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Antimicrobial Effect on Different Dyes and Inks

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ABSTRACT

The present study has been undertaken to find out the antimicrobial activity of methanol base ink. The antimicrobial activity was screened against ecoli, pseudomonas and staphylococcus. The dilutions of inks were prepared with different concentration, tested with respect to the bacterial strains and their inhibition was calculated. The methanolic extract of the ink of CAMLIN ink especially white board marker ink and parker gel pen ink with its blue and black colour respectively were prepared and the antimicrobial activity was determined by agar well diffusion method on different bacterial strains. It is been observed that with 10% dilution of samples Pseudomonas shows the maximum degradation of ink, in 20% dilution of samples staphylococcus and pseudomonas both shows the approximate equal degradation of ink, in 30% dilution of samples Pseudomonas shows maximum degradation, in 40% dilution of samples almost all bacteria shows the equally degradation in their mean positions, and in 50% dilution of sample only staphylococcus shows the degradation. Hence, it is very important to use the inks which show null or less sensitivity towards the microbes during the preparation of any important documents.

KEYWORDS: *Agar media; EColi; Pseudomonas; Staphylococcus; Inhibition Zone*

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INTRODUCTION

In the world of the business and literacy most of the transaction takes place through the documents. Documents place very important role in the today's society and roughly 50 percent cases related to the disputed documents. Documents denotes any matter expressed or described upon any surfaces by means of letter, figures, or marks or by more than one of these means; intended to be used, or which may be used, as a evidence of that matter.[1] Hence, it is important to determine which ink was used in writing of that document and help in roughly estimation of the age of that document. Examination of the writing material is one of the oldest branches of the forensic science. Analysis the age of the ink is one of the most challenging tasks which were done during the examination of the ink. There are many methods due to which age estimation of paper should be done. It may be roughly estimated from its condition, watermarks, composition etc.[2] The use of pen could be also help in finding the age of the documents in some rare cases. The age of the writing can be determined from the age of the ink. The changes in the colour of the ink due to the oxidation and the chloride or sulphate diffusion can give an approximated age of the writing.[3]The bacterial effect on the ink was one of the most helpful methods to determine the age of the writing. Ink analysis is also important for testing the paper whether it is belong to that time or not which it tried to be called. Like all ink have some properties for the resistance of particular microbe for example the markers used during the surgery having the antimicrobial activity and many more inks were made which having the antimicrobial activity. The microbes are the main reasons which degraded the paper faster and damaged the documents.[4] History of ink as old as pen or may be even more older. Chinese knew about ink in 23rd Century B.C. and made plants, animals, and mineral ink used in the paintings on silk and the paper. The best ink which are made by them are of pine sap, about 100 year old; also made ink from mixture of hide glue, carbon black, lampblack, and bone black pigments.[5] In India, about 4th B.C., ink was discovered and named it "Masi" made up from the burnt bones, tar and pitch. Greek and Romans made ink from soot, glue and water.[6]

Papyrus was first prepared writing material dating back to 3500 BC. In ancient time the plant named "Reddy Plant" or "Papyrus Plant" grew abundant in Egypt. Egyptian papyruses were produced in large stalks and were cut into the pieces about 2 feet long and split from the centre. Then it was covered by the wheat flour and water and after that it was used for the writing surfaces. During 2nd century, Egyptian rulers were not allowed papyrus to be exploitive due to high demand. It was believed that substances are precious enough to develop rivalry other country to Egypt. The king of Pergamum, the various ancient city of Asia is credited to invention of the parchment from 197-159 BC. Parchment was originally made from the split skin of the sheep. The grain half or wool side of the skin were made into strong leather while flesh part or lining side were used to make parchment.[7,8] Paper making was introduced in Spain around the year of 1150, spread to Italy, France, Germany

and England by 15th century. The first American paper was established by William Rittenhouse in 1690. Europe used same principle as Chinese for paper making substituting the metal wires for bamboo. Modern paper defined as flatted sheet usually vegetable fibers lay down on the fine screen from water suspension. Cellulose fibers are found most plants which are one major component of paper.

Methodology

The main aim of this study is to check the microbial effect on the ink or dyes. The material which is required to done this study is different inks which are diluted in the ethanol. After that for study of the bacterial effect, incubation of bacteria was done. The first step is to prepare the culture media in which the incubation was done.

1. Preparation of Culture Media (NUTRIENT AGAR):

Nutrient agar is generally used for the cultivation of the microbes. It is popular because it can grow a variety of bacteria and fungus in it and contain many nutrients which is essential in bacterial growth.

1.1. Preparation:

A. For preparing the nutrient agar, the requirements are 0.5% Peptone, 0.3% beef extract, 1.5% agar,0.5% NaCl and Distilled water
B. Mix all the components in the distilled water and stirred properly
C. Autoclave the dissolve mixture at 121 degree Celsius for 15-20 minutes
D. Once the nutrient agar has been autoclaved, allow it to cool but not solidify.
E. Pour the nutrient agar into each plates and that done in the sterile condition in the laminar and leave the plates until the agar has solidified

2. Bacterial Inoculation:

There are many methods of inoculation used in bacteriology. But in this experiment, STREAK PLATING TECHNIQUE was used. The purpose of this procedure is obtained well isolated colonies from a specimen.

- Inoculated the specimen on the agar media with the help of the sterile inoculation loop and then spread the specimen on the culture media in required portion.
- Streak the plate with the help of the streaking method, place the lid and incubated the streaked plate at the required temperature in an inverted position to prevent condensation.

3. Agar Well Preparation:

- Place swab into the broth culture, gently remove the excess liquid by pressing and then streak the agar plate to form a bacterial lawn.
- For obtaining the uniform growth, streaking should be done in one direction, rotate the plate 90 degree and streak the plate again in that direction. Repeat this atleast 3 - 4 times.
- Allow the plate for dry for approx 5 to 10 minutes and then make the well with the help of the flame sterilised forceps and then seal the well with the help of the agar
- Place the diluted ink in the wells and allow plates to be incubated overnight at an incubation temperature of 37 degree Celsius.

4. Ink Dilution Preparation:

During this experiment, 5 different kind of inks i.e. green colour marker ink, blue colour marker ink , black colour marker ink, blue colour gel pen ink and black colour gel pen ink. Then we dilute these inks in the methanol and prepare 10%, 20%, 30%, 40%, and 50% dilution.

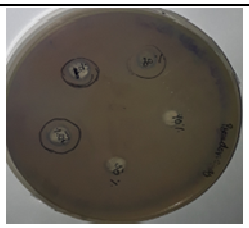
Table No. 1: - Depicting the ink samples with their dilutions

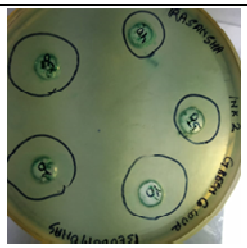
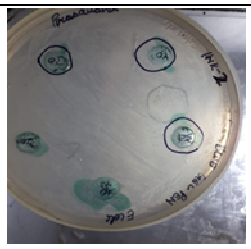
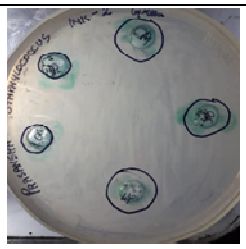
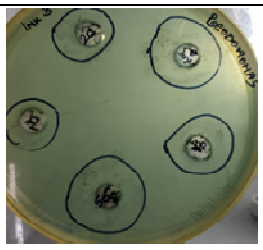
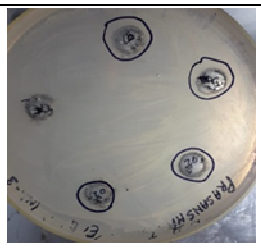
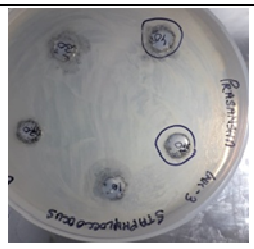
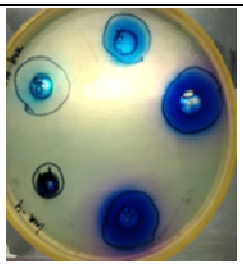
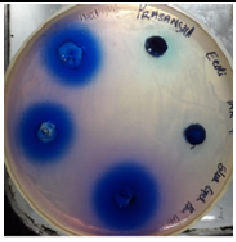
Serial No.	Ink sample	Dilutions of Ink at 10%, 20%, 30%, 40% and 50%
1.		
2.		

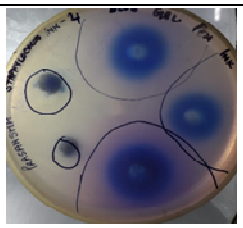
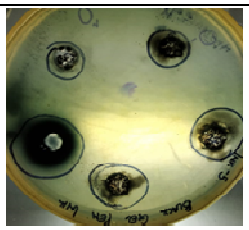
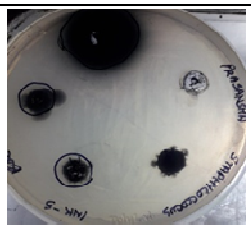
3.		
4.		
5.		

Result

Table 2: Depicting the microbial activity of ink at different dilutions

S. No.	Sample	Bacteria	Dilutions 10 %	20 %	30 %	40 %	50 %	Inhibition zone (IZ in cm)
1.	**Blue Marker Ink**	**Pseudomonas**	1.5	1.4	1.3	-	-	
		Ecoli	1.2	1.3	1.4	1.4	1.45	
		Staphylococcus	1.3	1.4	1.5	1.6	2.0	

		Pseudomonas	2.4	2.2	2.0	1.4	1.5	
2.	**Green Marker Ink**	**Ecoli**	0.3	0.5	0.7	1.2	1.4	
		Staphylococcus	0.2	0.5	0.7	0.9	1.2	
		Pseudomonas	1.7	2.0	2.4	2.2	2.5	
3.	**Black Marker Ink**	**Ecoli**	0.8	0.7	0.5	0.3	-	
		Staphylococcus	-	0.2	0.6	0.8	1.0	
		Pseudomonas	2.4	1.8	2.0	1.6	1.0	
4.	**Blue Gel pen Ink**	**Ecoli**	3.0	2.5	2.0	-	-	

		Staphylococcus	0.5	1.5	1.8	2.2	2.4	
5.	**Black Gel pen Ink**	**Pseudomonas**	1.2	1.5	2.0	2.2	2.5	
		Ecoli	-	0.7	1.2	1.5	2.2	
		Staphylococcus	-	-	0.7	0.8	2.0	

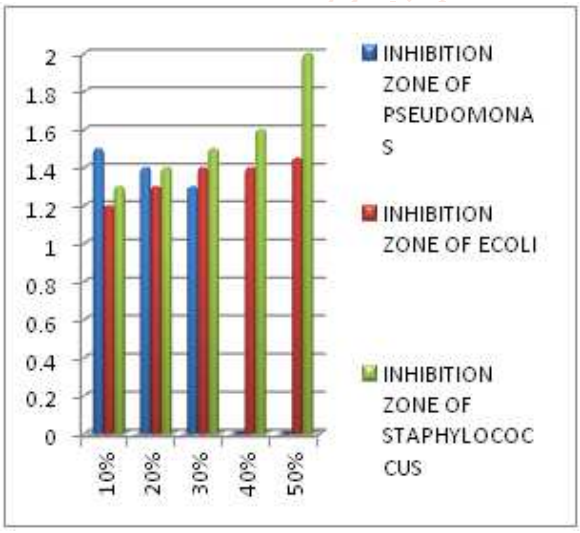


Figure No. 1:- Blue Marker Ink

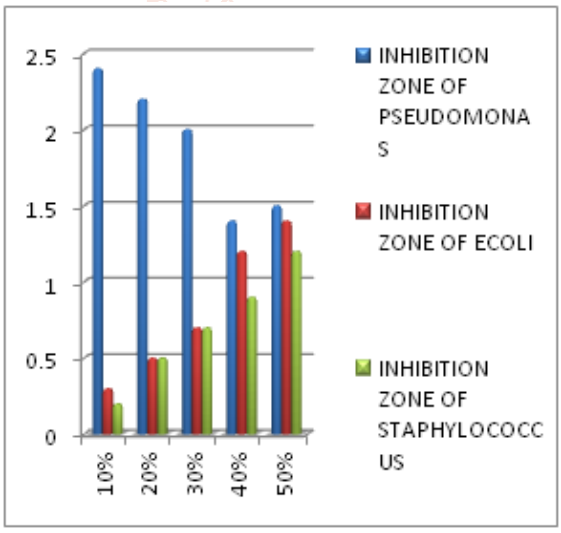


Figure No. 2:- Green Marker

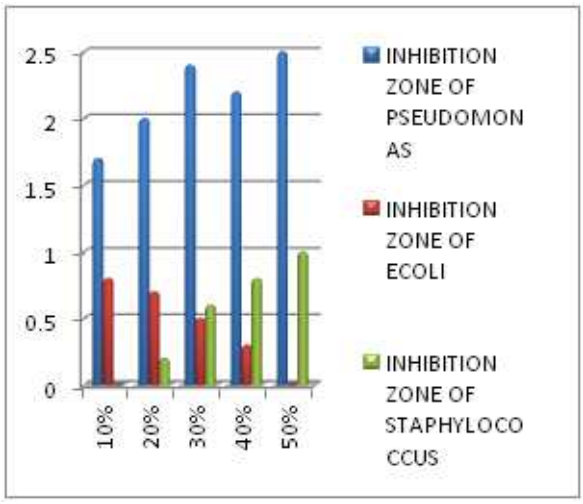


Figure No. 3:- Black Marker Ink

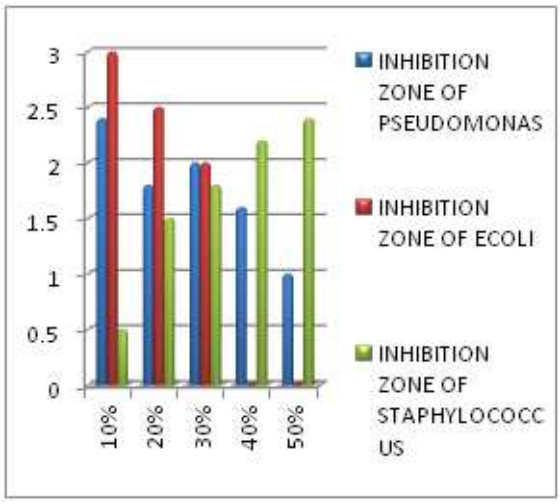


Figure No. 4:- Blue Gel pen Ink

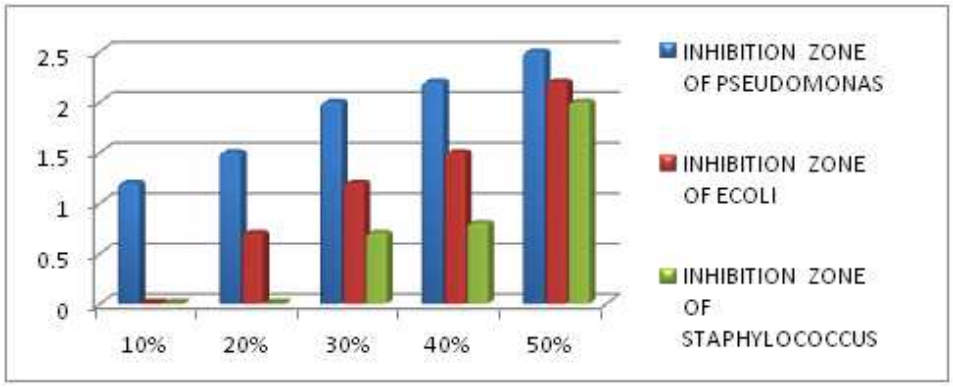


Figure No. 5:- Black Gel pen Ink

Figure No. 1 to 5:- Showing the Graphical representation of inhibition zone with relation to different microbes and their effect on the sample

Discussion

This research paper focuses on the relationship of the microbes with reference to the paper. Microbial effect on the ink is one of the main reasons which degrade or tamper the documents. So it is very important to use the inks which show null or less sensitivity towards the microbes. The antimicrobial detection on 5 different inks i.e. green colour marker ink, blue colour marker ink, black colour marker ink, blue colour gel pen ink and black colour gel pen ink can provide fruitful results which can aid forensic experts in dealing with the microbial effect on the ink in the laboratory.The microbes chosen to study the effect on different inks are Pseudomonas, EColi and Staphylococcus. The sample was used in different dilution rate i.e. 10%, 20%, 30%, 40% and 50% respectively. This study can narrow down the channel of investigation and also help in depicting the age of the documents and ink.

Conclusion

In the above table, shows that in sample 1 the inhibition zone of staphylococcus is more in 30%, 40% and 50% dilution and in 10% and 20% is equal to the inhibition zone of the pseudomonas. In sample 2, 3 and 5 the inhibition zone of pseudomonas is more. In sample 4, 10% and 20% dilution, the inhibition zone of EColi is more than the other but in 30% inhibition zone of pseudomonas and EColi are same. In 40% and 50% EColi do not show any kind of bacterial inhibition and staphylococcus shows maximum. In sample 5, there is continuous and uniform increasement of inhibition zone of pseudomonas but in sample 3, there is uneven and discontinues increasement of inhibition zones of all bacterial. In sample 1, there is continuous and uniform increasement of inhibition zone of the staphylococcus but in sample 5, 10% and 20% do not shows any kind of degradation by the staphylococcus but in 30%, 40 and 50% shows the continuous increasement in the inhibition zone.

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References

[1] Koppenhaver, K., n.d. *Forensic Document Examination: Principles and Practice By*. New Jersey: humana press.

[2] Bird, C., Found, B. and Rogers, D., 2010. Forensic Document Examiners' Skill in Distinguishing Between Natural and Disguised Handwriting Behaviors. *Journal of Forensic Sciences*, 55(5), pp.1291-1295.

[3] Mitchell, C., 1920. Estimation of the age of ink in writing. *The Analyst*, 45(532), p.247b.

[4] Messner, K., Alberighi, L., Banik,, G., Srebotnik, E. and Sobotka, W., n.d. Comparison of Possible Chemical and Microbial Factors Influencing Paper Decay by Iron-Gall Inks. *Springers*.

[5] Historyofpencils.com. 2020. *History of Ink And Pen - Writing With Ink*. [online] Available at: <http://www.historyofpencils.com/writing-instruments-history/history-of-ink-and-pen/> [Accessed 10 May 2020].

[6] En.wikisource.org. 2020. *The History Of Ink/The History Of Ink - Wikisource, The Free Online Library*. [online] Available at: <https://en.wikisource.org/wiki/The_History_of_Ink/The_History_of_Ink> [Accessed 10 May 2020].

[7] The Pixartprinting blog. 2020. *The History Of Paper: From Its Origins To The Present Day | Pixartprinting*. [online] Available at: <https://www.pixartprinting.co.uk/blog/history-paper/> [Accessed 10 May 2020].

[8] En.wikibooks.org. 2020. *Papermaking/History Of Paper - Wikibooks, Open Books For An Open World*. [online] Available at: <https://en.wikibooks.org/wiki/Papermaking/History_of_paper> [Accessed 10 May 2020].