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(54) Title: AN ANTIMICROBIAL COMPOSITION

(57) Abstract: There is disclosed an antimicrobial composition that may be used in the treatment of diseases and conditions associated with farm animals and livestock. The composition may also be utilised as an inhibitor of microbial growth on crops, plants or foodstuffs, such as grains and the like. The composition comprises an antimicrobially effective amount of clove bud oil, eucalyptus oil, lavender oil, tea tree and orange oil but may be diluted with water prior to application.



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AN ANTIMICROBIAL COMPOSITION

The present invention is concerned with an antimicrobial composition and, in particular, with an antimicrobial composition which is particularly useful in treating diseases and conditions associated with farm animals or livestock and which can also be utilised as an inhibitor of microbial growth on crops, plants or other foodstuffs.

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There are many microbial diseases or conditions which affect livestock of one type or another. Of particular importance are those conditions which can result or contribute to lameness or even death of an animal. Examples of such conditions include Contagious Pustular Dermatitis (Orf) in sheep, footrot in sheep and goats, in addition to foul of the foot and digital dermatitis in cattle. Orf is a virus which grows on the surface layers of the skin and in the mucous membrane of the mouth and nose. It penetrates through small abrasions in the skin. Even very minor grazes allow the virus to enter. It is a painful and disfiguring disease of sheep and goats and causes much distress to affected ewes and lambs which may result in death. Normally, when Orf occurs in individual animals, the entire flock should be treated.

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Footrot is also an infectious disease of livestock including sheep, goats or cattle and it is spread from animal to animal via pasture containing bacteria from the feet of infected animals. Footrot is caused by two different bacteria - *Fusobacterium necrophorum* (found wherever sheep are found) and *Bacteroides nodosus* of which there are many different strains. Some cause a virulent form of footrot whilst others

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are less invasive and are termed intermediate or benign, although the latter term is misleading because it still causes considerable pain and lameness.

5 Digital dermatitis on the other hand occurs in cattle and is thought to be caused by the bacteria *Bacteriodes* or *Treponema* but this has not yet been completely established. Other notable conditions include sandcracks and mud fever.

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Conditions of the hoof or foot of the animal are normally treated either by using antibiotics or chemical treatment baths. Orf, however, is not susceptible to antibiotics being a viral disease. A
15 number of proprietary products claim to effect a cure, but there is no evidence to support these claims. Within a few weeks the disease can in some cases clear up on its own and all that can reasonably be done is to aim to reduce the risk of secondary bacterial
20 infection with the use of topical antibiotic creams powders or aerosols. In serious cases antibiotics may be necessary. Vaccines are available but can only be used in flocks where the disease has occurred previously since it is an attenuated virus. Those that
25 are available do not confer a long lasting immunity.

As regards the diseases of the hoof of the animal, foot treatments currently being used in foot baths include various chemicals such as:

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(i) Formalin which is used in dilution of around 3% (1:30 parts water). However, this compound is painful to broken skin and stronger solutions can cause scald or severe
35 cracking of skin. The product must be fresh, as it is ineffective if there is

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organic matter in the solution.

Furthermore, it gives off toxic fumes and cannot be used indoors. It is corrosive and toxic;

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(ii) zinc sulphates which are more expensive and require a lengthy stand in time of between 30-60 minutes. The product also requires a large foot bath;

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(iii) organic acids normally diluted 1:50 parts water. They are generally of a very low pH and hence very acidic. They can be damagingly painful to the animal and corrosive to tissue. Organic acids are completely unselective in their treatment;

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(iv) copper sulphate: this is widely used and is very toxic to sheep. It stains the fleece and is corrosive to metal and poisonous to fish.

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All of the above products present serious problems of disposal and of pain for the animal. Therefore there exists a need for improved treatments for treating or alleviating many microbial diseases of livestock which are not only less expensive than those currently available but which include non-toxic ingredients which are non-painful to the animal being treated and which are safe to the environment and the user alike.

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Similarly, microbial growth is a major cause of spoilage of many stored crops and of plants causing diseases including, for example, moulds and rusts and mildew. Many of these diseases are significant in horticultural systems.

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Therefore, according to a first aspect of the invention there is provided an antimicrobial composition comprising an antimicrobially effective amount of clove bud oil, eucalyptus oil, lavender oil, tea tree oil and orange oil. The formulation has been found to be particularly effective in treating diseases associated with livestock/farm animals and especially those conditions such as Orf or ringworm and those affecting the foot or hoof of the animal. The composition has been found to be extremely effective in treating those diseases caused by a viral, fungicidal or bacterial agent.

Not only has the composition been found to be useful in treating various microbial related animal diseases, but it has also been found that the composition is versatile enough in terms of its antimicrobial action to be effective against microbial diseases in plants and yet is sensitive enough so as not to kill the plants or crop. The composition is therefore particularly broad spectrum in its mode of action and can be used for treating many types of such conditions. Livestock diseases which are most notably dealt with include Contagious Pustular Dermatitis in sheep (otherwise known as Orf) scald and fibromas, footrot in sheep, cattle and goats in addition to digital dermatitis and foul of the foot in cattle. Mud fever, especially occurring in horses is also particularly infectious, and this is also advantageously dealt with by the composition of the invention. Other diseases in crops and plants which are most notably treated include mildew, mould growth, rusts, leaf spots and such like.

The formulation is, advantageously, based on a unique blend of essential oils, all of which are safe to use

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and which can be disposed of effectively. Neither the Council of Europe's Committee of Experts on flavouring substances nor any other European Committee has recommended limits on the use of these ingredients.

5 Similarly, each of the ingredients has been evaluated by the US Flavouring Extract Manufacturers' Association (FEMA) who have classified them as GRAS (generally recognised as safe).

10 Preferably, the composition comprises approximately by volume, 16-36% clove bud oil, 34-54% eucalyptus oil, 3.5-13.5% lavender oil, 2-6% tea tree oil, and 7.5-27.5% orange oil. When the composition is for application to animals or livestock it, preferably,
15 includes a thickening agent which allows it to adhere better to the animal. Suitable thickening agents include, for example, a gelling agent, but preferably comprises lanolin or its derivatives, including lanolin oil, lanolin wax, lanolin alcohols, lanolin fatty
20 acids, isopropyl lanolate acetylated lanolin, acetylated lanolin alcohols, lanolin alcohol linoleate, and lanolin alcohol riconoleate. In an even preferred embodiment the composition is further diluted with water. Such a composition preferably
25 comprises a surfactant. In this aspect the composition comprises 5-15% clove bud oil, 10-23.5% eucalyptus oil, 1-6% lavender oil, 0.5-2.5% tea tree oil, 3-9.5% orange oil, 0.5-2.5% lanolin, 0.5-2.0% surfactant and 40-80% water. The composition may be
30 even further diluted prior to application depending on the nature and severity of the condition or disease being treated.

As aforementioned the compositions according to the
35 invention are also particularly useful in treating microbial diseases of livestock and particularly those

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of the foot or hoof of an animal, and particularly those conditions associated with a bacterial, viral or fungicidal agent. For treating these types of diseases the composition according to the invention, may be provided in the form of a foot bath. However, the composition may also be applied in any other suitable formulation, such as a cream or as a spray or aerosol for direct application to the infected area or area to be treated. The application of the composition to the animal in question can also confer a preventative or prophylactic effect as it can prevent secondary infection of conditions such as sandcracks or fissures which can often develop in livestock and which frequently become infected.

Providing the composition in the form of a spray as opposed to a foot bath when the composition is for application to a livestock animal offers significant advantages as the composition is applied directly to the affected area. As aforementioned, the properties of the composition are such that there is no pain or discomfort to the animal when the composition is applied either by means of a foot bath or a direct spray. In fact the composition when applied to the affected area advantageously confers a soothing affect to the animal.

The composition may be formulated with conventional propellants for dispersing as aerosols from conventional pressurised containers but preferably a non-propellant based system is used, such as a trigger pump spray bottle to produce an aerosol using the pump mechanism which builds the necessary pressure to produce the aerosol.

As is known, oil of eucalyptus is obtained from

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various species of eucalyptus and the resulting oils do not possess a uniform analysis. It is believed, however, that the properties of the eucalyptus oil used in the formulation according to the present invention are not dependent on a particular source of oil of eucalyptus and one may use oil derived from *Eucalyptus globulus* and/or *Eucalyptus dives*. Eucalyptus oil is rich in cineole and desirably eucalyptus oil according to the invention comprises cineole and preferably 1-8 cineole in an amount of from approximately 35 to 90% by volume.

In another aspect, there is also provided a composition according to the invention for use in controlling or alleviating microbial diseases, which in one embodiment may be associated with farm animals or livestock. Particularly preferred conditions to be treated are those associated with the hoof of ungulate animals. As aforementioned, the composition of the invention is particularly advantageous in treating bacterial, viral or fungicidal diseases or conditions, such as footrot, Orf, foul of the foot and digital dermatitis. However, it has also been surprisingly found that the composition is of broader spectrum being active against not only those diseases mentioned above and specified in the following examples, but also in treating ringworm in livestock animals such as cattle. The composition may also be adapted for internal administration to the animal for use as a wormer, for example. As aforementioned, the composition can also be used to treat for example mould in crops which are being stored, for example, in treating mildew or rust or leaf spots on plants or other similar conditions such as moulds, rusts or mildew, including white blister, powdery mildew, downy mildew, grey mould/botrytis, shothole, fungal leaf

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spots, blackspot, white rot or sooty mould.

Thus, in a further aspect there is provided a method for controlling or alleviating the presence of microorganisms at a locus, which method comprises
5 applying thereto an effective amount of a composition according to the invention. Preferably, those microorganisms are present on an animal and which are responsible for any of Contagious Pustular Dermatitis
10 (Orf), footrot, digital dermatitis, or foul of the foot, scald or ringworm in any of said ungulate animals. As aforementioned, the composition may be applied in the form of a spray or in the case of hoof associated conditions, as a foot bath as appropriate.
15 Alternatively, the microorganisms may be present on plants or stored crops or foodstuffs causing plant diseases such as moulds, rusts or mildew, including white blister, powdery mildew, downy mildew, grey mould/botrytis, shothole, fungal leaf spots,
20 blackspot, white rot or sooty mould.

The term "microorganism" as defined herein should be taken to mean any bacterial, virus, fungi or single cellular/unicellular or proteinaceous agent capable of
25 transmitting disease or symptoms thereof to an animal or mammal, a plant or foodstuff.

The present invention may be more clearly understood with reference to the following examples of the
30 invention which are provided purely by way of example only.

Example 1

35 A foot bath or spray for application to affected animals was prepared in any of 200L, 100L or 50L

batches as desired in accordance with the following composition.

5

FUNG. FOOT SPRAY/BATH - FOR SHEEP/CATTLE			
500mls Spray 5L bath			
To Make	200L 40 x 5L 400 x 500mls	100L 20 x 5L 200 x 500mls	50L 10 x 5L 100 x 500mls
Clove Bud	20L	10L	5L
Eucalyptus	33.5L	16.75L	8.4L
Lavender	6.5L	3.25L	1.6L
10 Tea Tree	3.25L	1.6L	800mls
Orange Oil	13.5L	6.75L	3.4L
Aqualose LL 100	3.25L	1.6L	800mls
Surfacare	2.5L	1.25L	630mls
15 Water	117.5L	58.8L	29.37L

1. SUMMARY

20 On 8 May 2001 (Day 0), fifty 12 week old indoor reared Suffolk cross Mule lambs were allocated to five groups of 10 animals following foot scoring for Ovine Interdigital Dermatitis (OID). Groups were treated with Barrier Foot Bath or Barrier Foot Spray while the remaining 10 lambs were left as untreated controls.

25 Feet were scored on Day 0, 2, 7, 9 and 14. Lambs were re-treated on Day 7. All treatments reduced the group foot score over the 14 day period compared to the untreated controls.

30 **2. OBJECTIVE**

To evaluate the efficacy of two Barrier Animal Health formulations (Barrier Foot Bath and Barrier Foot

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Spray) in a field situation against scald (Ovine Interdigital Dermatitis or OID) in lambs against untreated controls.

5 3. **MATERIALS AND METHODS**

3.1 **Site Details**

10 The sheep were located at Roundhay Farm, Yardley
Hastings, Northampton and were housed throughout the
trial. The building used was well ventilated and
drained and lambs were stocked at approximately 1 m sq
per lamb. They were bedded on dry straw. Proprietary
creep pellets were fed *ad lib* from purpose built creep
15 feed hoppers.

3.2 **Animals**

20 The trial was conducted on 50 Suffolk cross Mule
lambs. They were born in February and had been weaned
off their mothers at approximately 7 weeks of age.
(This was due to movement restrictions imposed by FMD)
At the start of the trial they were on average 12
weeks old and weighed in the range 30-40Kgs. The
25 lambs had been housed since birth and scald was
identified as a problem in a significant proportion of
the group prior to the start of the trial. They had
not been treated for scald in the previous 14 days.

30 All lambs were in good health, (with the exception of
lameness due to scald), throughout the trial. Each
lamb was identified with an individual numbered ear
tag and the groups were subsequently marked with
different coloured spray marker on the neck. The lambs
35 feet were cleaned and trimmed as necessary before
scoring and subsequent treatment.

3.3 Inclusion Criteria

For inclusion in the trial, a lamb had to have at least one foot affected by scald. In addition, any lambs with feet that showed signs of physical damage or footrot were excluded from the trial.

3.4 Treatments

Lambs were tagged, their feet scored (see protocol at Appendix 1) and then randomly allocated to one of the 5 groups. Each group was then allocated to a treatment, 10 animals (40 feet) per group. Treatments were applied on 8 May 2001 (Day 0) between 1.15pm and 2.15pm and again on 15 May 2001 (Day 7) between 12.30pm and 1.30pm. On each occasion feet were cleaned with water and trimmed as necessary before treatments were applied.

GROUP/COLOUR	TREATMENT	BATCH NUMBER
1 - Green	None - untreated controls	N/A
2 - Red	Barrier Foot Bath 1:200 in water	61050701 Exp 05/01/2001
3 - Blue	Formalin 5% - Strathclyde Nutrition Formaldehyde 35%; methanol 10%	107/1141
4 - Purple	Barrier Foot Spray	62050701 Exp 05/01/2002

All treatments were applied according to the manufacturers instructions. The footbath treatments were carried out in a polythene bath containing 100 litres of water, giving an average depth of 55mm to

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cover the whole foot above the coronet. Both water and product were measured accurately into the bath with calibrated containers.

5 **3.5 Weather Conditions**

The 8 May was relatively warm (15-18 Degrees centigrade) with a light breeze. There was some shade on site and the animals were not hot stressed at any
10 time. On 15 May it was cooler and there had been overnight rain.

3.6 Adverse Reactions

15 Observations after treatment confirmed there were no adverse reactions immediately after treatment in the Barrier Footbath, or Barrier Footspray nor were any reactions noted during the following 48 hours.
However, in the 5% formalin treated group, the lambs
20 with badly affected feet were in obvious discomfort afterwards. Several refused to put the bad foot down and take weight on it, most stood up against the wall with drooped heads and ears. This lasted for 20-30 minutes, after which there appeared to be no
25 persistent effect.

3.7 Animals removed from the trial

No animals were removed from the trial. Four feet of
30 score 4 had to be removed from the control group on Day 2 because they were deteriorating further and welfare considerations demanded treatment.

35 **3.8 Monitoring**

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The feet were scored 'blind' (i.e. group unknown at time of scoring) on all occasions (Day 0, Day 2, Day 7, Day 9 and Day 14).

5 3.9 Other Treatments

No other treatments were given to the lambs during the course of the trial.

10 4. RESULTS

For the purposes of analysis and discussion, the results have been collated in two ways:

15 4.1 Total Foot Scores for Each Treatment Group

Table 1 shows the total foot score by group on Day 0, 2, 7, 9 and 14. These results demonstrate that all treatments had an effect on the total foot scores of the groups.

Table 1 - TOTAL FOOT SCORE FOR EACH GROUP

	GROUP	Day 0	Day 2	Day 7	Day 9	Day 14
25	1- Control	42	45	43	37	47
	2 - Barrier Foot Bath	39	36	39	31	34
	3 - Formalin 5%	40	35	47	27	31
30	4 - Barrier Foot Spray	42	39	49	35	38

(* significant at $p \leq 0.01$)

The barrier products reduced the incidence of the condition. Formalin fared only slightly better but

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caused significant pain to the animals.

5. DISCUSSION AND CONCLUSION

5 Therefore all treatments had a positive effect on group foot score, reducing the total score compared to the controls over the trial period.

Example 2

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Trials commenced 1 November 2000

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Various grain feeds (untreated) were selected for trials which were to last for 12 weeks. We diluted the Antimicrobial Composition and sprayed the feed in exactly the same way as would have been done in their Mill. Currently AP201 ammonium propionate mould inhibitor (normally diluted 1:10) is used on many feeds. It is sprayed at the final stages using a fine mist directly to the feed as it is carried along a conveyor system for packing.

20

A total of eleven trials were carried out using different dilution rates varying from 1:10 to 1:200.

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After we applied our composition, the feed was stored in warm and moist conditions. After 12 weeks, the samples were tested for the presence of moulds and the results are shown in Table 1. In this test 11 samples were used of the feed. 5 grams of each sample were reconstituted with 10ml of sterile recovery medium. 1 gram of the resulting suspension was spread onto selective mould enhancing growth medium and incubated at 22°C for 6 days. The trials had to be carried out over a twelve week period as that is the length of time feed manufacturers would like the feed to remain

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mould free.

Plant Protection

5 The same antimicrobial composition was tested on
various moulds between 1 April 2001 and 15 July 2001.
We tested the composition - diluted 20mls in 10L water
and applied the mixture to 38 various Roses, which
were suffering with Powdery Mildew. The sites were at
10 Snetterton and Easton where we found several affected
bushes and climbers.

We were very surprised at the speed in which the
plants recovered. Six weeks after application, none
15 of the plants showed signs of Mildew.

We went on to test the same product on fruit trees
which included apple, pear, cherry and plum. The
trees were affected with Bacterial Leaf Spots and
20 Rusts. We removed the affected leaves and applied our
formula to the point of run off to all of the green
foliage. Four weeks later there was absolutely no
sign of Leaf Spots or Rust returning and all of the
trees went on to fruit very well.

25 Throughout the entire summer, many other plants
including trees, shrubs, bulbous plants, vegetables
and indoor plants were tested and on every occasion,
the formula seemed to work very well were any type of
30 fungi existed. It also seemed to prevent new fungi
developing for around 12 weeks.

Table 1

Sample	Mould c.f.u. per gram sample
A	< 1
B	< 1
C	< 1
D	< 1
E	< 1 (1:50 dilution)
F	< 1
G	< 1
H	4 (1:50 dilution)
J	< 1
K	< 1
L	20 (1:200 dilution)

CLAIMS

1. An antimicrobial composition comprising an antimicrobially effective amount of clove bud oil,
5 eucalyptus oil, lavender oil, tea tree oil, and orange oil.
2. A composition according to claim 1 comprising,
approximately, by volume, 16-36% clove bud oil, 34-54%
10 eucalyptus oil, 3.5-13.5% lavender oil, 2-6% tea tree oil and 17.5-27.5% orange oil.
3. A composition according to claim 1 or 2 further comprising a thickening agent.
15
4. A composition according to claim 3 wherein said thickening agent is lanolin.
5. A composition according to claim 4 comprising,
20 approximately, by volume, 16-36% clove bud oil, 34-54% eucalyptus oil, 3.5-13.5% lavender oil, 2-6% tea tree oil, 17.5-27.5% orange oil and 2-6% lanolin.
6. A composition according to claim 5 which is
25 further diluted with water.
7. An antimicrobial composition comprising 5-15% clove bud oil, 10-23.5% eucalyptus oil, 1-6% lavender oil, 0.5 to 2.5% of tea tree oil, 3-9.5% orange oil,
30 0.5%-2.5% lanolin, 0.5-2.0% surfactant and 40-80% water.
8. A composition according to claim 7 wherein said lanolin is Aqualose LL100.
35
9. A composition according to any preceding claim

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wherein said surfactant is Surfacare T20.

10. A composition according to any preceding claim which is diluted further prior to application.

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11. A composition according to any preceding claim for use in therapy of an animal body.

12. A composition according to any of claims 1 to 10 for use in controlling or alleviating fungal, viral or bacterial diseases.

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13. A composition according to claim 11 for use in controlling fungal, viral or bacterial diseases associated with farm animals or livestock.

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14. A composition according to claim 13 for use in controlling fungal, viral or bacterial diseases associated with the foot and hoof of said animals.

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15. A composition according to any preceding claim for use in controlling any of Contagious Pustular Dermatitis (Orf), footrot, scald, digital dermatitis, foul of the foot or ringworm.

25

16. A composition according to claim 12 for use in controlling fungal, viral or bacterial diseases of crops, plants or foodstuffs.

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17. A composition according to claim 12 for use as a mould inhibitor.

18. A composition according to any preceding claim which is formulated for application as a spray.

35

19. A method for controlling the presence of

microorganisms or disease causing organisms at a locus, which method comprises applying thereto an effective amount of a composition according to any of claims 1 to 10.

5

20. A method according to claim 19 wherein said composition is applied in the form of a spray or foot bath as appropriate to an animal for a disease or condition associated with said animal.

10

21. A method according to claim 19 or 20 wherein said microorganisms are those responsible for any of Contagious Pustular Dermatitis (Orf), footrot, scald, digital dermatitis, foul of the foot or ringworm.

15

22. Use of a composition according to any of claims 1 to 10 in the manufacture of a veterinary medicament for treating any of Contagious Pustular Dermatitis (Orf), footrot, scald, digital dermatitis, foul of the foot or ringworm.

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