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(54) Title: STERILANT COMPOSITION

(57) Abstract

A sterilant composition consists essentially of an aqueous solution of from 0.2 % to 1.0 % w/v of potassium iodide (KI) and an amount of iodine (I_2) in % w/v excess of the % w/v amount of the potassium iodide up to the maximum solubility of iodine in the potassium iodide solution. Preferably the ratio by weight of potassium iodide to iodine is 1:1.5. The composition is useful for cold sterilisation of surgical and dental instruments.

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STERILANT COMPOSITION

This invention relates to a sterilant composition which is particularly suitable for use in single-stage "cold" sterilization of instruments for use for example in surgery such as endoscopes which are heat sensitive, and dentistry, where the instruments require rapid turnover. There is a need for a simple relatively low cost sterilising fluid for use in particular in the above mentioned fields where steam sterilization may be inappropriate for a number of reasons.

Cold sterilizing procedures are of course already known. However, they have a number of disadvantages relating mainly to the nature of the chemicals used and there has been a need for some time for a cold sterilant which is non-toxic, non-carcinogenic, non-flammable and non-irritant. Such a composition should of course primarily be able to perform as an effective sterilant which means that in addition to destroying bacteria and other living life forms it must also be a rapid acting sporicide. An accepted definition of a sterilant is a solution which will reduce both the bacterial and the spore concentration by 5 log cycles. Additionally, for the sterilant composition to be useful as a cold sterilant this level of sporicidal action has to be achieved within a period of the order of thirty minutes.

From a practical point of view, also, the sterilant composition should be relatively low-cost, easily prepared and should also have a satisfactory shelf life. It should ideally be of such low cost that it can also be used in many situations as a more general application germ killer, i.e. a disinfectant.

Several attempts have been made in the past to provide a sterilant composition which meets all of the above criteria but none has been commercially successful.

An aqueous solution made from 5% iodine and 10% potassium iodide in water has been known for many decades as Lugol's Solution for use as a disinfectant and also, in well diluted form, has been prescribed for oral administration for the treatment of thyrotoxicosis. Additionally, JP-A-3 127 706 describes the use of a 0.1M iodine solution corresponding approximately to a four-fold diluted Lugol's solution (I_2 1.27% and KI 2.50%) for killing living spores. However, Lugol's solution only reduces the spore count by about 4 log cycles within thirty minutes and never kills all the spores, and after a week there is a significant reversion, i.e. appearance of additional colonies. Accordingly, Lugol's Solution does not appear to be a promising starting point for producing an effective fast acting cold sterilant composition. It has also been believed that an acidic pH, for example pH4, is desirable to enhance sporicidal activity.

Additionally, various iodophors have been described for use as disinfecting agents but none are effective as sterilizing agents. Quoting from Goodman and Gilman's "The Pharmacological Basis of Therapeutics": "An iodophor is a loose complex of elementary iodine with a carrier that serves not only to increase the solubility of iodine but also to provide a sustained-release reservoir of the element."

"The most widely used iodophor is polyvinylpyrrolidone (povidone). A 10% solution contains 1% of available iodine, but the free iodine concentration is less than 1 ppm." "Because of the low concentration the immediate bactericidal action is only moderate compared to that of iodine solutions."

It will be seen therefore that although iodophors have potential for providing elemental iodine in solution the actual concentration produced is limited so that action against living life forms is inhibited, and effectiveness against spores is of course still more limited.

GB 1,472,408 describes an iodophor solution containing polyvinyl alcohol, boric acid, iodine and potassium iodide. From a compositional point of view none of the examples use an excess of iodine over potassium iodide. Moreover, although the compositions have anti-bacterial properties, even though stated otherwise in the specification they are not effective sterilants; hat is they are not effective against spores within a short period of time.

EP 0,487,066 describes a disinfectant composition comprising a solution of hydroxypropyl cellulose which acts as an iodophor, iodine and an iodide, which may be potassium iodide. Similarly, this is a disinfectant and is not a sterilant.

US 4,526,751 describes the use of a iodophor solution of the povidone type referred to above which additionally contains potassium iodide and potassium bromide and which is a disinfectant. As in the case of other iodophor solutions it does not act as a sterilant.

JP 56,077,209 A describes an iodophor solution using an amino acid as a solubilising agent. Again this acts as a disinfectant but not as a sterilant.

However, it has surprisingly been found that selection of the proportion of the iodine to potassium iodide content in a simple aqueous solution of those two components results in a solution which is a very effective sporicide, typically reducing the spore concentration by 7 log cycles within thirty minutes, maintaining this activity down to a five-fold dilution and moreover having a good shelf life and without the

necessity of any additives such as a buffer in the solution to reduce the pH. Shelf life is particularly important as the less frequently the solution has to be prepared the more economical it becomes in terms of the cost of materials and manpower hours used in its preparation.

According to the present invention there is provided a sterilant composition which consists essentially of an aqueous solution of from 0.2% to 1.0% w/v of potassium iodide (KI) and an amount of iodine (I_2) in % w/v excess of the % w/v amount of the potassium iodide up to the maximum solubility of iodine in the potassium iodide solution.

Preferably the ratio of potassium iodide to iodine used to make the solution is substantially 1:1.5, and a packaged composition can conveniently be supplied for use in cold sterilising surgical and dental instruments comprising potassium iodide and iodine in a weight ratio of 1:1:5, either as an aqueous solution or in a form suitable for mixing with sterile water to form the sporicidal solution.

Particular advantages of the sterilant composition of the present invention are that it is simple to make, its materials are relatively inexpensive, it requires no additions such as pH buffers, it has a good shelf life, and it does not have many of the health and environmental disadvantages of prior chemical "cold" sterilants.

Specific examples of compositions within the scope of the present invention will now be described.

For the preparation of the solutions laboratory grade iodine and potassium iodide were employed. For the tests of sporicidal activity the spore suspension was "Spordex" and was a suspension of *Bacillus subtilis* spores 2.8×10^8 per ml supplied by Steris.

Where reference is made to a sodium acetate buffer solution two possible buffers are exemplified,

namely a 0.2M sodium acetate buffer (pH4.0) prepared from 0.2M sodium acetate (27.22 g/L) 180 ml plus 0.2M acetic acid (12ml/L) 820 ml and a 1.0M sodium acetate buffer (pH 4.0) prepared from 1.0M sodium acetate (136.1 g/L) 180 ml + 1.0M acetic acid (60 ml/L) 820 ml.

As a matter of procedure, the potassium iodide was first dissolved either in sterile distilled water or the buffer solution in the appropriate w/v amount and then the iodine was dissolved in the potassium iodide solution. Although the iodine dissolves readily the solution is stirred at room temperature for a minimum of two hours or overnight as appropriate. As controls corresponding solutions of potassium iodide but with no iodine were used.

For the purposes of the tests all solutions were stirred at room temperature, and all experiments were performed at room temperature. The temperature in the laboratory was recorded daily on a maximum/minimum thermometer and over the period of the tests the record shows a maximum temperature of 23.5°C and a minimum of 17°C.

For the purposes of determining sterilant activity before the beginning of each experiment a viable count of the commercial *Bacillus subtilis* spore suspension was carried out. Plates were prepared with nutrient agar. A 0.1ml sample was withdrawn and diluted in nutrient broth. A viable count by spread-plate method was carried out in duplicate using 0.1ml of 10^{-5} and 10^{-6} dilutions.

Two 0.5 ml samples of *Bacillus subtilis* spores were each mixed with 0.5 ml human serum and centrifuged at 3000 rpm for thirty minutes. The supernatants were decanted. The test samples were resuspended in 5.0ml sterilant. Viable counts at a range of dilutions were performed every ten minutes for thirty minutes. The spread plates were incubated

overnight at 37°C and the colonies were counted. A mean value of organisms/ml was calculated. The plates were then kept at room temperature for a week and a recount performed to determine if any reversion occurred after exposure to the sterilant.

A solution of 1.5% w/v iodine dissolved in 1% potassium iodide was first tested and then maintaining the original ratio of iodine to potassium iodide the sporicidal activities of solutions diluted by 50%, 80% and 90% were compared. Both the original solution and the diluted solutions were sporicidal although the 90% dilution was the least effective sporicide. The results are shown in the following Tables 1-5 and in the graphs of the accompanying Figure 1 of drawings.

The solutions maintained their effectiveness as sporicides after more than two weeks at room temperature.

Tests were also performed with solutions of the iodine and potassium iodide in 0.2M sodium acetate buffer giving pH 4.0 and also the 1M sodium acetate buffer referred to above. Generally, it was found that the buffer had little or no effect on the sporicidal activity.

TABLE 1 The effect of 0.75% w/v Iodine dissolved in 0.5% w/v Potassium iodide in water

	TEST				CONTROL			
TIME (mins)	DIL^N	COUNT		CONC/ML	DIL^N	COUNT		CONC/ ML
viable count (stock soln)	10^{-5} 10^{-6}	154 24	* 12	1.71×10^8 (3)				
0	10^{-3} 10^{-4}	392 49	361 29	3.83×10^6	10^{-4} 10^{-5}	142 22	138 15	1.63×10^7
10	10^{-1} 10^{-2} 10^{-3}	30 7 1	20 5 1	6.17×10^3				
20	10^{-1} 10^{-2}	0 3	2 0	8.00×10^2				
30	10^0 10^{-1}	0 0	0 0	0	10^{-4} 10^{-5}	180 17	179 15	1.70×10^7

* contaminated
( ) figures in brackets represent the number of counts from which the concentration has been calculated - where no figure is listed this can be taken as ALL the counts (ie. 4 or 6)

TABLE 2 The effect of 0.3% w/v Iodine dissolved in 0.2% w/v Potassium iodide in water

	TEST				CONTROL			
TIME (mins)	DIL^N	COUNT		CONC/ML	DIL^N	COUNT		CONC/ ML
viable count (stock soln)	10^{-5} 10^{-6}	154 24	* 12	1.71×10^8 (3)				
0	10^{-3} 10^{-4}	365 26	365 26	3.12×10^6	10^{-4} 10^{-5}	143 10	98 8	1.05×10^7
10	10^{-1} 10^{-2} 10^{-3}	10 0 0	5 1 0	4.17×10^2				
20	10^{-1} 10^{-2}	0 0	0 0	0				
30	10^0 10^{-1} 10^{-2}	1 0 0	0 0 0	1.67×10^0	10^{-4} 10^{-5}	131 9	121* 9	1.08×10^7

TABLE 3 The effect of 0.15% w/v Iodine dissolved in 0.1% w/v Potassium iodide in water

	TEST				CONTROL			
TIME (mins)	DIL^N	COUNT		CONC/ML	DIL^N	COUNT		CONC/ ML
viable count (stock sol^n)	10^{-5} 10^{-6}	99 25	103 23	1.71×10^8				
0	10^{-2} 10^{-3} 10^{-4}	TMC 391 43	TMC 392 32	3.83×10^6 (4)	10^{-4} 10^{-5}	168 17	146 13	1.53×10^7
10	10^{-1} 10^{-2}	647 1	528 1	5.88×10^4 (2)				
20	10^{-1} 10^{-2}	35 1	2 1	1.42×10^3				
30	10^0 10^{-1} 10^{-2}	60 24 0	57 0 1	7.63×10^2	10^{-4} 10^{-5}	156 *	153 *	1.55×10^7 (2)

* contaminated
( ) figures in brackets represent the number of counts from which the concentration has been calculated - where no figure is listed this can be taken as ALL the counts (ie. 4 or 6)

TABLE 4 The effect of 1.5% w/v Iodine dissolved in 1.0% w/v Potassium iodide in water

	TEST				CONTROL			
TIME (mins)	DIL^N	COUNT		CONC/ML	DIL^N	COUNT		CONC/ ML
viable count (stock sol^n)	10^{-5} 10^{-6}	172 13	221 19	1.78×10^8				
0	10^{-3} 10^{-4}	252 98	286 22	4.34×10^6	10^{-4} 10^{-5}	132 17	164 23	1.74×10^7
10	10^{-1} 10^{-2}	11 2	10 2	1.52×10^3				
20	10^{-1} 10^{-2}	1 0	2 0	7.50×10^1				
30	10^0 10^{-1} 10^{-2}	0 0 *	0 0 0	0	10^{-4} 10^{-5}	170 32	177 12	1.97×10^7

* contaminated

TABLE 5: Sporicidal activity of 1.5% w/v iodine dissolved in 1% w/v potassium iodide in water - two-fold, five-fold and ten-fold dilutions.

Table No.	Salt Sol^n %KI	$\%I_2$	LIQUID PHASE	pH	LOG CYCLES DECREASED IN 30 MINS	TIME TO DECREASE 5 LOG CYCLES	AMOUNT OF REVERSION*
1	0.5	0.75	water	5	7	30	not determined
2	0.2	0.3	water	5	7	20	not determined
3	0.1	0.15	water	5	5	30	++
4	1	1.5	water	5	7	20	+

* reversion (number of colonies after one week):

\+ = $< 10^2$

++ 10^3 to 10^4

CLAIMS

1. A sterilant composition which consists essentially of an aqueous solution of from 0.2% to 1.0% w/v of potassium iodide (KI) and an amount of iodine (I_2) in % w/v excess of the % w/v amount of the potassium iodide up to the maximum solubility of iodine in the potassium iodide solution.

2. A sterilant composition as claimed in claim 1 wherein the ratio of potassium iodide to iodine used to make the aqueous solution is substantially 1:1.5.

3. A sterilant composition as claimed in claim 1 of claim 2 which contains no additives.

4. A sterilant composition as claimed in claim 1 or claim 2 which contains no pH buffer.

5. A sterilant composition as claimed in claim 1 or claim 2 which additionally contains a pH buffer.

6. A sterilant composition as claimed in claim 5 wherein the buffer is a sodium acetate buffer at pH 4.0.

7. Use of a sterilant composition according to any one of claims 1 to 6 for cold sterilisation of surgical and dental instruments.

8. A packaged composition for use in cold sterilising surgical and dental instruments comprising potassium iodide and iodine in a weight ratio of 1:1.5.

AMENDED CLAIMS

[received by the International Bureau on 11 August 2000 (11.08.00);
original claims 1 and 8 amended; remaining claims unchanged (1 page)]

1. A sterilant composition which consists essentially of an aqueous solution of from 0.2% to 1.0% w/v of potassium iodide (KI) and an amount of iodine (I_2) in % w/v excess of the % w/v amount of the potassium iodide up to the maximum solubility of iodine in the potassium iodide solution, the composition containing no other iodine carrier.

2. A sterilant composition as claimed in claim 1 wherein the ratio of potassium iodide to iodine used to make the aqueous solution is substantially 1:1.5.

3. A sterilant composition as claimed in claim 1 or claim 2 which contains no additives.

4. A sterilant composition as claimed in claim 1 or claim 2 which contains no pH buffer.

5. A sterilant composition as claimed in claim 1 or claim 2 which additionally contains a pH buffer.

6. A sterilant composition as claimed in claim 5 wherein the buffer is a sodium acetate buffer at pH 4.0.

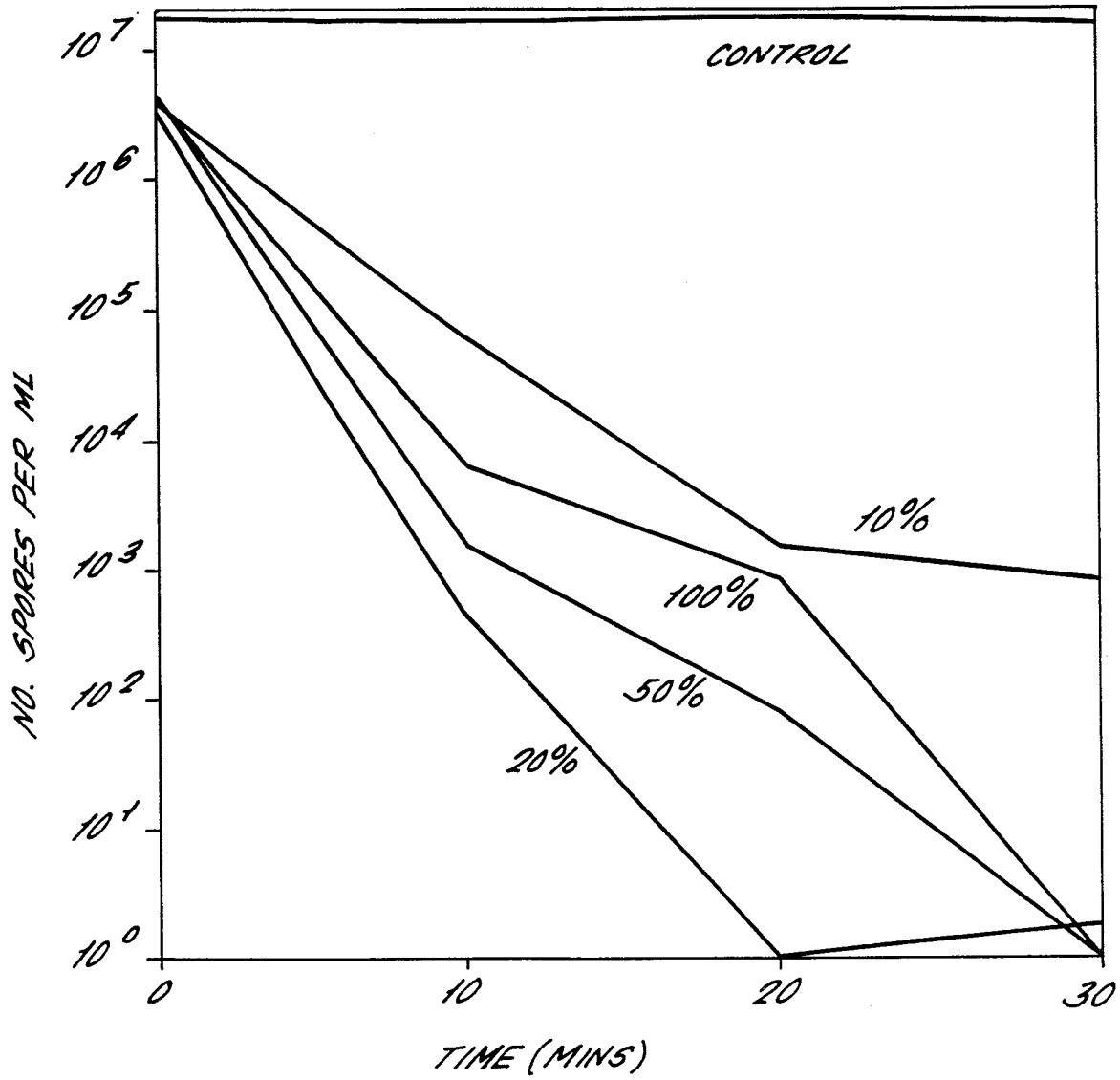
7. Use of a sterilant composition according to any one of claims 1 to 6 for cold sterilisation of surgical and dental instruments.

8. A packaged composition for use in cold sterilising surgical and dental instruments comprising an aqueous solution of potassium iodide and iodine in a weight ratio of 1:1.5, the composition containing no other iodine carrier.

STATEMENT UNDER ARTICLE 19(1)

Claims 1 and 8 have been amended so that the compositions claimed contain no iodine carrier other than potassium iodide. As is explained at page 2, line 24 to page 3, line 29 of the present application, other carriers for iodine are known, which may be referred to as iodophors. The purpose of the amendments is to exclude from the claims any compositions containing such other iodine carriers.

Claim 8 has also been amended to make it clear that only aqueous solutions are claimed.


CONTROL
10%
100%
50%
20%
NO. SPORES PER ML
10^7
10^6
10^5
10^4
10^3
10^2
10^1
10^0
0
10
20
30
TIME (MINS)

INTERNATIONAL SEARCH REPORT

Interr. ıal Application No
PCT/GB 00/00922

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 A01N59/12 //(A01N59/12,59:12)

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
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Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

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C. DOCUMENTS CONSIDERED TO BE RELEVANT

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Y	GB 2 311 222 A (KELEMEN MARY VIKTORIA ;FERGUSON ANDREW JOHN DUNCAN (GB)) 24 September 1997 (1997-09-24) the whole document --- -/---	1-7

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

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Date of the actual completion of the international search	Date of mailing of the international search report
7 June 2000	16/06/2000

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C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

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