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Antimicrobial property of Hen's Egg amniotic fluid

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ABSTRACT

Medical biotechnology is a new branch of biotechnology, which focuses on finding cures for diseases, and novel methods to inhibit pathogenic growth. The function of amniotic fluid is to protect the embryo from infection of foreign organisms and physical disturbances throughout its development within the egg. Minimum Inhibitory Concentration of this extra embryonic fluid is higher compared with standard antibiotics. In our research we studied the antimicrobial activity of amniotic fluid of the hen's egg. Our aim was to check the antimicrobial activity of the same fluid in a laboratory setting, against a wide spectrum of bacteria, compare the zone of inhibition for the microbes with standard antibiotics, and to determine the storage capacity of the fluid in it the immobilized state. In our study the amniotic fluid extracted from hen's egg are effective against *Streptococcus*, *Staphylococcus*, *E. coli* and *Klebsiella* and exhibiting MIC at 5% in 10 minutes. The extraction of amniotic fluid, its mode of action and immobilization by using natural dyes for increasing its effectiveness will be envisaged.

Keywords: Hens egg, amniotic fluid, antimicrobial property, Minimum inhibitory concentration, immobilisation

INTRODUCTION

In recent times, we have become acutely aware of emerging and increasingly devastating diseases, sparking the search for novel antimicrobial substances. The embryonic fluid present in eggs is known to have significant microbe-inhibiting properties, best demonstrated by the ability of temporary organs or embryonic membranes, especially the amnion, to protect the embryo. The exploitation of this mechanism of protection from invading organisms is the inspiration behind our study. {1,6,7,8,17}

Angiogenesis or anti-angiogenesis is evaluated quantitatively or semiquantitatively {2,5,7,8,9,11,13,14} We have chosen to study the antimicrobial activity of amniotic fluid of the hen's egg. Our aim was to check the antimicrobial activity of the same fluid in a laboratory setting, against a wide spectrum of bacteria, compare the zone of inhibition for the microbes with standard antibiotics, and determine the storage capacity of the fluid in it the immobilized state. {3,12,15,16,17}

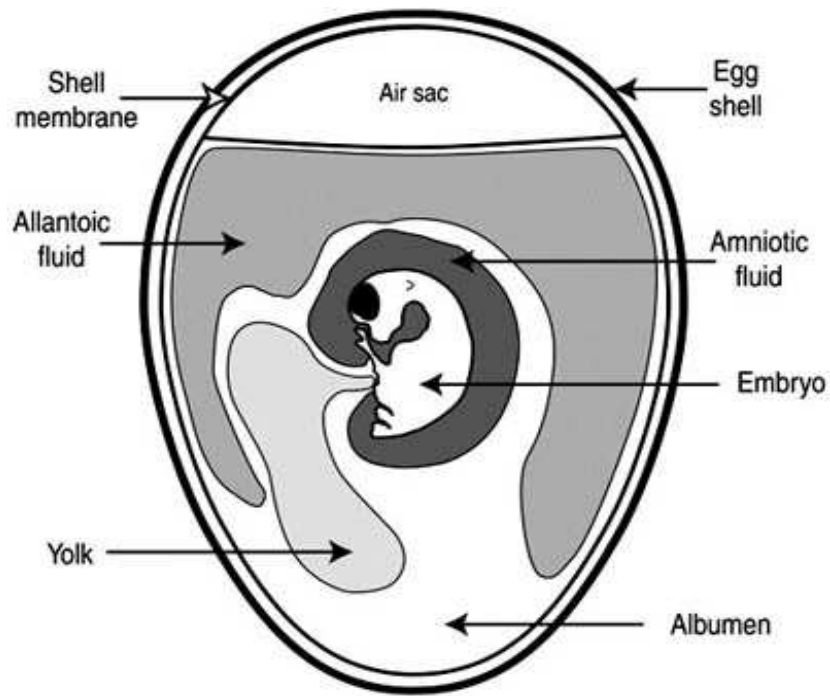


Fig.1 Embryonated egg

MATERIALS AND METHODS

Collection of embryonated eggs and extraction of amniotic fluid:

Embryonated eggs at different stages of development were collected stored in refrigerator at 4°C for further processing.

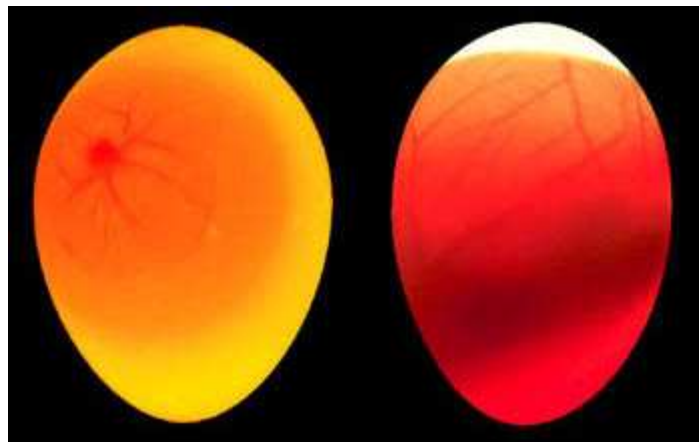


Fig.2 embryo: day 3 embryo :day 9



Fig.4 Collection of Amniotic fluid under aseptic condition

MIC Determination:

Minimum Inhibitory Concentration of Amniotic fluid was determined by taking some pathogenic strains like *Streptococcus*, *Staphylococcus*, *E. coli* and *Klebsiella* sps.

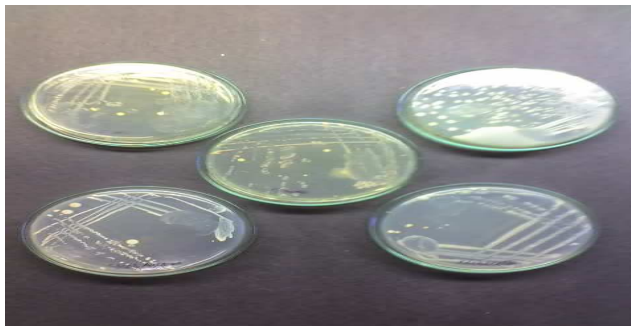


Fig.5 Pure cultures of pathogens (MTCC cultures)

Immobilization:

The amniotic fluid was immobilized within sodium-alginate beads, a process which helps retain the activity of the proteins present in the fluid, and preserve it. The fluids trapped within the cross linked gel could be mobilized once again using a suitable salt solution {2,6,8,12}.

RESULTS AND DISCUSSION**Collection and processing of embryonated eggs**

Fifty Hens embryonated eggs at different stages of development were collected from a Balaji hatcheries Chittoor, and stored in an Refrigerator for 24 hours, Amniotic fluid from these eggs was located by using candling process and then collected at different stages, using disposable syringes, and stored in sterile vials at 4oC for further processing.



Fig.6 collection of embryonated eggs

Table-1 MIC DETERMINATION

SNO	Microorganism used for study	AMNIOTIC FLUID CONCENTRATION			
		0.1%	1%	5%	10%
1	<i>Staphylococcus</i>	-	--	---	---
2	<i>E. coli</i>	-	--	---	---
3	<i>Klebsiella</i>	-	--	---	---
4	<i>Streptococcus</i>	-	--	--	---

+ (growth) - (inhibition of growth)

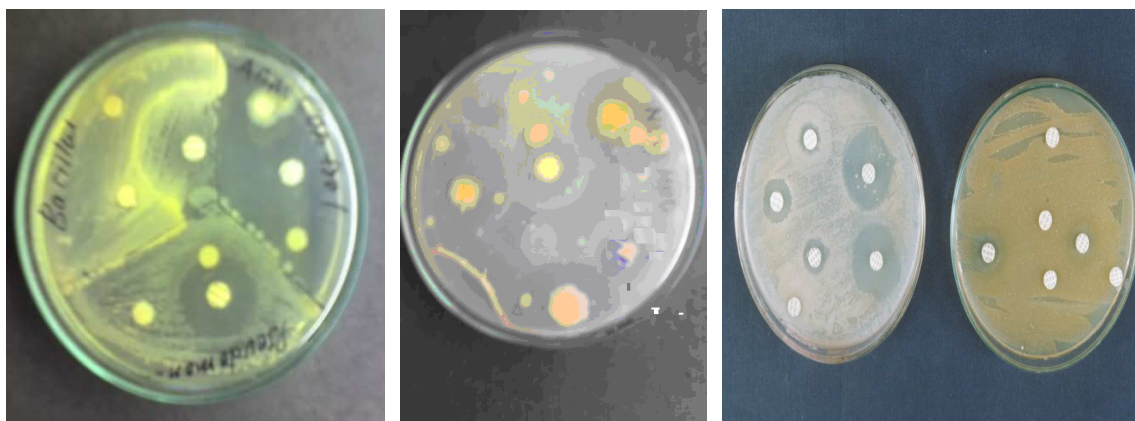


Fig.7 Antimicrobial assay

Immobilisation

The amniotic fluid was immobilized within sodium-alginate beads, a process which helps retain the activity of the proteins present in the fluid, and preserve it. The fluids trapped within the cross linked gel could be mobilized once again using a suitable salt solution. Immobilisation of Amniotic fluid was carried out by using natural dyes extracted from beet root, carrot, rose and turmeric, further the scope to study extensively on this aspect.



Fig.8 Immobilized beads

CONCLUSION

In our study the amniotic fluid extracted from hen's egg are effective against *Streptococcus*, *Staphylococcus*, *E. coli* and *Klebsiella* while exhibiting MIC at 5% in 10 minutes. The extraction of amniotic fluid, its mode of action and immobilization methods for increasing its effectiveness will be envisaged.

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