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864696

# THE UNITED STATES OF AMERICA

TO ALL TO WHOM THESE PRESENTS SHALL COME:

UNITED STATES DEPARTMENT OF COMMERCE

United States Patent and Trademark Office

*July 05, 2017*

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**APPLICATION NUMBER: 62/356,910**

**FILING DATE: *June 30, 2016***

**RELATED PCT APPLICATION NUMBER: *PCT/US17/39640***

**THE COUNTRY CODE AND NUMBER OF YOUR PRIORITY APPLICATION, TO BE USED FOR FILING ABROAD UNDER THE PARIS CONVENTION, IS *US62/356,910***



Certified by

*Michelle M. Lee*

Under Secretary of Commerce  
for Intellectual Property  
and Director of the United States  
Patent and Trademark Office

Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

<b>Application Data Sheet 37 CFR 1.76</b>		Attorney Docket Number	79470-US-PSP
		Application Number	
Title of Invention	SYNERGISTIC COMBINATION OF 3-IODO-2-PROPYNYL-BUTYLCARBAMATE AND DIAMINE		
The application data sheet is part of the provisional or nonprovisional application for which it is being submitted. The following form contains the bibliographic data arranged in a format specified by the United States Patent and Trademark Office as outlined in 37 CFR 1.76. This document may be completed electronically and submitted to the Office in electronic format using the Electronic Filing System (EFS) or the document may be printed and included in a paper filed application.			

**Secrecy Order 37 CFR 5.2:**

Portions or all of the application associated with this Application Data Sheet may fall under a Secrecy Order pursuant to 37 CFR 5.2 (Paper filers only. Applications that fall under Secrecy Order may not be filed electronically.)

**Inventor Information:**

Inventor	1			Remove
Legal Name				
Prefix	Given Name	Middle Name	Family Name	Suffix
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Legal Name				

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<b>Application Data Sheet 37 CFR 1.76</b>	Attorney Docket Number	79470-US-PSP
	Application Number	
Title of Invention	SYNERGISTIC COMBINATION OF 3-iodo-2-propynyl-butylcarbamate and diamine	

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Inventor	4	<input type="button" value="Remove"/>		
<b>Legal Name</b>				

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	Maciej		Szymeczko	
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<b>Legal Name</b>				

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<b>Application Data Sheet 37 CFR 1.76</b>	Attorney Docket Number	79470-US-PSP
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All Inventors Must Be Listed - Additional Inventor Information blocks may be generated within this form by selecting the **Add** button.

### Correspondence Information:

Enter either Customer Number or complete the Correspondence Information section below.  
For further information see 37 CFR 1.33(a).

An Address is being provided for the correspondence information of this application.

Customer Number	21898		
Email Address		<input type="button" value="Add Email"/>	<input type="button" value="Remove Email"/>

### Application Information:

Title of the Invention	SYNERGISTIC COMBINATION OF 3-IODO-2-PROPYNYL-BUTYLCARBAMATE AND DIAMINE		
Attorney Docket Number	79470-US-PSP	Small Entity Status Claimed	<input type="checkbox"/>
Application Type	Provisional		
Subject Matter	Utility		
Total Number of Drawing Sheets (if any)		Suggested Figure for Publication (if any)	

### Filing By Reference:

Only complete this section when filing an application by reference under 35 U.S.C. 111(c) and 37 CFR 1.57(a). Do not complete this section if application papers including a specification and any drawings are being filed. Any domestic benefit or foreign priority information must be provided in the appropriate section(s) below (i.e., "Domestic Benefit/National Stage Information" and "Foreign Priority Information").

For the purposes of a filing date under 37 CFR 1.53(b), the description and any drawings of the present application are replaced by this reference to the previously filed application, subject to conditions and requirements of 37 CFR 1.57(a).

Application number of the previously filed application	Filing date (YYYY-MM-DD)	Intellectual Property Authority or Country

### Publication Information:

Request Early Publication (Fee required at time of Request 37 CFR 1.219)

**Request Not to Publish.** I hereby request that the attached application not be published under 35 U.S.C. 122(b) and certify that the invention disclosed in the attached application **has not and will not be** the subject of an application filed in another country, or under a multilateral international agreement, that requires publication at eighteen months after filing.

### Representative Information:

Representative information should be provided for all practitioners having a power of attorney in the application. Providing this information in the Application Data Sheet does not constitute a power of attorney in the application (see 37 CFR 1.32). Either enter Customer Number or complete the Representative Name section below. If both sections are completed the customer Number will be used for the Representative Information during processing.

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<b>Application Data Sheet 37 CFR 1.76</b>		Attorney Docket Number	79470-US-PSP
		Application Number	
Title of Invention	SYNERGISTIC COMBINATION OF 3-iodo-2-propynyl-butylcarbamate and diamine		
Please Select One:			
<input checked="" type="radio"/> Customer Number		US Patent Practitioner	<input type="radio"/> Limited Recognition (37 CFR 11.9)
Customer Number	21898		

### Domestic Benefit/National Stage Information:

This section allows for the applicant to either claim benefit under 35 U.S.C. 119(e), 120, 121, 365(c), or 386(c) or indicate National Stage entry from a PCT application. Providing benefit claim information in the Application Data Sheet constitutes the specific reference required by 35 U.S.C. 119(e) or 120, and 37 CFR 1.78.

When referring to the current application, please leave the "Application Number" field blank.

Prior Application Status	<input type="text"/>	<input type="button" value="Remove"/>
Application Number	Continuity Type	Prior Application Number
<input type="text"/>	<input type="text"/>	Filing or 371(c) Date (YYYY-MM-DD)
Additional Domestic Benefit/National Stage Data may be generated within this form by selecting the <b>Add</b> button.		
		<input type="button" value="Add"/>

### Foreign Priority Information:

This section allows for the applicant to claim priority to a foreign application. Providing this information in the application data sheet constitutes the claim for priority as required by 35 U.S.C. 119(b) and 37 CFR 1.55. When priority is claimed to a foreign application that is eligible for retrieval under the priority document exchange program (PDX)<sup>i</sup> the information will be used by the Office to automatically attempt retrieval pursuant to 37 CFR 1.55(i)(1) and (2). Under the PDX program, applicant bears the ultimate responsibility for ensuring that a copy of the foreign application is received by the Office from the participating foreign intellectual property office, or a certified copy of the foreign priority application is filed, within the time period specified in 37 CFR 1.55(g)(1).

Application Number	Country <sup>i</sup>	Filing Date (YYYY-MM-DD)	Access Code <sup>i</sup> (if applicable)
<input type="text"/>	<input type="text"/>	<input type="text"/>	<input type="text"/>
Additional Foreign Priority Data may be generated within this form by selecting the <b>Add</b> button.			<input type="button" value="Add"/>

### Statement under 37 CFR 1.55 or 1.78 for AIA (First Inventor to File) Transition Applications

<p>This application (1) claims priority to or the benefit of an application filed before March 16, 2013 and (2) also contains, or contained at any time, a claim to a claimed invention that has an effective filing date on or after March 16, 2013.</p> <p><input type="checkbox"/> NOTE: By providing this statement under 37 CFR 1.55 or 1.78, this application, with a filing date on or after March 16, 2013, will be examined under the first inventor to file provisions of the AIA.</p>
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Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

<b>Application Data Sheet 37 CFR 1.76</b>	Attorney Docket Number	79470-US-PSP
	Application Number	
Title of Invention	SYNERGISTIC COMBINATION OF 3-iodo-2-propynyl-butylcarbamate and diamine	

## Authorization or Opt-Out of Authorization to Permit Access:

When this Application Data Sheet is properly signed and filed with the application, applicant has provided written authority to permit a participating foreign intellectual property (IP) office access to the instant application-as-filed (see paragraph A in subsection 1 below) and the European Patent Office (EPO) access to any search results from the instant application (see paragraph B in subsection 1 below).

Should applicant choose not to provide an authorization identified in subsection 1 below, applicant **must opt-out** of the authorization by checking the corresponding box A or B or both in subsection 2 below.

**NOTE:** This section of the Application Data Sheet is **ONLY** reviewed and processed with the **INITIAL** filing of an application. After the initial filing of an application, an Application Data Sheet cannot be used to provide or rescind authorization for access by a foreign IP office(s). Instead, Form PTO/SB/39 or PTO/SB/69 must be used as appropriate.

### 1. Authorization to Permit Access by a Foreign Intellectual Property Office(s)

**A. Priority Document Exchange (PDX)** - Unless box A in subsection 2 (opt-out of authorization) is checked, the undersigned hereby **grants the USPTO authority** to provide the European Patent Office (EPO), the Japan Patent Office (JPO), the Korean Intellectual Property Office (KIPO), the State Intellectual Property Office of the People's Republic of China (SIPO), the World Intellectual Property Organization (WIPO), and any other foreign intellectual property office participating with the USPTO in a bilateral or multilateral priority document exchange agreement in which a foreign application claiming priority to the instant patent application is filed, access to: (1) the instant patent application-as-filed and its related bibliographic data, (2) any foreign or domestic application to which priority or benefit is claimed by the instant application and its related bibliographic data, and (3) the date of filing of this Authorization. See 37 CFR 1.14(h)(1).

**B. Search Results from U.S. Application to EPO** - Unless box B in subsection 2 (opt-out of authorization) is checked, the undersigned hereby **grants the USPTO authority** to provide the EPO access to the bibliographic data and search results from the instant patent application when a European patent application claiming priority to the instant patent application is filed. See 37 CFR 1.14(h)(2).

The applicant is reminded that the EPO's Rule 141(1) EPC (European Patent Convention) requires applicants to submit a copy of search results from the instant application without delay in a European patent application that claims priority to the instant application.

### 2. Opt-Out of Authorizations to Permit Access by a Foreign Intellectual Property Office(s)

A. Applicant **DOES NOT** authorize the USPTO to permit a participating foreign IP office access to the instant application-as-filed. If this box is checked, the USPTO will not be providing a participating foreign IP office with any documents and information identified in subsection 1A above.

B. Applicant **DOES NOT** authorize the USPTO to transmit to the EPO any search results from the instant patent application. If this box is checked, the USPTO will not be providing the EPO with search results from the instant application.

**NOTE:** Once the application has published or is otherwise publicly available, the USPTO may provide access to the application in accordance with 37 CFR 1.14.

Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

<b>Application Data Sheet 37 CFR 1.76</b>	Attorney Docket Number	79470-US-PSP
	Application Number	
Title of Invention	SYNERGISTIC COMBINATION OF 3-IODO-2-PROPYNYL-BUTYLCARBAMATE AND DIAMINE	

## Applicant Information:

Providing assignment information in this section does not substitute for compliance with any requirement of part 3 of Title 37 of CFR to have an assignment recorded by the Office.

<b>Applicant</b>	1	<input type="button" value="Remove"/>
<p>If the applicant is the inventor (or the remaining joint inventor or inventors under 37 CFR 1.45), this section should not be completed. The information to be provided in this section is the name and address of the legal representative who is the applicant under 37 CFR 1.43; or the name and address of the assignee, person to whom the inventor is under an obligation to assign the invention, or person who otherwise shows sufficient proprietary interest in the matter who is the applicant under 37 CFR 1.46. If the applicant is an applicant under 37 CFR 1.46 (assignee, person to whom the inventor is obligated to assign, or person who otherwise shows sufficient proprietary interest) together with one or more joint inventors, then the joint inventor or inventors who are also the applicant should be identified in this section.</p>		
<input type="button" value="Clear"/>		
Assignee	Legal Representative under 35 U.S.C. 117	Joint Inventor
<input type="checkbox"/> Person to whom the inventor is obligated to assign.		<input type="checkbox"/> Person who shows sufficient proprietary interest
If applicant is the legal representative, indicate the authority to file the patent application, the inventor is:		
<input type="text"/>		
Name of the Deceased or Legally Incapacitated Inventor: <input type="text"/>		
If the Applicant is an Organization check here. <input checked="" type="checkbox"/>		
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Address 2		
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Country	US	Postal Code
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Phone Number		Fax Number
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Email Address	FFUIMPC@DOW.COM	
Additional Applicant Data may be generated within this form by selecting the Add button.		
<input type="button" value="Add"/>		

## Assignee Information including Non-Applicant Assignee Information:

Providing assignment information in this section does not substitute for compliance with any requirement of part 3 of Title 37 of CFR to have an assignment recorded by the Office.



Under the Paperwork Reduction Act of 1995, no persons are required to respond to a collection of information unless it contains a valid OMB control number.

<b>Application Data Sheet 37 CFR 1.76</b>	Attorney Docket Number	79470-US-PSP
	Application Number	
Title of Invention	SYNERGISTIC COMBINATION OF 3-iodo-2-propynyl-butylcarbamate and diamine	

<b>Assignee</b>	1
-----------------	---

Complete this section if assignee information, including non-applicant assignee information, is desired to be included on the patent application publication. An assignee-applicant identified in the "Applicant Information" section will appear on the patent application publication as an applicant. For an assignee-applicant, complete this section only if identification as an assignee is also desired on the patent application publication.

If the Assignee or Non-Applicant Assignee is an Organization check here.

Prefix	Given Name	Middle Name	Family Name	Suffix

**Mailing Address Information For Assignee including Non-Applicant Assignee:**

Address 1				
Address 2				
City		State/Province		
Country <sup>i</sup>		Postal Code		
Phone Number		Fax Number		
Email Address				

Additional Assignee or Non-Applicant Assignee Data may be generated within this form by selecting the Add button.

**Signature:**


**NOTE:** This Application Data Sheet must be signed in accordance with 37 CFR 1.33(b). However, if this Application Data Sheet is submitted with the INITIAL filing of the application and either box A or B is not checked in subsection 2 of the "Authorization or Opt-Out of Authorization to Permit Access" section, then this form must also be signed in accordance with 37 CFR 1.14(c).

This Application Data Sheet **must** be signed by a patent practitioner if one or more of the applicants is a **juristic entity** (e.g., corporation or association). If the applicant is two or more joint inventors, this form must be signed by a patent practitioner, **all** joint inventors who are the applicant, or one or more joint inventor-applicants who have been given power of attorney (e.g., see USPTO Form PTO/AIA/81) on behalf of **all** joint inventor-applicants.

See 37 CFR 1.4(d) for the manner of making signatures and certifications.

Signature	/Tifani M. Edwards, Reg. No. 62,109/		Date (YYYY-MM-DD)	2016-06-30	
First Name	Tifani	Last Name	Edwards	Registration Number	62109

Additional Signature may be generated within this form by selecting the Add button.

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<b>Application Data Sheet 37 CFR 1.76</b>	Attorney Docket Number	79470-US-PSP
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Title of Invention	SYNERGISTIC COMBINATION OF 3-iodo-2-propynyl-butylcarbamate and diamine	

This collection of information is required by 37 CFR 1.76. The information is required to obtain or retain a benefit by the public which is to file (and by the USPTO to process) an application. Confidentiality is governed by 35 U.S.C. 122 and 37 CFR 1.14. This collection is estimated to take 23 minutes to complete, including gathering, preparing, and submitting the completed application data sheet form to the USPTO. Time will vary depending upon the individual case. Any comments on the amount of time you require to complete this form and/or suggestions for reducing this burden, should be sent to the Chief Information Officer, U.S. Patent and Trademark Office, U.S. Department of Commerce, P.O. Box 1450, Alexandria, VA 22313-1450. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. **SEND TO: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450.**

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The information provided by you in this form will be subject to the following routine uses:

1. The information on this form will be treated confidentially to the extent allowed under the Freedom of Information Act (5 U.S.C. 552) and the Privacy Act (5 U.S.C. 552a). Records from this system of records may be disclosed to the Department of Justice to determine whether the Freedom of Information Act requires disclosure of these records.
2. A record from this system of records may be disclosed, as a routine use, in the course of presenting evidence to a court, magistrate, or administrative tribunal, including disclosures to opposing counsel in the course of settlement negotiations.
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5. A record related to an International Application filed under the Patent Cooperation Treaty in this system of records may be disclosed, as a routine use, to the International Bureau of the World Intellectual Property Organization, pursuant to the Patent Cooperation Treaty.
6. A record in this system of records may be disclosed, as a routine use, to another federal agency for purposes of National Security review (35 U.S.C. 181) and for review pursuant to the Atomic Energy Act (42 U.S.C. 218(c)).
7. A record from this system of records may be disclosed, as a routine use, to the Administrator, General Services, or his/her designee, during an inspection of records conducted by GSA as part of that agency's responsibility to recommend improvements in records management practices and programs, under authority of 44 U.S.C. 2904 and 2906. Such disclosure shall be made in accordance with the GSA regulations governing inspection of records for this purpose, and any other relevant (i.e., GSA or Commerce) directive. Such disclosure shall not be used to make determinations about individuals.
8. A record from this system of records may be disclosed, as a routine use, to the public after either publication of the application pursuant to 35 U.S.C. 122(b) or issuance of a patent pursuant to 35 U.S.C. 151. Further, a record may be disclosed, subject to the limitations of 37 CFR 1.14, as a routine use, to the public if the record was filed in an application which became abandoned or in which the proceedings were terminated and which application is referenced by either a published application, an application open to public inspections or an issued patent.
9. A record from this system of records may be disclosed, as a routine use, to a Federal, State, or local law enforcement agency, if the USPTO becomes aware of a violation or potential violation of law or regulation.

**SYNERGISTIC COMBINATION OF**  
**3-IODO-2-PROPYNYL-BUTYLCARBAMATE AND DIAMINE**

This invention relates to combinations of biocides, the combinations having greater  
5 activity than would be observed for the individual antimicrobial compounds.

Use of combinations of at least two antimicrobial compounds can broaden potential  
markets, reduce use concentrations and costs, and reduce waste. In some cases, commercial  
antimicrobial compounds cannot provide effective control of microorganisms, even at high  
use concentrations, due to weak activity against certain types of microorganisms, or relatively  
10 slow antimicrobial action, or instability under certain conditions such as high temperature and  
high pH. Combinations of different antimicrobial compounds are sometimes used to provide  
overall control of microorganisms or to provide the same level of microbial control at lower  
use rates in a particular end use environment. Additionally, synergy has been found to be an  
unpredictable phenomenon. Often like compounds display varying synergistic profiles when  
15 combined with a particular active. It may be that no synergy is evidenced or it may be that  
synergy exists but over a different synergistic range. Because of this observation, it is  
difficult, if not impossible to draw conclusions regarding the synergistic profile of one  
compound based upon the synergistic profile of a like compound. Thus more synergistic  
combinations and their synergistic ranges must be discovered.

20 On such example of synergy is found in U.S. Pat. App. Pub. No. 2007/0078118. This  
reference discloses synergistic combinations of N-methyl-1,2-benzisothiazolin-3-one (MBIT)  
with other biocides. There still exists a need for additional combinations of antimicrobial  
compounds having enhanced activity to provide effective control of microorganisms. The  
problem addressed by this invention is to provide such combinations of antimicrobial  
25 compounds.

In the present invention there is provided a synergistic antimicrobial composition  
comprising 3-iodo-2-propynyl-butylcarbamate (also known as iodopropynyl butylcarbamate  
and IPBC) (CAS registry number is 55406-53-6 ) and N-(3-Aminopropyl)-N-  
dodecylpropane-1,3-diamine (also known as diamine) (CAS registry number is 2372-82-9).

30 The invention further provides a method of inhibiting the growth of or controlling the  
growth of microorganisms in an aqueous media, the method comprising the step of adding a  
synergistic antimicrobial composition comprising IPBC and diamine. Also, a coating, and a  
dry film containing 3-iodo-2-propynyl-butylcarbamate and diamine are provided.

The following is a detailed description of the invention.

As used herein, the following terms have the designated definitions, unless the context clearly indicates otherwise.

5 The term "antimicrobial compound" refers to a compound capable of inhibiting the growth of or controlling the growth of microorganisms; antimicrobial compounds include bactericides, bacteriostats, fungicides, fungistats, algaecides and algistats, depending on the dose level applied, system conditions and the level of microbial control desired. Such term "antimicrobial compound" as used herein is synonymous with the term "biocide".

10 The term "microorganism" includes, for example, fungi (such as yeast and mold), bacteria and algae.

The following abbreviations are used throughout the specification: ppm = parts per million by weight (weight/weight), mL = milliliter, ATCC = American Type Culture Collection, SAG = Culture Collection of Algae at Goettingen University, CCAP = Culture Collection of Algae and Protozoa, DSMZ = Deutsche Sammlung von Mikroorganismen und Zellkulturen, and MIC = minimum inhibitory concentration.

20 Unless otherwise specified, temperatures are in degrees centigrade (°C), and references to percentages are by weight (wt.%). Percentages of antimicrobial compounds in the composition of this invention are based on the total weight of active ingredients in the composition, i.e., the antimicrobial compounds themselves, exclusive of any amounts of solvents, carriers, dispersants, stabilizers or other materials which may be present.

As used herein, "IPBC" is 3-Iodo-2-propynyl-butylcarbamate (CAS registry number 55406-53-6).

As used herein, "Diamine" is N-(3-Aminopropyl)-N-dodecylpropane-1,3-diamine (CAS registry number is 2372-82-9)

25 When a ratio is the herein to be "X:1 or higher," it is meant that the ratio is Y:1, where Y is X or greater, and when a ratio is the herein to be "X:1 or lower," it is meant that the ratio is Z:1, where Z is X or less. The same logic follows for ratios that are "1:X or higher" and "1:X or lower".

30 The present invention is a composition that contains both IPBC and diamine. It has been surprisingly found that compositions that contain both IPBC and diamine are synergistically effective as biocides. In the present invention, the weight ratio of the IPBC to diamine is from 1:10 to 10:1 alternatively from 1:1 to 1:10 and further alternatively from 10:1 to 1:1.

In some embodiments of the invention, the antimicrobial combination of this invention is useful for inhibiting the growth of or controlling the growth of microorganisms in an aqueous media. Such aqueous media includes but is not limited to industrial water and water containing/contaminated media, such as cooling water, air washer, heat exchangers, boiler water, pulp and paper mill water, other industrial process water media such as: ballast water, wastewater, metalworking fluids, oil and gas, latex, paint, coatings, adhesives, inks, tape joint compounds, pigment, water-based slurries, personal care and household products such as detergent, filtration systems (including reverse osmosis and ultrafiltration systems), toilet bowl, textiles, leather and leather production system, or a system used therewith. In one embodiment the antimicrobial composition is used as an in-can preservative.

Typically, the amount of the biocide combinations of the present invention for inhibiting the growth of or controlling the growth microorganisms is from 10 ppm to 5,000 ppm active ingredient. In some embodiments of the invention, the active ingredients of the composition are present in an amount of at least 20 ppm, alternatively at least 50 ppm, alternatively at least 100 ppm, alternatively at least 150 ppm, alternatively at least 200 ppm. In some embodiments, the active ingredients of the composition are present in an amount of no more than 2,000 ppm, alternatively no more than 1,000 ppm, alternatively no more than 500 ppm, alternatively no more than 400 ppm, alternatively no more than 300 ppm, alternatively no more than 250 ppm, alternatively no more than 200 ppm, alternatively no more than 100 ppm, alternatively no more than 50 ppm. Concentrations mentioned above are in a liquid composition containing the biocide combinations.

The present invention also encompasses a method for inhibiting the growth of or controlling the growth of microorganisms in the use areas described above, especially in in-can preservative applications, by incorporating the claimed biocide combination into the materials.

The composition of the present invention contains IPBC and diamine. It is contemplated that some embodiments may contain one or more additional antimicrobial compound.

The following are examples of the present invention.

The synergism of the biocides combination of the present invention was determined using the method described by Kull, F.C., et. al in *Applied Microbiology* 9:538-541 (1961). The formula to calculate the synergy index (SI) is

$$Qa/QA + Qb/QB = SI$$

Where

QA= concentration of compound A in ppm, acting alone produced an end point (growth / no growth)

Qa = concentration of compound A in ppm, in the mixture, which produced an end point (growth / no growth)

5 QB= concentration of compound B in ppm, acting alone produced an end point (growth / no growth)

Qb= concentration of compound B in ppm, in the mixture, which produced an end point (growth / no growth)

Synergism within two biocides is demonstrated when the SI has a value less than 1.

10 The mixtures showed an additive effect if SI is equal to 1 and antagonistic if SI is greater than 1.

The Minimum Inhibitory Test (MIC) is designed to evaluate the lowest concentration of a biocide, biocide blend or biocide combination to prevent microorganisms from growing in a defined broth.

15 The MIC and synergy testing was carried out as follows:

1. The test was executed with a Hamilton MLStarPlus robot using automated turbidity reading with BioTek Synergy H4 plate reader.
2. Biocide plates were prepared in 2.2 ml deep well plates by transferring and diluting biocides from stock solutions to first row of the plates. The concentrations of biocides in stock bottles were adjusted to be 20x more concentrated than the highest desired concentration.
- 20 3. Then 15 subsequent serial dilutions with dilution factor 1.3 were performed resulting in 16 different concentrations for each system.
4. In the next step serially diluted biocide systems were transferred to the media blocks containing 850  $\mu$ l of tryptic soy broth, comprising, Casein (pancreatic digest) 17 g/L, Soya peptone (papaic digest) 3 g/L, Sodium chloride 5 g/L, Dipotassium phosphate 2.5 g/L, Dextrose 2.5 g/L (“TSB”) containing different concentrations of Mowiol 18-88 partially hydrolyzed polyvinylacetate surfactant, commercially available from Kuraray Europe GmbH in each well. In case of single biocide systems 100 $\mu$ l were transferred and in case of biocide combinations 50 $\mu$ l of each biocide dilution were transferred to the media, resulting in 950  $\mu$ l of final volume of media + biocides and 9.5 times dilution of the biocides from the biocide plate. At this point, the concentrations of all biocides in media were 1.053 x final concentration.
- 25
- 30

5. After preparation and mixing of the described systems, 3 aliquots of 190 µl were prepared in 96-well microtiter plates.
6. Preparation of the microbe suspension:

Bacterial cultures:

5      *Pseudomonas aeruginosa*      DSM # 939      ATCC# 15442  
         *Staphylococcus aureus*      DSM # 799      ATCC# 6538

10      The culture was maintained as a glycerol stock at -80°C in cryovials. A cryovial was thawed and then 100 µl spread on a TSB agar plate. After incubation for 1 day at 30°C the bacteria were harvested with buffer at pH 7.3. A total viable count on TSB plate was carried out and bacterial suspension was diluted in buffer in order to deliver ~2x10<sup>7</sup> CFU/ml.

Yeast culture:

*Candida albicans*      DSM #1386      ATCC# 10231

15      The cultures were maintained as glycerol stocks at -80°C in cryovials, are thawed and then 100 µl spread on MEA (malt extract agar) petri dishes.

         The yeast strain plates were incubated at 28°C for 1-2 days then harvested with buffer pH 5.0.

         Based on total viable count results, the inoculum was prepared.

- 20      7. Each test sample (190 µl) was inoculated with the 10 µl of microbe suspension to provide a level of ~1 x 10<sup>6</sup> CFU/ml of the bacteria species.
8. The test samples were mixed and incubated at 30°C for 2 days (48 hours) when tested against bacteria and 3 days (72 hours), respectively, when tested against yeast.



9. Growth of the micro-organisms leads to turbidity after incubation, clarity indicates no growth. Reading of the results was carried out by measuring absorbance at 600 nm for each sample at the beginning of the test ( $t_{\text{zero}}$ ) and after incubation ( $t_{\text{endpoint}}$ ).  $t_{\text{endpoint}}$  was chosen at 48 hours for bacteria and 72 hours for yeast. The difference in absorbance between  $t_{\text{endpoint}}$  and  $t_{\text{zero}}$  was used to assign a score (“1” if  $\Delta > 0.2$ , confirming growth, and “0” if  $\Delta \leq 0.2$ , confirming no growth) from which the MIC values were derived. The lowest concentration that showed no growth (score of “0”) in the broth after incubation is taken as the MIC value.

The results of single biocide and combination of two biocides tested against bacteria and yeast are presented in Tables 1 and 2.

Table 1: MIC results for single biocides and combinations of two biocides (in ppm):

Active ingredients and ratio	<i>Staphylococcus aureus</i> (DSMZ#799)	<i>Pseudomonas aeruginosa</i> (DSMZ#939)	<i>Candida albicans</i> (DSMZ#1386)
IPBC (single)	21.6	1400.0	6.6
Diamine (single)	16.7	47.8	61.5
IPBC/Diamine 1:1	5.8 : 5.8	28.3 : 28.3	2.6 : 2.6
IPBC/Diamine 1:10	1.3 : 12.9	6.2 : 62.2	1.0 : 9.8
IPBC/Diamine 10:1	9.8 : 1.0	223.1 : 22.3	5.1 : 0.5

Table 2: Calculated synergy indices for the Combinations in Table 1

Active ingredients and ratio	<i>Staphylococcus aureus</i> (DSMZ#799)	<i>Pseudomonas aeruginosa</i> (DSMZ#939)	<i>Candida albicans</i> (DSMZ#1386)
IPBC/Diamine 1:1	0.62	0.61	0.44
IPBC/Diamine 1:10	0.83	1.30	0.31
IPBC/Diamine 10:1	0.51	0.63	0.78

CLAIMS

1. A synergistic antimicrobial composition comprising 3-iodo-2-propynyl-butylcarbamate and diamine.
2. The synergistic antimicrobial composition of claim 1, wherein the weight ratio of the 3-iodo-2-propynyl-butylcarbamate to diamine is from 1:10 to 10:1.
3. A method of inhibiting the growth of or controlling the growth of microorganisms in an aqueous media.
4. The method of claim 3 wherein the aqueous media is a coating.
5. A coating composition comprising the synergistic antimicrobial composition of claim 1.
6. A coating composition comprising the synergistic antimicrobial composition of claim 2.
7. A dry film made by a process comprising applying a layer of the coating composition of claim 4 to a substrate and drying the coating composition or allowing the coating composition to dry.

**ABSTRACT OF THE DISCLOSURE**

A synergistic antimicrobial composition containing 3-iodo-2-propynyl-butylcarbamate and diamine is provided. Also, a coating, a method of inhibiting the growth of or controlling the growth of microorganisms in an aqueous media, and a dry film containing 3-iodo-2-propynyl-butylcarbamate and diamine are further provided.

## Electronic Acknowledgement Receipt

<b>EFS ID:</b>	26227801
<b>Application Number:</b>	62356910
<b>International Application Number:</b>	
<b>Confirmation Number:</b>	2139
<b>Title of Invention:</b>	SYNERGISTIC COMBINATION OF 3-IODO-2-PROPYNYL-BUTYL CARBAMATE AND DIAMINE
<b>First Named Inventor/Applicant Name:</b>	Patrick T. Felder
<b>Customer Number:</b>	21898
<b>Filer:</b>	Tifani M. Edwards/Janice Soulas
<b>Filer Authorized By:</b>	Tifani M. Edwards
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**File Listing:**

Document Number	Document Description	File Name	File Size(Bytes)/ Message Digest	Multi Part /.zip	Pages (if appl.)
1	Application Data Sheet	79470-US-PSP_20160630_TME_Application_Data_Sheet_Fillable_PDF.pdf	1822655	no	9
			3c8875c324fe1d22091cef0966e01808d4bfa593		
<b>Warnings:</b>					
<b>Information:</b>					
2		79470-US-PSP.pdf	70032	yes	8
			28182d58af2c9ac8bb5d9eb7ddcc4c39cd3fd603		
	<b>Multipart Description/PDF files in .zip description</b>				
	<b>Document Description</b>		<b>Start</b>	<b>End</b>	
	Specification		1	6	
	Claims		7	7	
	Abstract		8	8	
<b>Warnings:</b>					
<b>Information:</b>					
3	Fee Worksheet (SB06)	fee-info.pdf	30246	no	2
			1ceedacb1ef58a4651274cf1e6e94564fbaad4c		
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