COMPARATIVE EVALUATION OF ANTIMICROBIAL EFFICACY OF Ig Y IMMUNOGLOBULINS AND PROBIOTICS ON THE LEVELS OF S. MUTANS – AN IN-VITRO STUDY

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ABSTRACT :

Background & Objectives: Dental caries is one of the most prevalent disease worldwide and the awareness against the prevention of caries should be enhanced. The present study evaluated the effectiveness of Probiotic lozenge (Bifilac) with Ig Y Immunoglobulins (Nodecay tablets) as caries preventive agents. The aim of this study was to evaluate and compare the antimicrobial efficacy of IgY Immunoglobulins and Probiotics on Streptococcus mutans at various concentrations.

Methods: Group A: Control, Group B: Probiotic (Bifilac Probiotic), Group C: IgY (Nodecay tablet)

Results: IgY immunoglobulins and probiotics showed resistance to S.mutans at a concentration of 25 mg\ml.

Interpretation & Conclusion: No statistically significant difference noted in antimicrobial efficacy between group B and C. Ig Y are specially prepared against s. mutans and probiotics are commonly used now a days. No decay tablets are available as lozenges, while probiotics when used in the form of lozenges can also aid in a considerable protection against dental caries. As both the drugs shows susceptibility to s. mutans in the present study both can be used as an effective alternative for prevention of dental caries in children.

KEYWORDS: Dental caries, S.mutans, IgY Immunoglobulins, Probiotics, Nodecay chewable tablets.

INTRODUCTION:

Dental caries is one of the most prevalent dental problems across the world. With the gradual change in the life style and food habits adults and children are more prone for caries development. Approximately nearly 36% of world population are experiencing teeth decay. Therefore there is an increasing demand to develop new treatment method for dental caries.[1]

Streptococcus mutans is considered one of the key organisms in causing caries, as it is isolated in large amounts from the caries lesion. They are more active especially in acidic environment as they are aciduric in nature. S. mutans produces glucosyltransferases which is the major virulence factor needed for the synthesis of water insoluable glucans. Hence the control of growth and development of s.mutans is important for the control of dental caries.[2]

During the last decade, the use of probiotics has gained interest within the dental research community with focus on caries development, periodontal health and halitosis. Bacteriotherapy in the form of probiotics is the natural way to maintain health and to protect the oral tissues from diseases.[3]

Ig Y immunoglobulins exist in the serum of chickens and eggs.[4] IgY immunotherapy has advantages like lack of reactivity with human complement system. Passive immunotherapy with Ig Y produces rapid and local onset of action, high specific activity & can be used for both infants and adults. IgY immunotherapy has several attractive features including: lack of reactivity with the human complement system and human Fc- receptors thereby preventing non-specific inflammation.[5]

The present study evaluated the effectiveness of

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probiotic lozenge (Bifilac) with Ig Y Immunoglobulins (Nodecay tablets)

AIM: The aim of this study is to evaluate and compare the antimicrobial efficacy of IgY IMMUNOGLOBULINS and PROBIOTICS on Streptococcus mutans at various concentrations

MATERIALS AND METHODS:

Three groups

Group A: Control

Group B: Probiotic (Bifilac Probiotic)

Group C: IgY (Nodecay tablet)

DRUG DILUTIONS:

Samples from group B and C were diluted using a saturated salt solutions of acidic pH to get five different dilutions (five different dilutions 25mg \ ml, 20mg\ml,10 mg\ ml, 5 mg \ ml, 2.5 mg\ml).[3]

S.No	DRUG B	DILUTIONS mg/ml	DRUG C	DILUTIONS mg/ml
1	B1	25	C1	25
2	B2	20	C2	20
3	B3	10	C3	10
4	B4	05	C4	05
5	B5	2.5	C5	2.5

Table 1

Fresh plates of test bacteria (s.mutans) were isolated from the dental caries and cultured on blood agar medium, this serves as the control group. Colonies of s. mutans from fresh culture was suspended in 5ml bacteriological peptone broth in labelled sterile tubes. They were incubated for six hours. Blood agar plates were prepared and used for the susceptibility study. Sterile filter paper disks of 6mm soaked in different dilutions were used.[6]

Reading the plates and interpreting the results

After 24 hours of incubation, the diameter of zone of complete inhibition are measured using a ruler.[7]

RESULTS :

S.No	DRUG B	Zone of Inhibition (in mm)	Interpre-tation	DRUG C	Zone of Inhibition (in mm)	Interpre-tation
1	B1	07	S	C1	11	S
2	B2	06	R	C2	06	R
3	B3	06	R	C3	06	R
4	B4	06	R	C4	06	R
5	B5	06	R	C5	06	R

S - Susceptible I - Intermediate R - Resistant

Table 2

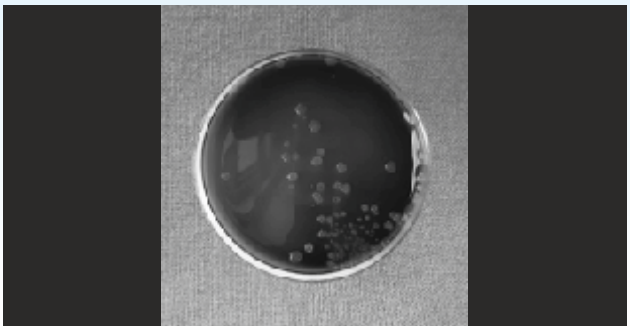

Fig 1

GROUP A

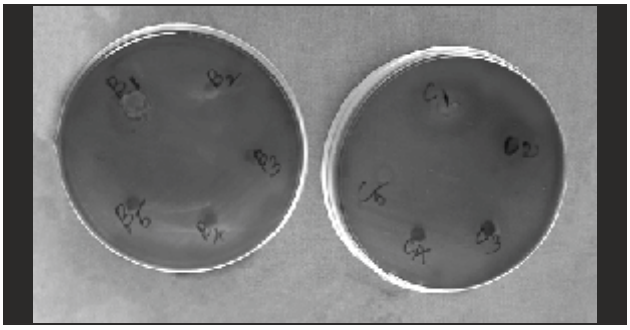

Fig 2

INTERPRETATION:

Group A (Control group showed a colony forming unit of 4 x 10^3 colony forming units after 24 hours of incubation.

After the incubation it was observed that C1 shows a inhibitory zone of size 11 mm at 25 µg\ml, and was resistant to all other concentrations from 20 to 0.25 µg\ml.

Whereas, the drug B showed a zone size of 07 mm at at 25 µg\ml, and was resistant to all other concentrations from 20 to 0.25 µg\ml.

STATISTICS:

Group Statistics:

	Group	n	Mean	Std deviation	Std error of mean
Zone of inhibition	Group B	5	6.20	.447	.200
Zone of inhibition	Group C	5	7.00	2.236	1.000

Table 3

Independent Sample t test

F	Sig.	t	Sig.	Mean	Difference	Std. Error	95% Confidence Interval of the Difference		
Zone of inhibition	Equal variances assumed	4.37	.070	-.784	.455	-.800	1.020	-3.152	1.552
	Equal variances not assumed			-.784	.474	-.800	1.020	-3.551	1.951

Table 4

INFERENCE:

This invitro study evaluated the viability of s.mutans in the presence of probiotic (bifilac) and IgY immunoglobulins (Nodecay tablets).

The results of our study shows that IgY immuno-globulins and probiotics showed resistance to S.mutans at a concentration of 25 mg\ml.

Group B - Mean ZOI = 6.20 ± 0.45

Group C - Mean ZOI = 7 ± 2.24

No statistically significant difference noted in antimicrobial efficacy between group B and C (Significance p= 0.070)

DISCUSSION:

S.mutans is the main causative agent of dental caries because of its ability to adhere to the tooth structure and its ability to produce insoluable glucans. The agent which effectively reduces the level of s mutans should be recognized in order to prevent the caries. Several agents which helps in reduction of s mutans levels were chlorhexidine in the form of mouth wash, gel and varnishes[8], povidone iodine mouth rinses, fluoride varnish.[9] The effectiveness of antimicrobials like augmentin and amoxillin against s mutans were proved by Staves et al.[10] Some of the natural agents like curcumin,[11] propolis, green tea,[12] has also proved effective in reducing the levels of s mutans.

Passive immunization of antibody to Streptococcus mutans glucan binding protein B (GBP-B) has been shown to induce protection against experimental dental caries in rats.[13]

IgY immunoglobulins were proved to be effective against s. mutans in the control of dental caries, periodontitis, oral candidiasis.[14] Similarly study by Nguyen[2] Gandhimathi[15] Mary[16] also proved that anti CA GTF IgY were reducing the level of s mutans colinazition.

It has been shown that administration of probiotic drops containing strains of Lactobacillus reuteri during the first year of life had an impact on caries prevalence and Early childhood caries development could be reduced through daily administration of probiotic chewing tablets as adjunct to daily use of fluoride toothpaste in preschool children.[17] Probiotics effectively reduces plaque index and s. mutans levels.[18]

Probiotics and Ig Y Immunoglobulins were proved to reduce the levels of s.mutans and to the best of our knowledge there are no literature comparing the effectiveness of these two drugs. Hence this invitro study was done to compare Bifilac probioticTM with Nodecay tabletsTM containing Ig Y.

Probiotic bacteriotherapy is the natural way to maintain health and to protect the oral tissues from diseases. Probiotics are defined as live microorganisms that confer a health benefit on the host when administrated in appropriate amounts.[18] Probiotics are emerging as a novel approach for the treatment of oral disorders, probiotic species act through a variety of mechanisms like exclusion and competition with potential pathogens, production of antimicrobial substance against oral pathogens, local and systemic immunomodulation's, enhancement of mucosal barrier functions.[19] Studies by Sugumari et al proved the effectiveness of probiotics against periodontal pathogens.

Leslie and Clem first coined the term IgY in 1969. Specific IgY antibodies against CA-Gtf (cell associated glycosyltransferase) of s mutans are obtained by immunizing hens with CA-Gtf antigens of s. mutan, causing them to produce antibodies as a part of their normal immune response.[20] The antibodies thus produced are transferred to the egg

yolk through maternal transfer. Egg yolk contains 99% fat and only 1% protein. These IgY antibodies, are extracted from the egg yolk laid by these hens, purified. Once formulated it is given orally to humans. Ig Y Immunoglobulins are reported to reduce the number of s mutans in the oral cavity by forming antigen antibody complexes in dental plaque and saliva.[21]

In this study Nodecay chewable tablets containing IgY immunoglobulins and Bifilic probiotic lozenges with Lactobacillus sporogenes, Streptococcus faecalis, Clostridium butyrium, and Bacillus mesentericus were used.

Elavarasu evaluated the antibacterial activity of Bifilac against P. gingivalis at various concentrations such as 2.5, 5, 10, and 20 µg/ml, of which 20 µg/ml showed a zone of 22 mm in accordance with this, bifilac lozenges and no decay chewable tablets were used in five different dilutions where only 25mg \ ml were susceptible and all the lower concentrations were resistant.[3]

The study results proved that a minimum of 25mg \ ml and above of both drugs should be used inorder to produce a significant reduction of s.mutans. Eventhough both the drugs showed suspectibility against s.mutans when used at the concentration of 25mg/ml but the results were not statistically significant.

CONCLUSION:

Ig Y are specially prepared against s. mutans and probiotics are commonly used now a days. No decay tablets are available as lozenges, while probiotics when used in the form of lozenges can also aid in a considerable protection against dental caries. As both the drugs shows susceptibility to s. mutans in the present study both can be used as an effective alternative for prevention of dental caries in children.

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