

Investigation of the Antimicrobial Activity of Neem and Bush Cane Liquid Extracts On Emulsion Paint

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ABSTRACT

This research work focused on locally sourced antimicrobial emulsion paint. Two antimicrobial agents were obtained from leaves of neem (dogoyaro) and bush cane trees (opete). The leaves were then dried for about two weeks, ground into particles and sieved with 75micrones and 100microns sieves. The particles were used as fillers in polyvinyl acetate to produce the PVA- paste and allowed to dry at room temperature. The PVA-neem and PVA-bush cane composites were used with the ingredients to produce PVA-neem and PVA-bush cane paint respectively. Results indicated that both paint samples (PVA-neem and PVA-bush cane paint) are good antimicrobial paints.

Keywords; *Emulsion paint, Neem leave particles, Bush cane leaves particles, antimicrobial agents, polyvinyl acetate.*

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1.1 BACKGROUND OF STUDY

Samples of the first known paintings made between 20,000 and 25,000 years ago, survive in caves in France and Spain. Primitive painting tended to depict humans and animals, and diagrams have also been found. Early artists relied on easily available natural substances to make paint such as natural earth pigments, charcoal, berry juice, blood, lard, and milk-weed sap. Later, the ancient Chinese, Egyptians, Hebrews, Greeks, and Romans used more sophisticated materials to produced paints for limited decoration, such as painting walls. Oils were used as vanishes, pigments such as yellow and red ochres, chalk, arsenic sulfide yellow, and malachite green were mixed with binders such as gun Arabic, lime, egg albumen and beeswax.

The twenty-first century has seen the changes in paint composition and manufacture. Today, synthetic pigments and stabilizers are commonly used to mass produce uniform batches of paints. New synthetic vehicle developed from polymers such as polyurethane and styrene-butadiene emerged during the 1940s. Alkyd resins more synthesized, and they have dominated production since. Before 1930, pigment was ground with stone mills, and these were replaced by steel balls. Today sand mills and high-speed dispersion mixers are used to ground dispersible pigments. Perhaps the greatest paint-related advanced has been its proliferation.

1.2. STATEMENT OF THE RESEARCH PROBLEM

The statement of the problem is the production of epoxy paints using extracts from neem leaves (dogoyaro leaves) and Bush cane leaves (Italiu) as antimicrobial agents.

1.3 OBJECTIVES OF THE STUDY

1.3.1 The Main Objective

The main aim of this work is the investigation of antimicrobial activity of neem and bush cane extracts on emulsion paint.

1.3.2 Specific Objectives

The objectives are as follows

1. To use leaf extracts as antimicrobial agents in epoxy paints.
2. To investigate the effect of leaf particles against microbial attacks.

1.4 JUSTIFICATION OF STUDY

The epoxy paint produced from this research work will provide lasting and effective protection against harmful bacteria, mould, fungi and viruses, ultimately helping the paint to minimise staining, bad odours and material degradation on any surface it is applied to. Once applied to any surface, it will not leech, cause discoloration or affect the surface.

1.5 SCOPE OF RESEARCH WORK

The scope of this research project work is organized to cover all vital aspect of epoxy paint formulation and production using locally obtained raw materials as antimicrobial additive, which will reduce the cost of epoxy paint production and also improve the quality of the paint produced.

2.1.1 REVIEW OF NEEM LEAVES EXTRACTS

Azadirachta Indica (neem) is a multipurpose tree with multiple health benefits. Antimicrobial activity of bark extract was found to be moderate on bacteria and fungi (effective at 1000 and 2000µg/ml), whereas seed extract exhibited least antimicrobial activity. Minimum inhibitory concentration (MIC) of leaf and bark extract was found to be in the range of 500 to 2000µg/ml for all the tested microorganisms, where as the seed extract did not inhibit the microorganisms at all the concentrations tested except *Candida albicans* (1000µg /ml). Our results suggest that aqueous extracts of *Azadirachta Indica* leaf and bark exhibit high antimicrobial activity.

Drug resistance is a serious global problem, and spread of resistance poses additional challenges for clinicians and the pharmaceutical industry. Use of herbal medicines in the developed world continue to rise because they are rich source of novel drugs and their bioactive principles form the basis in medicine, nutraceuticals, pharmaceutical intermediates and lead compounds in synthetic drugs (De, 2002; and Ncube N S et al 2008). Screening medicinal plants for biologically active compounds offers clues to develop newer antimicrobial agents. These compounds after possible chemical manipulation provide new and improved drugs to treat the infectious diseases (Natarajan et al 2003, Shah et al 2006). Plant based products extracts are cheaper alternatives to the development of synthetic drugs.

Azadirachta Indica (*A. Indica*) belongs to the family Meliaceae, commonly known as neem. It is used in traditional medicine as a source of many therapeutic agents. *A. indica* (leaf, bark and seed) are known to contain antibacterial, antifungal activities against different pathogenic microorganisms and antiviral activity against vaccinia, chikungunya, measles and coxsackie B viruses (Biswas K 2002). Different parts of neem (leaf, bark and seed oil) have been shown to exhibit wide pharmacological activities including; antioxidant, antimalarial, antimutagenic, anticarcinogenic, antiinflammatory, antihyperglycaemic, antiulcer and antidiabetic properties (Talwar et al 1997). The biological activities are attributed to the presence of many bioactive compounds in different parts. Antimicrobial activity has been investigated for neem leaves, bark and seed, but there are no studies on the comparative evaluation of aqueous extract of leaves, bark and seed. Hence, the current study was designed to investigate the comparative antimicrobial activity of neem leaves, bark and seed aqueous extract against human pathogenic bacteria and fungi. A number of factors such as, thickness and uniformity of the gel, size of the inoculum, temperature and pH that affect the accuracy and reproducibility of the agar diffusion method were also taken into consideration to obtain reliable results.

2.1.5 Determination of antimicrobial activity

The aqueous extracts of leaf, bark and seed of *A. Indica* were screened for antimicrobial activity by agar well diffusion method. Agar surface was cut with the help of sterile cork borer having a diameter of 6.0 mm size. All bacterial and fungal strains were grown in nutrient broth (NB) and Sabouraud dextrose broth (SDB) for 4-6 hours at specified temperatures. The turbidity of the broth culture was adjusted to 0.5 McFarland units. This gives a suspension containing approximately $1-2 \times 10^6$ colony forming units (CFU)/ml (Mackie & Mac Cartney 1996).

An aliquot (0.02 ml) of microbial culture was added to molten MHA at 45°C and poured into the petriplate. After solidification of the agar, appropriate wells were made on agar surface by using sterile cork borer (3 wells per 90 mm diameter plate). Different concentrations of the extracts were prepared using dimethyl sulfoxide (DMSO) and 50µl of each concentration was added to the wells. Bacterial cultures were incubated at 37°C for 24 hours and fungal cultures at 25°C for 48 hours. Antimicrobial activity was determined by measuring the zone of inhibition surrounding the well. The assays were carried out under aseptic conditions. Ciprofloxacin (5µg/disc) and Amphotericin B (100µg/disc) were used as positive controls for bacteria and fungi respectively and DMSO as a negative control. Each concentration included duplicates and the results are average of two independent experiments.

2.1.6. Determination of Minimum inhibitory concentration (MIC)

The minimum inhibitory concentration (MIC) of the aqueous extracts was determined by micro broth dilution method (Andrews, JM 2001). For MIC, two-fold serial dilutions of the extracts were prepared (500, 1000 and 2000 µg/ml) in microtitre wells. Incubation of the microtitre plates was carried out at 37°C for 18-24 hours for bacteria and at 25°C for 48 hours for fungi. After incubation, microtitre wells were observed for any visible growth. The bacterial suspensions were used as positive control and extracts in broth were used as negative control. The MIC was interpreted as the lowest concentration of the extract that did not show any visible growth when compared to control tubes.

Name of the microorganism	Minimum inhibitory concentration (MIC) in µg/ml		
	Leaf	Bark	Seed
<i>Staphylococcus aureus</i>	500	1000	ND
<i>Pseudomonas aeruginosa</i>	500	500	ND
<i>Proteus mirabilis</i>	500	500	ND
<i>Enterococcus faecalis</i>	500	500	ND
<i>Aspergillus fumigatus</i>	2000	1000	ND
<i>Candida albicans</i>	1000	1000	1000

ND-- not detected

Table 2: MIC values of the *A. Indica* aqueous extracts of leaf, bark and seed

Antibiotic resistance is a major concern and development of new agents from plants could be useful in meeting the demand for new antimicrobial agents with improved safety and efficacy (Srivastava et al 2000). In this study, we have shown that the aqueous extracts of neem leaf exhibited highest antimicrobial activity compared with the bark and seed. The difference in the antimicrobial efficacy could be due to variable distribution of phytochemical compounds in different parts. Margolone, margolonone and isomargolonone are tricyclic diterpenoids isolated from stem bark are shown to exhibit antibacterial activity (Pennington et al 1981). Nimbidin and nimbolide from seed oil show antifungal, antimalarial and antibacterial activity including inhibition of *Mycobacterium tuberculosis* (Rojanpo et al 1985, Khalid et al 1989). However presence of high concentrations of azadirachtins, quercetin and β sitosterol in *A. Indica* leaves might be responsible for strong antibacterial and antifungal activity compared with bark and seed (Subapriya R. Nagini S 2005).

Although crude extracts from various parts of neem have medicinal applications from time immemorial, very little work has been done on the biological activity and plausible medicinal applications of isolated compounds. Hence drug-development programmes could be undertaken to investigate the bioactivity, mechanism of action, pharmacokinetics and toxicity of compounds isolated from neem plant. Newer antimicrobials from plant extracts could also be useful in food, dairy and pharmaceutical industries to prevent contamination by limiting the microbial growth. The tests performed in the current study, compared the antimicrobial efficacy of aqueous extracts of neem leaf, bark and seed which showed high, moderate and low antimicrobial activities respectively.

2.2.1 REVIEW OF BUSH CANE (OPETE) LEAVES EXTRACTS AS ANTIMICROBIAL ADDITIVES

Costus afer, of the family Costaceae, a perennial rhizomatous herb, is commonly called "spiral ginger", "ginger lily" or "bush cane"⁸, "eti" by the Isokos and Urhobos and "monkey sugarcane" in Warri and most parts of Delta State, Nigeria. Most rural dwellers use this medicinal plant to treat upper respiratory tract and gastrointestinal infections⁹, gonorrhoea¹⁰ and syphilis¹¹. *Costus afer* is rich in phytochemical constituents. The leaves have been shown to contain an abundance of alkaloids and flavonoids and trace amounts of saponins, tannins and glycosides¹¹. Flavonoids and alkaloids have been reported to be antibacterial, antiviral, anti-inflammatory and antineoplastic¹².

2.2: Summary of analysis of variance of fresh juices on isolates

Bacterial Isolate	Sum of squares	Degrees of Freedom	Mean	F-Statistics	Error	Significance (P-value)
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<i>K. varians</i>	4309.91	6	718.32	853.84	0.84	<0.001
<i>S. pyogenes</i>	3041.84	6	506.97	128.53	3.94	<0.001
<i>K. pneumoniae</i>	4283.14	6	713.86	293.94	2.43	<0.001
<i>E. aerogenes</i>	4062.57	6	677.10	424.45	1.60	<0.001
<i>S. enterica</i>	3976.00	6	662.67	568.00	1.17	<0.001

This study revealed that no zone of inhibition (between $6.0\pm 1.0\text{mm}$ and $22.0\pm 1.0\text{mm}$) of the concentrated extracts (Table 3) of *A. cordifolia* compared with the control. However, they were more reactive against some of the isolates than the fresh juices. The highest susceptibility was shown by *E. aerogenes* to the extract from the leaves at a concentration of 575.0mg/ml while *K. varians* was least susceptible to the leaves extract at a concentration of 128.5mg/ml. Susceptibility of *K. varians* to all but the stem pith extracts at 134.0mg/ml was observed, but ANOVA (Table 4) shows that there is no significant difference in the zones of inhibition obtained ($P>0.05$). On the other hand, a significant difference in the zones of inhibition given by the other extracts was observed ($P<0.05$).

Bacteria	<i>C. afer</i> stem juice	<i>C. afer</i> leaves	<i>A. cordifolia</i> pith	<i>A. cordifolia</i> pith and <i>C. afer</i> stem juice	<i>A. cordifolia</i> leaves	<i>A. cordifolia</i> leaves and <i>C. afer</i> leaves	Gentamycin Control (35mg/ml)
<i>K. varians</i>	NI	11.0 ± 1.0	$39.3\pm 2.5^*$	9.0 ± 2.0	11.0 ± 2.0	13.0 ± 1.0	39.3 ± 0.6
<i>S. pyogenes</i>	9.3 ± 1.5	9.0 ± 2.0	29.7 ± 2.1	7.0 ± 2.0	9.0 ± 2.0	7.3 ± 2.5	39.0 ± 1.0
<i>K. pneumoniae</i>	8.7 ± 1.5	NI	30.3 ± 2.0	6.7 ± 1.5	10.6 ± 2.5	7.6 ± 1.6	40.0 ± 1.0
<i>E. aerogenes</i>	NI	NI	25.0 ± 2.0	NI	9.3 ± 2.5	NI	37.0 ± 1.0
<i>S. enterica</i>	NI	NI	$33.7\pm 2.5^*$	NI	NI	NI	28.0 ± 1.7

Bacteria	<i>C. afer</i> stem juice	<i>C. afer</i> leaves	<i>A. cordifolia</i> pith	<i>A. cordifolia</i> pith and <i>C. afer</i> stem juice	<i>A. cordifolia</i> leaves	<i>A. cordifolia</i> leaves and <i>C. afer</i> leaves	Gentamycin Control (35mg/ml)
<i>K. varians</i>	NI	11.0 ± 1.0	$39.3\pm 2.5^*$	9.0 ± 2.0	11.0 ± 2.0	13.0 ± 1.0	39.3 ± 0.6
<i>S. pyogenes</i>	9.3 ± 1.5	9.0 ± 2.0	29.7 ± 2.1	7.0 ± 2.0	9.0 ± 2.0	7.3 ± 2.5	39.0 ± 1.0
<i>K. pneumoniae</i>	8.7 ± 1.5	NI	30.3 ± 2.0	6.7 ± 1.5	10.6 ± 2.5	7.6 ± 1.6	40.0 ± 1.0
<i>E. aerogenes</i>	NI	NI	25.0 ± 2.0	NI	9.3 ± 2.5	NI	37.0 ± 1.0
<i>S. enterica</i>	NI	NI	$33.7\pm 2.5^*$	NI	NI	NI	28.0 ± 1.7

Table 4: Summary of analysis of variance of concentrated leaves extracts on isolates

E. aerogenes ($22.0\pm 1.0\text{mm}$) was inhibited only at a concentration of 575.0mg/ml by the extract from the leaves. An irregular result was observed with the pith extract where at 536.0mg/ml there was resistance but at 268.0mg/ml ($12.5\pm 1.7\text{mm}$) and 134.0mg/ml ($5.5\pm 1.0\text{mm}$) it was susceptible.

S. enterica showed *in vitro* susceptibility to the extract from the leaves at all concentrations, but the stem pith extracted inhibited the bacteria only at a concentration of 536.0mg/ml ($8.5\pm 2.0\text{mm}$). None of the bacterial isolates tested was susceptible to the concentrated leaves and stem juices of *C. afer*.

2.3. HISTORICAL BACKGROUND OF PAINT AND COATINGS

The oldest evidence of painting was left by primitive peoples. Cave dwellers and hunters left paintings of the animals that they hunted. Paintings in caves have survived because of their protected locations. Most cave paintings used the colors black, red, and yellow. Chemical analysis of these early paintings has shown that the main pigments used were iron and manganese oxides. To form applicable paints, the pigments were possibly mixed with egg white, animal fats, plant sap, or water. The resulting mixture of binder-containing pigments could then be applied to cave walls. During the period 3000–600 BC, many paint-making advances were made by the Egyptians. They not only developed pigments with a wider range of colors but are also credited with producing the first synthetic pigment (Egyptian Blue) and developing the first lake pigments. Preservative paints and varnishes were also used during this time. Drying oils as part of varnishes were used during the period 600 BC–400 AD by the Greeks and Romans. In the tenth century AD, Theophilus describes a varnish made by

heating amber resin with linseed oil. Varnish was used to protect painting on wood during the Middle Ages. Pigments were suspended in a varnish like the one described by Theophilus in order to make a more durable paint. For hundreds of years, paint formulations were handed down from one generation to the next and were often carefully guarded. Paints were produced in small batches, with the procedure being a relatively expensive one and the product not affordable to many. However, the demand for paint and coatings became great enough that by the late eighteenth and early nineteenth century it became profitable to make paint for wider consumption. The first paint and varnish factories were established during the nineteenth century. The industrial revolution and the mass production of the automobile strongly influenced the growth of the paint and coatings industry. The need for anti-corrosive coatings as well as other special-purpose coatings helped to accelerate the rate of scientific discovery. Titanium dioxide, the white pigment that would replace white lead, was introduced in 1918. After the middle of the twentieth century, the natural oils that had been used in paint formulations were replaced by synthetic resins. Today's coatings manufacturers offer a wide variety of products to protect, decorate, and perform special functions on the surfaces of products ranging from children's toys to spacecraft. In the later part of the twentieth century, society's growing environmental awareness has presented a new challenge to the paint and coatings industry to produce coating products that meet the demands of manufacturers and consumers and at the same time comply with the government environmental constraints. Certain chemicals have been shown to be toxic and hazardous to humans and/or their environment. Regulatory agencies are setting strict standards with which coatings manufacturers need to comply. This has led to a greater interest in developing coatings such as those that use water instead of volatile organic compounds in their formulation and powdered coatings that are absolutely solvent-free.

2.4. CLASSIFICATION OF PAINTS

Currently there is a wide range of paints available on the market, knowledge on the basic characteristics and properties of each family of paints, allow us to make the optimal decision in the selection of paints based on technical and economic requirements required by the job. Paints can be classified into different types on the basis of:

- Their Function
- Their Application And Uses
- The number of coats applied in the paint system
- The degree of emission of volatile compounds:
- The chemical backbone resin that paint is composed
- Appearance

2.4.1. Classification based on their functions:

Primers: The primers are the first layers of paint applied on the surface, the primers are designed and formulated to protect the surface against oxidation and corrosion and to be the basis for a good anchorage and adhesion for subsequent layers of paint.

Sealants: Sealants are the layers of paint that are located between the primers and topcoats, are generally used when apply putty, to seal and isolate putty to finish coat as well as improve adherence and support for the following layers of paint.

Topcoats: Topcoats refer to the entire set of paints that are used to colour the surface, are paints which are to be resistant to abrasion, ultraviolet light, chemicals, moisture, etc. ... due that kind of paints are in direct contact with the outside.

2.4.2 Classification Based On Application and Uses:

The paints are designed according to the sector which will go, for example primers used in the automotive sector are totally different from the primers used in the manufacture of ships, due to the different functional requirements (the paints of ships must be very resistant to humid environments and extremely saline) and the different materials on which the paint is applied (cars widely used aluminium and plastics of different compositions, while ship used primarily steel). Based on their areas of applications and uses, paints can be classified as

- Paints for the automotive industry
- Paints for general industry
- Paints for the construction
- Decorative paints (home)

2.4.3 Classification Based On the number of coats applied in the paint system:

Direct paint: Direct paint are those paints that are applied directly on the material or substrate, these paints offer some resistance to both oxidation and ultraviolet radiation and other external agents, providing directly the colour, gloss and aesthetic finish.

Monolayer Paint: Monolayers paints are known to the classical application of 2 coats of paint, primer plus enamel, the primer layer protects the material from oxidation and corrosion and promotes adhesion of the next layer of paint, the top layer commonly called enamel or direct gloss is provided by the colour, gloss and resistance to light and environmental agents.

Bilayer: Bilayers correspond to the paint coating system consisting of three layers, primer plus basecoat plus varnish, in this case the final finish is achieved by means of two different layers, basecoat is the first layer that provides colour and metallic effect, final transparent varnish layer provides gloss and protection against external agents.

Trilayer:

Finally we found trilayers paints, in which the paint application system consists of 4 layers, the first coat of primer and the last 3 layers correspond to the finish, with the latter three layers is achieved pearlescent or chameleon effects (colour change depending on the light incidence and the angle at which we see), this type of paints are used primarily in tuning automotive sector.

2.4.4 Classification Based On the degree of emission of volatile compounds:

For ecological reasons and job security, it has developed new ranges of paint that are designed to reduce the amount of solvents that are emitted and used during mixing, application and curing paint, due it produces a source of emission of volatile organic compounds (VOC's) harmful to both humans and the environment.

Powder paints are more environmentally friendly due it does not require or contain any concentration of solvents, water-based paints contain a tiny concentration of solvent which is negligible, follow the high-solids content paints which require less amount of diluent to the solvent based paints. Examples of such paints are

- Powder paint
- Water-based paint
- Paint with high solids content
- Solvent-based paint

2.4.5 Classification Based On the chemical backbone resin that paint is composed:

Due to the chemistry of the base resin or polymer base, each type of paint provides qualities and characteristics which can be improved by the addition of the fillers and additives, such as silicone-based paint are paints that repel water and facilitate cleaning of graffiti, silicate based paints are highly resistant to temperature are thus resins used in anti-heats paints. examples include;

- Epoxy paints
- Polyurethane Paints
- Acrylic Paints
- Alkyl paints
- Polyester Paint
- Vinyl paints
- Rubber paints
- Silicate paints
- Silicone paints Etc ...

2.4.6. Based on Appearance

- Eggshell
- Multicolored
- Matt
- Iridescent Texture
- Satin Finish
- Wrinkle Finish
- Semi-Gloss
- Luminous
- Fluorescent
- Gloss
- Crackle Finish

- Flat

2.5 TYPES OF PAINT

Paint is a fluid, or semi-fluid material which may be applied to surfaces in relatively thin layers, and which changes to a solid coating with time. The change to solid material may or may not be reversible, and many occur by evaporation of solvent or chemical reaction, or by a combination of the two. There are many types of paint, which include oil based paints (gloss paints), emulsion paints (water based paint), textured paints (textcote), cellulose paints, bituminous paints and rubber-based paints (latex paint).

2.5.1. Gloss paints (oil based paints): These are paints that may be classified according to whether the drying mechanism is predominantly solvent evaporation, oxidation or some chemical reaction. Gloss paints which dry essentially by solvent evaporation, rely on a fairly hard resin as the vehicle. Paints which dry by oxidation, the vehicle is usually an oil or an oil-based varnish, these usually contain driers to accelerate the drying of the oil. Paint based essentially on oil with suitable pigment such as titanium dioxide, extenders, and usually zinc-oxide and white lead, are conventional outside house paint because these materials give the combination of properties which meet this requirement. Oil paint is a type of slow drying paint that consists of particles of pigments suspended in a drying oil, commonly linseed oil. The viscosity of the paint may be modified by the addition of a solvent such as turpentine or white spirit, and varnish may be added to increase the glossiness of the dried oil paint film. It is the oldest and most traditional of the types of paint, generally suitable for all surfaces, but not the most economical for all occasions. Oil paints have been used in Europe since the 12th century for simple decoration but were not widely adopted as an artistic medium until the early 15th century. Common modern applications of oil paint are in finishing and protection of wood in buildings and exposed metal structures such as ships and bridges. Its hard-wearing properties and luminous colour make it desirable for both interior and exterior use on wood and metal. Due to its slow drying properties, it has recently been used in paint-on-glass animation. Thickness of coat has considerable bearing on time required for drying. For those instances when oil based paint would traditionally be preferable, but you desire a water based product, a number of companies have introduced “waterborne enamels” or “waterborne alkyds. “These paints look and behave much like oil-based options because they have good leveling qualities for a smooth finish.

ADVANTAGES OF OIL BASED PAINTS

- Attractive gloss
- Good “leveling” (brush strokes fill themselves in to create a smooth finish)
- Hard durable finish

2.5.2 Emulsion Paints (Water Based Paint)

These are paints with water-soluble vehicle and they include, calcimines, in which the vehicle is glue and casein in paints, in which the vehicle is casein or soya-bean protein.

This project research study is directed towards producing and formulating of emulsion paint (water – thinned paint) from local pigments and extenders as raw materials. The high demand for emulsion paint for protective and decorative purposes has encouraged the development of different equipments for the manufacturing operation.

This piece of research work is due to reducing the high cost of emulsion paint formulation and production, because of the imported raw materials. (E.g. Titanium dioxide), and thereby disclosing a local raw material from our natural domain which could also be used for the same purpose. An example of this locally obtained raw material for emulsion paint production is calcium carbonate in the form of calcite and dolomite

The majority of wall paint sold today is water-based, but oil-based paint remains popular for glossy woodwork, doors, and furniture, as well as demanding surfaces such as floors.

Be cautious when switching to a water-based paint if the surface has previously been coated with an oil based product, as the new paint may not stick. In this situation, Sherwin-Williams recommends washing the surface and then roughening it all over with a medium to smooth grit sandpaper – making it clean, dry, and dull in order to prevent peeling of the new coat.

3.1 Methodology

3.1.1 Materials

The materials/solvents used in this study include:

1. Calcium carbonate (CaCO₃)
2. Nitrosol
3. Ammonia

4. Polyvinyl acetate (P.V.A)
5. Titanium oxide (TiO₄)
6. Water
7. Kaolin
8. Hydrosol
9. Genipore

3.1.2 Apparatus

The instruments used include;

1. Reaction pot (20 litres)
2. Electronic weighting balance
3. Measuring bowl
- 7 Beaker (500ml)
- 8 Stirrer
- 9 Measuring cylinder
- 10 Conical flask (250ml)
- 11 Ceiling board
- 12 Stop watch

3.2. Methods

In this study, the formulation used for the production of antimicrobial emulsion paint at molecular level is represented in table 3.1 and 3.2

Table 3.1: formulation for the production of emulsion paint using neem leave extract as antimicrobial agent.

FORMULATION 1

CHEMICALS	WEIGHT (KG)
P.V.A	0.25
CALCIUM CABORNATE	4
NITROSOL	0.05
AMMONIA	0.05
ACRYLIC	0.25
TITANIUM OXIDE	0.1
FORMALINE	0.05
WATER	4
NEEM LEAVE EXTRACT	0.45

Table 3.2: formulation for the production of emulsion paint using bush cane leave extract as antimicrobial agent.

CHEMICALS	WEIGHT (KG)
P.V.A	0.25
CALCIUM CABORNATE	4.00
NITROSOL	0.05
AMMONIA	0.05
ACRYLIC	0.25
TITANIUM OXIDE	0.1
FORMALINE	0.05
WATER	4LITRES
BUSH CANE LEAVE EXTRACT	0.45

3.2.2 PAINT PRODUCTION PROCEDURE

Four litres of water was poured into a big bucket, previously dissolved titanium was added and stirred. This was followed by addition of calcium carbonate and the mixture was stirred for about 2 minutes with a motorized mixer for homogeneity. The diluted nitrosol was added, followed by ammonia and stirred. PVA paste was added to the several mixture and another liquid neem and bush cane added directly in a separate mixture.

3.3. TESTING

The standard drying (ASTM D711), viscosity(), cross-cut test, bending test, ash test and antimicrobial test were carried out and illustrated below.

3.3.1 Drying Test (ASTM D711)

25ml of the paint produced was painted on a ceiling board initially primed with white paint and allowed to dry at room temperature by taking the drying time using a stopwatch.

3.3.3 Cross-cut test (ISO 2409)

The resistance of the paint coatings to separation from substrates when the right angle lattice patterns were cut into the coatings was assessed based on ISO 2409 method.

3.3.4 Bend Test

The paints were applied on plastic slides of 10 cm x 4 cm x 1 cm with 3l painting brush and then allowed to dry at room temperature for 15 days before testing the bending ability of the different paint samples through an angle of 360°.

3.3.6 Ash Test (ASTM D2584)

2g of the paint samples were weighed into dried / pre-weighed porcelain crucibles in triplicate; this was heated in a muffle furnace at 700 OC for 1 hour. The crucibles were weighed after it has been cooled to room temperature in a desiccator. Ash residue remaining in the crucible was considered filler. The percentage ash was calculated using the following *expression*.

$$\text{Ash\%} = \frac{\text{weight of residue}}{\text{sample weight}} \times 100$$

3.3.7 Antimicrobial Test ()

Each sample of the three samples (Paint containing bush cane leave particles, paint containing neem leave particles, and commercial paint) was coated on a filter paper and dried for 24 hours. The coated filter papers were cut to 2 inch diameter. A set of each sample was leached for 24 hours before Antimicrobial testing was performed.

A 0.1ml of bacterial suspension was spread over a petri-dish containing solidified trypticase soy agar (TSA). Each coated sample was placed into the center of the plate and incubated for 24 hours at 30°C. During incubation, the bacteria grow and reproduce creating a mat of colonies completely covering the media's surface except for the vicinity of the painted sample where the Antimicrobial paint is and the growth of bacterial. After incubation, the samples with the zones of inhibition were recorded.

4.1 RESULTS AND DISCUSSION

4.1.1 Drying Time Results

The drying time results were tabulated as shown in Table 4.1. Results indicated that the drying time was lower than the control, indicating fast ability of the fillers (neem and bush cane).

Table 4.1: Drying time results of samples A,B and C

S/No	Paint Samples	Time (Minutes)
1.	A (neem paint)	20 - 25
2.	B (bush cane paint)	20 - 25
3	C (Commercial paint)	20 - 30

4.1.1. Ash test result

From the results obtained, it shows that the commercial paint had filler content of 69.25percent while the paint containing neem particles had 73.43percent filler content and the paint containing bush cane leave particles had filler content of 75.27percent. It shows that the paint containing bush cane leave particles had more toughness followed by the paint containing neem leave particles and then the commercial paint.

4.1.3. Antimicrobial test

S/N	SAMPLE	RESULT
1	Sample A (Neem paint)	70% Inhibition
2	Sample B (Bush cane paint)	80% Inhibition
3	Sample C (Commercial Paint)	20% Inhibition

4.2 DISCUSSION

Modification of emulsion paint by incorporating extracts from neem leaves and bush cane leaves improved the antimicrobial activities of the paint inhibiting the growth of most microbes that grow on walls of painted buildings. This paint solved the problem of microbial attacks on painted walls at the cheapest rate as the antimicrobial agents were sourced locally.

5.1 CONCLUSION

The process involved in paint production, qualities and performances of emulsion paint in particular are largely dependent on the properties of its constituents and the ratios of these constitutions include pigments, pigment extenders, additives and vehicles. Failure to make proper formation before production causes deterioration. Emulsion paint production from available materials must meet the present need for a high standard of performance so as to cater satisfactorily for the needs of the society, at a particular point in time and to demonstrate a credible record of an ability to challenge external and future influences.

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