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SHODH DARPAN

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I would like to thank management, editorial team, reviewers and initial team which have helped in making this journal a possibility and reality. We hope that the research featured here sets up many new milestones. I look forward to make this endeavor very meaningful.

- Sushil Kumar Sahu

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CONTENTS

RESEARCH PAPERS

A study of effect of brain Hemisphere dominance upon Mental Ability of Senior Secondary School Students

Dr. Jasbir Kaur[1]

Abstract

*The present study is intended to find out the interactional effect of brain hemisphere dominance with urban-rural belongingness upon mental ability of senior secondary school students. To fulfill the objectives of the present study 800 senior secondary school students were selected randomly from different schools located in urban (N = 400) and rural areas of Durg District of Chhattisgarh state. To explore the possibility of joint action effect of brain hemisphere domination urban rural belongingness on mental ability of selected senior secondary school students 3*2 factorial design was set up.*

The selected students from senior secondary school were classified into right brain hemisphere dominant, left brain hemisphere dominant and integrated brain hemisphere dominant with the help of SOLAT Scale by D. Venkatraman (1994).

The mental ability of the selected subjects was assessed by Culture Fait Test (Revised Edition 1980)

After undergoing a systemic and statistical analysis. Results and finding of the study shows that the students with integrated brain hemisphere dominance scored higher on the scale of mental abilities test them the students with right brain dominance and left brain dominance.

Introduction

Educators have been concerned over the years with identifying those factors which facilitate teaching learning process. Investigations have yielded no simple solution to this problem.

The biological understand of how the two hemisphere of our brain function has now reached general consensus. There is still much work to do, particularly on the differences observed between individuals and understanding all the brain process

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involved on cognition. Brain hemispheres play a flexible role in accomplishing variety of tastes. It is therefore important for the parents and teachers to understand the nature of student's brain and its functioning.

Hemisphericity is the cerebral dominance of an individual in retaining and processing modes of information in his own style of learning and thinking (Venkatraman, 1989)

There are some studies in which speed of processing has been directly linked to central nervous system functioning and to intelligence (Vernon, P.A. & Mori, M. (1992). Intelligence, reaction times and peripheral nerve conduction velocity as cited in Bee. H. (2000). The developing child (9^{th} ed.). Boston: Allyn and Bacon). Researches shows that the speed with which people are able to retrieve information is related to intelligence. Studies have shown that during perceptual tasks, right hemisphere is more activated so intelligent people may have more specialized right hemisphere (Barlow, 2001)

Studies have shown that students who are taught through methods that corresponds with their hemisphere style achieved higher test scores (Saleh, 2001) to amplify and unite the knowledge of left and right brain characteristics to achieve whole rain instruction is a current focus of many educators (Repress and Lutify, 2006)

Lavach (2009) examined the brain hemisphericity of students with different majors. He reported that humanities students showed preference for right hemispheric dominance. Natural preference for left hemisphere dominance.

So, far no such study has been conducted in which mental ability of the students was assessed in the light of hemispheric dominance and locales of the school.

Hence, the present study which analysis the effect of brain hemisphere dominance on mental ability of senior secondary school students is another attempt in this direction.

Significance of the study

The important if this study lies in the knowledge regarding relationship between brain hemisphere dominance and mental ability. Hence it is necessary to know the impact of right, left or integrated brain hemisphere dominance on mental ability. The results of

this study can also be used by parents while guiding their spous regarding choice of students as per their hemisphericity.

Conceptual and Operational Definitions

Brain hemisphericity is the cerebral dominance of our individual in retaining and processing modes of information in his own style of learning and thinking (Venkatraman, 1989)

Mental ability is the ability to learn, understand and make judgments or have opinions that are based on reason; (Cambridge Advance Learner's Dictionary, 2006)

In the present study, mental ability is considered as general intelligence of a person without taking cognizance climate.

Locale (or urban rural belongingness). The educators and the general public believe that students from smaller and rural schools receive an educators that is inferior to that of students from larger urban or sub urban schools.

Objectives of the study

To study the effect of brain hemispehere dominance upon mental ability in the light of locale of schools.

Hypothesis

Interaction Oriented

Brain hemisphere domination and locale (urban-rural belongingness) will show its joint action effect on mental ability of senior secondary students.

Tools

Independent Variable

Brain Hemisphere Dominance

For measuring Brain hemisphere dominance of selected subjects, SOLAT scale by D. Venkatraman (1994) was used. The reliability coefficient of correlation for right, left and integrated hemisphere function were found to be 0.89. 0.65 and 0.71 respectively. This

coefficient suggests that the SOLAT possesses reliability to a significant level. The construct validity of right, left and integrated hemisphere is 0.842, 0.621 and 0.678 respectively.

Dependent variable

Mental ability of the selected subjects was assessed by Culture Fair Test (Revised Hindi Edition, 1980). The concrete validityof the test was found to be 0.69 while the reliability coefficient reported was 0.85.

To explore the possibility of joint action effect of brain hemisphere dominance X locate (urban-rural belongingness) will show its joint action effect on mental ability of senior secondary students.

In order to verify hypothesis, a 3*2 factorial design was setup in which urban and rural students with right, left and integrated brain hemisphere domination were arranged according to their respectively cells. The mental ability of these identified senior secondary school students are presented in table 1 and 2 respectively.

	LOCALE		
	b_1 Urban	b_2 Rural	M
a_1 Right Dominant	M = 30.72 N = 102	M = 33.74 N = 130	32.24
a_2 Left Dominant	M = 32.48 N = 200	M = 33.82 N = 107	33.15
a_3 Integrated Dominant	M = 38.16 N = 98	M = 33.30 N = 103	35.73
M	33.79	33.62	

Table No. 1 : Brain Hemisphere Domination X Locale (urban-rural Belongingness) on mental Ability of Senior Secondary Students (N = 800)

Source of variation	SS	DF	MS	F
A	1405.687	2	702.843	28.80**
B	5.271	1	5.271	0.21(NS)
AB	1857.542	2	928.771	38.06**
Within treatment	19373.418	794	24.400	

Table No. 2 : Anova Summary

Effect of Brain Hemisphere Domination X Locale (Urban-rural) belongingness on mental ability of Senior Secondary School Students (N = 800)

Remarks – (1) ** Significant at .01 level (2) **NS** Not Significant

The F of 28.80, an indicator of the main effect of brain hemisphere dominance on mental ability is statistically significant at .01 level. It thereby reveals that the mental ability of students with integrated brain hemisphere are significantly greater than the mental ability scored of the students with dominant right and left brain hemisphere.

The F of 0.21, an indicator of the main effect of locale (urban-rural belongingness) upon mental ability turned out to be statistically insignificant and thereby rendered confirmation to the findings that it has nothing to do with mental ability of selected students.

The F of 38.06, an indicator of brain hemisphere domination and locale (A * B) interaction, turned out to be statistically significant at .01 level. While scanning table 2, it is clear that urban students with integrated brain hemisphere should more magnitude of mental ability than the urban-rural students with left and right brain hemisphere domination and rural students with integrated brain hemisphere domination.

Since interaction effect of brain hemisphere domination and urban-rural belongingness upon mental ability was found to be statistically significant, thus interaction hypothesis H is accepted.

Conclusion

On the basis of above mentioned discussion, it can be concluded that Brain hemisphere dominance plays a big part in mental ability. Students with integrated brain hemisphere

dominance have superior mental ability as compared to students with left and right hemisphere dominance.

Educational and social Implications

The results of the present study will be helpful for educators to evaluate mental ability of secondary school students in the light of brain hemisphere dominance. The results have educational as well as social implication in regard to making a curriculum which fulfils the need of the students on the basis of their brain hemisphere dominance which in turn beneficial for our society as our future generation will be much more learned due to this educational approach.

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Socio- Economic Situation in Bastar Tribal

Bhagwati Baghel [1], Dr. Swapan Kumar Kolay [2]

Abstract

The economic status of the tribes varies from region to region meanwhile there are few tribes which can be described as 'pure hunters' and there are many tribes who supplement their crude farming with hunting and fishing, and pasteurization. The occupational and economic status are ever influenced by alien factors like urbanisation and industralisation etc. As well the simple life of tribal is threatened by poverty and indebtedness which are creating low level of socio-economic status. However, tribals expanded more than their income in drinking on any occasion. According to above background present paper is an effort to understand the responsible factors of poor socio-economic status among tribals of Bastar. The result of the study shows that tribals are cheated by dominating group. By this they are facing land alliance, economic exploitation, socio-cultural exploitation, poverty and indebtedness problems.

***Key Words**: Pure hunters, Tribals, Poverty and indebtedness, Socio-economic status, Socio-cultural exploitation.*

Introduction

In this modern era large numbers of tribal people have been detribalized. Their natural habitat has been found to be incapable of maintaining their growing population and the sanctity of their moorings has been reduced on account of the pressing needs of feeding mouths which previously found forage for themselves in the forests or in the tiny terraces they built up for agriculture (Roy Burman, B. K., 1965). Tribe is after all a territorial group. Due to long association with the tribal people, the minority non-tribal communities also adopted many social customs and traditions of tribal culture which clearly had distinct advantage over their own tradition and social customs (Singh, K.S., 1995). The integrated village life to which the tribal people were tuned, no longer affords security to the tribals while the needs of modern life, of plantation and of luminal towns, are now being met by importing labour from the tribal areas which is

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creating economic problems among tribals particularly from those areas where contacts with civilization have disorganized social life. Singh and Jabbi (1996) write about Empirical studies in different parts of India have demonstrated that the health, education and employment status of the tribals in India is very low, lower than other social groups. Thus tribal life is still threatened through poverty and indebtedness.

To understanding of tribal economy and associated problems it is needful to identifying the problems of tribes in study area at different levels of economic and social development with regional problems by figure out the reasons of poor tribal economy. For the study purpose researchers had selected tribal village Asna, Bastar because majority of tribal population are inhabitants of *Gond, Muriya and Bhatra*.

With above background and objectives two types of samples were taken into account for our study. First, purposive sampling: this method was adopted in order to select the study areas. Second, random sampling: this was used for selecting the sample (respondents). Random samples have been taken as the number of households in a single village consists of more than 150 to 200. Therefore, we had used random sampling to collect data (50 households' altogether) for our study.

For collecting the data, we used both primary and secondary sources. Primary data was collected by interviewing people of Bhatra in tribal area. For gathering quantitative and qualitative data household survey was conducted using pre-tested schedules. Primarily, head of the households were interviewed to gather detail information regarding the family members. Moreover, some elders were also selectively interviewed in order to draw information concerning their opinion, condition and plights with the advent of tribal development problems. The secondary data were collected from related articles from journals, Government gazettes and published reports etc.

Result and Discussion

Today the tribals of Bastar are facing many problems related to their economy which are problems of poor food resources, inefficient agriculture, new occupation, land alienation, poor economy, administrative exactions and indebtedness also. These problems are most acute among the tribals of Bastar. The dominating group exploits the tribesmen taking advantage of their ignorance, simplicity, honesty and dread of the supernatural. This research paper is an effort to analyze reasons which effect tribals' economy.

Family Size

It is reveal from table 1 that in selected village has prevalence of nuclear family (78.18 percent) where only 16.36 percent families were large family. It's significant that the frequency of joint family was below 6 percent which shows the declining trend of the joint family is also prevalent in rural and tribal areas.

Type of Family	Total family		Family Member	
	Number	%	Number	%
Nuclear Family	43	78.18	121	59.61
Large Family	9	16.36	53	26.11
Joint Family	3	5.46	29	14.28
Total	**55**	**100**	**203**	**100**

Table 1: Type of Family in Selected Village

Low Level of Education

Illiteracy and ignorance are the main hurdles of tribal economy development. It has been observed from table 2, that large number of tribal population (60.48 percent) was illiterate while 39.52 percent were literate. It is clear that literacy status was not up to the mark because only few percentages of respondents (13.64 and 3.03 percentage) were obtained higher secondary school and graduation respectively. In most of the tribal areas percentage of adult literacy after

Literacy Status	Male		Female		Total	
	No	*%*	*No*	*%*	*No*	*%*
Illiterate	42	58.33	59	62.11	101	60.48
Literate (6+)	30	41.67	36	37.89	66	39.52
Level of Education						
Primary school	7	23.33	12	33.33	19	28.79
Secondary school	12	40.00	11	30.56	23	34.85
High school	5	16.67	8	22.22	13	19.70
Higher secondary school	4	13.33	5	13.89	9	13.64
Graduation and above	2	6.67	-	-	2	3.03
Total		**100**		**100**		**100**

Table 2: Educational Status of Tribals in Asna Village

independence , ranged between zero and five percent of late it has improved, particularly in the case of men, but the percentage of tribal literacy in still too low.

Traditional Occupation

Gare (1983) writes in research study, the exploitation of tribal in land and forest is major cause of tribal's economic underdevelopment and improvement. The tribal's' forest wealth has been ruined due to excessive cutting of timber by the forest labour co-operative societies. Hence, the tribes today survive on agriculture land and farm labour practices. Unemployment increased because those who never depended upon others for their livelihood were reduced to a dependent status (Mathur, H.M., 1976). When researcher observed the occupational status in the village, it has been seen 53.03 percent population were engaged in cultivation and labour work while only 3.79 percent were doing Government service. 4.55 percent respondents were Labour in the village. They had to work as wage earners under forest contractors or building contractors either in their own area or as migrants to other area. Bonded labour also become common. This tended them to such a low social and economic status as they had never experienced.

Type of Occupation	**Respondents**	
	No	**%**
Working Population	132	79.04
Non working Population	35	20.96
Total	**167**	**100**
Working Population		
Cultivation	70	53.03
Labour	42	31.82
Govt. service	5	3.79
Cultivation cum labour	8	6.06
Cultivation cum Govt. service	6	4.55
Other	1	0.76
Total	**132**	**100**

Table 3: Occupational Status

Traditional Practices of Economy

The majority of tribal hold small and limited agriculture lands and cultivate through traditional seeds method without irrigation. The tribal's' economic problems are related

with unproductive agriculture and hunger. It is always seen that tribal economy was self supporting subsistence and the tribal people were complacent, could satisfy their

meager wants and mainly depended on hunting, food-gathering, picking up minor forest produce, primitive and traditional methods of cultivation wherein they worked on their own land and had some cattle, etc. with tribal development programme, rights in forest were reduced, hunting, picking of forest produce controlled and during past year land was also alienated thus rendering many tribal landless labourers.

Type of Technology	Total Households	
	No	%
Traditional Method of Cultivation	6	10.91
Depend on Monsoon	30	54.55
Depend on Forest products	9	16.36
Depend on hunting and Food gathering	3	05.45
Traditional Art and Craft	7	12.73
Total	**55**	**100**

Table 4: Prevalence of Traditional Practices of Economy in the Village

Low Income and Economic Exploitation

Table 5 shows that 49.09 percent Household comes between Rs. 10000-20000/- of yearly income. However, only 5.45 percent households had good economy with more than Rs. 50001/- at annually. This shows very low economic range in the village. Amongst low income group economic exploitation is prevalence among tribal population. Money lenders, traders, roving merchants, etc., used to exploit him. Lower economic status leads to borrow/ credit money from resources in the village.

Annual Income Range (in Rs.)	Total Households	
	No	%
10000- 20000	27	49.09
20001- 30000	11	20.00
30001- 40000	9	16.36
40001- 50000	5	9.09
50001 ≤	3	5.45
Total	**55**	**100**

Table 5: Economic Status of Tribals

High Expenditure

It is observed from table 6 that expenditure is higher at the time of festivals, festivities and occasional celebrations in honour of guests to eat heavily into his lean resources

rather than their monthly/yearly income. It has been observed that tribals are very fond of drinking and this tendency turn their economic condition from sustainable to marginal. Table shows that large percentage of tribals expands 45001-55000 Rs. on food (67.27 percent), festivals (38.18) and drinking (58.18) which are higher than their income. We can say that extra expenditure creates economic deprivation in the village.

Expenditure Range (in Rs.)	**Food**		**Festival**		**Drinking**		**Education**		**Cloths**	
	No	***%***	***No***	***%***	***No***	***%***	***No***	***%***	***No***	***%***
5000-15000	1	1.82	5	9.09	4	7.27	29	52.73	27	49.09
15001-25000	5	9.09	9	16.36	7	12.73	17	30.91	9	16.36
25001-45000	9	16.36	13	23.64	11	20.00	5	9.09	9	16.36
45001-55000	37	67.27	21	38.18	32	58.18	3	5.45	7	12.73
55000<	3	5.45	7	12.73	1	1.82	1	1.82	3	5.45
Total	**55**	**100.00**	**55**	**100.00**	**55**	**100.00**	**55**	**100.00**	**55**	**100.00**

Table 6: Expenditure Status among Tribals in Study Village

Low Landholding Status

There is strong relationship between tribal population and land as; their entire livelihood depends on land, which they possess. The con cept of land holding has come up among the tribes since then they have shifted from primitive to shifting and now to sedentary agriculture. Their economic status depends upon the size and quality of land, which they hold and cultivate. It is realized that they have been cultivating the land since long but has not so far been registered in their name in terms of ownership of land (more than 60 percent landless).

Variables	**Total Households**	
	No	**%**
Landless	36	65.45
Landholder	19	34.55
Total	**55**	**100**
Landholder		
< 3 acres	9	47.37
3 to 5 acres	7	36.84
5 acres to above	3	15.79
Total	**19**	**100**

Table 7: Land Holding Status

Borrow the loan

Table 8 indicates that 67.27 percent household had borrowed money to fulfill their requirements. It is observed that public banking system did not provide loans for unproductive purposes or for ceremonies. Similarly the system lacks flexibility and is cumbersome for tribals. Hence they have to turn back to the traditional money lender whom they consider close at hand, more sympathetic and personalized as against an unsympathetic bureaucrat of the bank/office. This again places opportunities to exploit in the hand of the traditional *Sahukar.*

Variables	**Total Households**	
	No	**%**
Borrowed Money	37	67.27
Not Borrowed	18	32.73
Total	**55**	**100.00**

Table 8 : Crediting Status among Tribals

Land Alliance

34.55 percent households had their own Land while 65.45 percent households were landless. In study areas tribal farmers had small land holding ranging which was below 3 acre only (47.37 percent). But people who were landless, they considered that some time before they had sufficient land. A few had larger holdings. Unscrupulous money lenders and traders grabbed and misappropriated their lands and either made them landless or labourers on their lands. *Malguzars* and *Zamidars,* even from their own community, who had power, inflected hardships on them (Dube, S.C., 1949). Later, though legislation followed declaring all transfers of land form tribals to non- tribals illegal, whether for a consideration or otherwise, the loop holes were skillfully manipulated by the rich, defeating the very purpose of legislation (Chattopadhyayaya, K. P., 1949).

Sources	**Households**	
	No	**%**
Sahukar	34	61.82
Private Bank	2	3.64
Govt. Bank	17	30.91
Other sources	2	3.64
Total	**55**	**100.00**

Table 9 : Sources for Crediting in Study village

Below Poverty Line

Below Poverty Line is an economic benchmark and poverty threshold used by the government of India to indicate economic disadvantage and to identify individuals and households in need of government assistance and aid. Regarding this table 9 shows, very highest percentage of families come under below poverty line (83.64 percent) while only 16.36 percent families were above poverty line.

Status	**Respondents**	
	No	**%**
Above poverty line	9	16.36
Below poverty line	46	83.64
Total	**55**	**100**

Table 10 : Poverty Status in Village

Tribal's Problems with Administrative Institution

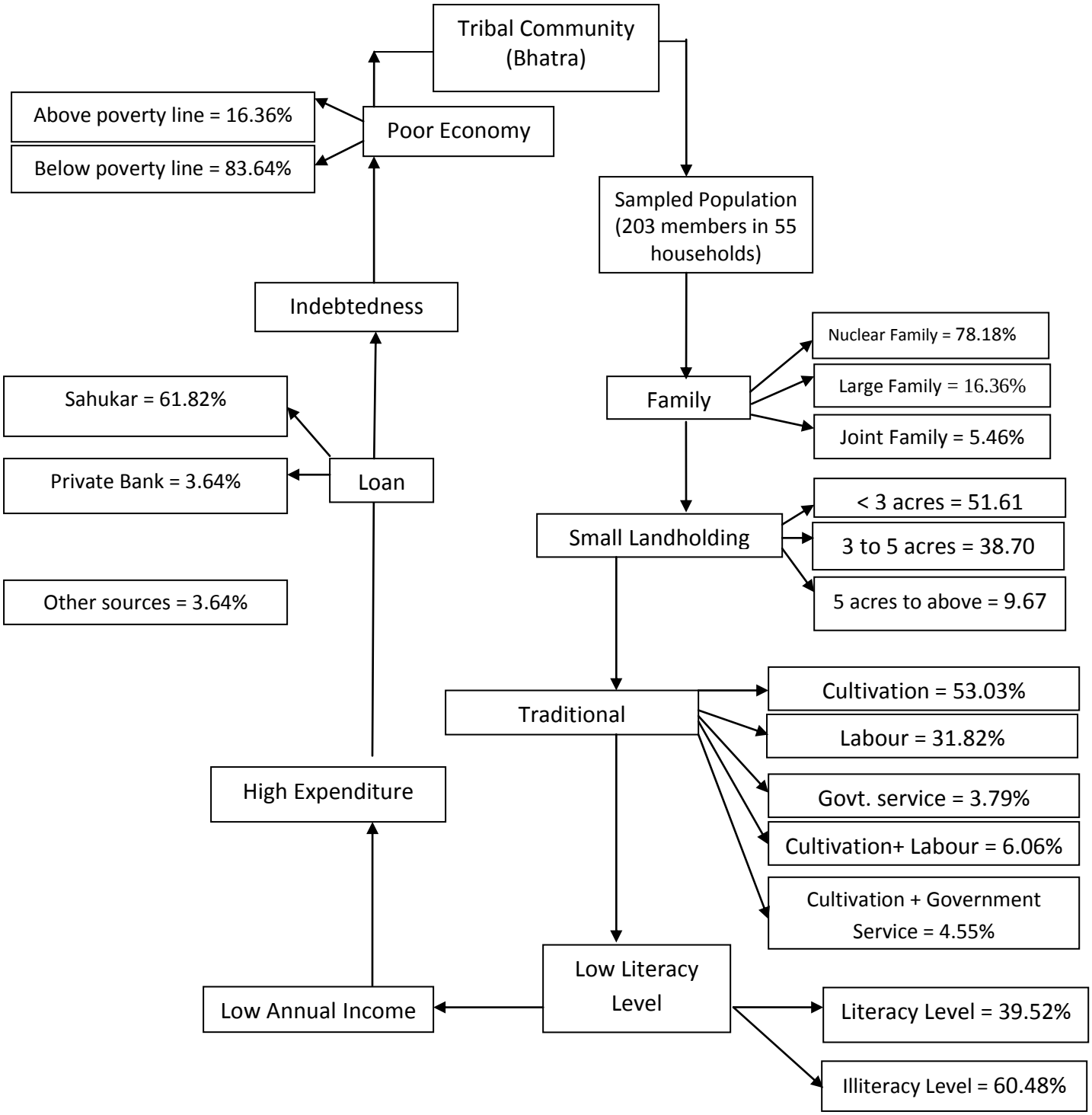
The offices and officers at the grass root or contact level who are supposed to reach them the economic benefits whether in cash or find, through loans, subsidies, things or services, have also created a bad record and bad impression on the tribal mind. They expected and accept bribes, share the benefits, delay the flow either due to the omnipresent red tape or willfully and very little reaches the tribals (Bailey, F.G., 1957). Most of the tribal communities have had their traditional habitat in forests. But the policy did not result so much in preserving the forest wealth but added a premium to the illegal gratification, which the tribals had to play even for satisfying their genuine, primary needs such as fuel and fodder. As per as loan is concerned, they face crises in their whole life and prefer local *Sahukar* (61.82 percent) while around 40 percent respondents seeking private bank, Government bank and other sources.

Indebtedness

The local creditors have no hesitation in advancing loans, for not only heavy compound interest, but also pre-emptive rights to purchase the standing crops at rock bottom price are their privilege (Thakkar, A. V., 1949).Naturally when tribal are in need of money, they are forced to depend in other particularly on the non-tribal money lenders. Under this system, a person who takes a loan from a money lender or land owner, is required to serve him just like a slave according to the term and conditions of loan. Quite often, if

he is unable to repay the loan, it descends to his son or even to his third generation. However pressing the claims of others may be, the creditor knows that the *Adivasi* will honour his word and the *Adivasi* knowing no arithmetic always leaves a comfortable arrear in favour of the creditor. Hence the tribal indebtedness in form of bonded labour is seen in the village. Thus the vicious circle becomes inescapable.

Vicious Circle



Trend Economy Leads Socio- Cultural Exploitation in Study Area

Social and cultural exploitation is also rampant. Tribal women/ girls are victims of this exploitation. Tribal culture is misconceived. Their traditional dress and free behaviour is wrongly conceived. The outsiders such as contractors, truck drivers, government employees, etc. belittle their culture and consider it cheap and vulnerable. Girls are lured or enticed and fall victim to allurements. In some area damage to the honesty and integrity of tribal man and women is to such an extent that they run after money and are ready to earn it any means. This is causality attributable to the social cost of development (Deogaonkar, S. G., 1992). Their traditional arts, crafts, skills and nobility of mind are disappearing. Social workers often say that a tribal village is considered as "developed" if the inhabitants have become dishonest, politicized and have a started practicing deceit. These negative fallouts of economic development are doing irreparable damage to the tribal culture.

Suggestions

Above all, the State Government must urgently restore its development and welfare presence in this and all the above mentioned following 'affected district' as well as 'affected villages' of Chhattisgarh

1. Fair Price Shops run by panchayats should be opened at the maximum distance of 5 kilometres from every hamlet, and should be opened at least 5 days a week.
2. A drive should be undertaken to ensure that all households of local tribal villagers are given ration cards and food grains at *Anthodia* rates (prevalent in Chhattisgarh) in all the 644 villages affected by the ongoing conflict.
3. MGNREGA job cards should be also given to all local households, and the State should strive to ensure that 100 days of work is given to every household that seeks employment under the scheme.
4. To service all the conflict affected areas, and government periodically send their representatives to evaluate the extent to which the steps are to be taken in the affected villages, to ensure the right to life with dignity and without fear, of all the affected villagers of this impoverished, troubled and conflict ridden region.

Conclusion

Tribal economy is always influence by outsiders where illiteracy, lack of awareness, unemployment, makes their socio-economical life vulnerable. The approach to the tribal economic development so far has been either aesthetic or political. Not much thought approach to be given toward developing a philosophy for action in tribal area that would harmonise synthesize tribal interests with larger regional and national interests. Voluntary social service agencies have done considerable humanitarian work in the tribal areas. But often their Idealism and sprit of service have not been matched by their understanding of tribal organization, values and problems. They failed to realize that their well intentioned 'reforming of economy' may be injurious to the tribes in terms of their social-cultural integration. It has suggested that many items given to tribals under welfare schemes can be produced in the tribal areas with tribal labour should be based on local resources and distributed through the agency of tribal people. This will reduce the costs, eliminate dependence on outside agency, assure timely supply and above all reduce scope for corruption.

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Prostate Cancer in Baster Region

Khagesh Bharti[1]

Abstract

Prostate cancer remains one of the most prevalent and lest understood of all human malignancies. Pathologic evidence suggests that neoplastic of prostate epithelium begin early in a mans adult life but do not become clinically evident or relevant until decades without treatment. In other cases the cancer grows aggressively responds poorly to therapy and cases death within a few years, the natural history of this enigmatic disease is heterogeneous ranging form a benign and indolent course to one that rapidly progresses causing significant morbidity and mortality.

Introduction

Prostate cancer is malignant tumor that originates in the prostate gland. As with any cancer, if it is advanced or left untreated in early stages it may eventually spread through the blood any lymph fluid to other organs. Fortunately prostate cancer tends to be slow growing compared to other cancer. As many as 90% of all prostate cancers remain dormant and clinically unimportant for decades. Most older man eventually develop at lest microscope evidence of prostate cancer. The prostate gland is an organ that surrounds the urinary urethra in men. It secretes fluid that mixes with sperm to make semen.[**Harvey Simon 2006]**

Prostate cancer (PCA) is the second most common cause of cancer and the sixth leading cause of cancer death among men worldwide. The worldwide PCA burden is expected to grow to 1.7 million new cases and 499 000 new deaths by 2030 simply due to the growth and aging of the global population (**Ferlay et al., 2010)**.

Review of Literature

Prostate cancer is a slow-growing tumor of older men, constituting the most common type of non-skin cancer and the second leading cause of cancer-related deaths in

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American men. In the late 80s and early 90s great attention was given to screening asymptomatic men by measuring concentration of prostate specific antigen (PSA), which eventually led to a significant increase in the detection of clinically insignificant tumors. Despite this increase, mortality due to prostate cancer has decreased every year since 1992. Though the exact pathogenesis is not clear, epidemiological evidence supports a relationship between prostate cancer and serum levels of testosterone. [**Ross RK, Bernstein L, Lobo RA, Shimizu H, Stanczyk F, Pike M, *et al.* 5 Alpha reductase activity and risk of prostate cancer among Japanese and US white and black males. Lancet 1992;339:8879]**

Other risk factors include advanced age, family history, African-American ethnicity, poor diet and cadmium exposure. The frequency of prostate cancer increases exponentially with advanced age and the natural progression to prostate cancer tends to be more aggressive in younger men and those with a family history of the disease. Although controversial, strategies for decreasing prostate cancer mortality have focused on early detection. The most important tumor marker for detection and post-treatment monitoring of prostate cancer is PSA level. However, PSA levels have fallacies. Increased levels of serum PSA can occur in prostate cancer, benign prostate hyperplasia and prostitutes. Conversely, treatment with 5 alpha reeducate inhibitors lowers PSA levels to approximately 50%. However, PSA assessment still remains the best method to detect cancers at a pre symptomatic stage. Newer methods for increasing the sensitivity and specificity of PSA screening tool are currently being adopted.[**Pienta KJ, Esper PS. Risk factors for prostate cancer. Ann Intern Med 1993;118:793-803]**

Material and Method

Material

Study Area

The present study was carried out in total 10 blood sample form patient for prostate cancer study form late Bali ram memorial hospital, jagdalpur [Chhattisgarh.]

Sample collection

The blood sample was callected form people of jagdalpur, chhattisgarh form late baliram memorial hospital. Through plate vail pancture method in PSA vacutainer. Collection of blood samples should be implemented according to the current practices. Serum only may be used separate serum as soon as possible to avoid any hemolysis. Extensive hemolyesis may effect test performance. Extensive hemolysis my affect test performance specimens with observable particular matter should be clarified by centrifugation prior to testing. Suspended fibrin particles or aggregates may yield falsely positve result. Do not use haemolyzed contaminated or tubid sample specimens. Fresh sample specimens are suitable for performing the test.

A. Preparation of Reagent for PSA

1. PSA calibrators 0.4ml/vial- icons A-F
   six [6] vials of references PSA-antigen at levels of 0[A] 5[B], 10[C],
   25[D], 50[E] and 100[f] ng/ml. store at 2-8C. presservative has been added.
2. PSA Enzyme Reagent – 11ml/vial –icon
   One [1] vial containing enzyme labeled antibody biotinylated monoconal mouse IgG in buffer, dye and preservative.store at 2-8c.
3. Streptavidin coated plate- 96 wells –icon
   one 96- well microplate coated with streptavidin and packaged in an alminum bag with a drying agent, store at 2-8C.
4. Wash solution concentrate- 20ml-icon
   One [1] containing a surfactant in buffered saline. A Preservation has been added. Store at 2-30C.
5. Substrate A-7ml/vial-icon S
   One [1] bottle containing tetramethy lbenzidine [TMB] in buffer. Store at 2-8c.
6. Substrate B – 7ml/vial – iconS
   One [1] bottle cantaining hydrogen peroxide [H2O2] in buffer. Store at 2-8c.
7. Stop solution-8ml/vial –icons
   one [1] bottle containing a strong acid [1N HCL]. Store at 2-30C .

B. Preparation of the Sample

The specimens shall be blood seram in type and the useful precutions in rhe collection of venipuncture samples should be observed. For accurate comparison to estabilshed normal values, a fasting moming serum sample should be optained. The blood should be collected in a plain redtop venipuncture tube without additives or anti-coagulants. Allow the blood to clot. Centrifuge the specimen to separate the serum form the cells. Sample my be refrigerated at 2-8c for a maximum period of five [5] days. If the specimen[s] cannot be assayed within this time the sample may be stored at temperatures of 20c for up to 30 days. Avoid use of contaminated devices. Avoid repetive freezing and thawing. When assayed in duplicate 0.050ml of the specimen is required.

Reagents

1. Wash Buffer
   Dilute contents of wash concentrable to 1000ml distilled or deionized water in a suitable storange container. Store at room temperature 20-27C for up 60 days.

2. Working substrate solution
   Pour the contents of the amber vial labeled solution 'A' into the clear vial labeled solution 'B' Place the yellow cap on the clear vial for easy identification. Mix and label accordingly. Store at 2-8C

Glass wares -

1. Test tube 2. Beaker 3. Microteep 4. Micropipette 5. Ependroff tubes

Additional materials required -

1. Water bath / incubator should be maintained at 37c
2. Eillasa with filter at 570-630 nm at 37c or with yellow/ red filter.
3. PSA is stable in serum for 7 days at 2-8c as the this may be released in the samples even without any microbial contamination.

Principle

The essential reagents required for an immunoenzymometric assay include high affinity and specifcity antibodies [enzyme and immobolized], with different and distinct epitope recognition in excess and native antigen. In this procedure, the immobilization takes place during the assay at the surface of a microplate well through the interaction of

streptavidin coated on the well and exogenously added biotinylated monoconal anti-PSA antibody.

Upon mixing monoconal biothinlated antibody the enzyme labeled antibody and a seram containning the native antigen reaction result between the native antigen and the antibodies without comout compertition or steric hindrance to form a soluble sandwich complex the interaction is illustrated by the following equation.

Enz [ab][p]+ Agpsa + btnAB[m] ------ enz ab[p] –Agpsa –btnAB[m]

Procedure

Present study involves following protocols -

1. Format the microplates wells for each serum reference control and patient specimen to be assayed in duplicate replace any unused microwell strips back in to the aluminum bag seal and store at 2-8c .
2. Pipette 0.025 ml [25ul] of the appropriate serum referance, control or specimen in to the assigned wall.
3. Add 0.100ml [100ul] of the PSA enzyme reagent to each well. It is very important to dispence all reagents close to the bottem of the coated well.
4. Swirl the microplate gently for 20-30 seconds to mix and cover.
5. Incubate 30 minutes at room temperature.
6. Discard the contents of the microplate by dacantation or aspiration. If decanting tap and blot the plate dry with absorbent paper.
7. Add 350ul of wash buffer [see reagent preparation section] decent [lap and blot] or aspirate. Repeat two 2 additional times for a total of three 3 washes. An automatic or manual plate washer can be used. Follow the manufactures instruction for proper usage. If a seqeeze bottle is employed, fill each well by depressing the container [avoiding air bubbles] to dispence the wash. Decant the wash and repeat two 2 additional times.
8. Add 0.100ml [100ul] of working substrate solution to all wells (see reagent preparation solution). Always add reagent in the same order to minimize reaction time differences between wells.

9. Incubate at room temperature for fifteen [15] mintes.
10. Add 0.050ml [50ul] of stop solution to each well and mix gently for 15-20 seconds. Always aad reagent in the same order to minimize reaction time differences between wells.
11. Read the absorbance in each well at 450nm [using a reference wavelength of 620-630nm to minimize well imperfections] in a microplate reader. The results should be read within thirty [30] minutes of adding the stop solution.

Calculation of Results

1. Record the absorbance obtained form the printout of the microplate reader as outlined in example 1.
2. Plot the absorbance for each duplicate serum reference versus the corresponding PSA concentration in ng/ml on liner graph paper. Do not average the duplicates of the serum reference before plotting.
3. Draw the best-fit curve through the plotted points.
4. To determine the concentration of PSA for an unknown locate the average absorbance of the duplicate for each unknown on the vertical axis of the graph find the intersecting point on the carve and read the concentration [in ng/ml] form the horizontal axis of the graph [the duplicate of the unknown my be average absorbance [1.142] intersects the dose response curve at [23.6ng/ml] PSA concentration[see figure].

Sample	Well number	Abs[a]	Mean abs[b]	Value ng/ml
Cal a	A1/b1	0.019/0.019	0.019	0.0
Cal b	C1/d1	0.279/0.567	0.276	5.0
Cal c	E1/f1	0.587/0.559	0.563	10.0
Cal d	G1/f1	1.248/1.179	1.213	25.0
Cal e	A2/ b2	0.051/1.947	1.999	50.0
Cal f	C2/ d2	2.892/0.776	2.833	100.0
patient	E3/ f3	1.186/ 1.099	1.142	23.6

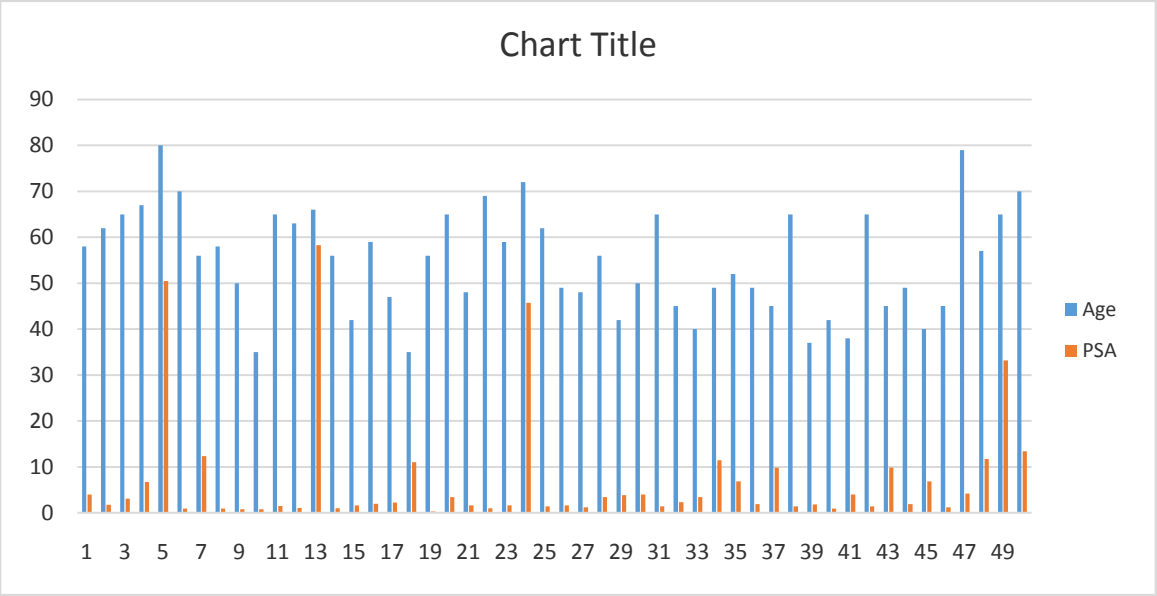
Result & Discussion

By the study performed in the topic diagnostic significance of adenosine demitasse in prostate cancer was conducted at department of biochemistry, late Bali Ram kashyap memorial govt. medical college jagdalpur in 50 patients of man of age group 45-70. The result examined by this study is that out of which 50 males, there was significant increase in parameters in both sexes as compared to controls.

Case	Sex	Age	PSA
1	M	58	4.00
2	M	62	1.76
3	M	65	3.09
4	M	67	6.72
5	M	80	54.3
6	M	70	0.91
7	M	56	12.34
8	M	58	0.90
9	M	50	0.80
10	M	35	0.76
11	M	65	1.47
12	M	63	1.04
13	M	66	58.25
14	M	56	0.98
15	M	42	1.6
16	M	59	1.96

Case	Sex	Age	PSA
17	M	47	2.20
18	M	35	0.02
19	M	56	0.30
20	M	65	3.4
21	M	48	1.6
22	M	69	0.98
23	M	59	1.62
24	M	72	1.12
25	M	62	42.2
26	M	49	1.61
27	M	48	1.2
28	M	56	3.4
29	M	42	3.8
30	M	50	4.0
31	M	65	1.37
32	M	45	2.27
33	M	40	3.4

Case	Sex	Age	PSA
34	M	49	11.4
35	M	52	6.8
36	M	49	1.88
37	M	45	9.8
38	M	65	1.39
39	M	37	1.8
40	M	42	0.9
41	M	38	4.0
42	M	65	1.39
43	M	45	9.8
44	M	49	1.88
45	M	40	6.8
46	M	45	1.2
47	M	79	4.2
48	M	57	11.7
49	M	65	33.5
50	M	70	13.4



Conclusion

The International Agency for Research on Cancer has provided data on Global cancer burden and time trends of various cancers in different parts of the world. The cancer registries under the NCRP network of the Indian Council of Medical Research have provided data on the magnitude of the cancer problem in India. They are also serving as the base in taking measures for prevention and control of cancer in India. The data from cancer registries have indicated that cancer of the prostate is one of the most common malignancies among elderly men with a rising time trend in many areas. Although many studies on prostate cancer have been conducted in different parts of the world, the etiology of this disease is largely unknown. Age >50 years, being an African- American and a family history of prostate cancer are considered to be as established risk factors for prostate cancer. Other factors such as vasectomy, marital history, dietary habits, tobacco use, alcohol intake, etc. have conflicting evidence on the association of prostate cancer.

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Analysis of Thalassemia by Electroforesis in the Region of Bastar

Nisha Netam[1]

Abstract

The diagnosis of b thallassemia by capillary electrophoresis in conjunction with laser-induced fluorescence using poly (ethylene oxide) (PEO) solutions in the presence of electroosmotic flow (EOF). During the electrophoretic separation, PEO solution entered a capillary from the anodic vial by EOF. The separation of a mixture of the polymerase chain reaction (PCR) products (330 and 334 base pairs) from a healthy person and a β-thalassemia patient was accomplished within 15 min at 15 kV using 1.5% PEO containing 2 M urea at 30 _C. The electropherogram patterns instead of migration times were used to diagnose β-thalassemia, with an accuracy of 100% for the analyses of blood samples from suspected patients. The results shown in this study indicate the potential of this simple, rapid, and cost-effective method for the diagnosis of β-thalassemia.

Introduction

Blood is a fluid that sustains life. The components of blood include red blood cells, white blood cells, platelets, and plasma. The red blood cells which are also called erythrocytes (erythro means red; cytes = cells) have the important responsibility of carrying the oxygen throughout the body. Haemoglobin exists in the red blood cells. If a person has too few red blood cells, as determined by a red blood cell count, or if there is not enough haemoglobin in the red blood cell, he or she is diagnosed with anaemia. Because haemoglobin carries oxygen, anaemia often causes people to fatigue.

The term thalassemia is derived from the Greek, thalassa (sea) and haima (blood). Beta-thalassemia includes three main forms: Thalassemia Major, variably referred to as Cooley's Anemia" and "Mediterranean Anemia", Thalassemia Intermedia and Thalassemia Minor also called "β-thalassemia carrier", "β-thalassemia trait" or "heterozygous β-thalassemia". Apart from the rare dominant forms, subjects with

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thalassemia major are homozygotes or compound heterozygotes for beta0 or beta+ genes, subjects with thalassemia intermedia are mostly homozygotes or compound heterozygotes and subjects with thalassemia minor are mostly heterozygotes. According to Thalassemia International Federation, only about 200,000 patients with thalassemia major are alive and registered as receiving regular treatment around the world [3]. The most common combination β-talassemia with abnormal Hb or structural Hb variant with thalassemic properties is HbE /β-thalassemia which is most prevalent in Southeast Asia where the carrier frequency is around 50%.

Although over 700 structural haemoglobin variants have been identified, only three (Hb S, Hb C, and Hb E) reach high frequencies. The homozygous state for the sickle-cell gene results in sickle-cell anaemia, while the compound heterozygous state for the sickle-cell and Hb C genes results in Hb SC disease which, although milder than sickle-cell anaemia, also has important public health aspects. Haemoglobin E, which is the commonest structural haemoglobin variant globally, is innocuous in its heterozygous and homozygous states but, because it is synthesized at a reduced rate, it can interact with β-thalassaemia to produce a condition called Hb E β-halassaemia, which is extremely common and is presenting an increasingly important health problem in many parts of Asia.

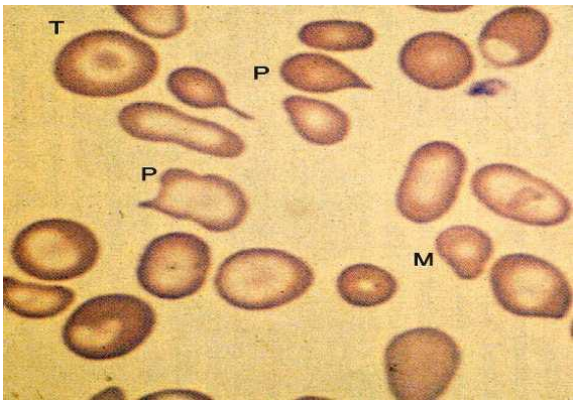

Fig. 1: Peripheral smear from a patient with beta-zero thalassemia major showing more marked microcytosis [M] andanisopoikilocytosis [P] than in thalassemia minor. Target cells [T] and hypochromia are prominent.

Objective

Analysis of blood through electrophoresis for the identification of Thallaseamia (genetic disease).

The diagnosis of b thallassemia by capillary electrophoresis in conjunction with laser-induced fluorescence using poly (ethylene oxide) (PEO) solutions in the presence of electroosmotic flow (EOF). During the electrophoretic separation, PEO solution entered a capillary from the anodic vial by EOF. The separation of a mixture of the polymerase chain reaction (PCR) products (330 and 334 base pairs) from a healthy person and a β-thalassemia patient was accomplished within 15 min at 15 kV using 1.5% PEO containing 2 M urea at 30 _C. The electropherogram patterns instead of migration times were used to diagnose β-thalassemia, with an accuracy of 100% for the analyses of blood samples from suspected patients.

Review of Literature

Globally, iron excess occurs mainly in persons with geographically specific genetic mutations that permit the absorption from the diet of more iron than is physiologically needed. Two main types of hereditary iron overload are well recognized: *1*) hereditary hemochromatosis, which is seen in populations derived from northern Europe, and *2*) the thalassemias and related hemoglobinopathies of South and Southeast Asia, the Middle East, and the Mediterranean . Thalassemia mutations are extremely common: up to 25% of Thai people are carriers of α+-thalassemia, and, in regions of Thailand, Laos, and Cambodia, up to 60% of people are carriers of hemoglobin E (HbE), a hemoglobinopathy caused by a mutation of the B-globin gene. In southern China, which has a population of >350 million, 5% of people are carriers for α-thalassemia and 4% are carriers for β-thalassemia or HbE .Heterozygotes for a-thalassemia 1, B-thalassemia, and HbE typically are asymptomatic and have mild microcytic, hypochromic anemia. In contrast, in thalassemia homozygotes and compound heterozygotes such as HbE/β-thalassemia compound heterozygotes, ineffective erythropoiesis in an expanded marrow stimulates iron absorption even if iron stores are adequate, and this stimulation increases the risk of iron excess when transfusions are given. If heterozygotes have some degree of ineffective erythropoesis and absorb more dietary iron, they may, to a lesser degree than homozygotes and compound heterozygotes, also be at risk of iron excess. In

heterozygotes for hemochromatosis, one study reported that iron absorption from a meal with added iron was 3-fold that in controls; another study found no differences in absorption, despite large populations that are heterozygous for thalassemia in countries implementing iron fortification, there are few data on the absorption and utilization of iron in these persons. Earlier studies in β-thalassemia heterozygotes are difficult to interpret because they used oral solutions of 59Fe given with ascorbic acid that overestimate dietary iron absorption and made comparisons without adjustment for differences in iron status.

described to date. The large majority of mutations are simple nucleotide substitutions or deletions or insertions of oligonucleotides leading to frame shift. Rarely the β-thalassemias are the results of gross gene deletions. Homozygosity for β-thalassemia usually leads to the severe transfusion-dependent phenotype of thalassemia major. Treatment with a regular transfusion program and chelation therapy, aimed at reducing the transfusion iron-overload allows for normal growth and development and extends the life expectancy into the third to fifth decade. Bone marrow or cord blood transplantation (BMT) from an HLA (human leukocyte antigen) -identical sib represents an alternative to traditional transfusion and chelation therapy. More rarely the homozygous state for β-thalassemia leads to the phenotype of thalassemia intermedia. Individuals with thalassemia intermedia present later, and have milder anemia and only rarely require transfusion. However also patients with thalassemia intermedia are at risk to develop iron overload secondary to increased intestinal iron absorption because of ineffective erythropoiesis.

Chemicals

All chemicals for preparing buffer and PEO (M. W.8, 000,000) solutions were from Aldrich (Milwaukee, WI, USA). Ethidium bromide (EtBr) that is cheap and weakly fluorescent was obtained from Molecular Probes (Eugene, OR, USA). TB buffers were prepared from Tris adjusted with boric acid to pH 9.0 and 10.0, respectively. In this manuscript, the molarity of TB buffer refers to that of Tris. Buffers of 800 mM TB (pH 10.0) and 200 mM TB (pH 9.0) were used to fill capillaries and prepare 1.5% PEO (with/without urea), respectively.

METHODOLOGY

Study Area

The present study was carried out from newly diagnosed patients from; Late Baliram Kashyap Memorial Govt. Medical College, Jagdalpur, C.G.

Equipment and Accessories Required

1. CAPILLARYS 2 FLEX – PIERCING System SEBIA, PN 1227.
2. Sample racks supplied with CAPILLARYS 2 FLEX – PIERCING.
3. CAPILLARYS 2 FLEX – PIERCING racks for tubes 11mm, SEBIA, PN 1360, 5 units.
4. Container kit supplied with CAPILLARYS 2 FLEX – PIERCING: Rinse (to fill with distilled or deionised water), wash solution and waste container.
5. Collection tubes with 13mm diameter and their corresponding caps (maximal length of tubes with cap : 90 mm, maximal diameter of cap : 17 mm) : for example, BD Vacutainer, Terumo Venosafe 5 mL, Greiner Bio – one Vaculate 1 2 3 or 4 mL or Sarstedt S-Monovette 4mL tubes (13x75 mm), or collection tubes with 11 mm diameter and their corresponding caps (maximal length of tubes with cap : 90 mm, maximal diameter of cap : 17 mm) : for example Sarstedt S-Monovette 2,7 mL or Kabe Labortechnik Primavette S 2, 6 mL tubes (11x66 mm), or collection tubes with equivalent dimensions approved for clinical assays.
6. Tubes and caps for Controls, SEBIA, PN 9202, (20 units) or PN 9205 (500 units); conical tubes and their caps to analyze blood controls with the CAPILLARYS 2 FLEX – PIERCING instrument.
7. Wadge adapters for tubes for controls SEBIA, PN 9203, 10 units (or supplied with CAPILLARYS 2 FLEX – PIERCING).
8. Boxes for controls storage, SEBIA, PN 2082: 2 boxes for storage of dilution segments containing hemolyzed Controls.

Samples for Analysis

Sample collection and storage

Fresh anticoagulated whole blood samples collected in tubes K2EDTA OR K3EDTA as anticoagulant are recommended for analysis. Avoid anticoagulants containing

iodoacetate. Blood must be collected according to established producers used in clinical laboratory testing. Samples may be stored for up to 7 days between 2 and 8 ºC.

NOT E: Samples should not be stored at room temperature (15 to 30 ºC)!

Progressive haemoglobins (Hb) degradation may occur for stored between 2 to 8^0C.

When than blood sample is stored for more than 7 days at 2 – 8 ^{0}C.

1. a weak fraction, corresponding to methemoglobin, appears in the Hb S migration zone,
2. when Hb C is present, a fraction corresponding to degraded Hb C appears more anodic than Hb A2 which does interfare with it (Z(E) zone, see the table in paragraph "Interpretation").
3. when Hb O-Arab is present, a fraction corresponding to degraded Hb O-Arab appears in the Hb S migration zone (Z(S) zone, see the table in paragraph "Interpretation").
4. when Hb E is present, a fraction corresponding to degraded Hb E appears in the Z(D) zone (see the table in paragraph " Interpretation").
5. when Hb S is present, a fraction corresponding to degraded Hb S appears in the Hb F migration zone (Z (F) zone, see the table in paragraph "Interpretation").
6. when Hb A is present, a fraction corresponding to degraded Hb A ("aging fraction" of Hb A) appears more anodic (Z11 zone, see the table in paragraph "Interpretation").

When Hb F is present (in blood samples from newborn babies), a fraction appears in the Hb A migration zone (Z (A) zone, see the table in paragraph "Interpretation") due to the degradation.

When stored for more than 10 days, viscous aggregates in red blood cells are observed; it is necessary to discard them before the analysis.

Sample Preparation

1. Use directly whole blood samples.
2. Check that all tubes contain 1 mL minimum of blood and are perfectly closed.
3. Vortex for 5 seconds blood samples stored at 2 – 8 ^{0}C for one week.

Particular cases

Analysis of samples without any Hb A or Hb A2 (these samples are perfectly quantified but not identified by 2 ones):

To identify haemoglobin fractions in a sample without any haemoglobin A or haemoglobin A2 it is recommended to prepare this sample according to the following procedure:

1. Vortex for 5 seconds the whole blood sample.
2. In a conical tube for control, mix one volume (50 uL) of whole blood to analyse with one volume (50 uL) of Normal Hb A2 Control and cap the tube.
3. Vortex for 5 seconds.
4. Place the tube with a wedge adapter on a sample rack of the CAPILLARYS 2 FLEX-PIERCING instrument.
5. Slide the sample rack into the CAPILLERYS 2 FLEX-PIERCING instrument.
6. Perform the analysis of this sample according to the standard procedure like a usual blood sample.

The results are then automatically considered by the software for the data analysis.

IMPORTANT: For a sample without any Hb A or Hb A2 prepared according to this procedure, the result obtained with the mixed sample will enable presumption variant identification due to the prepared of the haemoglobin fractions in the appropriate identification zones. Do not report the relative quantification from the mixed sample result.

The relative quantification of haemoglobins should be reported utilizing the initial, unmixed sample result (without any dilution in the blood control).

Samples to Avoid

1. Avoid coagulated blood samples.
2. Avoid aged improperly stored blood samples; the automated hemolysis of samples may be disturbed by viscous aggregates in red blood cells. Then degradation products (as artefacts) may affect the electrophoretic pattern.
3. In these 2 previous cases, aggregates in red blood cells may affect the collection of the sample by the probe.
4. Do not analyze directly tubes containing less than 1mL of blood sample, the analysis should be affected (see particular cases).

5. The CAPILLARYS HEMOGLOBIN (E) procedure performed with the CAPILLARYS 2 FLEX-PIERCING instrument has not been evaluated in the neonate / newborn population (age range – birth to 28 days). SEBIA does not make any claim for validation of neonatal samples under the age of 28 days and reporting results on such samples in the own responsibility of the biologist. Each laboratory should refer to its internal procedures for validation and reporting patients under the age of 28 days.

Procedure

The CAPILLARYS 2 FLEX-PIERCING instrument is a multiparameter instrument for haemoglobins analysis on parallel capillaries. The haemoglobins assay uses 8 capillaries to run the samples.

The sequence of automated steps is as follows:

1. Bar code reading of sample tubes (for up to 8 tubes) and sample racks;
2. Mixing of blood samples before analysis;
3. Sample hemolysis and dilution from primary tubes in dilution segments;
4. Capillary wasting;
5. Injection of hemolyzed samples;
6. Hemoglobin separation and direct detection of the separated hemoglobins on capillaries.

The manual steps include:

1. Placement of sample tubes (with caps) in sample racks in position 1 to 8;
2. Placement of new dilution segments in sample-racks;
3. Placement of racks on the CAPILLARYS 2 FLEX-PIERCING instrument;
4. Removal of sample-racks after analysis.

Preparation of Capillarys Analysis

1. Switch on CAPILLARYS 2 FLEX-PIERCING instrument and computer.

2. Set up the software, enter and the instrument automatically starts.
3. The CAPILLARYS HEMOGLOBIN (E) kit is intended to run with "HEMOGLOBIN" analysis program from the CAPILLARYS 2 FLEX-PIERCING instrument. To select "HEMOGLOBIN (E)" analysis program and place the CAPILLARYS HEMOGLOBIN (E) buffer and hemolyzing solution vials in the instrument, please read carefully the CAPILLARS 2 FLEX-PIERCING instrument manual.
4. The sample rack contains 8 positions for sample tubes. Place up to 8 capped sample tubes with whole blood on each sample rack (positions 1 to8); the bar code of each tube must be visible in the opening of the sample rack.
5. Position a new dilution segment on each sample rack will be ejected if the segment is missing.
6. Slide the complete sample carrier(s) into the CAPILLARYS 2 FLEX-PIERCING instrument through the opening in the middle of the instrument. Up to 13 sample racks can be introduced successively and continuously into the instrument. **When analysing a control blood sample, it is advised to use the sample rack No. F0 intended for control blood sample with specific tubes, caps and the wedge adapter for tubes for controls.**
7. Remove analysed sample racks from the plate on the left side of the instrument.
8. Take off carefully used dilution segments from the sample rack and discard them.

Dilution – Migration – Description of the Automated Steps

1. Bar codes are read on both sample tubes and sample racks.
2. Mixing of tubes.
3. Samples are diluted in hemolysing solution and the sample probe is rinsed after each sample.
4. Capillaries are washed.
5. Diluted samples are injected into capillaries.
6. Migration is carried out under constant voltage for about 8 minutes and the temperature is controlled by Peltier effect.
7. Hemoblobins are detected directly by scanning at 415 nm and an electrophoretic profile appears on the screen of the instrument.

Result Analysis

1. At the end of the analysis relative quantification of individual haemoglobin fractions is performed automatically and profile can be analysed; the haemoglobin fractions, Hb A, Hb F and Hb A2 are automatically identified; the Hb A fraction is adjusted in the middle of the review window. The resulting electrophoregrams are evaluated visually for pattern abnormalities.
2. The potential positions of the different haemoglobin variants (identified in zones called Z1 to Z15) are shown on the screen of the instrument and indicated on the ticket. The table in paragraph “Interpretation” shows know variants which may be present in each corresponding zone.
3. When the software identifies a hemoglobin fraction in a defined zone, the name of this zone is framed.
4. Patterns are automatically adjusted with regard to Hb A and Hb A2 fractions their interpretation:
    a) When Hb A and/or Hb A2 fractions are not detected on an electrophoretic pattern, a yellow warning signal appears, the adjustment is performed using the position of the Hb A fraction on the two previous patterns obtained with the same capillary; then, there is no fraction identified (except when Hb C is detected in this case, Hb A2 and Hb C fractions are identified);
    b) when Hb F is detected on an electrophoretic pattern, without any detection of Hb A, the yellow warning signal does not appear, the adjustment is then performed using the position of the Hb F fraction, and Hb F and/or Hb A and/or Hb A2 fractions are identified;
    c) when the adjustment is not possible, a red warning signal appears, Hb F and Hb A2 fractions are then not identified (Call SEBIA);
    d) when optical density (OD) is insufficient on a migration control electrophoretic pattern (obtained with the Normal Hb A2 Control, identified with its bar code label on the sample rack No. 0), a warning message is displayed in order to consider or remove this analysis for the determination of Hb A fraction position. Then, a purple warning signal appears on the review window and Hb A and Hb A2 fractions are not identified.

e) In all cases, the different migration zones (Z1 to Z15) do not appear neither on the screen of the instrument, nor on the ticket result.

f) On the electrophoretic pattern, the curves of Hb A2 and Hb C fractions, are calculated and redrawn by fitting with adjustment (or fitted) and are overlaid with the native curve. This display allows the Hb A2 fraction quantification if Hb C is present in the sample.

End of Analysis Sequence

At the end of each analysis sequence, the operator must initiate the "shut down" procedure of the CAPILLARYS 2 FLEX-PIERCING instrument in order to store capillaries in optical conditions.

Filling of Reagent Containers

The CAPILLARYS 2 FLEX-PIERCING instrument has a reagent automatic control.

Result & Discussion

Result

By the study performed by us in the topic "ANALYSIS OF THALASSEMIA BY SEBIA ELECTROPHORESIS IN THE REGION OF BASTAR" in 22 patients of age group 0 – 1, 5 – 20, 20 - 40, and 41 – 55. The result we examined by this study is that the mean value of age group of the 22 patients is 23.95. Direct detection at 415 nm in capillaries yields relative concentrations (percentages) of individual haemoglobin zones. Reference values for individual major electrophoretic haemoglobin zones in the CAPILLARYS 2 FLEX-PIERCING instrument have been established from a healthy population of 113 adults (men and women) with normal haemoglobin values using HPLC technique:

Haemoglobin A: comprised between 96.7 and 97.8 %

Haemoglobin F: ≤ 0.5% (*)

Haemoglobin A2: comprised between 2.2 and 3.2 %

Most haemoglobinopathies are due to substitution by mutation of a single amino acid in one of four types of polypeptide chains. The clinical significance of such a change depends on the type of amino acid and the site involved. In clinically significant disease, either the α-chain or the β-chain is affected.

More than 1400 variants of adult haemoglobin have been described. The first abnormal haemoglobin studied and the most frequently occurring have an altered net electric charge leading to an easy detection by electrophoresis.

There are five main abnormal haemoglobins which present a particular clinical interest: S, C, E, O-Arab and D.

The CAPILLARYS HAEMOGLOBIN (E) kit is intended for the identification of haemoglobinopathies and thalassemias.

Haemoglobin S

Haemoglobin S is the most frequent. It is due to the replacement of one glutamic acid (an acidic amino acid No. 6) of the β-chain by valine(a neutral amino acid): when compared to Hb A, its isoelectric point is elevated and its total negative charge decreased with analysis pH. Its electrophoretic mobility is therefore increased in the capillary and this haemoglobin is faster than A fraction. With alkaline buffered CAPILLARY HAEMOGLOBIN (E) procedure, haemoglobin S migrates A and A2 fractions, next to Hb A2.

Haemoglobin C

One glutamic acid of the β-chain is replaced by lysine (a basic amino acid No. 6): its mobility is strongly reduced. When compared to Hb A, its isoelectric point is highly elevated and its total negative charge decreased with the analysis pH. Its electrophoretic mobility is therefore increased in the capillary and this haemoglobin is faster than A fraction which allows its differentiation. Haemoglobin C, E and O-Arab are not superimposed on the electrophoretic pattern and are easily identified.

Haemoglobin E

One glutamic acid of the β-chain is replaced by lysine. With CAPILLARYS HAEMOGLOBIN (E) procedure, haemoglobin E migrates just anodically behind haemoglobin A2 and is totally separated from it then, when haemoglobin E is present, A2 fraction can be measured to detect β-thalassemia.

According to our study we found that Hb A value is comprises between 25 – 70 %, Hb F comprises between 0.5 – 12 %, Hb S zone have 25 – 62 % and Hb A2 value comprises

between 2.0 – 3.4 % but in normal patient the value of Hb A value comprises between 96.8 – 97.8 % and Hb A2 value comprises between 2.2 – 3.2 %.

According to my serve in small region of Bastar on an anaemic patient I found that most of the patients which are trait belong to schedule tribe (ST).

Patients	Age	Gender	Caste	Electrophoresis Thalassemia Result
Patient 1	8	F	ST	Normal
Patient 2	26	F	ST	Normal
Patient 3	32	F	Gen	Trait
Patient 4	17	F	ST	Normal
Patient 5	22	F	OBC	Normal
Patient 6	25	F	ST	Normal
Patient 7	16	F	ST	Trait
Patient 8	23	F	ST	Normal
Patient 9	22	F	ST	Normal
Patient 10	20	F	ST	Normal
Patient 11	24	F	SC	Normal
Patient 12	23	F	OBC	Normal
Patient 13	51	M	Gen	Normal
Patient 14	28	M	SC	Normal
Patient 15	40	M	ST	Trait
Patient 16	7	M	OBC	Diseased
Patient 17	23	F	ST	Normal
Patient 18	34	F	OBC	Normal
Patient 19	23	F	ST	Trait
Patient 20	21	F	ST	Normal
Patient 21	22	F	Gen	Normal
Patient 22	20	F	SC	Normal

Table : Different Age Groups & their Caste

Discussion

According to Michael B Zimmermann, Suthat Fucharoen, Pattanee Winichagoon, Pornpan Sirankapracha, Christophe Zeder, Sueppong Gowachirapant, Kunchit Judprasong, Toshihiko Tanno, Jeffery L Miller, and Richard F Hurrell. Iron utilization was approximately15% lower in α-thalassemia 1 or β-thalassemia heterozygotes than in controls. When corrected for differences in serum ferritin, absorption was significantly higher in the α- and β-thalassemia groups, but not the HbE heterozygotes, than in controls. HbE/β-thalassemia compound heterozygotes had lower iron utilization and higher iron absorption and body iron than did controls. Nontransferrin-bound iron and growth differentiation factor 15 were higher in the compound heterozygotes, but not in the other groups, than in the controls.

Transfusion and Chelation Therapy

According to my studies α-thalassemias are characterised by the α-chains consequently affecting the synthesis of all normal haemoglobins the excess of synthesis of the β and γ-chains induces the formation of tetramers withot any α-chain:

- Haemoglobin Bart`s = γ4
- Haemoglobin H = β4

Haemoglobin S present a low isoelectric point with a CAPILLARYS HAEMOGLOBIN (E) procedure, it migrates more anodic then Haemoglobin A (and may appear as one or several fractions).

β-thalassemias are characterised by the decrease of synthesis of the β-chains. Therefore Haemoglobin F and A2 percentages are increased with respect to Haemoglobin A and β-chain variants. With CAPILLARYS HAEMOGLOBIN (E) procedures, values obtained for different normal haemoglobin fractions allow the detection of β-thalassemias.

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Production, Optimization and Purification of Polypeptides Antibiotic-Bacteriocin and Polymyxin A from Lactobacillus Delbrueckii and Bacillus Polymyxa

Richa Saraf[1]

Abstract

Antibiotic is one of the important commercially exploited secondary metabolites produced by bacteria and used in the wide range. Most of the antibiotics used today are isolated from the microbes. Samples were collected from Hyderabad. ***Lactobacillus delbruekii*** *was isolated from soil and* ***Bacillus poymyxa*** *was isolated from textile effluent.* ***Lactobacillus delbruekii*** *and* ***Bacillus poymyxa*** *were characterized based on colony morphology, cell shape and biochemical tests. They were screened for the production of bacitracin and polymyxin on nutrient media against test organisms viz:* ***E.Coli, pseudomonas, Proteus vulgaries & Staphylococcus aureus.***

Introduction

There are varieties of microorganisms present in the soil including bacteria that can be established in any natural environment.

Bacteria are most important and omnipresent. They are minute, unicellular, primitive and non-chlorophyll containing microorganism. [A.Gowsalya, 2014]

Antibiotics revolutionized medicine in the 20th century, and have together with vaccination led to the near eradication of diseases such as tuberculosis in the developed world. [The World Health Organization. April 30, 2014].

Antibiotics work in one of a few ways: by either interfering with the bacteria's ability to repair its damaged DNA, by stopping the bacteria's ability to make what it needs to grow new cells, or by weakening the bacteria's cell wall until it bursts.[Maria Trimarchi, 1998]

Several antibiotics are also effective against fungi and protozoans, and some are toxic to humans and animals, even when given in therapeutic dosage.[V. Ponnusami, 2014]

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Antibiotics may cause vaginal infections (what we commonly call yeast infections), as well as upset stomach and diarrhea, among other problems. [K.R. Sugumaran, 2014]

The increase in bacterial strains that are resistant to conventional antibacterial therapies has prompted the development of bacterial disease treatment strategies that are alternatives to conventional antibacterial.[Marquez B (2005)]

Lactic acid bacteria lack the ability to synthesize cytochromes and poryphirins and therefore cannot generate ATP, they obtain ATP only through sugar fermentation since lactic acid bacteria does not use oxygen in their energy production. (Michaela *et al*., 2009).

These bacteria are differentiated from the other microorganisms by the ability to convert hexoses to lactic acid. Lactic acid bacteria can be generally divided into two groups based on the sugar fermentation pathways, those organisms which produce exclusively lactic acid as end product are termed as Homofermentative lactic acid bacteria, and organisms which produce acetic acid, ethanol, CO2 in addition to lactic acid are termed as Heterofermentative LAB. (Cleveland et al.,

In recent years the use of Lactic acid bacteria as "Probiotics" is gaining more importance since because mainly lactobacillus and bifidobacteria may have several therapeutic functions (Berg, 1996).

Lactic acid bacteria (LAB) can produce antimicrobial substances with the capacity to inhibit the growth of pathogenic and spoilage microorganisms.(Marshal et al 1997)

Especially, yoghurt bacteria are effective for the prevention and treatment of some diseases related with pathogen microorganisms by several mechanisms such as production of substances above mentioned which are active at low pH. In addition, they appear to be thee limination of colonization with pathogenic bacteria and the treatment of gastrointestinal tract infections, including those sustained by *Clostridium difficile* and *Helicobacter pylori* (Petti et al., 2008).

Many LAB are resistant to antibiotics. This resistance attributes are often intrinsic and non transmissible. On the other hand, intrinsically antibiotic-resistant strains may

benefit patients whose normal intestinal microbiota has become unbalanced or greatly reduced in numbers due to the administration of various antimicrobial agents (Erdogrul et al, 2006).

Although, there is a lot of research about antimicrobial activity and antibiotic resistance of LAB,. These yoghurt bacteria were tested for their antimicrobial activities against Gram-positive and Gram negative food pathogenic and spoilage microorganisms. Their resistances towards some antibiotics were also observed.[AsliAkpinar et al ,2011].

Bacillus species are gram-positive aerobic or facultative anaerobic, sporulating rod shaped bacteria that are widely spread in nature, being implicated in food poisoning.[Graumann P.,2007].

Bacillus species exhibit a wide range of physiologic abilities that allow the organism to flourish in every environment and compete favorably with other organisms within the environment, due to its ability to form spores produce metabolites that are heat stable, cold, radiation, and desiccation disinfect ants and have antagonistic effect on other microorganisms.[Fleming A.2008].

Since that time to date, there has been continues search for more effective antibiotics that can stand the emerging menace of drug resistance among microorganisms worldwide [Mallampalli A., 2008].

Resistance to antibiotics has resulted in morbidity and mortality from treatment failures and increased health care costs. The increases in antibiotics resistant have been attributed to inappropriate use, inadequacies on the part of the manufacturers and leads to the steady decline of effective antibiotics annually worldwide [Blomberg B., 2008].

Review of literature

Bacteria are omnipresent; we can isolate bacteria from soil, air, water. Lactic acid bacteria exhibited antimicrobial activity. Certain *Bacillus polymyxa* strains that associate with many plant species have been used effectively in the control of plant pathogenic fungi and bacteria. In this we review the possible mechanism of action by which *Bacillus*

polymyxa promotes plant growth and suppresses some plant diseases. The potential application of secondary metabolites as consumer friendly bio preservatives either the form of protective cultures are as additives is significant besides being less potentially toxic or carcinogenic than current antimicrobial agents, lactic acid bacteria and their byproducts have been shown to be more effective and flexible in several applications. The further studies will be focused on the optimization and characterization of these antibacterial compounds.

Biochemical Tests

A. Fermentation of Carbohydrates (Glucose Fermentation)

Procedure

1. Fermentation medium was prepared.
2. Broth was taken into fermentation tubes, was autoclaved at 12lb pressure for 15 minutes.
3. Each of the specified fermentation tubes of media was labeled with the name of the organism to be inoculated.
4. Sugar fermentation broths was inoculated with each bacterium and kept one uninoculated tube as a comparative control.
5. All the inoculated and uninoculated tubes were incubated at 35^{o}C for 24-48 hours.

B. Catalase Test

Principle

During aerobic respiration in the presence of oxygen, microorganisms produced hydrogen peroxide(H_2O_2) which is lethal to the cell. The enzyme catalase present in some microorganisms breaks down H_2O_2 to water and oxygen as shown below and helps them in their survival.

2 H_2O_2-----catalase---> 2 H_2O + O_2 (g)

Catalase test is performed by adding H_2O_2 to tryticase soy agar slant culture . release of free oxygen gas bubbles is a positive catalase test.

Procedure

1. Trypticase agar slant was prepared.
2. Medium was poured in culture tube and flask.
3. Autoclaved at 15lb pressure for 15 minutes.
4. Trypticase agar slant was inoculated with culture.
5. Uninoculated trypticase soy agar slant was kept as control.
6. Culture was incubated at 37^{o}c for 24-48 hours.
7. H_2O_2 was allowed to flow over the growth of each slant culture.

C. MR-VP (Methyl Red and Voges Proskauer) test

Procedure

1. MR-VP broth was prepared.
2. 5ml broth was poured in each tube and sterilized by autoclaving at 15 lb pressure for 15 minutes.
3. MR-VP tubes were inoculated with isolated bacteria and kept one as comparative uninoculated control.
4. All tubes were incubated at 35^{o}c for 48 hours.
5. Drops of Methyl red indicator was added to each test tube.

D. Citrate Utilization Test

Procedure

1. Simmon's citrate agar slants were prepared.
2. Simmon's citrate agar slants were inoculated by isolated bacteria by means of a stab and streak inoculation. One tube was kept as an uninoculated comparative control.
3. All slants were incubated at 37^{o}c for 48 hours.

E. Mannitol test

Procedure

1. Fermentation medium was prepared.
2. Broth was taken into fermentation tubes, was autoclaved at 12lb pressure for 15 minutes.
3. Each of the specified fermentation tubes of media was labeled with the name of the organism to be inoculated.
4.

5. Sugar fermentation broths was inoculated with each bacterium and kept one uninoculated tube as a comparative control.
6. All the inoculated and uninoculated tubes were incubated at 35^{o}C for 24-48 hours

Antibiotic Production from Lactobacillus Delbrueckii

A. Crude Antibiotic

1. From each plate of MRS agar, colonies with different morphologies were picked up randomly and transferred into the tubes containing MRS broth and were incubated in aerobic conditions at 37°C for 48 hours.
2. After 48 hours of incubation antibiotic was produced and pH was decreased to 6 because of acid production.
3. It was crude antibiotic. It was then centrifuged at 10000 rpm for 10 minutes.
4. After centrifugation supernatant was taken and pellet was discarded.
5. Antimicrobial assay of this crude antibiotic was done against test organisms.

(Chandra Mouli Lalam, P. Y. Naidu, T. Srinivasan)

B. Ion exchange chromatography

Procedure

1. 6 elutes with 25 Mm Tris-HCL was prepared,25mM, 50mM, 75mM, 100mM, 125mM, 150mM Nacl in 6 different test tubes.
2. Column preparation: column was washed with ethanol, and then washed with distilled water.
3. DEAE was added to the column, leave it for setting
4. The buffer was eluted
5. Enzyme was added in to the column leave it for settling.
6. The sample was collected in to the test tubes.
7. Elute -1 was added in to the column, leave it for settling, sample was collected in to the same test tube.
8. Elute -2 was added in to the column, leave it for settling, sample was collected in to the same test tube.
9. Like this, the elution process for elutes 3, 4, 5 & 6 also was performed.
10. Antimicrobial assay of these elute was done with different test organisms.

Production of Antibiotic from Bacillus Polymyxa

A.Crude Antibiotic

1. Broth was inoculated by bacillus polymyxa and incubated in aerobic conditions at 37°C for 48 hours.
2. After 48 hours of incubation antibiotic was produced.
3. It was crude antibiotic. It was then centrifuged at 10000 rpm for 10 minutes.
4. After centrifugation supernatant was taken and pellet was discarded.
5. Antimicrobial assay of this crude antibiotic was done against test organisms.
6. Purification of crude antibiotic by adsorption through activated charcoal

Procedure

1. 2% of activated charcoal was added in crude antibiotic and stirred continuously on an ice pack for 10-15 minutes to allow complete adsorption.
2. Solution was then filtered by using filter paper.
3. The activated charcoal particles containing the antibiotics were washed with a mixture of ethanol: acidified water (30: 70, pH 2.0).
4. The elute collected was the purified antibiotic.
5. Further, the purified antibiotic are tested upon cultures of bacteria by agar diffusion method and the zone of inhibition was measured and compared to that of crude antibiotics.

B. Solvent (Acetone) precipitation

Procedure

1. 5 volume of ice cold acetone (-20^{o}c) was added to the sample (dialyzed sample) in an acetone safe container and was vortex thoroughly and leave at -20^{o}c for at least 30 minutes to allow maximum precipitation to occur.
2. Sample was spun down for 10 minutes at 13000-15000rpm.
3. Supernatant was discarded and white pellet was air dried although not completely as this will make resolubilisation more difficult.
4. Finally the pellet was resolubilised in phosphate buffer (pH 7) and store at 4^{o}c for further use.

5. Antimicrobial assay of this purified antibiotic was done with different test organisms.

C. Thin Later Chromatography

Procedure

1. Microscopic slides were taken and was sterilized.
2. Solvent and silica gel plate was prepared.
3. Silica gel was poured in to slides.
4. Kept it to air dry.
5. After this it was kept in hot air oven at100^{o}c for 1 hour to dry it.
6. 10 µl crude and purified antibiotic was poured in to silica plate.
7. Kept it in to the solvent for the movement of solute

Results and Discussion

S.No.	Biochemical test	Colony-2 (isolated from soil)	Colony-4 (isolated from textile effluent)
1	Gram staining	Gram +ve, Bacillus	Gram +ve, Bacillus
2	Acid fast	-ve	NA
3	Endospore	-ve	+ve
4	Starch hydrolysis	-ve	+ve
5	Glucose fermentation	-ve	NA
6	Catalyst test	-ve	NA
7	Mannitol fermentation test	-ve	+ve
8	VP test	NA	+ve
9	Citrate utilization test	NA	-ve
	Isolated organisms	*Lactobacillus delbrueckii*	*Bacillus polymyxa*

Table No .4.3 : Biochemical Tests

S.No.	Name of isolated organisms	Name of test organisms				
		Staphylococcus aureas	Bacillus polymyxa	Proteus vulgaries	pseudomonas	E. Coli
1	Lactobacillus delbrueckii	+ve	+ve	-ve	-ve	-
2	Bacillus polymyxa	+ve	-	+ve	-ve	-ve

Table No. 4.4 : Screening of Isolated Colony

Table No.4.5 : Optimization of Isolated Bacteria

S. No.	Optimization at pH	Zone of inhibition of bacteriocin against Test organisms			
		Staphylococcus aureus	Proteus vulgaries	Bacillus polymyxa	pseudomonas
1	5.0	No zone	No zone	17 mm	No zone
2	6.0	No zone	No zone	14 mm	No zone
3	7.0	No zone	No zone	13 mm	No zone
4	8.0	No zone	No zone	15 mm	No zone
5	9.0	No zone	No zone	13 mm	No zone

Table no. 4.5.1 : Antimicrobial assay of Lactobacillus delbrueckjj after pH Optimization at 5.0, 6.0, 7.0, 8.0, 9.0

S.No.	Optimization at C-source	Zone of inhibition of bacteriocin against Test organisms			
		Staphylococcus aureus	Proteus vulgaries	Bacillus polymyxa	pseudomonas
1	Sucrose	28.00 mm	No zone	25.00 mm	26.75 mm
2	Fructose	20.00 mm	No zone	25.00 mm	25.25 mm
3	Lactose	26.75 mm	No zone	25.50 mm	26.50 mm
4	Maltose	28.00 mm	No zone	15.00 mm	26.50 mm
5	Mannitol	25.75 mm	No zone	23.50 mm	

Table No. 4.5.2 : Antimicrobial assay of Lactobacillus delbrueckjj after corbon source Optimization

S.No.	Optimization at Nitrogen Source	Zone of inhibition of bacteriocin against Test organisms			
		Staphylococcus aureus	Proteus vulgaries	Bacillus polymyxa	Pseudo-monas
1	Tryptone + beef extrct	22 mm	No zone	15 mm	No zone
2	Peptone + yeast extract	20 mm	No zone	15 mm	No zone

Table no 4.5.3 : Antimicrobial assay of Lactobacillus delbrueckjj after nitrogen source Optimization

S.No.	Optimization at incubation time	Zone of inhibition of bacteriocin against Test organisms			
		Staphylococcus aureus	Proteus vulgaries	Bacillus polymyxa	pseudomonas
1	48	40 mm	26 mm	14 mm	No Zone
2	72	19 mm	No zone	18 mm	No Zone

Table No. 4.5.4 : Antimicrobial assay of Lactobacillus delbruecki after 72 hour incubation (Optimization)

S.No.	Optimization at pH	Zone of inhibition of polymyxin against Test organisms			
		Staphylococcus aureus	Proteus vulgaries	E.Coli	Pseudomonas
1	5.0	No Zone	15mm	No Zone	16mm
2	6.0	No Zone	16mm	No Zone	20mm
3	7.0	No Zone	14mm	No Zone	13mm
4	8.0	13mm	No Zone	No Zone	16mm
5	9.0	No Zone	14mm	No Zone	15mm

Table No.4.6.1 : Antimicrobial assay of polymyxin after pH Optimization

S.No.	Optimization at C-source	Zone of inhibition of polymyxin against Test organisms			
		Staphylococcus aureus	Proteus vulgaries	E.Coli	Pseudomonas
1	Dextrose	No Zone	13mm	18mm	No Zone
2	Sucrose	No Zone	No Zone	17mm	No Zone
3	Fructose	15mm	No Zone	No Zone	No Zone
4	Lactose	No Zone	No Zone	No Zone	No Zone
5	Maltose	No Zone	No Zone	No Zone	No Zone

Table no.4.6.2 : Antimicrobial assay of polymyxin after corbon source Optimization

S.No.	Optimization at Nirogen-source	Zone of inhibition of polymyxin against Test organisms			
		Staphylococcus aureus	Proteus vulgaries	E.Coli	pseudomonas
1	Tryptone	No zone	22mm	15mm	16mm

Table no 4.6.3 : Antimicrobial assay of polymyxin after nitrogen source Optimization

S. No.	Optimization at incubation time	Zone of inhibition of polymyxin against Test organisms			
		Staphylococcus aureus	Proteus vulgaries	E.Coli	pseudomonas
1	24	60 mm	14 mm	No zone	15 mm
2	48	14mm	20mm	15mm	16mm
3	72	21.5	13.25	14.5	No Zone

Table No. 4.6.4 : Antimicrobial assay of polymyxin after incubation (Optimization)

S .No.	Time in hours	OD
1	1 hr.	0.88
2	2 hrs.	0.80
3	3 hrs.	0.99
4	4 hrs.	1.00
5	5 hrs.	1.02
6	6 hrs.	1.02
7	7 hrs.	0.91
8	8 hrs.	0.83
9	9 hrs.	0.72
10	10 hrs.	0.72

Table no 4.7 : OD of Lactobacillus Delbrueckii

S .No.	Time in hours	OD
1	1 hr.	0.76
2	2 hrs.	0.74
3	3 hrs.	1.00
4	4 hrs.	1.00
5	5 hrs.	0.99
6	6 hrs.	0.98
7	7 hrs.	0.80
8	8 hrs.	0.83
9	9 hrs.	0.72
10	10 hrs.	0.72

Table No.4.8 : OD of Bacillus Polymyxa

Discussion

Soil samples were collected from rhizosphere and water sample was collected from textile effluent. Then serial dilution was performed. Dilution no.6^{th} and 7^{th} was then spreads on agar plate. After 24 hours of incubation bacterial colonies were observed. After gram staining of these colonies, colony-2 and colony-4 both are gram positive, Bacillus. Colony-4 was endospore positive and gave the positive result of starch, glucose, VP and mannitol test. Colony-4 was ferment the glucose and the colour of the medium was change from red to yellow due to the decreased pH. Colony-2 was given the negative starch hydrolysis catalase test. It cannot ferment the glucose. Both the colonies also showed negative citrate test as they are unable to use citrate as a carbon source and it was indicated by no change of the colour of the citrate agar, which remain green as such , from these tests it is confirmed that the colony-2 is Lactobacillus dellbrueckii, and colony-4 is Bacillus polymyxa. The zone of inhibition formed by the Lactobacillus was maximum 40mm in diameter against Staphylococcus aureus and Bacillus polymyxa formed 26.6mm of inhibition zone against proteas vulgaties and staphylococcus aureus

and no zone of inhibition against E.coli. Optimization of production media was also done for both organisms. Optimization of pH, incubation time carbon source and nitrogen source optimization was done. After that antimicrobial assay was done. Lactobacillus delbrueckii showed maximum antimicrobial activity during 48 hour incubation and carbon source optimization. Bacillus polymyxa showed maximum antimicrobial activity after 24 hour incubation and carbon and nitrogen source optimization. At pH optimization both bacteria showed antimicrobial activity against few test organisms only such as for P. vulgaries and S. aureus. Purification of crude antibiotic was done by Acetone purification method, ion exchange chromatography, activated charcoal purification method. After ion exchange chromatography first, second and third elute gave maximum antimicrobial activity than the other elutes. From these work carried out it is confirmed that both Bacteriocin and Polymyxin have antimicrobial activity and showed maximum result when they purified on an ion exchange column. These are the antibiotics which can be taking with new emerging capacity of multi drug resistant bacteria.

Conclusion

Antibiotic, chemical substance produced by a living organism, generally a microorganism that is detrimental to other microorganisms. Antibiotics commonly are produced by soil microorganisms and probably represent a means by which organisms in a complex environment, such as soil, control the growth of competing microorganisms. Microorganisms that produce antibiotics useful in preventing or treating disease include the bacteria and the fungi. *Lactobacillus delbruekii was isolated from soil and Bacillus polymyxa was isolated from textile effluent.* Both are gram positive, Bacillus. The cell-free supernatant from the strains of lactic acid bacteria exhibited antimicrobial activity. The potential application of secondary metabolites as consumer bio preservative is significant. This investigation has revealed the potentials of Bacillus species especially on antibiotic production, it is therefore necessary for isolation and purification of the chemical substances (Metabolites) for detail studies in order to determine their composition and structures. After optimization of both the antibiotics at different pH , nitrogen source, carbon source and incubation time the *Lactobacillus delbruekii* and

Bacillus polymyxa, had wide range of antimicrobial activity against. However, *Bacillus polymyxa*, had little or no effect on the tested organisms. This study suggests that some Bacillus species have potential to produce high quality antibiotics that can be use to control microbial growth in future. Antibiotics from *Lactobacillus delbruekii* showed strong pathogen inhibition ability, which has potential market application.

Significance and Impact of the Study provided a reliable and rapid method for isolation and structural characterization of polypeptide antibiotics from these bacteria.

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Effectiveness of Edutainment on Academic Achievement of Students from Tribal Areas of Bastar

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Abstract

The purpose of this study is to examine the effectiveness of edutainment on academic achievement of students from Tribal areas of Bastar, one of the Districts in Chhattisgarh in central India. A total of 120 students from standard XI of Govt Higher Secondary School have been involved in this study. They shared the same level of knowledge and talents. The respondents were divided into two different groups-control group and experimental group. Each group consisted of 60 students. Both groups were given a set of pre-test. Then the edutainment program was used to support the teaching and learning process in experimental group. While the control group was taught using traditional methods, the experimental group was taught using edutainment program prepared by the scholar. After that, the respondents from both groups were given post-test and a set of questionnaire. The collected data were analyzed using manual statistical methods. Results showed, 96.7% of students (58) agreed that they liked to use edutainment program as it increased their motivation to learn and helped them to learn Communication Skills in English in a better way. Most students agreed that edutainment program is learner-centered and students' friendly and it is suitable to be used in the classroom as it enhanced teaching-learning process.

Keywords : *Edutainment (Education + Entertainment), Edutainment program, Students' achievement, Students' attitude, Students' understanding, Teaching method.*

Introduction

Bastar is one of the Districts in Chhattisgarh in central India. Jagdalpur is its headquarters. Of the total population more than 70% are tribals like Gond, Madia, Muria, Dhruva, Bhatra, Halba, etc. According to 2011 census the average literacy rate of the region is 54.94%. Most of the students in Government schools are first / second

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generation learners. Automatically the achievement level of these students is very low. The reasons for this poor standard in education are lack of motivation, interest, enthusiasm, etc. Edutainment can be the best means for developing these factors and thereby to increase their level of achievement. In recent times, electronic games, home computers and the internet have taken an important place in the life of the students, especially of adolescents. Edutainment has become immensely popular and educationally successful among parents, teachers and children.

What is Edutainment?

Edutainment (Education through entertainment OR Educational entertainment) is content designed to educate and entertain. It includes content that is primarily educational but has incidental entertainment value, and content that is mostly entertaining but contains educational value. It is being used by governments in various countries to disseminate information via television productions, film, museum exhibits, and computer software which use entertainment to attract and maintain an audience to influence viewers' opinions and behaviors.

Types of Edutainment

Educational toys and games, as major part of edutainment, inspire the students to learn in a better way. Communication Satellite plays a quite important role in educational purposes. Radio, Television and Films are efficient means for curriculum transaction, of upgrading teachers and bringing about significant changes in educational practices. Computerized education and internet have been proved effective teaching and learning methods in classrooms. Public Access Areas like Museums and Zoos can serve as efficient means to get firsthand experience and knowledge for the students.

Literature Reviews

As per the study made by Saroj Pandey, 2007, Edutainment as a method of teaching-learning can best be suited to the new constructivist approach in education suggested by National Curriculum Framework 2005. The study made by Esta de Fossard & Robert Lande, 2008, summarized the advantages of entertainment-education as the Nine Ps (9 Ps).

Research Objectives

1. To identify how edutainment can affect the students' achievement in the tribal areas of Bastar
2. To identify whether edutainment can change students' attitude towards study in class in the tribal areas of Bastar

Methodology

1. Participants

This research was conducted in a Government Higher Secondary School located in Adawal, 6 kms away from Jagdalpur in the District of Bastar in Chhattisgarh. Students of standard XI (11) were selected to involve in the research. Out of them 120 students were randomly selected, of almost the same IQ level. The students were divided equally into two different groups: experimental group and control group.

2. Instruments

The main instrument used by the scholar in the study was a set of questionnaires consisting of 2 sections. The first section of the questionnaire covered the questions about the background of the respondents, while the second section consisted of questions that related to student's understanding, interest and teaching methods. The students were given the questionnaire after the learning process using edutainment program prepared by the scholar.

3. Procedure

The first step in this research was to teach both groups of students 'Communication Skills in English'. The control group was taught traditionally using Blackboard, charts, etc. Meanwhile, the experimental group was taught using the edutainment program prepared by the scholar. After the learning session ended, the questionnaire was distributed to the students. Permission to conduct the research was first secured from the school Principal and the sessions took place in the classroom during the English period with the help of Laptop and Projector Screen as well as audio-visual aids. All the respondents were assured that their participation in the study was voluntary and at any time they can opt out.

4. Data Analysis

The data collected during this research was analyzed using manual statistical methods. Two tailed Z-test was used to elicit whether there was a significant difference among the two groups: control group and experimental group. Significance level was decided by taking p values into consideration. The P value / alpha value was set at 0.05.

Findings and Discussion

1. Edutainment Program

This research was conducted using the Edutainment program prepared by the researcher.

2. Pre-test and Post-test Result

Both groups had learnt 'Communication Skills in English' using traditional teaching methods. After the class ended, they had been given a set of questions that tested their understanding towards the topic. After two weeks, the students were being taught again the same topic. This time the experimental group was taught using the edutainment program. The result of the students' achievement is shown in Fig. (a) and Fig. (b). Based on Fig. (a) and Fig. (b), the results indicated that the highest number of students in control group (pre-test) got 14 marks (22 students) while the highest number of students in control group (post-test) got 13 marks (20 students). The highest mark for the students in experimental group (pre-test) was 12 and 15 (both with the number of students 16) while the highest number of students in experimental group (post-test) got 15 marks (30 students). There is a big difference when compared to the result of post test for experimental group. After having a class using edutainment program, half of the students (30 students) got the score with 15 marks.

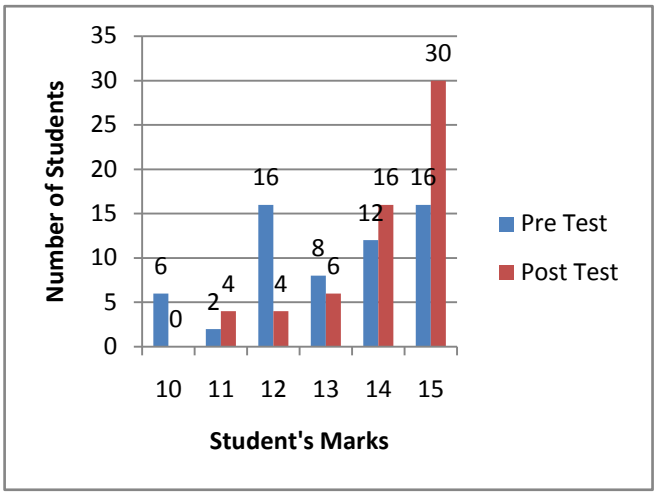

Fig. (a): Pre-test and post-test result for control group experimental

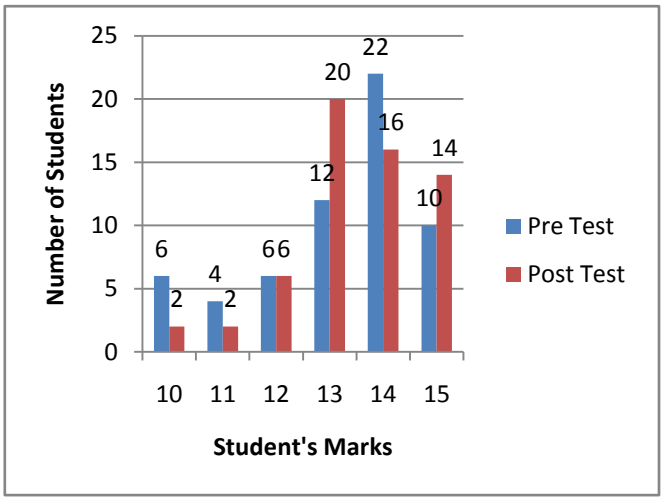

Fig. (b): Pre-test and post-test result for group

3. Analysis of Z-test

Z-test was used to compare the mean score for the responses. As seen in Table I, at the beginning the pre-test means of control group and experimental group were 13.16 and 13.10 respectively. These results showed that the sample's pre-treatment knowledge levels were much closer to each other and there was not a statistical difference between the two groups (p = + / -1.96, p limit is 0.05).

Value	Pre-test control group	Pre-test experimental group
Mean	13.16	13.10
Standard deviation	0.583	0.574
Significance value of z test	-1.992	-2.8

Table 1: Z-test result of pre-test for control group and experimental group

At the end of the treatment, the post-test means of control group and experimental group were 13.46 and 14.23 (as shown in Table II). A statistical significant difference was found between the groups (p = 0.005, p<0.005).

Value	Post-test control group	Post-test experimental group
Mean	13.46	14.23
Standard deviation	0.624	0.774
Significance value of z test	1.875	9.2

Table 2: Z-test result of post-test for control group and experimental group

As per the z-test, the first step was to state the Null Hypothesis.

"There is no effectiveness of edutainment on the academic achievement of students from tribal areas of Bastar."

The Alternative Hypothesis was,

"There is effectiveness of edutainment on the academic achievement of students from tribal areas of Bastar."

The Alpha level (Significance level) was set at .05. The z-score associated with the current significance level is 1.96.

As this is a two-tailed test, the scholar had to consider the left as well as the right tails. Therefore, a value less than -1.96 or greater than 1.96 would lead to the rejection of Null Hypothesis.

In case 1 (Control group), in the pre-test and post-test, the values fall between the specified range. So the Null Hypothesis is accepted.

In case 2 (Experimental group), in the pre-test, the z-value is within the range and therefore the Null Hypothesis is accepted. In the post-test, the z-score is greater than 1.96. Therefore, the Null Hypothesis is rejected and the Alternative Hypothesis is accepted.

4. Questionnaire analysis

The questionnaire was given to the respondents in the experimental group after they had a lesson using edutainment program. The questionnaire was divided into two main sections- Section A that focused on the background of the respondents and Section B that covered the students' response towards edutainment program. In this paper, the discussion of findings is focused on the second section of the questionnaire.

5. Students' Understanding

Fig. (c) shows the evaluation result regarding the students' understanding towards 'Communication Skills in English'. As we can see from the graph, 96.7 % (58 students) of the respondents agreed that they liked edutainment program as it increased their motivation to learn and helped them to learn 'Communication Skills in English' in a better way; 93.3% (56 students) said that the edutainment program increased their understanding of the 'Communication Skills in English' and knowledge of grammatically correct language. 90% (54 students) of the students agreed that edutainment program gave them chance to increase their vocabulary. When asked about the usage of edutainment program in expressing ideas in appropriate manner, 80% (48 students) said that edutainment helped them for expressing ideas in appropriate manner.

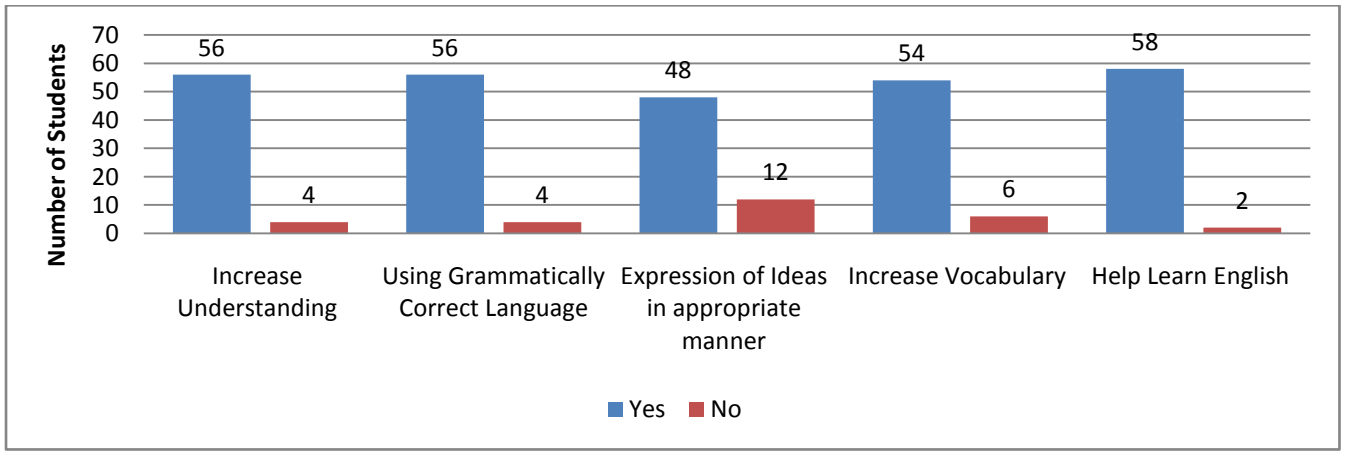

Fig. (c): Evaluation result of students' understanding towards communication skills in English

6. Students' Attitude

The respondents also had been asked about their attitude towards edutainment program. In Fig. (d), 96.7% (58 students) said that they liked to use this program as it helped to increase their interest to learn 'Communication Skills in English' and also increased their motivation towards learning; 93.3% (56 students) said that teaching-learning process became more interesting when they learned the topic using edutainment program; more than 85% (52 students) agreed that the edutainment program helped them in self learning.

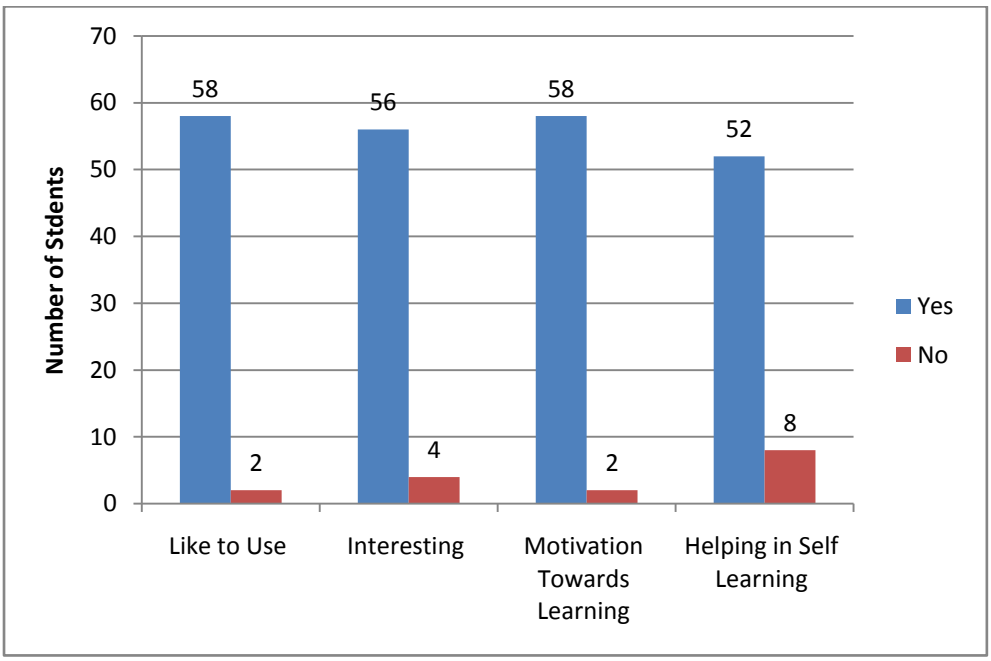

Fig. (d) : Evaluation result regarding students' attitude towards edutainment program

Conclusion

Educating through edutainment program is educating the students for a better tomorrow and for a bright future. It is challenging, but is a great need of the day. This study proved that Edutainment program is successful in increasing the achievement level of students of Bastar.

"50% Education + 50% Entertainment → 100% Edutainment = 100% Achievement" (Fig.(e))

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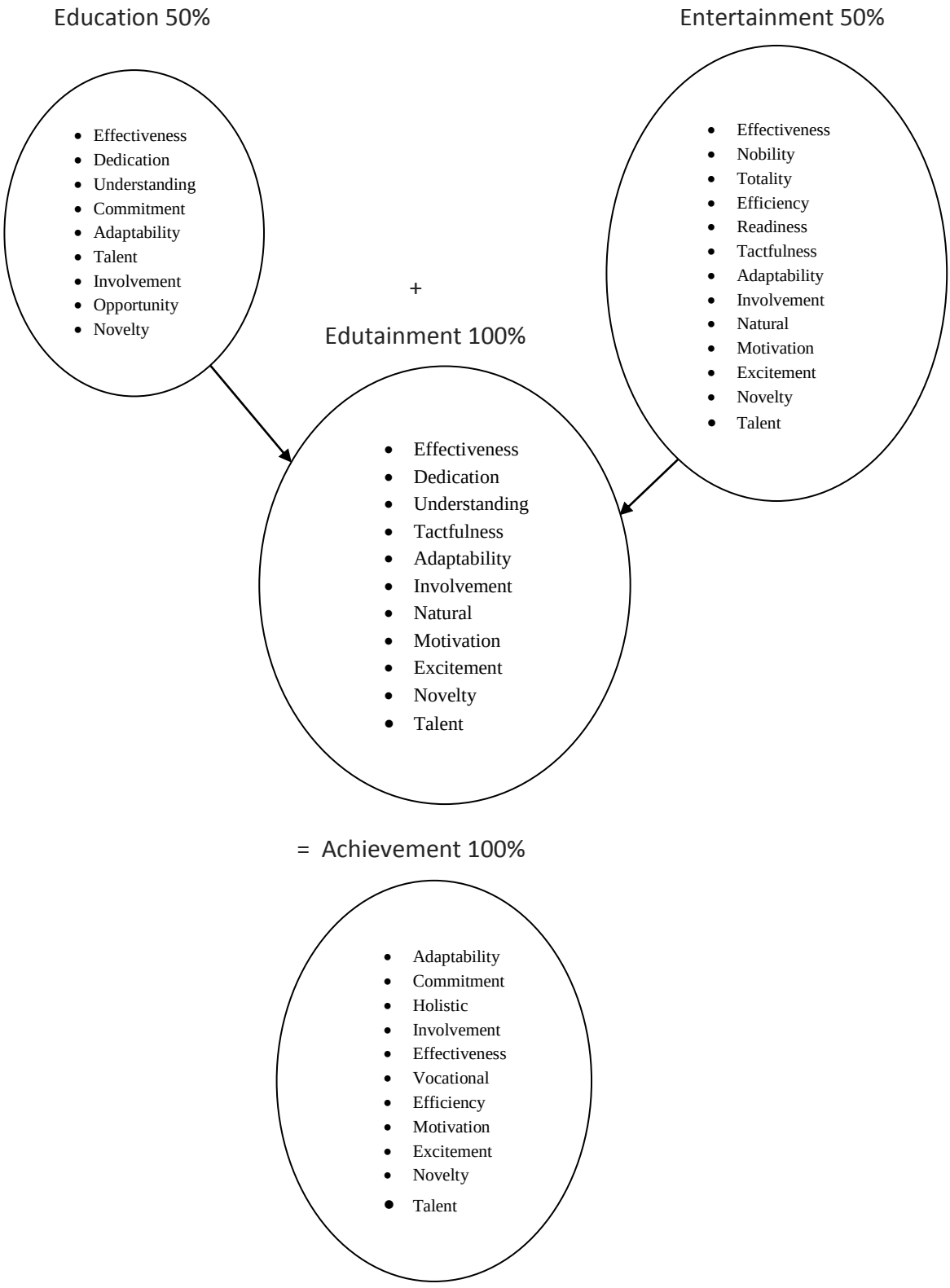

Fig. (e): Schematic representation of the relationship between Education, Entertainment, Edutainment and Achievement

REVIEW ARTICLES

A Review of Programmed Cell Death- Apoptosis

Rahul Singh Thakur[1]

Abstract

Apoptosis, is generally characterized by distinct morphological characteristics and energy-dependent biochemical mechanisms. Apoptosis is considered a vital component of various processes including normal cell turnover, proper development and functioning of the immune system, hormone-dependent atrophy, embryonic development and chemical-induced cell death. Inappropriate apoptosis (either too little or too much) is a factor in many human conditions including neurodegenerative diseases, ischemic damage, autoimmune disorders and many types of cancer. The ability to modulate the life or death of a cell is recognized for its immense therapeutic potential. Therefore, research continues to focus on the elucidation and analysis of th

e cell cycle machinery and signaling pathways that control cell cycle arrest and apoptosis. To that end, the field of apoptosis research has been moving forward at an alarmingly rapid rate. Although many of the key apoptotic proteins have been identified, the molecular mechanisms of action or inaction of these proteins remain to be elucidated. The goal of this review is to provide a general overview of current knowledge on the process of apoptosis including morphology, biochemistry, mechanism of apoptosis in health and disease, detection methods, as well as current approaches in altering cell death pathway.

Introduction

The term apoptosis (a-po-toe-sis) was first used in a now-classic paper by Kerr, Wyllie, and Currie in 1972 to describe a morphologically distinct form of cell death, although certain components of the apoptosis concept had been explicitly described many years previously

(Kerr et al., 1972; Paweletz, 2001; Kerr, 2002). Our understanding of the mechanisms involved in the process of apoptosis in mammalian cells transpired from the investigation of programmed cell death that occurs during the development of the nematode *Caenorhabditis*

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elegans (Horvitz, 1999). In this organism 1090 somatic cells are generated in the formation of the adult worm, of which 131 of these cells undergo apoptosis or "programmed cell death." These 131 cells die at particular points during the development process, which is essentially invariant between worms, demonstrating the remarkable accuracy and control in this system. Apoptosis has since been recognized and accepted as a distinctive and important mode of "programmed" cell death, which involves the genetically determined elimination of cells. However, it is important to note that other forms of programmed cell death have been described and other forms of programmed cell death may yet be discovered (Formigli et al., 2000; Sperandio et al., 2000; Debnath et al., 2005).

Apoptosis occurs normally during development and aging and as a homeostatic mechanism to maintain cell populations in tissues. Apoptosis also occurs as a defense mechanism such as in immune reactions or when cells are damaged by disease or noxious agents (Norbury and Hickson, 2001). Although there are a wide variety of stimuli and conditions, both physiological and pathological, that can trigger apoptosis, not all cells will necessarily die in response to the same stimulus. Irradiation or drugs used for cancer chemotherapy results in DNA damage in some cells, which can lead to apoptotic death through a *p53*-dependent pathway. Some hormones, such as corticosteroids, may lead to apoptotic death in some cells (e.g., thymocytes) although other cells are unaffected or even stimulated.

Some cells express Fas or TNF receptors that can lead to apoptosis via ligand binding and protein cross-linking. Other cells have a default death pathway that must be blocked by a survival factor such as a hormone or growth factor. There is also the issue of distinguishing apoptosis from necrosis, two processes that can occur independently, sequentially, as well as simultaneously (Hirsch, 1997; Zeiss, 2003). In some cases it's the type of stimuli and/or the degree of stimuli that determines if cells die by apoptosis or necrosis. At low doses, a variety

of injurious stimuli such as heat, radiation, hypoxia and cytotoxic anticancer drugs can induce apoptosis but these same stimuli can result in necrosis at higher doses. Finally, apoptosis is a coordinated and often energy-dependent process that involves the activation of a group of

cysteine proteases called “caspases” and a complex cascade of events that link the initiating stimuli to the final demise of the cell.

Morphology of Apoptosis

Light and electron microscopy have identified the various morphological changes that occur during apoptosis (Hacker, 2000). During the early process of apoptosis, cell shrinkage and pyknosis are visible by light microscopy (Kerr et al., 1972). With cell shrinkage, the cells are

Figure A representing a photomicrograph of a section of exocrine pancreas from a B6C3F1 mouse. The arrows indicate apoptotic cells that are shrunken with condensed cytoplasm. The nuclei are pyknotic and fragmented. Note the lack of inflammation.

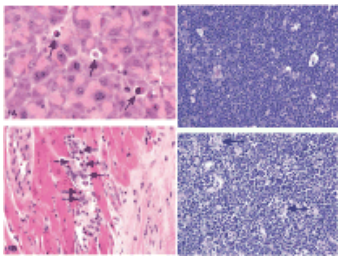
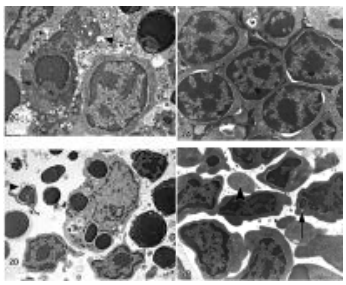


Figure B representing a transmission electron micrograph (TEM) of the normal thymus tissue .The lymphocytes are closely packed, have large nuclei and scant cytoplasm.



smaller in size, the cytoplasm is dense and the organelles are more tightly packed. Pyknosis is the result of chromatin condensation and this is the most characteristic feature of apoptosis. On histologic examination with hematoxylin and eosin stain, apoptosis involves single cells or small clusters of cells. The apoptotic cell appears as a round or oval mass with dark eosinophilic cytoplasm and dense purple nuclear chromatin fragments (Figure A). Electron microscopy can better define the subcellular changes. Early during the chromatin condensation phase, the electron-dense nuclear material characteristically aggregates

peripherally under the nuclear membrane although there can also be uniformly dense nuclei (Figure B).

Mechanisms of Apoptosis

The mechanisms of apoptosis are highly complex and sophisticated, involving an energy-dependent cascade of molecular events . To date, research indicates that there are two main apoptotic pathways: the extrinsic or death receptor pathway and the intrinsic or mitochondrial pathway. However, there is now evidence that the two pathways are linked and that molecules in one pathway can influence the other (Igney and Krammer, 2002). There is an additional pathway that involves T-cell mediated cytotoxicity and perforin-granzyme-dependent killing of the cell. The perforin/granzyme pathway can induce apoptosis via either granzyme B or granzyme A. The extrinsic, intrinsic, and granzyme B pathways converge on the same terminal, or execution pathway. This pathway is initiated by the cleavage of caspase-3 and results in DNA fragmentation, degradation of cytoskeletal and nuclear proteins, cross-linking of proteins, formation of apoptotic bodies, expression of ligands for phagocytic cell receptors and finally uptake by phagocytic cells. The granzyme A pathway activates a parallel, caspase-independent cell death pathway via single stranded DNA damage (Martinvalet et al., 2005).

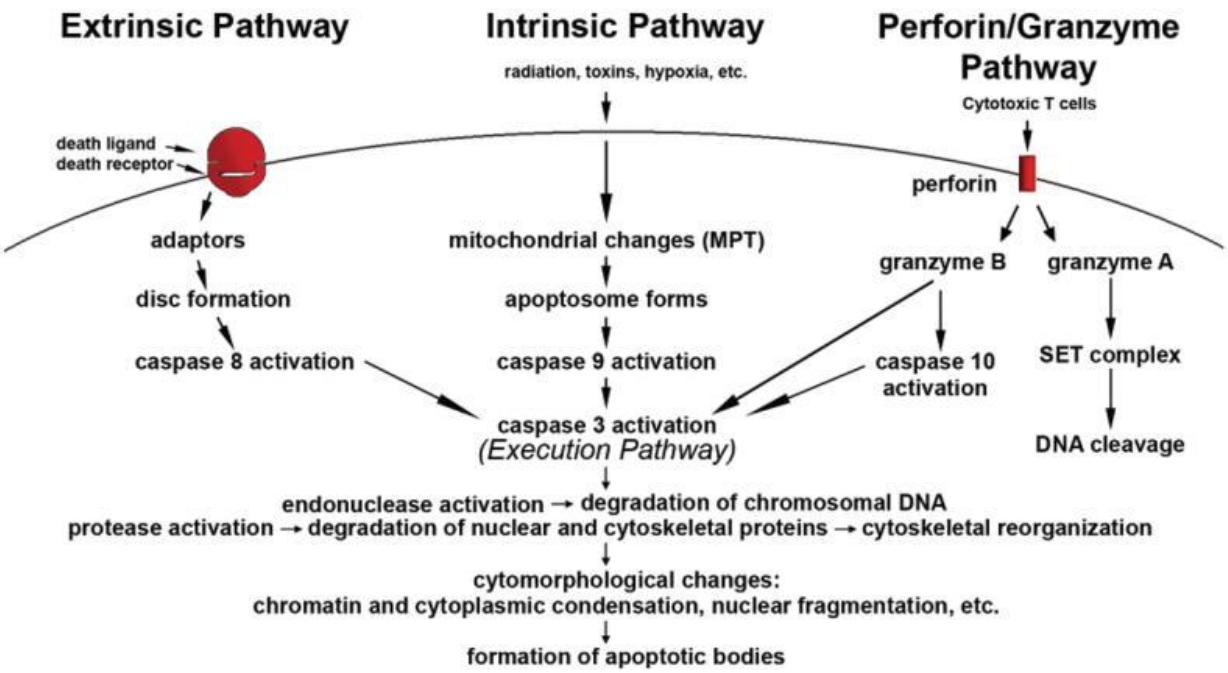


Figure C. Schematic representation of apoptotic events. The two main pathways of apoptosis are extrinsic and intrinsic as well as a perforin/granzyme pathway. Each requires specific triggering signals to begin an energy-dependent cascade of molecular events. Each pathway activates its own initiator caspase (8, 9, 10) which in turn will activate the executioner caspase-3. However, granzyme A works in a caspase-independent fashion. The execution pathway results in characteristic cytomorphological features including cell shrinkage, chromatin condensation, formation of cytoplasmic blebs and apoptotic bodies and finally phagocytosis of the apoptotic bodies by adjacent parenchymal cells, neoplastic cells or macrophages.

Assays for Apoptosis

Apoptosis occurs via a complex signaling cascade that is tightly regulated at multiple points, there are many opportunities to evaluate the activity of the proteins involved. As the activators, effectors and regulators of this cascade continue to be elucidated, a large number of apoptosis assays are devised to detect and count apoptotic cells. However, many features of apoptosis and necrosis can overlap, and it is therefore crucial to employ two or more distinct assays to confirm that cell death is occurring via apoptosis. One assay may detect early (initiation) apoptotic events and a different assay may target a later (execution) event.

The second assay, used to confirm apoptosis, is generally based on a different principle. Multiplexing, which is the ability to gather more than one set of data from the same sample, is another methodology for apoptosis detection that is becoming increasingly popular. There are a large variety of assays available, but each assay has advantages and disadvantages which may make it acceptable to use for one application but inappropriate for another application (Watanabe et al., 2002; Otsuki et al., 2003). Therefore, when choosing methods of apoptosis detection in cells, tissues or organs, understanding the pros and cons of each assay is crucial.

Understanding the kinetics of cell death in each model system is also critical. Some proteins, such as caspases, are expressed only transiently. Cultured cells undergoing apoptosis in vitro

will eventually undergo secondary necrosis. Apoptotic cells in any system can die and disappear relatively quickly. The time from initiation of apoptosis to completion can occur as quickly as 2–3 hours. Therefore a false negative can occur if the assay is done too early or too late. Moreover, apoptosis can occur at low frequency or in specific sites within organs, tissues and cultures. In such cases, the ability to rapidly survey large areas could be useful. In general, if detailed information on the mechanism of cell death is desired, the duration of toxin exposure, the concentration of the test compound and the choice of assay endpoint become critical.

A detailed description of all methodologies and assays for detecting apoptosis is beyond the scope of this article. However, some of the most commonly employed assays are mentioned and briefly described. Apoptosis assays, based on methodology, can be classified into six major groups and a subset of the available assays in each group is indicated and briefly discussed:

- Cytomorphological alterations
- DNA fragmentation
- Detection of caspases, cleaved substrates, regulators and inhibitors
- Membrane alterations
- Detection of apoptosis in whole mounts
- Mitochondrial assays.

Current Approaches in Altering Cell Death Pathways

Inhibitor of Apoptosis Protein-Based Therapeutics : Releasing the Apoptotic Brakes

Inhibitor of apoptosis proteins (IAPs) are acknowledged today as a major control point in the execution of cell death. IAPs comprise a family of caspase-inhibiting proteins characterized by a shared conserved sequence region, termed the BIR domain (Salvesen and Duckett, 2002). Currently, eight endogenous IAPs are known in humans, all of which inhibit apoptosis. Not surprisingly, some of them have been found to be overexpressed in a variety of cancers. So far, the main physiological roles of IAPs seem to be 1) the establishment of a threshold at

which caspases are kept inactive, and 2) providing a pool of active caspases which can rapidly execute death after release. Growing evidence also suggests the participation of IAP proteins in other cellular functions apart from inhibiting caspases, including protein degradation, cell cycle control, and signal transduction (Deveraux and Reed, 1999). Survivin, for instance, is an IAP member containing a single BIR domain which is expressed to high levels in cancer cells, but not in normal cells (Ambrosini et al., 1997). However, it is still unclear whether this is due to an antiapoptotic role of survivin conferring a survival advantage to the tumor cell or due to other reasons. Survivin is proposed to play a role in cell division (Reed and Bischoff, 2000; Uren et al., 2000b) and might therefore be expressed at higher levels in actively dividing cancer cells than in normal differentiated cells.

Conclusions

Apoptosis is regarded as a carefully regulated energy-dependent process, characterized by specific morphological and biochemical features in which caspase activation plays a central role. Although many of the key apoptotic proteins that are activated or inactivated in the apoptotic pathways have been identified, the molecular mechanisms of action or activation

of these proteins are not fully understood and are the focus of continued research. The importance of understanding the mechanistic machinery of apoptosis is vital because programmed cell death is a component of both health and disease, being initiated by various physiologic and pathologic stimuli. Moreover, the widespread involvement of apoptosis in the pathophysiology of disease lends itself to therapeutic intervention at many different checkpoints. Understanding the mechanisms of apoptosis, and other variants of programmed cell death, at the molecular level provides deeper insight into various disease processes and may thus influence therapeutic strategy.

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Study of Analysis Genetic Disorders in Sickle Cell Anaemic Patients by using Sebia Electrophoresis Method

Anjali joshi[1]

Abstract

Sickle cell anaemia primarily affects people with African, Mediterranean, Middle Eastern, and Indian ancestry. Sickle cell anemiea is an inherited disease in which the body is unable to produce normal haemoglobin, an iron containing protein. Abnormal haemoglobin can morph cells that can become lodged in narrow blood vessels, blocking oxygen from reaching organs and tissues. The effects of sickle cell anaemia are bouts of extreme pain, infectious, fever, jaundice, stroke, slow growth, organ and failure.

Introduction

Of the 330,000 babies born with a major hemoglobinopathy worldwide, 275,000 have SCD, making it the major global hemoglobinopathy (Aygun & Odame, 2012; Modell & Darlison, 2008; Weatherall, 2011). SCD patients in the developed world account for only 10% of the world's SCD patient population (Aygun & Odame, 2012). In 2008, Aliyu et al. (2008) reported United Nations estimates that there are between 20 and 25 million people worldwide living with SCD, of which 12–15 million live in Africa. It is estimated that 75–85% of children born with SCD are born in Africa, where mortality rates for those under age 5 range from 50% to 80% (Aygun & Odame, 2012; Makani et al., 2011).

Sickle cell anaemia primarily affects people with African, Mediterranean, Middle Eastern, and Indian ancestry. Sickle cell anemiea is an inherited disease in which the body is unable to produce normal haemoglobin, an iron containing protein. Abnormal haemoglobin can morph cells that can become lodged in narrow blood vessels, blocking oxygen from reaching organs

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and tissues. The effects of sickle cell anaemia are bouts of extreme pain, infectious, fever, jaundice, stroke, slow growth, organ and failure. (Vorvick *et al*., 2010).

In India, newborn screening programmes for sickle cell disorders among tribal and non-tribal populations have only recently been initiated during the last three to five years in south Gujarat, Maharashtra, Chhattisgarh, Odisha and Madhya Pradesh. In the Kalahandi district in Odisha, 1668 newborns were screened and 19.03 per cent of tribals were sickle heterozygous and 36 babies with sickle cell anaemia were identified.(Italia K,2015)

Normal red blood cells are usually round and soft and travel easily through small vessels; however sickle cells are abnormally shaped and stiff, thus causing them to have difficulty travelling through small vessels. Sickle cells can clog vessels depriving tissues of oxygen. As spoken of in two articles, sickle cells have a shorter life span than normal red blood cells. Normal red blood cells can live 120 days compared to sickle cells that only last 10 to 20 days, resulting in anaemia. (Nabili, 2008)

Sickle cell anaemia occurs when a person inherits two sickle cell gene, one from each parent, that cause the red blood cells to change and become crescent shaped. The underlying problem involves hemoglobin, a protein molecule which is a component of the red blood cells that carries oxygen from the lungs to the body's tissues and returns carbon dioxide from the tissues to the lungs (Nabili, 2008) Like most illnesses, sickle cell anaemia has one primary cause. In order for sickle cell anaemia to occur, a sickle cell gene must be inherited from both the mother and the father, so that the child has two sickle cell genes (Shiel, 2006).

Symptoms include chronic anaemia, acute chest syndrome, stroke, splenic and renal dysfunction, pain crises, and susceptibility to bacterial infections. Pediatric mortality is primarily due to bacterial infection and stroke. In adults, specific causes of mortality are more varied, but individuals with more symptomatic disease may exhibit early mortality. Disease expression is variable and is modified by several factors, the most influential being genotype. Other factor sinclude p-globin cluster haplotypes, a-globin gene number, and fetal hemoglobin expression. In recent years, newborn screening, better medical care, parent

education, and penicillin prophylaxis have successfully reduced morbidity and mortality due to (Ashley , et. al 2000)

The p-globin gene is located on the short arm of chromosome 11. It is a member of the globin gene family, a group of genes involved in oxygen transport. Other members of this gene family include the a-, y-,5-, £-, and ^-globin genes. The globin genes are developmentally regulated, such that certain genes are expressed at specific times during human development.

Two f3-globin protein chains combine with two a-globin protein chains and a heme to form the predominant hemoglobin (Hb) found in human adults.(Kark JA, 1987)

A blood test can check for hemoglobin S — the defective form of hemoglobin that underlies sickle cell anaemia. In the United States, this blood test is part of routine newborn screening done at the hospital. But older children and adults can be tested, too.

In adults, a blood sample is drawn from a vein in the arm. In young children and babies, the blood sample is usually collected from a finger or heel. The sample is then sent to a laboratory, where it's screened for hemoglobin S.

Review of Literature

According to ANVISA 2002. Sickle cell anaemia, as clinical expression of the homozygosis of the S hemoglobin gene, is an important genetic abnormality in Brazil, especially in the regions which received large numbers of African slaves. In our country it is estimated that 3 for every 500 Brazilian- Africans have sickle cell anaemia trace and one for every 500 blacks in Brazil is born with some form of the disease. Although there is a higher prevalence in African-descendants, the Caucasian population, especially those from the Mediterranean (Greece, Italy, etc.), Middle East, India, have many cases of sickle cell disease.

According to estimates from the World Health Organization, every year in Brazil we have about 3,500 children born with sickle cell disease. Twenty percent of them will not reach five

years of age, because of complications directly associated with the disease itself. Only early diagnosis and proper treatment can change this scenario. According to data from APAE-Salvador, from August of 2000, when the "foot test" started, neonatal screening for hemoglobin diseases, allowed us to know the true incidence of such disease: 1,655 live newborns with sickle cell anaemia and 1 child with sickle cell trace for every 17 births.

According to BARRY et.al, in 1972 study a high prevalence of hemiplegia was found in 89 patients with sickle-cell disease seen over a 5-year period. Twenty-three patients (26%) had neurological manifestations; hemiplegia occurred in 15 (17%). During the same period, hemiplegia occurred in only 5% of patients with sickle-hemoglobin C disease and in 1.7% of patients with sickle-cell trait; the latter is the same as that in the negro population with normal hemoglobin (1.8%). A review of the English-language literature shows that the high frequency of hemiplegia in the natural history of sickle-cell disease has not been sufficiently emphasized, despite its previous documentation in isolated case reports. Unless better methods for controlling the clinical expressions of this hemoglobinopathy can be found, stroke will remain one of the commonest serious complications in patients with sickle-cell disease.

According to RICHARD P. PERRINE at.al, in 1978. For study 270 Saudi Arabs with homozygous sickle cell anaemia, using chart review, a register (since 1969), and home visiting in 42 cases. Average follow-up for the total group was 10 years. Seventy-four percent of those diagnosed by age 3 years presented on screening or with merely anaemia; 26% presented with illness, abnormal physical findings, or pain. Compared with American or Jamaican blacks, serious complications occurred only 6% to 25% as frequently; leg ulcers did not occur at all; the mortality under age 15 years was 10% as great; mean levels of blood hemoglobin were higher (10 g/dl), reticulocyte count was lower (5% to 6%), and mean fetal hemoglobin (HbF), which was inversely correlated with reticulocytes, was higher (22% to 26.8%). The high HbF is believed to account for the very mild clinical manifestations.

According to Girot R in 2005 Sickle cell disease is a genetic autosomal recessive disease of hemoglobin. The disease results from a mutation of the sixth codon of the beta-globin gene, which induces the synthesis of an abnormal hemoglobin called hemoglobin S (HbS). The polymerisation of deoxy HbS molecules causes a chronic hemolytic anaemia and vaso-occlusive phenomenons. The disease affects mainly people from West Indies and Sub saharian Africa. Due to recent movements of these populations over the past years, sickle cell disease has spread across all continents. Painful crises, severe infections such as septicemia, meningitis, osteomyelytis, acute anaemia episodes, and severe vaso-occlusive events, mainly neurological, are the most frequent complications affecting children. Recent progresses in the care of patients have deeply modified the prognosis. The mean life expectancy of patients is now above 40 years. The conventional treatment includes antibiotics and immunizations, analgesics, and blood transfusion. The effects of chronic blood transfusion, hydroxyurea and bone marrow transplantation are the subject of current comparative evaluations.

According to Brambilla DJ (1994) .Sickle cell disease affects millions of people worldwide. Today, with proper health care, many SCD patients have a good quality of life (QoL) and are in fairly good health most of the time. These people can live up to their forties or fifties, or longer. Despite the 'common' underlying genetic basis and a similar pathophysiology, patients with SCD present a highly variable clinical phenotype due to Single Nucleotide Polymorphisms (SNPs) variability throughout the genome. Patients with SCD are at high risk for developing multisystem acute and chronic complications associated with significant morbidity and mortality.

According to Ballas SK (2005) .The most common complication of SCD is an acute episode of severe pain referred to as an acute vaso-occlusive crisis (VOC). AVOC is defined as pain resulting from tissue ischemia caused by vaso-occlusion most commonly in the bone(s) and bone marrow. Other common acute complications of SCD include fever related to infection, acute renal failure, hepatobiliary complications, acute anaemia, splenic sequestration, acute chest syndrome (ACS), and acute stroke.

According to Booth C, 2010 Patients with SCD have an increased risk for severe bacterial infection, resulting primarily from reduced or absent splenic Function.

Methodology

Study Area

The present study was carried out from newly diagnosed patients from; Late Baliram Kashyap Memorial Govt. Medical College, Jagdalpur, C.G.

Sample Collection

The blood sample was collected from people of jagdalpur, Chhattisgarh from late Baliram memorial hospital.

Procedure

The CAPILLARYS 2 FLEX-PIERCING instrument is a multiparameter instrument for haemoglobins analysis on parallel capillaries. The haemoglobins assay uses 8 capillaries to run the samples.

Preparation of Capillarys Analysis

1. Switch on CAPILLARYS 2 FLEX-PIERCING instrument and computer.
2. Set up the software, enter and the instrument automatically starts.
3. The CAPILLARYS HEMOGLOBIN (E) kit is intended to run with “HEMOGLOBIN” analysis program from the CAPILLARYS 2 FLEX-PIERCING instrument. To select “HEMOGLOBIN (E)” analysis program and place the CAPILLARYS HEMOGLOBIN (E) buffer and hemolyzing solution vials in the instrument, please read carefully the CAPILLARS 2 FLEX-PIERCING instrument manual.

4. The sample rack contains 8 positions for sample tubes. Place up to 8 capped sample tubes with whole blood on each sample rack (positions 1 to8); the bar code of each tube must be visible in the opening of the sample rack.
5. Position a new dilution segment on each sample rack will be ejected if the segment is missing.
6. Slide the complete sample carrier(s) into the CAPILLARYS 2 FLEX-PIERCING instrument through the opening in the middle of the instrument. Up to 13 sample racks can be introduced successively and continuously into the instrument. **When analysing a control blood sample, it is advised to use the sample rack No. F0 intended for control blood sample with specific tubes, caps and the wedge adapter for tubes for controls.**
7. Remove analysed sample racks from the plate on the left side of the instrument.
8. Take off carefully used dilution segments from the sample rack and discard them.

Result

By the study performed by us in the topic **Study Of Analysis Genetic Disorders In Sickle Cell Anaemic Patients By Using Sebia Electrophoresis Method** in 20 patients of age group 5– 10, 11 – 21, 21 - 30, and 31 –40. The result we examined by this study is that the mean value of age group of the 20 patients is 4.25. Direct detection at 415 nm in capillaries yields relative concentrations (percentages) of individual haemoglobin zones. Reference values for individual major electrophoretic haemoglobin zones in the CAPILLARYS 2 FLEX-PIERCING instrument have been established from a healthy population of 113 adults (men and women) with normal haemoglobin values using HPLC technique:

Haemoglobin A: comprised between 96.7 and 97.8 %

Haemoglobin F: ≤ 0.5% (*)

Haemoglobin A2: comprised between 2.2 and 3.2 %

Most haemoglobinopathies are due to substitution by mutation of a single amino acid in one of four types of polypeptide chains. The clinical significance of such a change depends on the type of amino acid and the site involved. In clinically significant disease, either the α-chain or the β-chain is affected.

More than 1400 variants of adult haemoglobin have been described. The first abnormal haemoglobin studied and the most frequently occurring have an altered net electric charge leading to an easy detection by electrophoresis.

There are five main abnormal haemoglobins which present a particular clinical interest: S, C, E, O-Arab and D.

The CAPILLARYS HAEMOGLOBIN (E) kit is intended for the identification of haemoglobinopathies.

Haemoglobin S

Haemoglobin S is the most frequent. It is due to the replacement of one glutamic acid (an acidic amino acid No. 6) of the β-chain by valine(a neutral amino acid): when compared to Hb A, its isoelectric point is elevated and its total negative charge decreased with analysis pH. Its electrophoretic mobility is therefore increased in the capillary and this haemoglobin is faster than A fraction. With alkaline buffered CAPILLARY HAEMOGLOBIN (E) procedure, haemoglobin S migrates A and A2 fractions, next to Hb A2.

Haemoglobin C

One glutamic acid of the β-chain is replaced by lysine (a basic amino acid No. 6): its mobility is strongly reduced. When compared to Hb A, its isoelectric point is highly elevated and its total negative charge decreased with the analysis pH. Its electrophoretic mobility is therefore increased in the capillary and this haemoglobin is faster than A fraction which allows its differentiation. Haemoglobin C, E and O-Arab are not superimposed on the electrophoretic pattern and are easily identified.

Age of group	No. of cases	HbA	HbS	HbA2	HbF
5-10	2	2	2	2	0
11-20	6	6	6	6	2
21-30	9	9	9	9	3
31-40	3	3	3	3	0
Total	20	20	20	20	5

Table No. 1 : Pattern of Hb Electrophoresis in Sickle Cell Disease (SS)

Discussion

According to BARRY et.al, in 1972 study a high prevalence of hemiplegia was found in 89 patients with sickle-cell disease seen over a 5-year period. Twenty-three patients (26%) had neurological manifestations; hemiplegia occurred in 15 (17%). During the same period, hemiplegia occurred in only 5% of patients with sickle-hemoglobin C disease and in 1.7% of patients with sickle-cell trait; the latter is the same as that in the Negro population with normal hemoglobin (1.8%). A review of the English-language literature shows that the high frequency of hemiplegia in the natural history of sickle-cell disease has not been sufficiently emphasized, despite its previous documentation in isolated case reports. Unless better methods for controlling the clinical expressions of this hemoglobinopathy can be found, stroke will remain one of the commonest serious complications in patients with sickle-cell disease.

But in my study his synthesis were no found in the patients possibly because of age factor or some other reasons.

According to RICHARD P. PERRINE at.al, in 1978. For study 270 Saudi Arabs with homozygous sickle cell anaemia, using chart review, a register (since 1969), and home visiting in 42 cases. Average follow-up for the total group was 10 years. Seventy-four percent of those diagnosed by age 3 years presented on screening or with merely anaemia; 26% presented with illness,

abnormal physical findings, or pain. In these studies conducted 2 or 3 patients' number had similar symptoms of illness & pain.

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प्रभावी शिक्षण – सक्रिय अधिगम विधि (ALM)

डॉ0 श्रीमती जस्सी जोस[1]

सारांश

शिक्षण प्रविधि ऐसा साधन है जिनका प्रयोग शिक्षण के समय विद्यार्थियों को पाठ में रूचि लेने, पाठ को स्पष्ट करने तथा उसे हृदयगम कराने के लिए किया जाता है। इस दृष्टि से शिक्षण के उद्देश्य को प्राप्त करने के लिए जहां एक ओर शिक्षक किसी न किसी शिक्षण विधि की सहायता लेता है । वहीं दूसरी ओर उसे विद्यार्थियों के मस्तिष्क में ज्ञान को अंकित करने के लिए शिक्षण प्रविधि का सहारा लेना पड़ता है, इस प्रकार संक्षेप में शिक्षण प्रविधियों का प्रयोग इसलिए किया जाता है कि शिक्षण रोचक, प्रभावशाली तथा सफल बन जाए ।

मनोविज्ञान के प्रत्येक क्षेत्र में बडी तीव्रता से वृद्धि हो रही है क्योंकि समाज में परिवर्तन भी तीव्र गति से हो रहा है। शिक्षा के उद्देश्यों एवं मूल्यों में परिवर्तन हो रहा है। नवीन पाठ्यवस्तुओं का भी विकास हो रहा इन सभी परिवर्तनों के परिणामस्वरूप शिक्षण के क्षेत्र में शिक्षण की कई विधियों एवं प्रविधियों का विकास हो रहा है। शिक्षण को सरल एवं रोचक बनाने के लिए शिक्षण प्रविधियों का प्रयोग किया जाता है ।

शिक्षण प्रविधियाँ शिक्षण की प्रक्रिया में आवश्यक कड़ी के रूप में प्रयुक्त होती है । अध्यापक अपनी शिक्षण विधि के आधार पर शिक्षण का एक व्यापक स्वरूप निश्चित कर लेता है किंतु शिक्षण उद्देश्य की प्राप्ति के लिए उसे अनेक शिक्षण प्रविधियों को अविलम्ब ग्रहण करना पड़ता है।

शिक्षण प्रविधि का अर्थ

शिक्षण प्रविधि ऐसा साधन है जिनका प्रयोग शिक्षण के समय विद्यार्थियों को पाठ में रूचि लेने, पाठ को स्पष्ट करने तथा उसे हृदयगम कराने के लिए किया जाता है। इस दृष्टि से शिक्षण के उद्देश्य को प्राप्त करने के लिए जहां एक ओर शिक्षक किकसी न किसी शिक्षण विधि की सहायता लेता है वहीं दूसरी ओर उसे विद्यार्थियों के मस्तिष्क में ज्ञान को अंकित करने के लिए शिक्षण प्रविधि का सहारा लेना पड़ता है,

इस प्रकार संक्षेप में शिक्षण प्रविधियों का प्रयोग इसलिए किया जाता है कि शिक्षण रोचक, प्रभावशाली तथा सफल बन जाए। सीखना तब होता है जब बच्चे सक्रिय रूप से सीखने की पूरी प्रक्रिया से जुड़े हों

[1] सहायक प्राध्यापक (शिक्षा विभाग), क्राईस्ट कॉलेज, जगदलपुर, जिला–बस्तर (छ0ग0), भारत

और उसका अर्थ और ज्ञान का सृजन स्वयं कर रहे हों। केवल चुपचाप बैठकर सुनना और सूचनाओं को ग्रहण करना ही सीखना नहीं होता। सीखने वाले ही किसी चीज का अर्थ और ज्ञान बनाने वाले होते है। इसलिए आज वर्तमान समय में शिक्षण को प्रभावशाली बनाने के लिए कई सक्रिय अधिगम विधि एवं प्रविधियों का प्रयोग किया जा रहा है।

सक्रिय अधिगम प्रविधियों की विशेषतायें

- विद्यार्थियों को सबक सीखने के समय अवसर उपलब्ध कराता है ।
- स्वमूल्यांकन में मदद करता है।
- मूल्यांकन के भय से मुक्ति व तनावमुक्त अध्ययन में सहायता प्रदान करता है।
- सीखना एक सक्रिय आनंददायी प्रक्रिया बन जाती है।
- उपचारात्मक सहायता तत्काल सुलभ हो जाती है।
- उच्च स्तर के सोच के कौशल (अनुप्रयोग, विश्लेषण संश्लेषण मूल्यांकन आदि) का विकास।
- सर्वांगीण विकास का समान अवसर प्राप्त होता है।
- सीखने में लचीलापन आता है।
- आत्मसम्मान एवं आत्मविश्वास में वृद्धि होती है ।
- तार्किक शक्ति के विकास में सहायक।

इस प्रकार शिक्षण को प्रभावशाली बनाने के लिए एक शिक्षक कई सक्रिय अधिगम विधि एवं प्रविधियों का प्रयोग कर सकता है जो निम्नानुसार हैं –

(1) क्लोज टेस्ट (CLOZE TEST)

इसमें किसी एक पैराग्राफ का चयन कर उसे श्यामपट पर लिखा या अभ्यास के लिए दिया जा सकता है। लिखते समय कुछ महत्वपूर्ण जानकारियों का गायब कर उसकी जगह खाली स्थान रखा जाता है। पूरे वाक्य को पढ़कर विद्यार्थियों को सोच समझ कर तर्क के आधार पर अपने शब्द ज्ञान एवं भावार्थ के आधार पर खाली स्थानों में उचित शब्द लिखने के लिए चयन करना पड़ता है।

उदाहरण :

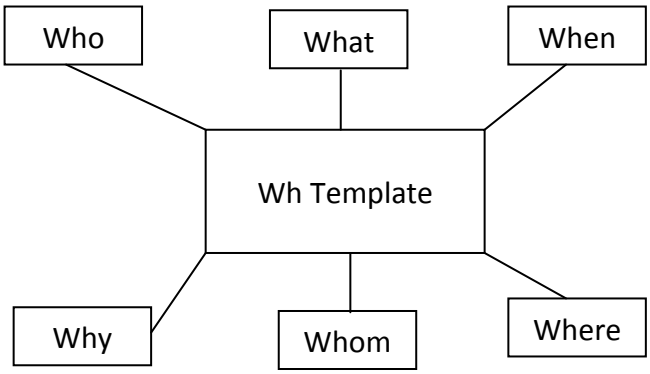
मैं आज ———गया, वहां मैंने दूध और कुछ, अंडे खरीदे। मैं यह जानता था कि आज बारिश होगी परंतु मैं ————साथ लाना भूल गया जिसकी वजह से मैं———गया।

(2) Wh टेम्पलेट (Wh – Template)

सीखने के क्रम में प्रश्न पूछना अथवा जिज्ञासा समाधान एक महत्वपूर्ण कड़ी होती है। इस कड़ी को बेहतर ढंग से संपन्न करवाने में wh टेम्पलेट महत्वपूर्ण भूमिका निभा सकती है। इस प्रविधि के

क्रियान्वयन करने हेतु अध्यापक सर्वप्रथम कक्षा में बच्चों को दोस्ताना माहौल तैयार कर प्रश्न पूछने का अवसर प्रदान करें।

कक्षा में किसी एक दीवार पर चार्ट में wh टेम्पलेट बनाकर स्थापित करें। जब भी कोई प्रकरण पढ़ाएँ, विद्यार्थियों को अपने आप से इस wh टेम्पलेट से उस प्रकरण के संबंध में विभिन्न प्रश्नों के जवाब स्वयं से पूछने को कहें।



(3) किम (K.I.M.)

कई बार हमें कुछ विशिष्ट आइडिया समय पर याद नहीं आते या कुछ शंका हो जाती है। ऐसे में यदि हम किसी तकनीक का उपयोग करें तो ऐसी स्थिति उत्पन्न नहीं होती।

प्रायः नए शब्द आदि में भी विद्यार्थियों को शंका हो जाती है और समय पर उसका अर्थ समझ में नहीं आ पाता । ऐसी स्थिति में भी यह तकनीक बहुत उपयोगी हो सकती है। इस हेतु उन्हें उस शब्द को पहले कॉलम में लिखते हुए दूसरे कालम में उसका अर्थ सरल वाक्य में और तीसरे में उस शब्द को याद रखने हेतु कोई चित्र या सूचना लिखी जा सकती है। चित्र बनाने में समय ना लगाते हुए सरल रेखा से चित्र बनाये जा सकते है अथवा अखबार आदि से काट कर चिपकाया भी जा सकता है। पहले कालम में key words लिखे जाते हैं। दूसरे कालम में उनसे संबंधित सूचनाएँ (information) लिखी जाती है। तीसरे कालम में उस information को याद रखने के लिए कुछ memory clues या चित्र बनाये जाते हैं।

(4) क्रोनो–लॉग किम (Chrono-Log)

कई बार हमें कई ऐसी चीजें ध्यान में रखनी पड़ती है जिनमें एक विशेष क्रम होता है। कभी–कभी इतिहास में या विज्ञान आदि में भी क्रम का बहुत महत्व होता है। ऐसी स्थिति में अध्यापक यदि इन घटनाओं को क्रम से क्रोनो–लॉग के रूप में लिख कर अपने सामने या कक्षा में एक विशेष स्थान पर प्रदर्शित कर सकें तो उन्हें देख–देख कर बच्चे ऐसी घटनाओं के क्रम को सही तरीके से बिना रटे याद कर सकते है।

किसी घटना या गतिविधि का एक निश्चित क्रम हो और क्रम से प्रस्तुत कर सकने पर ही उसका महत्व हो । इन क्षेत्रों को उनके सही क्रम में रखते हुए चार्ट पेपर में बच्चों अथवा शिक्षक के माध्यम से क्रोनो–लॉग का विकास किया जा सकता है।

(5) पाठ के आधार पर प्रश्न (Document Based Questions)

किसी पाठ या किसी अन्य गद्य को बच्चों को पढ़ने का अवसर दें। उन्हें इसे पढ़ने के उपरान्त कुछ प्रश्नों के उत्तर देने होंगे, यह स्पष्ट करें। बच्चों को गद्य पढ़ते समय नोट्स बनाने का सुझाव दें। गद्य को पढ़ने के बाद प्रश्नों को बच्चों को उपलब्ध करवाएँ। बच्चों को गद्य पढ़ने के उपरांत अपने मन से प्रश्नों के उत्तर लिखने को कहें। उत्तर लिखने के लिए समय निर्धारित करें। बच्चों को अपने बनाए नोट्स के आधार पर उत्तर लिखने का अवसर दें।

(6) प्रोजक्ट कार्य (Project Work)

विद्यार्थियों को समय–समय पर विभिन्न प्रकार के प्रोजेक्ट्स् स्वयं करने को दिए जाने चाहिए। इन प्रोजेक्ट्स् को उनके आंतरिक मूल्यांकन से जोड़ना चाहिए ताकि इसे वे गंभीरता पूर्वक लेते हुए कुछ नया सीखने का प्रयास करें।

निम्न विषयों पर विज्ञान में छोटे–छोटे प्रोजेक्ट्स् करवाए जा सकते हैं –

धातु– अधातु की सूची, मिट्टी के प्रकार, प्रदूषण के प्रकार, वन पर आधारित, जल स्त्रोत और उसका संरक्षण, स्वास्थ्य संबंधित जानकारियों का संकलन आदि ।

निष्कर्ष

शिक्षण के सक्रिय अधिगम विधि एंव प्रविधि पर नज़र डालने पर हम इस परिणाम पर पहुँचते हैं कि शिक्षण को सरल तथा रोचक बनाने के लिए शिक्षण में उपरोक्त सक्रिय प्रविधियों का महत्वपूर्ण स्थान है । इन प्रविधियों के माध्यम से शिक्षक विद्यार्थियों के सम्पर्क में आकर उन्हें क्रियाशील बनाता है । अतः शिक्षण को प्रभावशाली बनाने के लिए शिक्षण प्रक्रिया में सक्रिय अधिगम प्रविधियों को महत्वपूर्ण स्थान देना चाहिए।

संदर्भ ग्रन्थ

1. शिक्षा के तकनीकी आधार– आर. ए शर्मा, एन. आर. स्वरूप सक्सेना
2. सक्रिय अधिगम विधि –राज्य शैक्षिक अनुसंधान एवं प्रशिक्षण परिषद्, छत्तीसगढ़ . (2015–2016)

छत्तीसगढ़ की लोक संस्कृति

श्रीमती माग्रेट कुजूर[1]

प्रस्तावना

छत्तीसगढ़ व्यापक रूप से गावों व कस्बों का प्रदेश है। यहाँ की संस्कृति को लोक संस्कृति के रूप में देखना अधिक सार्थक है। छत्तीसगढ़ की संस्कृति लोक संस्कृति है। जो जनजातीय भू–भागों में अपनी पृथक सांस्कृतिक अस्मिता के साथ संरक्षित है। इसके आचार– विचार ,रीजि–रिवाज, जीवन शैली ,उत्सव ,मूल्य और मान्यताएँ भाषा कथाएँ, लोक गाथाएँ , लोक नृत्य , लोक संगीत, लोक नाट्य , लोकाचरण , लोक देवता, शिल्प और लोक चित्रकला का विराट संसार है, जो समष्टि रूप में, उस प्रदेश की समग्र वृहत्तम संस्कृति है।

विकास की दृष्टि से भले ही छत्तीसगढ़ प्रथम पंक्तियों में न गिना जाय , परंतु सांस्कृतिक रूप से अग्रगण्य है। दया, क्षमा , ममता सरलता एवं उदारता यहॉ के लोगो का प्रमुख गुण है। इसलिए कहा गया है– **"छत्तीसगढिया सबले बढ़िया" छत्तीसगढ़** अनेक संस्कृतियों को पोषण देता रहा है। जहॉ तक उसकी अपनी संस्कृति का प्रश्न है। वह अपनी आदिम विशेषताओं से अभी भी जुडा हुआ है। यहॉ के अधिकांश निवासी प्रकृति पर आस्था रखते हैं। पर सच तो ये है कि श्रम और ईमान से बढ़कर इनका कोई देवता नहीं है। मौखिक परम्परा से प्राप्त होने वाले युगीन साहित्य को लोक साहित्य कहा जाता है।समस्त लोक ही इसे अपनी कृति मानता है। गॉव के निरक्षर लोग अपनी सांस्कृतिक भूख मौखिक रचनाओं से शांत करते है। इसमें विशेष अंचल की माटी की सौंधी महक का अहसास होता है।

लोक संस्कृति लोक मानस धर्म से प्रभावित होता है। यह संस्कृति वैचारिक परंपराओं का पालन करती हुई धार्मिक परंपराओं में विश्वास करती है। इसलिए वाल्मीकि के राम या भवभूति के राम को इतना महत्व नहीं देती जितना कि तुलसी के राम को यह देती है। ऐसा इसलिए है, कि तुलसी के राम मर्यादा पुरूषोत्तम थे। उनका संबंध मुख्य रूप से उन्ही लोगो के साथ बना रहा जिनके साथ लोक जीवन जीने वाला साधारण जन रहता है। उदाहरण में कौआ बंदर , गिद्ध, नदी ,बेर, कंद–मूल सभी समर्पित है, इस प्रकार से ग्रामीण संस्कृति लोक संस्कृति का एक ऐसा माध्यम है जैसे जल की शीतलता का। छत्तीसगढ़

की लोक संस्कृति में ऐसी स्थिति हमें जगह–जगह देखने को मिलती है।प्रकृति,विकृति और संस्कृति को परिभाषित करते हुए कहा जाता है– कि आप आठ रोटी खा सकते हैं

[1] सहायक प्राध्यापक, हिन्दी शास. महाविद्यालय, धरमजयगढ़, जिला–रायगढ़ (छ.ग.), भारत

यह प्रकृति है, आप दस रोटी खा सकते हैं, यह विकृति है। आप दो रोटी किसी भूखे को देकर स्वतः केवल छः रोटी खाते है,यह संस्कृति है।इसी संस्कृति के कायल छत्तीसगढ़ी को परिभाषित करते हुए एक कवि ने लिखा है–

पत्थर लिखते गौरव गाथा,

माटी इतिहास बनाती है।

झरनों से फूट रही कविता

हर धारा गीत सुनाती है

सब लोग विनम्र सदाचारणी संस्कृति ही इनकी थाती है।

जितनी चौड़ी है महानदी

उतनी ही इनकी छाती है।

संबंधित साहित्यों का पुनरावलोकन

छत्तीसगढ़ एक समृद्धशाली राज्य है, यह प्रशन्नता का विषय है, कि बदलते परिवेश में भी छत्तीसगढ़ी लोक संस्कृति के प्रति जनमानस की रूचि अभी भी बनी हुई है। जनमानस विभिन्न अवसरों पर गाये जाने वाले लोक गीतों ,रीति–रिवाजों ,परम्पराओं ,कहानियों, लोकोक्तियों ,हाना ,आदि के ज्ञान से अछूता नहीं है। छत्तीसगढ़ी लोक साहित्य से संबंधित पर्याप्त अध्ययन हो चुका है जिनमें कुछ प्रमुख साहित्यकारों एवं उनकी कृतियों का विवरण इस प्रकार है–

1. डॉ. गंगाप्रसाद गुप्त "बरसैया"

इनके द्वारा रचित "**छत्तीसगढ़ का साहित्य इतिहास"** एक प्रमुख ग्रंथ है, जो छत्तीसगढ़ी साहित्यकारों के परिचय एवं साहित्य की विभिन्न विधाओं के वर्णन की दृष्टि से महत्वपूर्ण है।

2. डॉ. अनसुईया अग्रवाल

इनके द्वारा रचित **"छत्तीसगढ़ी लोकोक्तियां और जनजीवन "** एक प्रमुख ग्रंथ है जिसमें परिनिष्ठित छत्तीसगढी लोकोक्तियों का समाजशास्त्रीय अध्ययन किया है। लोकोक्तियों का व्यक्ति ,समाज, व्यावसाय, प्रकृति, धर्म ,नीति, उपदेश, जाति, आचरण, इत्यादि से संबंधित किया गया है। मानव समुदाय में लोकोक्तियों की उपादेयता ,उनकी प्रासंगिकता के अध्ययन की दृष्टि से भी यह ग्रंथ महत्वपूर्ण है। बोली के रूप में छत्तीसगढ़ी लोकोक्तियों का सामाजशात्रियों, आर्थिक एवं जातीय अध्ययन भी प्रस्तुत ग्रंथ में दिखाई देता है।

3. डॉ. नारायणलाल परमार

"छत्तीसगढ़ी लोकगीतों की भूमिका "छत्तीसगढ़ी लोकगीतों से संबंधित इनका महत्वपूर्ण ग्रंथ है, जिसमें सर्वप्रथम छत्तीसगढ़ी लोकसाहित्य पर दृष्टिपात करते हुए लोकसाहित्य का मूल्यांकन किया गया है। तत्पश्चात् छत्तीसगढ़ी लोकगीतों में नई चेतना का विश्लेशण करते हुए लोकगीतों की उपयोगिता बतायी गयी है।

4. डॉ. सुधीर शर्मा एवं नंदकिशोर तिवारी

छत्तीसगढ़ी साहित्य परिषद बिलासपुर छ0ग0 द्वारा प्रकाशित "**छत्तीसगढ़ी कहिनी"** छत्तीसगढ़ी लोकसाहित्य से संबंधित महत्वपूर्ण ग्रंथ है, जिसकी प्रस्तावना में छत्तीसगढ़ी लोककथाओं से सबंधित जानकारी प्रस्तुत करते हुए 13 छत्तीसगढ़ी कहानियों का समावेश किया गया है।

5. डॉ. मन्नुलाल यदु

छत्तीसगढ़ अस्मिता प्रतिष्ठान रायपुर छ0ग0 द्वारा प्रकाशित **"छत्तीसगढी कहावत कोष"** छत्तीसगढ़ी कहावतों के ज्ञान की दृष्टि से अतीव महत्वपूर्ण है जिसके प्रारंभिक भाग में कहावतों का स्वरूप एवं परिभाषित प्रस्तुत करते हुए कहावत तथा मुहावरों में अंतर समझाया गया है। इसके प्रथम खण्ड में न केवल छत्तीसगढ़ी कहावतों का विस्तृत संकलन प्रस्तुत किया गया है अपितु वे कहावतें व्याख्या में भी प्रस्तुत की गयी हैं। मध्य–मध्य में कहावतों से संबंधित दन्त कथायें भी दी गयी हैं। इस कोष मे लगभग 875 कहावतों का संग्रह है। इसी कोष के द्वितीय खण्ड में छत्तीसगढ़ी कहावतों का अन्य भाषाओं और बोलियों की कहावतों के साथ तुलनात्मक अध्ययन भी किया गया है।

6. डॉ. विनय कुमार पाठक

आपने अपनी कविता **" तॅय उठथस सुरूज उथे" और "एक किसिम के नियाव" में** छत्तीसगढ़ी भाषा के सच्चे चित्र को उकेरा है।

7. डॉ. सत्यभामा आडिल

"सीख– सीख के गोठ" आडिल जी की छत्तीसगढ़ी निबंध है, लोकोक्तियों को छत्तीसगढ़ी में हाना कहते हैं। आपने अपनी बात को पुष्ट करने के लिए "हाना" का प्रयोग किया है।

8. प्रकाश मनु एवं डॉ. सुनीता

"छत्तीसगढ़ की सर्वश्रेठ लोक कथाएं" नामक इस पुस्तक में छत्तीसगढ़ राज्य की ऐतिहासिक, पौराणिक, तथा भौगोलिक स्थितियों के साथ –साथ उसके वर्तमान को समझने की गंभीर कोशिश की गई है।

इस प्रकार स्पष्ट है कि छत्तीसगढ़ी लोकसाहित्य से संबंधित अनेकानेक ग्रंथ लिखे गये हैं, किन्तु वर्तमान

में लोकसाहित्य की स्थिति लोकसाहित्य निर्माण संबंधी समस्यायें एवं लोकसाहित्य के उत्थान हेतु उपायों पर दृष्टिपात कर साहित्य सृजन हो यह महती आवश्यकता है।

शोध की सार्थकता

छत्तीसगढ़ सांस्कृतिक विकास की संभावनाओं के लिए एक विशाल मंच है। यहां भिन्न–भिन्न प्रकार के लोकनाटय, लोकनृत्य, लोकगीत, लोकसंगीत, लोकचित्रकारी आदि यहां की संस्कृति में फल–फूल रही है। लेकिन जनमानस आधुनिक मनोरंजन के साधनों के कारण अपनी इस बृहद एवं व्यापक संस्कृति से दूर होती जा रही है। आधुनिक नगरीय संस्कृति ने रीति–रिवाजों को प्रभावित किया है। लेकिन आज भी जनमानस विभिन्न अवसरों पर गाये जाने वाले लोकगीतों के ज्ञान से अछूता नहीं है। यद्यपि एकल परिवार प्रणाली के कारण बुजुर्गों द्वारा सुनाई जाने वाली लोककथाओं का अस्तित्व धीरे–धीरे समाप्ति की ओर है किन्तु जनमानस को आज भी पुरानी लोककथाएं ज्ञात हैं। इस तरह छत्तीसगढ़ के गर्भ में जो बहुमूल्य संस्कृति बिखरी हुई है उसे उपर्युक्त विद्वानों ने अपने शोध अध्ययन के द्वारा समेटने का प्रयास किया है तथा इस संस्कृति को पुनर्जीवित कर बहुरंगी संस्कृति के प्रकाश से समाज को परिचित कराने का प्रयास किया है।

चयनित ग्रंथ सूची

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2 डॉ. अनुसूया अग्रवाल, "छत्तीसगढ़ी लोकोक्तियों और जनजीवन" भावना प्रकाशन दिल्ली. 2001.

3 डॉ. नारायण लाल परमार "छत्तीसगढी लोकगीतों की भूमिका " पहचान प्रकाशन रायपुर.2000.

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8 पूजा शर्मा "छत्तीसगढ़ की लोक संस्कृति" शिव बुक हाउस जयपुर. 2009

9 डॉ सत्यभामा आडिल, जनपदीय भाषा–साहित्य (छत्तीसगढ़ी)