metal
187 ions including silver zeolites, quaternary ammonium salts and copper ions (Chen and others
188 1996; Devlieghere and others 2000a; Han 2000; Ouattara and others 2000; Hoffman and
189 others 2001; Cooksey 2005). Other AM agents such as acetic acid from vinegar and benzoic
190 acid from cranberries are found in nature, but are classified as synthetic AM agents when
191 produced synthetically (Davidson and Taylor 2007).

192 Many synthetic AM compounds have been evaluated in synthetic polymeric materials by
193 various researchers. Table 1 summarises these synthetic AM agents incorporated into or
194 coated onto packaging materials as potential candidates for food packaging. Although Table
195 1 contains a large amount of information on the activity of AM agents successfully
196 incorporated into various synthetic polymers, comparison between the different AM agents
197 and/or AM films is difficult due to variations in strains of microorganisms and different
198 experimental conditions or equipment used by the various researchers. In order to compare
199 the results of various experiments involving AM agents, there is a need for a standardisation
200 of the test methods as suggested by Suppakul and others (2003a).

201

202 Numerous studies have concentrated on incorporating common food preservatives such as
203 organic acids, their salts and anhydrides into packaging films (see Table 1). Studies on
204 benzoic or sorbic acid incorporated into packaging materials have evaluated their action
205 against various microorganisms in laboratory media such as agar plates and/or in actual food
206 products. The packaging films incorporated with these organic acids or anhydrides have
207 demonstrated inhibitory effects against various spoilage and pathogenic microorganisms.
208 Weng and others (1999) showed that benzoic acid or sorbic acid incorporated into
209 poly(ethylene-co-methacrylic acid) (PEMA) film inhibited the growth of *A. niger* and
210 *Penicillium* sp. on solid media. Weng and Chen (1997) investigated the AM activity of
211 benzoic acid or benzoyl chloride incorporated into ionomer films. The AM activity of these
212 films was demonstrated by their ability to inhibit the growth of *Penicillium* sp. and *A. niger*.
213 In an earlier study, Weng and Hotchkiss (1993) incorporated benzoic acid or benzoic
214 anhydride into LDPE films which significantly suppressed the growth of *Rhizopus stolonifer*,
215 *Penicillium* sp. and *A. toxacarius* on potato dextrose agar and on the surface of Cheddar
216 cheese. Matche and others (2006) examined the AM activity of benzoyl chloride incorporated

217 into modified ethylene acrylic acid films against *Penicillium* sp. and *A. niger* sp. on solid
218 media for 15 days with the film demonstrating inhibition against both species. Silveira and
219 others (2007) incorporated sorbic acid into LDPE films with the aim of preserving fresh
220 pastry dough. It was found that 3% (w/w) sorbic acid incorporated into a 70 µm film reduced
221 2 and 1.5 log cycles of mesophilic and psychrotrophic bacteria respectively on the pastry
222 dough after 40 days of storage at 8°C compared to the control film. Limjaroen and others
223 (2005) coated sorbic acid onto polyvinylidene chloride copolymer films to control the growth
224 of *L. monocytogenes* on beef bologna and Cheddar cheese. It was found that sorbic acid
225 coated on the films inhibited microbial growth on cheese by log 0.6 CFUg⁻¹ after 28 days of
226 storage at 4°C. They further reported that the population of *L. monocytogenes* on beef
227 bologna was reduced by log 0.6 and 1.4 CFUg⁻¹ for films containing 1.5% and 3.0% (w/v)
228 respectively compared to the control film.

229

230 Other researchers have studied the AM activity of salts against several microorganisms as
231 shown in Table 1. Han and Flores (1997) developed LDPE films containing potassium
232 sorbate and found that these films successfully reduced the growth of *S. cerevisiae* *in vitro*
233 experiments. Vartiainen and others (2003b) demonstrated that potassium sorbate, sodium
234 benzoate and sodium nitrate incorporated into LDPE, poly(maleic acid-co-olefine), PET or
235 PS films inhibited the growth of *B. cereus* on culture media. Limjaroen and others (2003)
236 coated potassium sorbate onto polyvinylidene chloride copolymer films and reported that the
237 films inhibited the growth of *L. monocytogenes* on solid media.

238

239 In addition to the antibacterial activity, the antifungal activity of synthetic AM agents
240 incorporated in polymeric materials has been investigated (Halek and Anita 1989; López-
241 Malo and others 2007). Vartiainen and others (2003a) examined the inhibitory effects of

242 imazalil incorporated into LDPE against the growth of *A. niger* by the agar diffusion assay
243 with films containing 0.05-0.25% (w/w) imazalil demonstrating significant inhibitory
244 activity. Weng and Hotchkiss (1992) incorporated imazalil into LDPE film and evaluated the
245 antimycotic activity of this agent on the growth of *A. toxacarius* and *Penicillium* sp. on potato
246 dextrose agar (PDA) and Cheddar cheese. They reported that 2 g kg⁻¹ of imazalil suppressed
247 the growth of *A. toxacarius* on PDA whereas a film containing 1 g kg⁻¹ of imazalil reduced
248 the growth of *Penicillium* sp. The latter film inhibited the growth of both mould species on
249 the surface of Cheddar cheese. López-Malo and others (2002; 2005) examined the antifungal
250 activity of potassium sorbate, sodium benzoate and sodium bisulfite against the growth of
251 *Aspergillus flavus* inoculated on laboratory media with each of the agents imparting an
252 inhibitory effect. López-Malo and others (2007) investigated the antifungal activity of sodium
253 benzoate and cinnamon extract, separately or in combination, against the growth of *A. flavus*
254 on potato dextrose agar or a checkerboard array respectively. They found that both AM
255 agents demonstrated antifungal activity on *A. flavus*, with cinnamon extract being more
256 effective than sodium benzoate. They claimed that mixtures of cinnamon extract and sodium
257 benzoate showed promising antifungal activity.

258

259 Triclosan and hexamethylenetetramine are common synthetic AM agents that have been
260 evaluated in packaging systems with some commercial developments. Vermeiren and others
261 (2002) investigated the AM activity of triclosan incorporated into LDPE film and reported
262 that concentrations of 0.5 and 1.0% (w/w) demonstrated AM activity against *L.*
263 *monocytogenes*, *Sal. enteritidis*, *Staph. aureus*, *E. coli* O157:H7 and *Brocothrix*
264 *thermosphacta* in an agar diffusion assay. Cutter (1999) reported that triclosan was effective
265 against bacteria on the surface of beef. Recently, Camilloto and others (2010) studied the
266 activity of triclosan incorporated into LDPE against *Staph. aureus*, *E. coli*, *L. innocua* and *P.*

267 *aeruginosa* in the agar disc diffusion test and found that the AM film inhibited the growth of
268 *Staph. aureus* and *E. coli*. Chung and others (2003a) investigated the AM activity of triclosan
269 coated onto styrene-acrylate copolymer against *Enterococcus faecalis* on solid and in liquid
270 media and showed effective inhibition of the bacteria. Ji and Zhang (2009) reported that
271 triclosan incorporated into PVC film inhibited the growth of *Staph. aureus* and *E. coli* using
272 the plate-counting technique. Devlieghere and others (2000b) studied the AM activity of
273 hexamethylenetetramine impregnated into an LDPE film and found it to be effective against
274 spoilage microorganisms on cooked ham.

275

276 **2.2 Natural Antimicrobial Agents**

277 In recent years, natural AM agents have attracted much attention in the food and packaging
278 industries as a replacement for synthetic ones for food preservation. According to Davidson
279 and Zivanovic (Davidson and Zivanovic 2003), natural AM agents are classified by their
280 sources: AM agents derived from plant EOs (e.g. basil, thyme, oregano, cinnamon, clove and
281 rosemary); animal sources (e.g. lysozyme, lactoferrin); microbial sources (nisin, natamycin);
282 and naturally occurring polymers (chitosan). The EOs extracted from plant sources consist of
283 various mixtures including terpenoids, esters, aldehydes, ketones, acids and alcohols
284 (Dorman and Deans 2000). These plant EOs are volatile and generally possess relatively
285 strong odours (Bakkali and others 2008).

286

287 Extracts derived from various herbs and EOs contain a range of natural compounds such as
288 thymol, linalool and carvacrol which have a broad AM spectrum against different pathogenic
289 and spoilage microorganisms including Gram-negative species such as *E. coli*, *Yersinia*
290 *enterocolitica*, *P. aeruginosa* and *Sal. choleraesuis* (López and others 2007a; Suppakul and
291 others 2011b), Gram-positive bacteria such as *L. monocytogenes*, *Staph. aureus*, *B. cereus*

292 (Friedman and others 2002; López and others 2007b; Gutiérrez and others 2009), yeasts such
293 as *S. cerevisiae*, *Candida albicans*, *Debaryomyces hansenii* (Rupika and others 2006;
294 Suppakul and others 2008; Kuorwel and others 2011a) and moulds such as *Alternaria*
295 *alternate*, *A. niger*, *Botrytis cinerae*, *A. flavus*, *penicillium roqueforti* (López-Malo and others
296 2007; Rodríguez-Lafuente and others 2010). These additives are considered to be safe and
297 have the "Generally Recognised As Safe" (GRAS) status as designated by the American
298 Food and Drug Administration (Zaika 1988; Han 2005; Matan and others 2006).
299 Antimicrobial agents derived from plant sources are produced as secondary metabolites and
300 are associated generally with the volatile EO fractions. The mode of action of AM agents
301 and/or AM activity of plant EOs is related to their chemical structure namely, the presence of
302 hydrophilic functional groups such as the hydroxyl groups of phenolic components and/or
303 lipophilicity of the components in the EOs which depends on their concentration (Frag and
304 others 1989; Davidson and Naidu 2000; Dorman and Deans 2000; Friedman and others 2002;
305 Bagamboula and others 2004). [Essential oils and their principal constituents inhibit](#)
306 [microorganisms via a range of mechanisms such as: disruption of the cytoplasmic](#)
307 [membrane \(Knobloch and others 1989; Sikkema and others 1995; Helander and others 1998\);](#)
308 [leakage of intracellular constituents such as metabolites and ions \(Sikkema and others 1995;](#)
309 [Lambert and others 2001\); coagulation of cell content \(Gustafson and others 1998; Pauli](#)
310 [2001\); inhibition of protein synthesis \(Helander and others 1998\), enzymes associated with](#)
311 [cell wall synthesis \(Conner and Beuchat 1984\), DNA/RNA synthesis \(Ultee and others 1999;](#)
312 [Tassou and others 2000\), general/metabolite pathways \(Ultee and others 2002\); and/or the](#)
313 [destruction of the osmotic integrity of the cell membrane \(Ultee and Smid 2001\).](#) The AM
314 activity of different EOs is very difficult to compare given the variation of EOs compositions
315 amongst the plant species, differences in the geographic origin of the plants, harvesting

316 season, extraction methods and the part of plant that is used (Zaika 1988; Elgayyar and others
317 2001).

318 There are a number of test methods used to determine the AM activity of various EOs and
319 their principal constituents. These include diffusion methods (agar diffusion), dilution
320 methods (broth and agar dilution) and microatmosphere methods (Davidson and Zivanovic
321 2003; Guynot and others 2003; Nedorostova and others 2011; Tunç and Duman 2011). These
322 test methods provide preliminary information on the possible effectiveness of the tested
323 active constituents. The agar diffusion method has been widely used in the past, but the
324 results obtained from this technique are qualitative. Although the agar diffusion method can
325 indicate the AM activity of EOs and/or their principal components on solid media, the high
326 hydrophobicity of EOs is always a major problem (Davidson and Zivanovic 2003). As a
327 result, the agar disc diffusion assays do not generally demonstrate a clear zone of inhibition at
328 very low concentrations; however these do exhibit a clear inhibition zone at high
329 concentrations of hydrophobic, lipophilic AM agents (Friedman and others 2002; Sanla-Ead
330 and others 2011). Conversely, microatmosphere methods, which allow the determination of
331 the AM activity of EOs and/or their constituents in the vapour phase, can be used with
332 lipophilic AM films at low concentrations of AM agents (López and others 2007a; Fisher and
333 others 2009; Goñi and others 2009; Kloucek and others 2011). [Recently, Sanla-Ead and
334 others \(2011\) investigated the AM activity of cinnamaldehyde and eugenol incorporated into
335 cellulose-based packaging films against Gram-negative bacteria \(*E. coli*, *Sal. enteritidis*\),
336 Gram-positive bacteria \(*L. monocytogenes*, *Staph. aureus*\) and yeasts \(*C. albicans*, *C.
337 cerevisiae*\) using the vapour diffusion assay. The authors reported that cinnamaldehyde and
338 eugenol incorporated into cellulose-based packaging films demonstrated positive inhibitory
339 effects against the tested microorganisms.](#)

340

341 Table 2 summarises the AM activity of a range of common natural agents that have been
342 incorporated into or coated onto synthetic packaging films. Table 2 also lists other studies
343 that have evaluated the inhibitory effects of natural AM agents *in vitro* or directly on food
344 products without incorporating them into packaging films.

345

346 **2.2.1 Antimicrobial Activity of Basil Essential Oils**

347 Basil EOs contain primarily linalool and methylchavicol as the active volatile components
348 which are responsible for their AM activity (Simon and others 1990; Fyfe and others 1998;
349 Wan and others 1998; Bezic and others 2003; Suppakul 2004). Many studies have evaluated
350 the AM activity of basil EOs against various microorganisms both *in vitro* and on a range of
351 food products as shown in Table 2. Prasad and others (1986) investigated the AM activity of
352 the EOs of *O. basilicum* against various Gram-positive and Gram-negative bacteria with the
353 oils shown to be more effective against Gram-positive bacteria including *Bacillus*
354 *sacharolyticus*, *B. stearothermophilus*, *B. subtilis*, *B. thurengiensis*, *Micrococcus glutamicus*
355 and *Sarcina lutea* than the Gram-negative ones. Lachowicz and others (1998) evaluated the
356 AM effects of EOs of sweet basil against acid-tolerant food microflora. They reported greater
357 inhibitory effects of the tested EOs against the Gram-positive bacteria *Bacillus* sp., *Staph.*
358 *aureus* sp., *Micrococcus* sp., *Sarcina* sp., *Lactobacillus* sp. than against the Gram-negative
359 bacteria *E. coli*, *Salmonella* sp., *Enterobacter* sp. and *Pseudomonas* sp. In contrast to these
360 studies, Koga and others (1999) found that the Gram-positive bacteria were more resistant to
361 basil EOs than the Gram-negative ones.

362

363 Various researchers have reported also the inhibitory effect of basil EOs against fungi. Rai
364 and others (1999) evaluated the antifungal activity of the EOs of ten plant species (including
365 *O. basilicum*) and reported that the EOs of basil were active against all *Fusarium* species

366 including *F. acuminatum*, *F. solani*, *F. pallidoroseum* and *F. chlamyosporum*. Conner and
367 Beuchat (1984) reported positive AM activity of basil EOs against *Kloeckera apiculata* on
368 solid media. Basilico and Basilico (1999) investigated the inhibitory effects of some EOs,
369 including that of basil (*O. basilicum*), against the growth of *A. ochraceus* and subsequent
370 ochratoxin A production. They reported that at a level of 1000 ppm, only basil EO decreased
371 the fungal growth and the production of ochratoxin A for up to 7 days after which mould
372 growth occurred.

373

374 **2.2.2 Antimicrobial Activity of Linalool**

375 Linalool has been reported to possess both fungistatic and antibacterial properties against a
376 wide spectrum of microorganisms such as *Staph. aureus*, *L. innocua*, *E. coli*, *A. niger* and *S.*
377 *cerevisiae* (Lachowicz and others 1998; Friedman and others 2002; Suppakul and others
378 2003a). As shown in Table 2, numerous studies have evaluated the AM activity of linalool
379 incorporated into packaging films. For example, Suppakul and others (2006; 2008) reported
380 that linalool incorporated into LDPE film exhibited inhibitory activity against the growth of
381 *Staph. aureus*, *L. innocua*, *E. coli* and *S. cerevisiae* on culture media and on the surface of
382 Cheddar cheese. Rardniyom (2008) investigated the AM activity of linalool coated onto
383 LDPE and nylon films against the growth of *E. coli* and reported effective inhibitory activity
384 in liquid culture and on Cheddar cheese. Rupika and others (2006) reported that linalool
385 incorporated into LDPE films demonstrated significant inhibitory activity against the growth
386 of *L. innocua* and *E. coli* both *in vitro* and on the surface of Cheddar cheese. Suppakul and
387 others (2011b) reported that linalool and/or methylchavicol incorporated into LDPE films
388 demonstrated inhibitory activity against the growth of *E. coli* on agar disc media.

389

390 The AM activity of linalool against several microorganisms has also been reported in studies
391 conducted *in vitro* only. Kim and others (1995) investigated the AM activity of some EO
392 components including linalool against five food-borne pathogens (*E. coli*, *E. coli* O157:H7,
393 *Sal. typhimurium*, *L. monocytogenes* and *V. vulnificus*) and found a dose-related increase in
394 the zone of inhibition against all tested strains except for *L. monocytogenes*. Mazzanti and
395 others (1998) reported that linalool completely inhibited the growth of all yeasts (seven
396 strains of *C. albicans*, *C. krusei* and *C. tropicalis*), *Staph. aureus* and *E. coli* using the agar
397 disc diffusion method. Dorman and Deans (2000) investigated the antibacterial activity of 21
398 plant volatile oil components including linalool against 25 bacterial strains using the agar
399 well diffusion method. It was reported in this study that linalool was an effective AM agent
400 against a broad spectrum of 23 out of the 25 bacterial strains investigated.

401

402 **2.2.3 Antimicrobial Activity of Oregano and Thyme Essential Oils**

403 Oregano and thyme are popular culinary herbs with their EOs containing terpenoid
404 compounds, mainly the monoterpenoid phenols of thymol (5-methyl-2-[1-methylethyl]
405 phenol) and carvacrol (5-isopropyl-2-methyl phenol). These EOs have been claimed to
406 demonstrate potential health benefits, antioxidant activity and AM properties (Nychas 1995;
407 Baratta and others 1998; Youdim and Deans 2000; Olasupo and others 2004; Tepe and others
408 2004; Davidson and Taylor 2007). The AM activity of thyme and oregano EOs is primarily
409 attributed to their major components thymol and carvacrol respectively (Frag and others
410 1989; Cosentino and others 1999; Davidson and Naidu 2000; Dorman and Deans 2000;
411 Lambert and others 2001; Bagamboula and others 2004; Davidson and Taylor 2007).

412

413 The AM activity of oregano and thyme EOs against various microorganisms has been
414 investigated on media and on a range of food products as summarised in Table 2. For

415 example, Lin and others (2004) evaluated the AM activity of phenolic compounds derived
416 from oregano against *L. monocytogenes* on solid media, on beef and on fish products and
417 reported that the extracts exhibited AM activity against *L. monocytogenes* in the agar
418 diffusion assays. Friedman (2004) studied the antibacterial activity of ten different EOs
419 including that of oregano against *E. coli* and *Sal. enterica* in apple juice. They reported that
420 the selected EOs exhibited greater AM activity against *Sal. enterica* than *E. coli*.

421

422 The AM activity of oregano and thyme EOs was investigated in liquid culture and solid
423 media against various microorganisms. Becerril and others (2007) investigated the AM
424 activity of oregano EOs incorporated into a patented plastic packaging material against *E.*
425 *coli* and *Staph. aureus*, using a "kill time" assay. The authors claimed that oregano EOs
426 exhibited significant AM activity with a kill time of approximately 90 min for *E. coli* and 104
427 min for *Staph. aureus*. Marques and others (2008) studied the AM activity of three natural
428 AM agents: oregano, garlic and chitosan against the growth of *Sal. enterica* in liquid culture
429 at 10 and 20°C. They reported that all of the natural agents inhibited significantly the
430 microbial growth at both temperatures with oregano demonstrating the highest inhibitory
431 effects followed by garlic then chitosan. Rodriguez-Lafuente and others (2010) investigated
432 the AM activity of oregano and cinnamon EOs incorporated into packaging-paper against
433 *Alternaria alternata* using an *in vitro* antifungal assay. The authors reported that oregano and
434 cinnamon EOs inhibited the growth of *A. alternata* on solid media.

435

436 Nielsen and Rios (2000) reported that oregano EOs exhibit an inhibitory activity against
437 microorganisms commonly associated with bread spoilage. Tepe and others (2004) examined
438 the AM activity of *Thymus eigi* EOs and its main constituents carvacrol, thymol and *p-*
439 cymene against *B. catarrhalis*, *C. perfringens*, *B. cereus*, *Staph. aureus*, *S. pneumoniae*, *M.*

440 *smegmatis* and *P. aeruginosa* *in vitro* and found that these EOs demonstrate AM activity
441 against the tested microorganisms. More recently, Gutierrez and others (2008) evaluated the
442 synergistic effect of the EOs of thyme, oregano, lemon balm, marjoram, rosemary and sage
443 against *B. cereus*, *E. coli*, *L. monocytogenes* and *P. aeruginosa* using the spot test on agar
444 media. They reported a significant AM activity of oregano in combination with basil, thyme
445 or marjoram against *B. cereus*, *E. coli* and *P. aeruginosa*. Similarly, Gutierrez and others
446 (2009) determined the AM activity of the EOs of thyme, oregano, lemon balm and marjoram
447 against *Enterobacter* sp., *Listeria* sp., *Lactobacillus* sp. and *Pseudomonas* sp. using foods
448 based on lettuce, meat and milk. Their findings demonstrated that minimum inhibitory
449 concentrations were significantly lower in lettuce and beef media than in tryptic soy broth
450 and that oregano and thyme produced the most active EOs. Lopez and others (2007b)
451 reported AM activity of oregano, cinnamon and clove EOs incorporated into PP or PE/EVOH
452 against various Gram-negative bacteria (*E. coli*, *Y. enterocolitica*, *P. aeruginosa* and *Sal.*
453 *choleraesuis*), Gram-positive bacteria (*L. monocytogenes*, *Staph. aureus*, *B. cereus* and *E.*
454 *faecalis*), yeasts (*C. albicans*, *D. hansenii*, *Z. rouxii*) and moulds (*B. cinerae*, *A. flavus*, *E.*
455 *repens*, *p. roqueforti*, *P. islandicum*, *P. commune*, *P. nalgiovensis*). Similarly, Lopez and
456 others (2007a) reported AM activity of cinnamon, oregano and thyme EOs against various
457 Gram-negative bacteria (*E. coli*, *Y. enterocolitica*, *P. aeruginosa* and *Sal. choleraesuis*),
458 Gram-positive bacteria (*L. monocytogenes*, *Staph. aureus*, *B. cereus* and *E. faecalis*), yeasts
459 (*C. albicans*) and moulds (*A. flavus*, *P. islandicum*). The main constituents: cinnamaldehyde,
460 carvacrol and thymol also demonstrated inhibitory effect against *L. monocytogenes*, *Sal.*
461 *choleraesuis*, *A. flavus* and *C. albicans* using a modified vapour diffusion test.
462
463 Sagdiç and Özcan (2003) investigated the AM activity of various EOs including oregano and
464 thyme EOs against different microorganisms including *Bacillus amyloliquefaciens*, *B. cereus*,

465 *Enterobacter aerogenes*, *E. coli*, *Sal. enteritidis*, *Staph. aureus* and *Yersinia enterocolitica*.
466 They reported that oregano was particularly effective against all bacteria during incubation.
467 Oussalah and others (2006) studied the inhibitory effects of sixty different EOs including
468 oregano and thyme against *Pseudomonas putida*. The results of their study showed that many
469 EOs possess *in vitro* antibacterial activity against *P. putida* with oregano and thyme EOs
470 demonstrating the highest AM activity. Viuda-Martos and others (2008) evaluated the
471 effectiveness of the EOs of oregano, sage, clove, thyme, rosemary and cumin on the growth
472 of various microorganisms including *Lactobacillus curvatus*, *Lactobacillus sakei*, *Staph.*
473 *carnosus* and *Staph. xylosus*, *Enterobacter gergoviae* and *Enterobacter amnigenus*. They
474 found that each of the EOs demonstrated inhibitory activity against all bacteria tested with
475 oregano showing the highest AM activity and reported that the effects of thyme, sage and
476 rosemary were concentration dependent. Baydar and others (2004) studied the antibacterial
477 activity of EOs of thyme, oregano and savoury against various pathogenic bacteria including
478 *B. cereus*, *E. coli* and *L. monocytogenes*. They reported positive AM activity against the
479 tested bacteria and suggested that the inhibition may be attributed to the action of the
480 components carvacrol, γ -terpinene and *p*-cymene (a constituent of cumin or thyme EOs). The
481 results of these studies demonstrate that oregano and thyme EOs have the potential to be used
482 as AM agents in the food industry for better preservation of quality, enhancement of safety
483 and extension of shelf life. Nevertheless, additional information is required on the benefits of
484 these EOs before considering them as potential candidates for manufacturing of AM films
485 with commercial applications.

486

487 **2.2.4 Antimicrobial Efficacy of Carvacrol and Thymol**

488 Carvacrol and thymol are the major components of oregano and thyme EOs. They have
489 received substantial attention as useful natural AM agents due to their natural origin and

490 GRAS status, as well as them exhibiting a broad AM spectrum against different
491 microorganisms, and possessing heat stability when incorporated into packaging materials
492 (Deans and Ritchie 1987; Zaika 1988; Ultee and others 1998; Lorenzo and others 2003;
493 Couladis and others 2004; Azaz and others 2005; Han 2005; Matan and others 2006). Table 2
494 shows that carvacrol and/or thymol can be applied in food products to control microbial
495 contamination by various microorganisms including bacteria, yeasts and moulds.

496

497 Bagamboula and others (2004) determined the AM effect of carvacrol or thymol against
498 *Shigella* sp. (*S. sonnei* and *S. flexneri*) on lettuce. They observed a decrease in *Shigella* sp.
499 after washing the lettuce with 0.5% and 1% (v/v) thymol or carvacrol and found that at 1%
500 (v/v) of each agent, the population decreased to an undetectable level. They also reported
501 significant inhibition of *Shigella* sp. using the agar diffusion method. The AM activity of
502 carvacrol has also been reported by Ultee and others (2000) when studying the preservation
503 of rice against *B. cereus*. Roller and Seedhar (2002) investigated the effectiveness of
504 carvacrol against the natural flora of freshly cut melons and kiwifruit. They found that
505 carvacrol reduced significantly the viable count of natural flora on kiwifruit dipped in a
506 solution of the agent, but it was less effective on honeydew melons. Kiskó and Roller (2005)
507 explored the AM effectiveness of carvacrol against *E. coli* inoculated into unpasteurised
508 apple juice. They found that carvacrol reduced the bacteria to an undetectable level within the
509 first two days of storage. Ultee and Smid (Ultee and Smid 2001) found carvacrol to be
510 effective against *B. cereus* toxin production in soups to an undetectable level. Chiasson and
511 others (2004) reported effective AM activity of carvacrol and thymol against *E. coli* and *Sal.*
512 *typhimurium* in minced meat products. Seaberg and others (2003) reported inhibitory effects
513 of carvacrol against *L. monocytogenes* in ready-to-eat beef slices. Recently, Rardniyom
514 (2008) coated carvacrol onto LDPE and nylon films and reported that the AM film inhibited

515 the growth of *E. coli* on Cheddar cheese by log 2.3 and 1.8 CFU g⁻¹ on samples stored at 8
516 and 12°C respectively for 15 days.

517 In addition to studies on real foods, several studies have reported the inhibitory effect of
518 carvacrol and thymol both on solid and liquid media as shown in Table 2. On solid media,
519 using the agar diffusion test, López-Malo and others (2005) found that carvacrol and thymol
520 had a significant inhibitory effect against *A. flavus*. Singh and others (2006) investigated the
521 AM activity of thymol against various microorganisms using the agar well diffusion method
522 and showed that thymol inhibited completely the growth of *B. cereus* and *P. aeruginosa*.
523 Tepe and others (2004), reported the positive AM activity of carvacrol and thymol against *B.*
524 *catarrhalis*, *C. perfringens*, *B. cereus*, *Staph. aureus*, *S. pneumoniae*, *M. smegmatis* and *P.*
525 *aeruginosa in vitro*. Sivropoulou and others (1996) reported significant AM activity of
526 carvacrol and thymol against *Staph. aureus*. Dorman and Deans (2000) reported effective
527 AM activity of thymol and carvacrol against selected microorganisms including *B. cereus*,
528 *Staph. aureus*, *L. monocytogenes*, *E. coli*, *A. niger* and *S. cerevisiae* using the agar well
529 diffusion method. Olasupo and others (2003b) reported that carvacrol and thymol
530 demonstrated the highest AM activity against *E. coli* and *Sal. typhimurium* using liquid
531 culture compared to other agents including eugenol, nisin, cinnamic acid and diacetyl
532 compounds. Rupika and others (2005) found that carvacrol and/or thymol impregnated into
533 LDPE films had a significant inhibitory activity against *E. coli*, *Staph. aureus*, *L. innocua*, *P.*
534 *aeruginosa*, *A. niger* and *S. cerevisiae* using the agar disc diffusion assay. Han and others
535 (2005) investigated the effectiveness of carvacrol and thymol coated onto LDPE film against
536 *L. innocua* and *E. coli* in solid and liquid media and observed an inhibitory effect using the
537 agar diffusion method. In the liquid culture test, carvacrol and thymol incorporated into the
538 film reduced significantly the specific growth rate and the final cell concentration of *L.*
539 *innocua*.

540

541 Falcone and others (2005) reported that thymol inhibited significantly the growth of *S.*
542 *cerevisiae* and *B. cereus* in liquid media. They reported that the growth kinetics of *B. cereus*
543 in liquid media is a function of thymol concentration. Ultee and others (1998) investigated
544 the AM activity of carvacrol against *B. cereus* using a liquid culture media and reported that
545 the activity depends on the concentration, exposure time, temperature and pH. Periago and
546 others (2004) studied the AM activity of carvacrol and cymene against the growth of two
547 strains of *L. monocytogenes* and found that carvacrol and cymene reduced microbial growth
548 during the lag and exponential phases. They found that the combination of carvacrol and
549 cymene resulted in a larger decrease in viable counts of *L. monocytogenes* compared with the
550 separate application of these agents. Burt and others (2005) conducted a comparative study of
551 the AM activity of oregano and thyme EO components (carvacrol, thymol, *p*-cymene and γ -
552 terpinene) against the growth of *E. coli* O157:H7 and also found synergistic effects of these
553 components using the checkerboard assay. They reported that carvacrol and thymol
554 demonstrated individual and additive antibacterial activity against *E. coli* O157:H7, but no
555 observable AM activity by *p*-cymene and γ -terpinene was found. Although the vast majority
556 of studies involving essential oils or their extracts suggest a positive and broad spectrum of
557 AM activity, an important aspect that needs more attention is how to minimise the loss of
558 these volatile agents during processing, particularly at high temperatures. Gutiérrez and
559 others (2009) reported AM activity of carvacrol, thymol and cinnamadehyde incorporated
560 into PP against various Gram-negative (*E. coli*, *Yersinia enterocolitica*, *P. aeruginosa* and
561 *Sal. choleraesuis*), Gram-positive bacteria (*L. monocytogenes*, *Staph. aureus*, *B. cereus* and
562 *Enterococcus faecalis*), yeasts (*Candida Albicans*, *Debaryomyces hansenii*,
563 *Zygosaccharomyces rouxii*) and moulds (*Botrytis cinerae*, *A. flavus*, *Eurotium repens*,
564 *penicillium roqueforti*, *P. islandicum*, *P. commune*, *P. nalgiovensis*). Recently, Persico and

565 others (2009) claimed that carvacrol incorporated into a LDPE film demonstrated an AM
566 activity against *B. thermosphacta*, *L. innocua* and *Carnobacterium. sp* on agar medium.

567

568

569 **2.2.5 Other Natural Antimicrobial Agents**

570 Numerous studies have evaluated the inhibitory effects of other natural AM agents including
571 bacteriocins, plant extracts such as grapefruit seed extract (GFSE), enzymes and spices (see
572 Table 2). Bacteriocins such as nisin are ribosomally synthesised peptides produced by lactic
573 acid bacteria and possess bactericidal properties against a range of microorganisms (Siragusa
574 and others 1999). They were widely studied for their AM activity in packaging films.
575 Grower and others (2004) developed an AM film by coating nisin onto an LDPE film and
576 reported that these coatings were effective against *L. monocytogenes* on solid microbiological
577 media and on the surface of individually packed hotdogs. Natrajan and Sheldon (2000)
578 reported significant AM activity of nisin coated on three different packaging films: polyvinyl
579 chloride (PVC), linear low-density polyethylene (LLDPE) and nylon against *Sal.*
580 *typhimurium* on broiler drumstick skin stored at 4°C. The AM activity of the nisin film was
581 found to be at higher nisin concentrations and when the film was in direct contact with the
582 tested products for a longer period. Kim and others (2002) coated nisin onto LDPE film in
583 order to control naturally-occurring bacteria on packaged fresh oysters and ground beef
584 stored at 3 and 10°C. They claimed that nisin coated onto the film reduced microbial growth
585 at both temperatures in contrast to a non-coated LDPE film. The inhibitory effects of AM-
586 coated films on the growth of coliform bacteria were more evident at 10°C than at 3°C, while
587 the effect on the total aerobic bacteria count was consistently apparent at both temperatures.
588 Siragusa and others (1999) evaluated the AM effectiveness of nisin incorporated into LDPE
589 films against the growth of *B. thermosphacta* inoculated on the surface of a beef carcass.

590 They reported that the films reduced significantly the population of *B. thermosphacta* at the
591 end of a storage period at 4 and 12°C. Scannell and others (2000) investigated the AM
592 activity of nisin immobilised onto polyethylene (PE) and/or nylon films and found that the
593 films reduced the levels of *L. innocua* and *Staph. aureus* in sliced cheese and ham. Mauriello
594 and others (2004) investigated the anti-listerial effect of bacteriocin produced by
595 *Lactobacillus curvatus* 32Y incorporated PE and oriented nylon films. The films were coated
596 with the bacteriocin using three different methods: soaking, spraying and coating and all the
597 films inhibited the growth of *L. monocytogenes* on both solid media and pork steaks.
598 Mauriello and others (2005) coated nisin onto LDPE films in order to control the growth of
599 *Micrococcus luteus* in tryptone soya broth and in raw, pasteurised and UHT milk. The nisin
600 coated onto LDPE films was shown to have an inhibitory effect against the growth of the
601 bacteria in the broth and also reduced microbial counts in the milk products. Cooksey (2001)
602 coated nisin onto LDPE films and evaluated their inhibitory effect against *L. monocytogenes*
603 on packaged hotdogs and reported that coatings containing 2500 IU mL⁻¹ or greater of nisin
604 applied to the films effectively inhibited microbial growth on the hotdogs stored under
605 refrigeration for 60 days. Cutter and others (2001) investigated the AM activity of nisin
606 incorporated into PE food packaging films and reported a significant AM effect of the films
607 against *B. thermosphacta*.

608

609 The AM activity of plant extracts was investigated by several researchers and invariably
610 demonstrated an inhibitory effect against various microorganisms. For example, Hong and
611 others (2000) investigated the AM effectiveness of 5% (w/w) propolis extract or clove extract
612 incorporated into LDPE films against *E. coli*, *L. plantarum*, *S. cerevisiae* and *Fusarium*
613 *oxysporum*. All extracts demonstrated an inhibitory activity on the growth of *L. plantarum*
614 and *F. oxysporum*. Ha and others (2001) investigated the AM activity of GFSE incorporated

615 into multi-layered PE films against *M. flavus*, *E. coli*, *Staph. aureus* and *B. subtilis* on ground
616 beef. The coated films demonstrated an AM activity against all the microorganisms studied.
617 Lee and others (1998) developed an LDPE packaging film incorporated with GFSE and
618 reported that the film containing narigin, ascorbic acid, hesperidin and various organic acids
619 was shown to possess a wide spectrum of AM activity. However, although the LDPE films
620 containing GFSE had an inhibitory effect on the growth of *E. coli* and *Staph. aureus* on solid
621 media, they were unable to inhibit the growth of *Leuconostoc mesenteroides*, *S. cerevisiae*,
622 *Aspergillus oryzaei*, *A. niger* and *Penicillium chrysogenum*. Rodriguez and others (2008)
623 investigated the AM activity of an active packaging film incorporated with cinnamon EOs
624 against *Rhizopus stolonifer* both *in vitro* and on sliced bread. They reported that cinnamon
625 EOs inhibited the growth of *R. stolonifer* on solid media and on sliced bread.

626
627 The AM films incorporated with EOs and/or their principal constituents have the potential for
628 packaging of many food products such as bakery (Suhr and Nielsen 2003; Rodríguez and
629 others 2008; Rokchoy and others 2009; Mehyar and others 2011), dairy (Suppakul and others
630 2008; Kuorwel and others 2011a), meat, chicken and fish (Suppakul and others 2003a; Kerry
631 and others 2006; Wu and others 2010), and fresh produce (Rodríguez-Lafuente and others
632 2010). There are already some commercial applications for AM packaging systems such as
633 wasabi extract (or Japanese horseradish) used for Japanese rice lunch boxes (Koichiro 1993),
634 Piatech manufactured by Rhone-Poulenc (USA) and Daikoku Kasei Co (Japan) (Brody and
635 others 2001).

636

637 **3 Conclusions**

638 In recent years, an increasing interest has emerged in the development of various forms of
639 active packaging systems intended to protect food products from microbial contamination.

640 Many synthetic and natural AM agents incorporated into or coated onto synthetic polymer-
641 based packaging materials have demonstrated significant AM activity against various
642 microorganisms. Although incorporating synthetic AM agents directly into foods can
643 effectively inhibit the growth and survival of various microorganisms, consumers today
644 demand minimally processed, preservative-free food products with a longer shelf life. Thus,
645 natural AM agents such as basil, thyme and oregano EOs with their main components
646 linalool, thymol and carvacrol respectively are well suited to be utilised as preservatives in
647 foods and as potential alternatives for synthetic food additives. Packaging materials
648 containing AM agents demonstrate a potential for applications in AM packaging systems that
649 could reduce the risk of food-borne illness associated with microbial contamination in food
650 products. Although the commercial applications of these systems is not yet widespread, it is
651 anticipated that AM packages containing natural AM agents would be one of the new
652 developments in food packaging (cheeses, processed meats, fish, bakery, fruits and
653 vegetables and more) in the near future.

654 **Table 1: Antimicrobial activity of common synthetic AM agents**

655

AM Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
Benzoic acid	0.5 mol/L	PEMA	PDA	<i>A. niger</i> and <i>Penicillium</i> sp.	Inhibited microbial growth	Weng and others (1999)
Benzoic acid	0.5-2% w/w	LDPE	Agar media; Cheddar cheese	<i>R. stolonifer</i> , <i>Penicillium</i> sp. and <i>A. toxacarius</i>	Failed to inhibit mould growth	Weng and Hotchkiss (1993)
Benzoic anhydride	0.5-2% w/w	LDPE	Agar media; Cheddar cheese	<i>R. stolonifer</i> , <i>Penicillium</i> sp. and <i>A. toxacarius</i>	Demonstrated antimycotic activity on media and cheese	Weng and Hotchkiss (1993)
EDTA	5% w/w	LDPE	Agar diffusion	<i>B. subtilis</i> , <i>A. niger</i> and <i>E. coli</i>	Inhibited <i>B. subtilise</i> and <i>A. Niger</i> but not <i>E. coli</i>	Vartiainen and others (2003a)
Imazalil	1000-2000 mg/kg	LDPE	Agar media; Cheddar	<i>A. toxacarius</i> , <i>Penicillium</i> sp.	All concentrations delayed microbial growth on media	Weng and Hotchkiss (1992)

AM Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
			cheese		and cheese	
Imazalil	0.05-0.25% w/w	LDPE	Agar diffusion	<i>B. subtilis</i> , <i>A. niger</i> and <i>E. coli</i>	Inhibited <i>B. subtilise</i> and <i>A. Niger</i> but not <i>E. coli</i>	Vartiainen and others (2003a)
Potassium sorbate	2-3% w/v	PVC	Agar media	<i>L. monocytogenes</i>	Films inhibited microbial growth	Limjaroen and others (2003)
Propionic acid	0.5-2% w/w	LDPE	Agar media; Cheddar cheese	<i>R. stolonifer</i> , <i>Penicillium</i> sp. and <i>A. toxacarius</i>	Failed to inhibit mould growth	Weng and Hotchkiss (1993)
Propionic anhydride	0.5-2% w/w	LDPE	Agar media; Cheddar cheese	<i>R. stolonifer</i> , <i>Penicillium</i> sp. and <i>A. toxacarius</i>	Failed to inhibit mould	Weng and Hotchkiss (1993)
Sodium propionate	0.5-2% w/w	LDPE	Agar media; Cheddar cheese	<i>R. stolonifer</i> , <i>Penicillium</i> sp. and <i>A. toxacarius</i>	Failed to inhibit mould	Weng and Hotchkiss (1993)

AM Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
Sodium diacetate	0.5-3% w/v	PVC	Agar media	<i>L. monocytogenes</i>	No AM activity observed	Limjaroen and others (2003)
Sorbic acid	1.5-3% w/v	PVC	Agar media	<i>L. monocytogenes</i>	Inhibited microbial growth	Limjaroen and others (2003)
Sorbic acid	0.5-2% w/w	LDPE	Agar media; Cheddar cheese	<i>R. stolonifer</i> , <i>Penicillium</i> sp. and <i>A. toxacarius</i>	Failed to inhibit mould	Weng and Hotchkiss (1993)
Sorbic acid	0.5 mol L ⁻¹	PEMA	PDA	<i>A. niger</i> and <i>Penicillium</i> sp.	Inhibited microbial growth	Weng and others (1999)
Triclosan	500-1000 mg kg ⁻¹	LDPE	Agar diffusion; Chicken breasts	<i>L. monocytogenes</i> , <i>Sal. enteritidis</i> , <i>Staph. aureus</i> , <i>E. coli</i> O157:H7, <i>B. thermosphacta</i> , <i>B. cereus</i> , <i>L. sake</i> , <i>L. brevis</i> , <i>P.</i>	Inhibited <i>L. monocytogenes</i> , <i>Sal. enteritidis</i> , <i>Staph. aureus</i> , <i>E. coli</i> O157:H7 with slight inhibition of <i>B.</i>	Vermeiren and others (2002)

AM Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
				<i>Roqueforti</i> , <i>A. niger</i> and <i>C. albicans</i>	<i>thermosphacta</i> , but no activity against, <i>B. cereus</i> , <i>L. sake</i> , <i>L. brevis</i> , <i>P. Roqueforti</i> , <i>A. niger</i> and <i>C. Albicans</i>	
Triclosan	5% w/w	PVC	Plate count	<i>Staph. aureus</i> , <i>E. coli</i>	<i>Staph. aureus</i> , <i>E. coli</i>	Ji and Zhang (2009)

656

657 **Table 2: Antimicrobial activity of natural AM agents**

658

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
Carvacrol	1.33-2.65% w/w	LDPE	Liquid culture	<i>E. coli</i>	Reduced microbial growth	Rupika and others (2008)
Carvacrol	1-4% w/w	PP	Agar medium	<i>E. coli</i> , <i>Y. enterocolitica</i> , <i>P. aeruginosa</i> , <i>Staph.</i> <i>aureus</i> , <i>B. cereus</i> and <i>E.</i> <i>faecalis</i> , <i>C. albicans</i> , <i>D.</i> <i>hansenii</i> , <i>Z. Rouxii</i> , <i>A.</i> <i>flavus</i> , <i>E. repens</i> , <i>p.</i> <i>roqueforti</i> , <i>P. commune</i>	Carvacrol demonstrated AM activity against all the tested microorganisms	Gutiérrez and others (2009)
Carvacrol	1-4% w/w 1-21.8µL/L	PP, PE/EVOH	Agar medium or vapour diffusion	<i>L. monocytogenes</i> , <i>Sal.</i> <i>choleraesuis</i> , <i>A. flavus</i> and <i>C. albicans</i>	carvacrol inhibited the growth of all the tested microorganisms	Lopez and others (2007b; 2007a)

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media method	Target Microorganism(s)	Findings	References
Carvacrol	0.2-2% w/w	LDPE	Agar media	<i>E. coli</i> , <i>Staph. aureus</i> , <i>L. innocua</i> , <i>P. aeruginosa</i> , <i>A. niger</i> and <i>S. cerevisiae</i>	Inhibited <i>E. coli</i> , <i>Staph. aureus</i> , <i>A. niger</i> and <i>S. cerevisiae</i> but not <i>L. innocua</i> or <i>P. aeruginosa</i>	Rupika and others (2005)
Carvacrol	10% w/w	LDPE	Agar media	<i>B. thermosphacta</i> , <i>L. innocua</i> and <i>Carnobacterium. sp</i>	Carvacrol demonstrates <i>B. thermosphacta</i> , <i>L. innocua</i> and <i>Carnobacterium. sp</i>	Persico and others (2009)
Carvacrol	4% w/w	LDPE and nylon film	liquid food media; Cheddar cheese	<i>E. coli</i>	Multilayer films inhibited microbial growth	Rardniyom and others (2008)

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
Cinnamaldehyde	1-4% w/w	PP	Agar medium	<i>E. coli</i> , <i>Y. enterocolitica</i> , <i>P. aeruginosa</i> , <i>Staph. aureus</i> , <i>B. cereus</i> and <i>E. faecalis</i> , <i>C. albicans</i> , <i>D. hansenii</i> , <i>Z. Rouxii</i> , <i>A. flavus</i> , <i>E. repens</i> , <i>p. roqueforti</i> , <i>P. commune</i>	cinnamaldehyde demonstrated AM activity against all the tested microorganisms	Gutiérrez and others (2009)
Cinnamaldehyde	1-4% w/w 0.4-21.8µL/L	PP, PE/EVOH	Agar medium or vapour diffusion method	<i>L. monocytogenes</i> , <i>Sal. choleraesuis</i> , <i>A. flavus</i> and <i>C. albicans</i>	Cinnamaldehyde inhibited the growth of all the tested microorganisms	Lopez and others (2007b; 2007a)
Clove extract	20% w/w	LDPE	Liquid culture	<i>E. coli</i> , <i>L. plantarum</i> , <i>S. cerevisiae</i> and <i>F. oxysporum</i>	Effective against <i>L. plantarum</i> and <i>F. oxysporum</i> but not against	Hong and others (2000)

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
					<i>E. coli</i> and <i>S. cerevisiae</i>	
GFSE	0.1 or 1% w/w	LDPE	Agar media; Curled lettuce; Soybean sprouts	<i>E. coli</i> , <i>Staph. aureus</i> , <i>L. mesenteroides</i> , <i>S. cerevisiae</i> , <i>A. oryzaei</i> , <i>A. niger</i> , <i>P. chrysogenum</i>	Inhibited <i>E. coli</i> and <i>Staph. aureus</i> but not <i>S. cerevisiae</i> , <i>A. oryzaei</i> , <i>A. niger</i> , or <i>P. chrysogenum</i> .	Lee and others (1998)
GFSE	0.5% or 1% w/v	Multi-layered PE (coated)	Ground beef, Agar media	<i>M. flavus</i> , <i>P. aeruginosa</i> , <i>E. coli</i> , <i>Staph. aureus</i> and <i>B. subtilise</i> , <i>S. cerevisiae</i> , <i>A. Niger</i> , <i>P. chysogenum</i> , <i>L. mesenteroides</i>	AM activity against <i>M. flavus</i> , <i>E. coli</i> , <i>Staph. aureus</i> and <i>B. subtilise</i>	Ha and others (2001)
Lactoferrin	0.5-2.5% w/v	PVC	Agar media	<i>L. monocytogenes</i>	No AM activity	Limjaroen and others (2003)
Lacticin NK24	20 gL ⁻¹	LDPE	Fresh oysters;	Coliform, total aerobic	Inhibited microbial growth	Kim and others (2002)

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
			ground beef	bacteria		
Linalool	0.037% w/w	LDPE	Agar media	<i>E. coli</i>	Linalool incorporated into LDPE film inhibit the growth of <i>E. Coli</i> after 1 year of storage	Suppakul and others (2011b)
Linalool	0.338% w/w	LDPE	Agar media; Cheddar cheese	<i>E. coli</i> , <i>L. innocua</i> , <i>S. cerevisiae</i>	Inhibitory activity against <i>E. Coli</i> but not <i>L. innocua</i> or <i>S. cerevisiae</i> on agar media; reduced <i>E. coli</i> and <i>L. innocua</i> on cheese	Suppakul (2004; 2006; 2008)
Linalool	4% w/w	LDPE and nylon film	Liquid culture; Cheddar cheese	<i>E. coli</i>	Multilayer films inhibited microbial growth	Rardniyom and others (2008)

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
Linalool	0.54-1.19% w/w	LDPE	Agar and liquid media; Cheddar cheese	<i>E. coli, L. innocua</i>	Inhibited microbial growth	Rupika and others (2006)
Methylchavicol	0.028% w/w	LDPE	Agar media	<i>E. coli</i>	Methylchavicol inhibitory activity against the growth of <i>E. Coli</i> after 1 year of storage	Suppakul and others (2011b)
Methylchavicol	0.345% w/w	LDPE	Agar media; Cheddar cheese	<i>E. coli, L. innocua, S. cerevisiae</i>	Inhibitory activity against <i>E. coli</i> but not against <i>L. innocua</i> or <i>S. cerevisiae</i> on agar media	Suppakul (2004; 2006; 2008)
Nisin	0.05 or	LDPE	Beef carcass	<i>B. thermosphacta</i>	Inhibited microbial growth	Siragusa and others

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
	0.1% w/v					(1999)
Nisin	2-2.5% w/v	PVC	Agar media	<i>L. monocytogenes</i>	Inhibited microbial growth	Limjaroen and others (2003)
Nisin	157 mg/mL	LDPE	Agar media	<i>L. monocytogenes</i>	Inhibited microbial growth	Grower and others (2004)
Nisin	100 µg/mL	LLDPE, PVC, nylon	Broiler skin	<i>Sal. typhimurium</i>	Significantly reduced microbial population	Natrajan and Sheldon (2000)
Nisin	20 g/L	LDPE	Fresh oysters; ground beef	coliform, total aerobic bacteria	Suppressed coliform and bacterial growth	Kim and others (2002)
Nisin	0.03 or 0.6 g/mL	PE or polyamide	Sliced cheese; ham	<i>L. innocua</i> and <i>Staph. aureus</i>	Reduced microbial growth in cheese	Scannell and others (2000)
Propolis	20% w/w	LDPE	Liquid culture	<i>E. coli</i> , <i>L. plantarum</i> , <i>S. cerevisiae</i> and <i>F. oxysporum</i>	Inhibited <i>L. plantarum</i> and <i>F. oxysporum</i> but not <i>E. coli</i> or <i>S. cerevisiae</i>	Hong and others (2000)

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
Thymol	0.85-3.15% w/w	LDPE	Liquid culture	<i>E. coli</i>	Inhibited microbial growth	Rupika and others (2008)
Thymol	1-4% w/w	PP	Agar medium	<i>E. coli, Y. enterocolitica, P. aeruginosa, Staph. aureus, B. cereus and E. faecalis, C. albicans, D. hansenii, Z. Rouxii, A. flavus, E. repens, p. roqueforti, P. commune</i>	Thymol demonstrated AM activity against all the tested microorganisms	(Gutiérrez and others 2009)
Thymol	0.23-1.6% w/w	LDPE	Agar media	<i>E. coli, Staph. aureus, L. innocua, P. aeruginosa, A. niger and S. cerevisiae</i>	Inhibited <i>E. coli, Staph. aureus, A. niger</i> and <i>S. cerevisiae</i> but not <i>L. innocua</i> , and <i>P. aeruginosa</i>	Rupika and others (2005)

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
Thymol	1-4% w/w 1-21.8µL/L	PP, PE/EVOH	Agar medium or vapour diffusion method	<i>L. monocytogenes</i> , <i>Sal. choleraesuis</i> , <i>A. flavus</i> and <i>C. albicans</i>	Thymol inhibited the growth of all the tested microorganisms	Lopez and others (2007b; 2007a)
Basil EOs				<i>E. coli</i> , <i>Staph. aureus</i> , seven strains of <i>Candida</i>	Inhibited microbial growth	Mazzanti and others (1998)
Basil EOs			Solid media	25 strains of bacteria	Inhibited microbial growth	Dorman and Deans (2000)
Basil EOs			Solid media	<i>Staph. aureus</i>	Inhibited microbial growth	Baratta and others (1998)
Basil EOs			Solid media Food	<i>Bacillus</i> sp., <i>Staph. aureus</i> sp., <i>micrococcus</i> sp., <i>Sarcina</i> sp., <i>Lactobacillus</i> sp., <i>E. coli</i> , <i>Salmonella</i> ,	Inhibitory effects against Gram-positives (<i>Bacillus</i> sp., <i>Staph. aureus</i> sp., <i>micrococcus</i> sp., <i>Sarcina</i>	Lachowicz and others (1998)

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
				sp., <i>Enterobacter</i> sp., <i>Pseudomonas</i> sp.	sp. and <i>Lactobacillus</i> sp. Reduced effects against Gram-negatives (<i>E. coli</i> , <i>Salmonella</i> sp., <i>Enterobacter</i> sp. and <i>Pseudomonas</i> sp.	
Basil EOs				<i>Fusarium acuminatum</i> , <i>F. solani</i> , <i>F. pallidoroseum</i> and <i>F. chlamydosporum</i>	EOs effective against all <i>Fusarium</i> species	Rai and others (1999)
Cinnamon EOs	1-4% w/w 13.1- 131µL/L	PP, PE/EVOH	Agar medium or vapour diffusion method	<i>E. coli</i> , <i>Y. enterocolitica</i> , <i>P. aeruginosa</i> , <i>Staph. aureus</i> , <i>B. cereus</i> and <i>E. faecalis</i> , <i>C. albicans</i> , <i>D. hansenii</i> , <i>Z. Rouxii</i> , <i>A.</i>	Cinnamon EOs in PP or PE/EVOH demonstrated AM activity against all the tested microorganisms	Lopez and others (2007b; 2007a)

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
				<i>flavus</i> , <i>E. repens</i> , <i>p. roqueforti</i> , <i>P. commune</i>		
Cinnamon EOs	3-6% (w/w)	Paraffin-based paper	<i>In vitro</i> , Sliced bread	<i>A. alternata</i>	Cinnamon EOs inhibited the growth of <i>A. alternata</i> on solid media	Rodriquez-Lafuente and others (2010)
Cinnamon EOs	1-6% w/w	Paraffin-paper	<i>In vitro</i> , Sliced bread	<i>R. stolonifer</i>	Cinnamon EOs in paraffin film inhibited the growth of <i>R. stolonifer</i>	Rodríguez and others (2008)
Clove	1-4% w/w	PP, PE/EVOH	Agar medium	<i>E. coli</i> , <i>Y. enterocolitica</i> , <i>P. aeruginosa</i> , <i>Staph. aureus</i> , <i>B. cereus</i> and <i>E. faecalis</i> , <i>C. albicans</i> , <i>D. hansenii</i> , <i>Z. Rouxii</i> , <i>A. flavus</i> , <i>E. repens</i> , <i>p.</i>	Clove EOs demonstrated AM activity against all the tested microorganisms	Lopez and others (2007b; 2007a)

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
				<i>roqueforti, P. commune</i>		
Oregano EOs	0.8% (v/w)	surface dipping, O ₂ permeable films	beef meat fillets	<i>L. monocytogenes</i> , autochthonous flora	Reduced growth by 2-3 log ₁₀	Tsigarida and others (2000)
Oregano EOs	3-6% (w/w)	Paraffin-based paper	<i>In vitro</i> , Cherry tomato	<i>A. alternata</i>	Oregano EOs inhibited the growth of <i>A. alternata</i> on solid media	Rodriquez-Lafuente and others (2010)
Oregano EOs		dressing, MAP	fresh fish fillets	<i>Staph. aureus</i> , <i>Sal. enteritidis</i> , Residential flora	Bacterostatic and bactericidal effects	Tassou and others (1996)
Oregano EOs	800 ppm	surface spreading	thin-sliced beef	<i>L. monocytogenes</i>	Significant inhibition	Seaberg and others (2003)
Oregano EOs	0.05-1%	PE bags	minced beef	spoilage microbiota	Reduction in microbial	Skandamis and

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
	(v/w)				loads	Nychas (2001)
Oregano EOs		surface application	raw fish fillets	<i>Photobacterium phosphoreum</i>	No significant growth reduction	Mejlholm and Dalgaard (2002)
Oregano EOs	1-4% w/w 13.1-175µL/L	PP, PE/EVOH	Agar medium or Vapour diffusion method	<i>E. coli, Y. enterocolitica, P. aeruginosa, Staph. aureus, B. cereus</i> and <i>E. faecalis, C. albicans, D. hansenii, Z. Rouxii, A. flavus, E. repens, p. roqueforti, P. commune</i>	Oregano EOs in PP or PE/EVOH demonstrated AM activity against all the tested microorganisms	Lopez and others (2007b; 2007a)
Oregano EOs	50 µL	Suspensions of oils in apple juice	Apple juices	<i>E. coli, Sal. enterica</i>	Selected oils were bactericidal	Friedman and others (2004)
Oregano EOs	0.1-10%	dissolved in	Liquid	<i>Sal. enterica</i>	OEO showed strongest	Marques and others

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
	(v/v)	brain heart infusion broth	culture		AM activity	(2008)
Oregano EOs			<i>In vitro</i>	<i>B. cereus</i> , <i>E. coli</i> L. <i>monocytogenes</i>	Inhibited microbial growth	Baydar and others (2004)
Thyme EOs	50 µL	vapour contact	sponge cake analogues	<i>Eurotium</i> sp., <i>Aspergillus</i> sp., <i>Pencillium</i> sp.	Significant reduction in microbial growth	Guynot and others (2003)
Thyme EOs	135 or 270 µL/L	vapour contact	rye bread	<i>Pencillium</i> sp., <i>E. repens</i> , <i>A. flavus</i>	Significant reduction in microbial growth	Suhr and Nielsen (2003)
Thyme EOs	0.1-1% (v/v)	cheese-EO-mixture	soft cheese	<i>L. monocytogenes</i> <i>Sal. enteritidis</i>	Significant inhibition in low-fat cheese; no inhibition in full-fat cheese	Smith-Palmer and others (2001)
Thyme EOs	1:5 dilution	surface application	cooked poultry	<i>A. hydrophila</i> , <i>L. monocytogenes</i>	Inhibited growth of <i>A. Hydrophila</i>	Hao and others (1998)

Antimicrobial Agent	Amount added	Packaging Material	Test Type/Media	Target Microorganism(s)	Findings	References
Thyme EOs	1-4% w/w 26.2- 175µL/L	PP, PE/EVOH	Agar medium, Vapour diffusion method	<i>E. coli</i> , <i>Y. enterocolitica</i> , <i>P. aeruginosa</i> , <i>Staph.</i> <i>aureus</i> , <i>B. cereus</i> and <i>E.</i> <i>faecalis</i> , <i>C. albicans</i> , <i>D.</i> <i>hansenii</i> , <i>Z. Rouxii</i> , <i>A.</i> <i>flavus</i> , <i>E. repens</i> , <i>p.</i> <i>roqueforti</i> , <i>P. commune</i>	thyme EOs demonstrated AM activity against all the tested microorganisms	Lopez and others (2007b; 2007a)
Thyme EOs		surface application	raw fish fillets	<i>Photobacterium</i> <i>phosphoreum</i>	No significant growth reduction	Mejlholm and Dalgaard (2002)

659

660 **List of Abbreviations and Nomenclature**

661	AM	Antimicrobial
662	AP	Active Packaging
663	CFU	Colony Forming Units
664	EOs	Essential Oils
665	EVOH	Ethylene Vinyl Alcohol
666	GRAS	Generally Recognized As Safe
667	GFSE	Grapefruit Seed Extract
668	HDPE	High-Density Polyethylene
669	LB	Lactic-acid Bacteria
670	LDPE	Low-Density Polyethylene
671	LLDPE	Linear Low-Density Polyethylene
672	MAP	Modified Atmosphere Packaging
673	OEO	Oregano Essential Oil
674	PDA	Potato Dextrose Agar
675	PE	Polyethylene
676	PEG	Polyethylene Glycol
677	PEMA	Poly(Ethylene- <i>co</i> -Methacrylic Acid)
678	PET	Polyethylene Terephthalate
679	PP	Polypropylene
680	PS	Polystyrene
681	PVC	Polyvinyl Chloride
682	PVDC	Poly(Vinylidene Chloride) or Poly(Vinyl Dichloride)
683	TEO	Thyme Essential Oil

684 **4 References**

- 685 An DS, Hwang YI, Cho SH, Lee DS. 1998. Packaging of fresh curled lettuce and
686 cucumber by using low density polyethylene films impregnated with
687 antimicrobial agents. *J Korean Soc Food Sci Nutr* 27 (4):675-81.
- 688 Appendini P, Hotchkiss JH. 1997. Immobilization of lysozyme on food contact
689 polymers as potential antimicrobial films. *Packag Technol Sci* 10 (5):271-9.
- 690 Appendini P, Hotchkiss JH. 2002. Review of antimicrobial food packaging. *Innov*
691 *Food Sci Emerg Technol* 3 (2):113-26.
- 692 Azaz AD, Kurkcuoglu M, Satil F, Baser HC, Tumen G. 2005. In vitro antimicrobial
693 activity and chemical composition of some *Satureja* essential oils. *Flavour*
694 *Frag J* 20 (6):587-91.
- 695 Bagamboula CF, Uyttendaele M, Debevere J. 2004. Inhibitory effect of thyme and
696 basil essential oils, carvacrol, thymol, estragol, linalool and *p*-cymene towards
697 *Shigella sonnei* and *S. flexneri*. *Food Microbiol* 21 (1):33-42.
- 698 Bakkali F, Averbeck S, Averbeck D, Idaomar M. 2008. Biological effects of essential
699 oils - a review. *Food Chem Toxicol* 46 (2):446-75.
- 700 Baratta TM, Dorman TJH, Denes GS, Figueiredo CA, Barroso GJ, Ruberto G. 1998.
701 Antimicrobials and antioxidant properties of some commercial oils. *Flavour*
702 *Frag J* 13 (4):235-44.
- 703 Basílico MZ, Basílico JC. 1999. Inhibitory effects of some spice essential oils on
704 *Aspergillus ochraceus* NRRL 3174 growth and ochratoxin A production. *Lett*
705 *Appl Microbiol* 29 (4):238-41.
- 706 Baydar H, Sagdiç O, Özkan G, Karadogan T. 2004. Antibacterial activity and
707 composition of essential oils from *Origanum*, *Thymbra* and *Satureja* species
708 with commercial importance in Turkey. *Food Control* 15 (3):169-72.
- 709 Becerril R, Gómez-Lus R, Goñi P, López P, Nerín C. 2007. Combination of analytical
710 and microbiological techniques to study the antimicrobial activity of a new
711 active food packaging containing cinnamon or oregano against *E. coli* and *S.*
712 *aureus*. *Anal Bioanal Chem* 388 (5):1003-11.
- 713 Bezic N, Skocibusic M, Dunkic V, Radonic A. 2003. Composition and antimicrobial
714 activity of *Achillea clavennae* L. essential oil. *Phytother Res* 17 (9):1037-40.
- 715 Brody AL, Strupinsky ER, Kline LR. 2001. Antimicrobial packaging. In: Active
716 packaging for food applications. Lancaster, Penn.: Technomic Publishing Co.
717 p. 131-96.
- 718 Burt S. 2004. Essential oils: Their antibacterial properties and potential applications in
719 foods-a review. *Int J Food Microbiol* 94 (3):223-53.
- 720 Burt S, Vlieland R, Haagsman H, Veldhuizen E. 2005. Increase in activity of
721 essential oil components carvacrol and thymol against *Escherichia coli*
722 O157:H7 by addition of food stabilisers. *J Food Protect* 68 (5):919-26.
- 723 Camilloto GP, Ana Clarissa SP, Nilda de Fátima FS, Emiliane AA, Nélio JA, Sukarno
724 OF. 2010. Effect of active packaging incorporated with triclosan on bacteria
725 adhesion. *J Food Sci* 75 (8):E557-E64.
- 726 Cha DS, Chinnan MS. 2004. Biopolymer-based antimicrobial packaging: A review.
727 *Crit Rev Food Sci Nutr* 44 (4):223-37.
- 728 Chen MC, Yeh GHC, Chiang BH. 1996. Antimicrobial and physicochemical
729 properties of methylcellulose and chitosan films containing a preservative. *J*
730 *Food Process Pres* 20 (5):379-90.
- 731 Chiasson F, Borsa J, Ouattara B, Lacroix M. 2004. Radiosensitisation of *Escherichia*
732 *coli* and *Salmonella typhi* in ground beef. *J Food Protect* 67 (6):1157-62.

- 733 Chung D, Spyridon EP, Yam KL. 2003a. Evaluation of a polymer coating containing
734 triclosan as the antimicrobial layer for packaging materials. *Int J Food Sci*
735 *Technol* 38 (2):165-9.
- 736 Chung Y-C, Wang H-L, Chen Y-M, Li S-L. 2003b. Effect of abiotic factors on the
737 antibacterial activity of chitosan against waterborne pathogens. *Bioresource*
738 *Technol* 88 (3):179-84.
- 739 Coma V. 2007. Bioactive packaging technologies for extended shelf life of meat-
740 based products. *Meat Sci* 78 (1-2):90-103.
- 741 Conner DE, Beuchat LR. 1984. Effects of essential oils from plants on growth of food
742 spoilage yeasts. *J Food Sci* 49 (2):429-34.
- 743 Cooksey K. 2000. Utilisation of antimicrobial packaging films for inhibition of
744 selected microorganisms. In: Risch SJ, editor. *Food Packaging*. Washington:
745 American Chemical Society. p. 17-25.
- 746 Cooksey K. 2001. Antimicrobial food packaging materials. *Addit Polym* 2001 (8):6-
747 10.
- 748 Cooksey K. 2005. Effectiveness of antimicrobial food packaging materials. *Food*
749 *Addit Contam* 22 (10):980-7.
- 750 Cosentino S, Tuberoso CIG, Pisano B, Satta M, Mascia V, Arzedi E, Palmas F. 1999.
751 *In-vitro* antimicrobial activity and chemical composition of Sardinian *Thymus*
752 essential oils. *Lett Appl Microbiol* 29 (2):130-5.
- 753 Couladis M, Tzakou O, Kujundzic S, Sokovic M, Mimica-Dukic N. 2004. Chemical
754 analysis and antifungal activity of *Thymus striatus*. *Phytother Res* 18 (1):40-2.
- 755 Cran MJ, Rupika LAS, Sonneveld K, Miltz J, Bigger SW. 2010. Release of naturally
756 derived antimicrobial agents from LDPE films. *J Food Sci* 75 (2):E126-E33.
- 757 Cutter CN. 1999. The effectiveness of triclosan-incorporated plastic against bacteria
758 on beef surfaces. *J Food Protect* 62 (5):474-9.
- 759 Cutter CN, Willett JL, Siragusa GR. 2001. Improved antimicrobial activity of nisin-
760 incorporated polymer films by formulation change and addition of food grade
761 chelator. *Lett Appl Microbiol* 33 (4):325-8.
- 762 Davidson PM, Naidu AS. 2000. Phyto-phenols In: Naidu AS, editor. *Natural Food*
763 *Antimicrobial Systems*. Boca Raton, Flor.: CRC Press. p. 265-94.
- 764 Davidson PM, Zivanovic S. 2003. The use of natural antimicrobials. In: Zeuthen P,
765 Bogh-Sorensen L, editors. *Food Preservation Techniques*. Boca Raton, Flor.:
766 Woodhead Publishing Limited and CRC Press LLC. p. 5-30.
- 767 Davidson PM, Sofos JN, Branen AL. 2005. *Antimicrobials in foods*. Boca Raton,
768 Flor.: CRC press, Taylor & Francis Group. 706 p.
- 769 Davidson PM, Taylor MT. 2007. Chemical preservatives and natural antimicrobial
770 compounds. In: Doyle P, Beuchat LR, Montville TJ, editors. *Food*
771 *Microbiology: Fundamentals and Frontiers*. Washington: American Society for
772 Microbiology Press. p. 713-34.
- 773 de Oliveira TM, de Fátima Ferreira Soares N, Magela Pereira R, de Freitas Fraga K.
774 2007. Development and evaluation of antimicrobial natamycin-incorporated
775 film in gorgonzola cheese conservation. *Packag Technol Sci* 20 (2):147-53.
- 776 Deans JG, Ritchie G. 1987. Antimicrobial properties of plant essential oils. *Int J Food*
777 *Microbiol* 5 (2):165-80.
- 778 Devlieghere F, Vermeiren L, Bockstal A, Debevere J. 2000a. Study on antimicrobial
779 activity of a food packaging materials containing potassium sorbate. *Acta*
780 *Aliment* 29 (2):137-46.

- 781 Devlieghere F, Vermeiren L, Jacobs M, Debevere J. 2000b. The effectiveness of
782 hexamethylenetetramine-incorporated plastic for the active packaging of
783 foods. *Packag Technol Sci* 13 (3):117-22.
- 784 Devlieghere F, Vermeiren L, Debevere J. 2004a. New preservation technologies:
785 Possibilities and limitations. *Int Dairy J* 14 (4):273-85.
- 786 Devlieghere F, Vermeulen A, Debevere J. 2004b. Chitosan: Antimicrobial activity,
787 interactions with food components and applicability as a coating on fruit and
788 vegetables. *Food Microbiol* 21 (6):703-14.
- 789 Dorman HJD, Deans SG. 2000. Antimicrobial agents from plants: Antibacterial
790 activity of plant volatile oils. *J Appl Microbiol* 88 (2):308-16.
- 791 Du W-X, Olsen CW, Avena-Bustillos RJ, McHugh TH, Levin CE, Friedman M. 2008.
792 Storage stability and antibacterial activity against *Escherichia coli* O157:H7 of
793 carvacrol in edible apple films made by two different casting methods. *J Agric*
794 *Food Chem* 56 (9):3082-8.
- 795 Elgayyar M, Draughon FA, Golden DA, Mount JR. 2001. Antimicrobial activity of
796 essential oils from plants against selected pathogenic and saprophytic
797 microorganisms. *J Food Protect* 67 (7):1019-24.
- 798 Falcone P, Speranza B, Del Nobile MA, Corbo MR, Sinigaglia M. 2005. A study on
799 the antimicrobial activity of thymol intended as a natural preservative. *J Food*
800 *Protect* 68 (8):1664-70.
- 801 Farag RS, Daw ZY, Abo-Raya SH. 1989. Influence of some spice essential oils on
802 *Aspergillus parasiticus* growth and production of aflatoxins in a synthetic
803 medium. *J Food Sci* 54 (1):74-6.
- 804 Farkas J. 2007. Physical methods of food preservation. In: Doyle P, Beuchat LR,
805 Montville TJ, editors. *Food Microbiology: Fundamentals and Frontiers*.
806 Washington: American Society for Microbiology Press. p. 685-705.
- 807 Fisher K, Phillips C, McWatt L. 2009. The use of an antimicrobial citrus vapour to
808 reduce *Enterococcus* sp. On salad products. *Int J Food Sci Technol* 44
809 (9):1748-54.
- 810 Fitzgerald DJ, Stratford M, Narbad A. 2003. Analysis of the inhibition of food
811 spoilage yeasts by vanillin. *Int J Food Microbiol* 86 (1-2):113-22.
- 812 Friedman M, Henika PR, Mandrell RE. 2002. Bactericidal activities of plant essential
813 oils and some of their isolated constituents against *Campylobacter jejuni*,
814 *Escherichia coli*, *Listeria monocytogenes*, and *Salmonella enterica*. *J Food*
815 *Protect* 65 (10):1545-60.
- 816 Friedman M, Henika PR, Levin CE, Mandrell RE. 2004. Antibacterial activities of
817 plant essential oils and their components against *Escherichia coli* O157:H7
818 and *Salmonella enterica* in apple juice. *J Agric Food Chem* 52 (19):6042-8.
- 819 Fyfe L, Armstrong F, Stewart J. 1998. Inhibition of *Listeria monocytogenes* and
820 *Salmonella enteritidis* by combinations of plant oils and derivatives of benzoic
821 acid: The development of synergistic antimicrobial combinations. *Int J*
822 *Antimicrob Ag* 9 (3):130-6.
- 823 Goñi P, López P, Sánchez C, Gómez-Lus R, Becerril R, Nerín C. 2009. Antimicrobial
824 activity in the vapour phase of a combination of cinnamon and clove essential
825 oils. *Food Chem* 116 (4):982-9.
- 826 Grower JL, Cooksey K, Getty KJK. 2004. Development and characterization of an
827 antimicrobial packaging film coating containing nisin for inhibition of *Listeria*
828 *monocytogenes*. *J Food Protect* 67 (3):475-9.

- 829 Gustafson JE, Liew YC, Chew S, Markham JL, Bell HC, Wyllie SG, Warmington JR.
830 1998. Effects of tea tree oil on *Escherichia coli*. Lett Appl Microbiol 26
831 (3):194-8.
- 832 Gutierrez J, Barry-Ryan C, Bourke P. 2008. The antimicrobial efficacy of plant
833 essential oil combinations and interactions with food ingredients. Int J Food
834 Microbiol 124 (1):91-7.
- 835 Gutierrez J, Barry-Ryan C, Bourke P. 2009. Antimicrobial activity of plant essential
836 oils using food model media: Efficacy, synergistic potential and interactions
837 with food components. Food Microbiol 26 (2):142-50.
- 838 Gutiérrez L, Escudero A, Batlle Rn, Nerín C. 2009. Effect of mixed antimicrobial
839 agents and flavors in active packaging films. J Agric Food Chem 57
840 (18):8564-71.
- 841 Guynot ME, Ramos AJ, Setó L, Purroy P, Sanchis V, Marín S. 2003. Antifungal
842 activity of volatile compounds generated by essential oils against fungi
843 commonly causing deterioration of bakery products. J Appl Microbiol 94
844 (5):893-9.
- 845 Ha JU, Kim YM, Lee DS. 2001. Multilayered antimicrobial polyethylene films
846 applied to the packaging of ground beef. Packag Technol Sci 15 (2):55-62.
- 847 Halek WG, Anita G. 1989. Fungal inhibition by a fungicide coupled to an ionomeric
848 film. J Food Safety 9 (3):215-22.
- 849 Han J, Castell-Perez ME, Moreira RG. 2005. Antimicrobial films as a potential
850 technology to increase pathogen radiation sensitivity. Proceedings of IFT
851 Annual Meeting. New Orleans, Lou., July 15-20.
- 852 Han JH, Floros JD. 1997. Casting antimicrobial packaging films and measuring their
853 physical properties and antimicrobial activity. J Plastic Film Sheeting 13
854 (4):287-98.
- 855 Han JH, Floros JD. 1998. Simulating diffusion model and determining diffusivity of
856 potassium sorbate through plastics to develop antimicrobial packaging film. J
857 Food Process Pres 22 (2):107-22.
- 858 Han JH. 2000. Antimicrobial food packaging. Food Technol 54 (3):56-65.
- 859 Han JH. 2003. Antimicrobial food packaging. In: Ahvenainen R, editor. Novel Food
860 Packaging Techniques. Cambridge: Woodhead Publishing Limited. p. 50-70.
- 861 Han JH. 2005. Antimicrobial packaging systems. In: Han JH, editor. Innovations in
862 Food Packaging. San Diego, Calif.: Elsevier Academic Press. p. 92-108.
- 863 Helander IM, Alakomi HL, Latva KK, Mattila ST, Pol I, Smid EJ, Wright VA. 1998.
864 Characterisation of the action of selected essential oil components on gram-
865 negative bacteria. J Agric Food Chem 46 (9):3590-5.
- 866 Hoffman KL, Han IY, Dawson PL. 2001. Antimicrobial effects of corn zein films
867 impregnated with nisin, lauric acid and EDTA. J Food Protect 64 (6):885-9.
- 868 Hong S-I, Park J-D, Him D-M. 2000. Antimicrobial and physical properties of food
869 packaging films incorporated with some natural compounds. Food Sci
870 Biotechnol 9 (1):38-42.
- 871 Ji J, Zhang W. 2009. Bacterial behaviors on polymer surfaces with organic and
872 inorganic antimicrobial compounds. J Biomed Mater Res A 88A (2):448-53.
- 873 Joerger RD. 2007. Antimicrobial films for food applications: A quantitative analysis
874 of their effectiveness. Packag Technol Sci 20 (4):231-73.
- 875 Juneja VK, Sofos JN. 2005. Control of foodborne microorganisms. In: Rooney ML,
876 editor. Food Packaging: Principles and Practice. New York: Marcel Dekker
877 Inc. p. 13-281.

- 878 Kerry JP, O'Grady MN, Hogan SA. 2006. Past, current and potential utilisation of
879 active and intelligent packaging systems for meat and muscle-based products:
880 A review. *Meat Sci* 74 (1):113-30.
- 881 Kim JM, Marshall MR, Cornell JA, Preston JF, Wei CI. 1995. Antibacterial activity of
882 carvacrol, citral and gerniol against *Salmonella typhimurium* in culture
883 medium and on fish cubes. *J Food Sci* 60 (6):1364-8.
- 884 Kim Y-M, Paik H-D, Lee D-S. 2002. Shelf-life characteristics of fresh oysters and
885 ground beef as affected by bacteriocin-coated plastic packaging film. *J Sci*
886 *Food Agr* 82 (9):998-1002.
- 887 Kloucek P, Smid J, Frankova A, Kokoska L, Valterova I, Pavela R. 2011. Fast
888 screening method for assessment of antimicrobial activity of essential oils in
889 vapor phase. *Food Res Int in press*: DOI: 10.1016/j.foodres.2011.04.044.
- 890 Knobloch K, Pauli A, Iberl B. 1989. Antibacterial and antifungal properties of
891 essential oil components. *J Essent Oil Res* 1 (3):119-28
- 892 Koga T, Hirota N, Takumi K. 1999. Bactericidal activities of essential oils of basil
893 and sage against a range of bacteria and the effects of these essential oils on
894 *Vibrio parahaemolyticus*. *Microbiol Res* 154 (3):267-73.
- 895 Koichiro Y. 1993. Keeping freshness by antimicrobial packaging incorporating
896 wasabi extract. *Food Sci* 11 (35):102-7.
- 897 Krochta JM, De Mulder-Johnston C. 1997. Edible and biodegradable polymer films.
898 *Food Technol* 51 (2):61-74.
- 899 Kuorwel KK, Cran MJ, Sonneveld K, Miltz J, Bigger SW. 2011a. Antimicrobial
900 activity of natural agents against *Saccharomyces cerevisiae*. *Packag Technol*
901 *Sci in press*: DOI: 10.1002/pts.939.
- 902 Kuorwel KK, Cran MJ, Sonneveld K, Miltz J, Bigger SW. 2011b. Antimicrobial
903 activity of biodegradable polysaccharide and protein-based films containing
904 active agents. *J Food Sci* 76 (3):R90-R102.
- 905 Lachowicz KJ, Jones GP, Briggs DR, Bienvenu FE, Wan J, Wilcock A, Coventry MJ.
906 1998. The synergistic preservative effects of the essential oils of sweet basil
907 (*Ocimum basilicum l.*) against acid-tolerant food microflora. *J Appl Microbiol*
908 26 (3):209-14.
- 909 Lambert RJW, Skandamis PN, Coote PJ, Nychas GJ. 2001. A study of the minimum
910 inhibitory concentration and mode of action of oregano essential oil, thymol
911 and carvacrol. *J Appl Microbiol* 91 (3):453-62.
- 912 Lau OW, Wong SK. 2000. Contamination in food from packaging material. *J*
913 *Chromatogr A* 882 (1):255-70.
- 914 Lee D-S, Hwang Y, Cho S-H. 1998. Developing antimicrobial packaging film for
915 curled lettuce and soyabean sprouts. *Food Sci Biotechnol* 7 (2):117-21.
- 916 Limjaroen P, Ryser E, Lockhart H, Harte B. 2003. Development of a food packaging
917 coating material with antimicrobial properties. *J Plastic Film Sheeting* 19
918 (2):95-109.
- 919 Limjaroen P, Ryser E, Lockhart H, Harte B. 2005. Inactivation of *Listeria*
920 *monocytogenes* on beef bologna and Cheddar cheese using polyvinyl-idene
921 chloride films containing sorbic acid. *J Food Sci* 70 (5):M267-M71.
- 922 Lin YT, Labbe RG, Shetty K. 2004. Inhibition of *Listeria monocytogenes* in fish and
923 meat systems by use of oregano and cranberry phytochemical synergies. *Appl*
924 *Environ Microbiol* 70 (9):5672-8.
- 925 López-Malo A, Alzamora SM, Palou E. 2002. *Aspergillus flavus* dose-response curves
926 to selected natural and synthetic antimicrobials. *Int J Food Microbiol* 73 (2-
927 3):213-8.

- 928 López-Malo A, Maris Alzamora S, Palou E. 2005. *Aspergillus flavus* growth in the
929 presence of chemical preservatives and naturally occurring antimicrobial
930 compounds. *Int J Food Microbiol* 99 (2):119-28.
- 931 López-Malo A, Barreto-Valdivieso J, Palou E, Martín FS. 2007. *Aspergillus flavus*
932 growth response to cinnamon extract and sodium benzoate mixtures. *Food*
933 *Control* 18 (11):1358-62.
- 934 López P, Sánchez C, Batlle R, Nerín C. 2007a. Vapor-phase activities of cinnamon,
935 thyme, and oregano essential oils and key constituents against foodborne
936 microorganisms. *J Agric Food Chem* 55 (11):4348-56.
- 937 López P, Sánchez C, Batlle R, Nerín C. 2007b. Development of flexible antimicrobial
938 films using essential oils as active agents. *J Agric Food Chem* 55 (21):8814-
939 24.
- 940 Lorenzo D, Loayza I, Dellacassa E. 2003. Composition of the essential oils from
941 leaves of two *Hedyosmum spp.* and bolovia. *Flavour Frag J* 18 (1):32-5.
- 942 Marques A, Encarnação S, Pedro S, Nunes M. 2008. In vitro antimicrobial activity of
943 garlic, oregano and chitosan against *Salmonella enterica*. *World J Microbiol*
944 *Biotechnol* 24 (10):2357-60.
- 945 Matan N, Rimkeeree H, Mawson AJ, Chompreeda P, Haruthaithanasan V, Parker M.
946 2006. Antimicrobial activity of cinnamon and clove oils under modified
947 atmosphere conditions. *Int J Food Microbiol* 107 (2):180-5.
- 948 Matche RS, Kulkarni G, Raj B. 2006. Modification of ethylene acrylic acid film for
949 antimicrobial activity. *J Appl Polym Sci* 100 (4):3063-8.
- 950 Mauriello G, Ercolini D, La Storia A, Casaburi A, Villani F. 2004. Development of
951 polythene films for food packaging activated with an antilisterial bacteriocin
952 from *Lactobacillus curvatus* 32Y. *J Appl Microbiol* 97 (2):314-22.
- 953 Mauriello G, De Luca E, La Storia A, Villani F, Ercolini D. 2005. Antimicrobial
954 activity of a nisin-activated plastic film for food packaging. *Lett Appl*
955 *Microbiol* 41 (6):464-9.
- 956 Mazzanti G, Battinelli L, Salvatore G. 1998. Antimicrobial properties of the linalool-
957 rich essential oil of *Hyssopus officinalis L. var decumbens* (Lamiaceae).
958 *Flavour Frag J* 13 (5):289-94.
- 959 Mehyar GF, Al-Qadiri HM, Abu-Blan HA, Swanson BG. 2011. Antifungal
960 effectiveness of potassium sorbate incorporated in edible coatings against
961 spoilage molds of apples, cucumbers, and tomatoes during refrigerated
962 storage. *J Food Sci* 76 (3):M210-M7.
- 963 Mejlholm O, Dalgaard P. 2002. Antimicrobial effect of essential oils on the seafood
964 spoilage micro-organism *Photobacterium phosphoreum* in liquid media and
965 fish products. *Lett Appl Microbiol* 34 (1):27-31.
- 966 Miltz J, Passy N, Mannhwim CH. 1995. Trends and applications of active packaging
967 systems. In: Ackerman P, Jagerstad M, Ohlsson T, editors. *Food and*
968 *Packaging Materials - Chemical Interaction*. Cambridge: The Royal Society of
969 Chemistry. p. 201-10.
- 970 Miltz J, Rydlo T, Mor A, Polyakov V. 2006. Potency evaluation of a dermaseptin s4
971 derivative for antimicrobial food packaging applications. *Packag Technol Sci*
972 19 (6):345-54.
- 973 Natrajan N, Sheldon BW. 2000. Efficacy of nisin-coated polymer films to inactivate
974 *Salmonella typhimurium* on fresh broiler skin. *J Food Protect* 63 (9):1189-96.
- 975 Nedorostova L, Kloucek P, Urbanova K, Kokoska L, Smid J, Urban J, Valterova I,
976 Stolicova M. 2011. Antibacterial effect of essential oil vapours against different

- 977 strains of *Staphylococcus aureus*, including MRSA. Flavour Frag J *in press*:
 978 DOI: 10.1002/ffj.2068.
- 979 Nielsen PV, Rios R. 2000. Inhibition of fungal growth on bread by volatile
 980 components from spices and herbs, and the possible application in active
 981 packaging, with special emphasis on mustard essential oil. *Int J Food*
 982 *Microbiol* 60 (2-3):219-29.
- 983 Nychas GJE. 1995. Natural antimicrobials from plants. In: Gould GW, editor. *New*
 984 *Methods of Food Preservation*. London: Blackie Academic & Professional. p.
 985 58-89.
- 986 Olasupo NA, Fitzgerald DJ, Narbad A, Gasson MJ. 2004. Inhibition of *Bacillus*
 987 *subtilis* and *Listeria innocua* by nisin in combination with some naturally
 988 occurring organic compounds. *J Food Protect* 67 (3):596-600.
- 989 Ouattara B, Simard RE, Piette G, Bégin A, Holley RA. 2000. Inhibition of surface
 990 spoilage bacteria in processed meats by application of antimicrobial films
 991 prepared with chitosan. *Int J Food Microbiol* 62 (1-2):139-48.
- 992 Oussalah M, Caillet S, Saucier L, Lacroix M. 2006. Antimicrobial effects of selected
 993 plant essential oils on the growth of a *Pseudomonas putida* strain isolated from
 994 meat. *Meat Sci* 73 (2):236-44.
- 995 Ozdemir M, Floros JD. 2004. Active food packaging technologies. *Crit Rev Food Sci*
 996 *Nutr* 44 (3):185-93.
- 997 Padgett T, Han IY, Dawson PL. 1998. Incorporation of food-grade antimicrobial
 998 compounds into biodegradable packaging films. *J Food Protect* 61 (10):1330-
 999 5.
- 1000 Pauli A. 2001. Antimicrobial properties of essential oil constituents. *Int J Aromather*
 1001 11 (3):126-33.
- 1002 Periago P, Delgoda B, Fernandez P, Palop A. 2004. Use of carvacrol and cymene to
 1003 control growth and viability of *Listeria monocytogenes* cells and predictions of
 1004 survivors using frequency distribution functions. *J Food Protect* 67 (7):1408-
 1005 16.
- 1006 Persico P, Ambrogi V, Carfagna C, Cerruti P, Ferrocino I, Mauriello G. 2009.
 1007 Nanocomposite polymer films containing carvacrol for antimicrobial active
 1008 packaging. *Polym Eng Sci* 49 (7):1447-55.
- 1009 Prasad G, Kumar A, Singh AK, Bhattacharya AK, Singh K, Sharma VD. 1986.
 1010 Antimicrobial activity of essential oils of some *Ocimum* species and clove oil.
 1011 *Fitoterapia* 57 (6):429-32.
- 1012 Quintavalla S, Vicini L. 2002. Antimicrobial food packaging in the meat industry.
 1013 *Meat Sci* 62 (3):373-80.
- 1014 Rai MK, Qureshi S, Pandey AK. 1999. In vitro susceptibility of opportunistic
 1015 *Fusarium* spp. to essential oils. *Mycoses* 42 (1-2):97-101.
- 1016 Ramirez MX, Walters KB, Hirt DE. 2005. Relationship between erucamide surface
 1017 concentration and coefficient of friction of LLDPE film. *J Vinyl Addit*
 1018 *Technol* 11 (1):9-12.
- 1019 Rardniyom C. 2008. Development of multi-layer films containing natural
 1020 antimicrobial agents. [PhD dissertation]. Melbourne, Australia: Victoria
 1021 University. 145 p.
- 1022 Rardniyom C, Miltz J, Bigger SW, Cran MJ, Sonneveld K. 2008. Parameters affecting
 1023 the design of antimicrobial films. *Proceedings of 16th IAPRI World*
 1024 *Conference on Packaging*. Bangkok, Thailand, June 8-12.

- 1025 Raybaudi-Massilia RM, Mosqueda-Melgar J, Martín-Belloso O. 2009. Edible
1026 alginate-based coating as carrier of antimicrobials to improve shelf-life and
1027 safety of fresh-cut melon. *Int J Food Microbiol* 121 (3):313-27.
- 1028 Rodríguez-Lafuente A, Nerín C, Batlle R. 2010. Active paraffin-based paper
1029 packaging for extending the shelf life of cherry tomatoes. *J Agric Food Chem*
1030 58 (11):6780-6.
- 1031 Rodríguez A, Nerín C, Batlle R. 2008. New cinnamon-based active paper packaging
1032 against *Rhizopus stolonifer* food spoilage. *J Agric Food Chem* 56 (15):6364-9.
- 1033 Rojas-Graü MA, Solvia-Fortuny R, Martín-Belloso O. 2009. Edible coatings to
1034 incorporate active ingredients to fresh-cut fruits: A review. *Trends Food Sci*
1035 *Technol* 20 (10):438-47.
- 1036 Rokchoy S, Suppakul P, Jinkarn T. 2009. Antimicrobial effects of vanillin coated
1037 solution for coating paperboard intended for packaging bakery products. *Asian*
1038 *J Food Agro-Ind* 4 (2):138-47.
- 1039 Roller S, Seedhar P. 2002. Carvacrol and cinnamic acid inhibit microbial growth in
1040 fresh-cut melon and kiwifruit at 4°C and 8°C. *Lett Appl Microbiol* 35 (5):390-
1041 4.
- 1042 Rooney ML. 1995. Active food packaging. London: Blackie Academic &
1043 Professional. p. 260.
- 1044 Rupika LAS, Sonneveld K, Miltz J, Bigger SW. 2005. Development and evaluation of
1045 low-density polyethylene-based antimicrobial food packaging polymers
1046 containing thymol and carvacrol. Proceedings of 22nd IAPRI Symposium on
1047 Packaging. Campinas, Brazil, May 22-24.
- 1048 Rupika LAS, Miltz J, Sonneveld K, Cran MJ, Bigger SW. 2006. Effectiveness of
1049 antimicrobial films containing a natural antimicrobial agent. Proceedings of
1050 15th IAPRI World Conference on Packaging. Tokyo, Japan, October 2-5.
- 1051 Rupika LAS, Sonneveld K, Bigger SW, Cran MJ, Miltz J. 2008. Modelling microbial
1052 inactivation by films containing thymol or carvacrol. Proceedings of 16th
1053 IAPRI World Conference on Packaging. Bangkok, Thailand, June 8-12.
- 1054 Rydlo T, Miltz J, Mor A. 2006. Eukaryotic antimicrobial peptides: Promises and
1055 premises in food safety. *J Food Sci* 71 (9):R125-R35.
- 1056 Sagdiç O, Özcan M. 2003. Antibacterial activity of turkish spice hydrosols. *Food*
1057 *Control* 14 (4):141-3.
- 1058 Salleh E, Muhamadi I, Khairuddin N. 2007. Inhibition of *Bacillus subtilis* and
1059 *Escherichia coli* by antimicrobial starch-based film incorporated with lauric
1060 acid and chitosan. Proceedings of 3rd CIGR Section VI International
1061 Symposium on Food and Agricultural Products: Processing and Innovation.
1062 Naples, Italy,
- 1063 Sanla-Ead N, Jangchud A, Chonhenchob V, Suppakul P. 2011. Antimicrobial activity
1064 of cinnamaldehyde and eugenol and their activity after incorporation into
1065 cellulose-based packaging films. *Packag Technol Sci in press*: DOI:
1066 10.1002/pts.952.
- 1067 Scannell AGM, Hill C, Ross RP, Marx S, Hartmeier W, Arendt EK. 2000.
1068 Development of bioactive food packaging materials using immobilised
1069 bacteriocins Lacticin 3147 and Nisaplin®. *Int J Food Microbiol* 60 (2-3):241-
1070 9.
- 1071 Schelz Z, Molnar J, Hohmann J. 2006. Antimicrobial and antiplasmid activities of
1072 essential oils. *Fitoterapia* 77 (4):279-85.

- 1073 Seaberg AC, Labbe RG, Shetty K. 2003. Inhibition of *Listeria monocytogenes* by elite
1074 clonal extracts of oregano (*Origanum vulgare*). Food Biotechnol 17 (2):129-
1075 49.
- 1076 Sikkema J, Bont JAM, Poolman B. 1995. Mechanisms of membrane toxicity of
1077 hydrocarbons. Microbiol Rev 59 (2):201-22.
- 1078 Silveira MFA, Soares NFF, Geraldine RM, Andrade NJ, Gonçalves MPJ. 2007.
1079 Antimicrobial efficiency and sorbic acid migration from active films into
1080 pastry dough. Packag Technol Sci 20 (4):287-92.
- 1081 Simon JE, Quinn J, Murray RG. 1990. Basil: A source of essential oils. In: Janick J,
1082 Simon JE, editors. Advances in New Crops Research. Portland, Oreg.: Timber
1083 Press. p. 484-9.
- 1084 Singh G, Marimuthu P, De Heluani CS, Catalan CAN. 2006. Antioxidant and biocidal
1085 activities of *Carum nigrum* (seed) essential oil, oleoresin, and their selected
1086 components. J Agric Food Chem 54 (1):174-81.
- 1087 Singh TK, Drake MA, Cadwallader KR. 2003. Flavour of Cheddar cheese: A
1088 chemical and sensory perspective. Comp Rev Food Sci Food Safety 2 (4):139-
1089 62.
- 1090 Siragusa GR, Cutter CN, Willett JL. 1999. Incorporation of bacteriocin in plastic
1091 retains activity and inhibits surface growth of bacteria on meat. Food
1092 Microbiol 16 (3):229-35.
- 1093 Sivropoulou A, Papanikolaou E, Nikolaou C, Kokkini S, Lanaras T, Arsenakis M.
1094 1996. Antimicrobial and cytotoxic activities of *Origanum* essential oils. J
1095 Agric Food Chem 44 (5):1202-5.
- 1096 Skandamis PN, Nychas G-JE. 2001. Effect of oregano essential oil on microbiological
1097 and physico-chemical attributes of minced meat stored in air and modified
1098 atmospheres. J Appl Microbiol 91 (6):1011-22.
- 1099 Smith-Palmer A, Stewart J, Fyfe L. 2001. The potential application of plant essential
1100 oils as natural food preservatives in soft cheese. Food Microbiol 18 (4):463-70.
- 1101 Smith J, Hoshino J, Abe Y. 1995. Interactive packaging involving sachet technology.
1102 In: Rooney ML, editor. Active Food Packaging. London: Blackie Academic &
1103 Professional. p. 143-72.
- 1104 Suhr KI, Nielsen PV. 2003. Antifungal activity of essential oils evaluated by two
1105 different application techniques against rye bread spoilage fungi. J Appl
1106 Microbiol 94 (4):665-74.
- 1107 Suppakul P, Sonneveld K, Miltz J, Bigger SW. 2003a. Antimicrobial properties of
1108 basil and its possible application in food packaging. J Agric Food Chem 51
1109 (11):3197-207.
- 1110 Suppakul P, Sonneveld K, Miltz J, Bigger SW. 2003b. Active packaging technologies
1111 with an emphasis on antimicrobial packaging and its applications. J Food Sci
1112 68 (2):408-20.
- 1113 Suppakul P. 2004. Study of antimicrobial polymeric packaging films containing basil
1114 extracts. [PhD dissertation]. Melbourne, Australia: Victoria University 259 p.
- 1115 Suppakul P, Miltz J, Sonneveld K, Bigger SW. 2006. Characterization of
1116 antimicrobial films containing basil extracts. Packag Technol Sci 19 (5):259-
1117 68.
- 1118 Suppakul P, Sonneveld K, Bigger SW, Miltz J. 2008. Efficacy of polyethylene-based
1119 antimicrobial films containing principal constituents of basil. LWT - Food Sci
1120 Technol 41 (5):779-88.

- 1121 Suppakul P, Sonneveld K, Bigger SW, Miltz J. 2011a. Diffusion of linalool and
 1122 methylchavicol from polyethylene-based antimicrobial packaging films. LWT
 1123 - Food Sci Technol 44 (9):1888-93
- 1124 Suppakul P, Sonneveld K, Bigger SW, Miltz J. 2011b. Loss of AM additives from
 1125 antimicrobial films during storage. J Food Eng 105 (2):270-6.
- 1126 Tassou C, Koutsoumanis K, Nychas GJE. 2000. Inhibition of Salmonella enteritidis
 1127 and Staphylococcus aureus in nutrient broth by mint essential oil. Food Res Int
 1128 33 (3-4):273-80.
- 1129 Tassou CC, Drosinos EH, Nychas G-JE. 1996. Inhibition of resident microbial flora
 1130 and pathogen inocula on cold fresh fish fillets in olive oil, oregano, and lemon
 1131 juice under modified atmosphere or air. J Food Protect 59 (1):31-4.
- 1132 Tepe B, Daferera D, Sokmen M, Polissiou M, Sokmen A. 2004. *In vitro* antimicrobial
 1133 and antioxidant activities of the essential oils and various extracts of *Thymus*
 1134 *eigii*. J Agric Food Chem 52 (5):1132-7.
- 1135 Tsigarida E, Skandamis P, Nychas G-JE. 2000. Behaviour of *Listeria monocytogenes*
 1136 and autochthonous flora on meat stored under aerobic, vacuum and modified
 1137 atmosphere packaging conditions with or without the presence of oregano
 1138 essential oil at 5°C. J Appl Microbiol 89 (6):901-9.
- 1139 Tunç S, Duman O. 2011. Preparation of active antimicrobial methyl
 1140 cellulose/carvacrol/montmorillonite nanocomposite films and investigation of
 1141 carvacrol release. LWT - Food Sci Technol 44 (2):465-72.
- 1142 Ultee A, Gorris LGM, Smid EJ. 1998. Bactericidal activity of carvacrol towards the
 1143 food-borne pathogen *Bacillus cereus*. J Appl Microbiol 85 (2):211-8.
- 1144 Ultee A, Kets EPW, Smid EJ. 1999. Mechanisms of action of carvacrol on the food-
 1145 borne pathogen *Bacillus cereus*. Appl Environ Microbiol 65 (10):4606-10.
- 1146 Ultee A, Slump RA, Steging G, Smid EJ. 2000. Antimicrobial activity of carvacrol
 1147 towards *Bacillus cereus* on rice. J Food Protect 63 (5):620-4.
- 1148 Ultee A, Smid EJ. 2001. Influence of carvacrol on growth and toxin production by
 1149 *Bacillus cereus*. Int J Food Microbiol 64 (3):373-8.
- 1150 Ultee A, Bennik MHJ, Moezelaar R. 2002. The phenolic hydroxyl group of carvacrol
 1151 is essential for action against the food borne pathogen *Bacillus cereus*. Appl
 1152 Environ Microbiol 68 (4):1561-8.
- 1153 Vartiainen J, Skytta E, Ahvenainen-Rantala R, Enqvist J. 2003a. Antimicrobial and
 1154 barrier properties of LDPE films containing imazalil and EDTA. J Plastic Film
 1155 Sheeting 19 (4):249-61.
- 1156 Vartiainen J, Skytta E, Enqvist J, Ahvenainen R. 2003b. Properties of antimicrobial
 1157 plastics containing traditional food preservatives. Packag Technol Sci 16
 1158 (6):223-30.
- 1159 Vermeiren L, Devlieghere F, Beest MV, Kruijf ND, Debevere J. 1999. Developments
 1160 in the active packaging of foods. Trends Food Sci Technol 10 (3):77-86.
- 1161 Vermeiren L, Devlieghere F, Debevere J. 2002. Effectiveness of some recent
 1162 antimicrobial packaging concepts. Food Addit Contam 19 (suppl.):163-71.
- 1163 Viuda-Martos M, Ruiz-Navajas Y, Fernandez-Lopez J, Perez-Alvarez J. 2008.
 1164 Antibacterial activity of different essential oils obtained from spices widely
 1165 used in mediterranean diet. Int J Food Sci Technol 43 (3):526-31.
- 1166 Vojdani F, Torres AJ. 1989. Potassium sorbate permeability of methylcellulose and
 1167 hydroxypropyl methylcellulose multi-layer films. J Food Process Pres 13
 1168 (6):417-30.

- 1169 Wan J, Wilcock A, Coventry MJ. 1998. The effect of essential oils of basil on the
1170 growth of *Aeromonas hydrophila* and *Pseudomonas fluorescens*. J Appl
1171 Microbiol 84 (2):152-8.
- 1172 Weng Y-M, Hotchkiss JH. 1992. Inhibition of surface molds on cheese by
1173 polyethylene film containing the antimycotic imazalil. J Food Protect 58
1174 (5):367-9.
- 1175 Weng Y-M, Hotchkiss JH. 1993. Anhydrides as antimycotic agents added to
1176 polyethylene films for food packaging. Packag Technol Sci 6 (3):123-8.
- 1177 Weng Y-M, Chen M-J. 1997. Sorbic anhydride as antimycotic additive in
1178 polyethylene food packaging films. LWT - Food Sci Technol 30 (5):485-7.
- 1179 Weng Y-M, Chen M-J, Chen W. 1999. Antimicrobial food packaging materials from
1180 poly(ethylene-co-methacrylic acid). LWT - Food Sci Technol 32 (4):191-5.
- 1181 Wu J-G, Wang P-J, Chen SC. 2010. Antioxidant and antimicrobial effectiveness of
1182 catechin-impregnated PVA–starch film on red meat. J Food Qual 33 (6):780-
1183 801.
- 1184 Youdim KA, Deans SG. 2000. Effect of thyme oil and thymol dietary
1185 supplementation on the antioxidant status and fatty acid composition of the
1186 ageing rat brain. J Nutr 83 (1):87-93.
- 1187 Zaika LL. 1988. Spices and herbs: Their antimicrobial activity and its determination. J
1188 Food Safety 9 (2):97-118.