



"FINE NEEDLE SPLENIC ASPIRATE CULTURE IN CASES OF SEPTICEMIA

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ABSTRACT

Objectives To assess the value of splenic aspirate culture in cases of septicemia in adults and to compare its results with the conventional peripheral venous blood culture.

Methodology One year prospective study. 100 patients with symptoms of fever, signs of 'septicemia' were taken. Clinical diagnosis of septicemia was accepted when in an appropriate clinical set-up high fever with chills, tachycardia & tachypnoea was seen. In 50 cases each splenic aspirate & blood culture were performed simultaneously.

Results In septicemic patients the positivity of splenic & blood culture were 88.23% (15/17) & 42.85% (15/35) respectively. In non septicemic group also the positivity of splenic aspirate was three times to blood culture 4% & 1.7% respectively.

Conclusion. Early and superior results obtained by splenic aspirate culture as compared to conventional peripheral venous blood culture.

KEYWORDS : Septicemia, Tachycardia , Tachypnoea , Splenic Aspirate , Blood Culture

INTRODUCTION

Bacteremia and septicemia present a problem to clinicians when the specific microorganism can not be grown in blood culture. When there is focus of infection like abscess or sore throat, Pneumonia etc. the causative organism can be grown with success in 90% of cases. But when there is no clear cut focus of infection seen like in subacute bacterial endocarditis and septicemia without any obvious focus then blood culture is the only investigation which will reveal organism.[1]

There are many limitation of blood culture, some of these are (1) the positivity of blood culture is only 50-60% that too is first week. (2) when sample has been taken before the start of antimicrobial therapy, sometimes even 2-3 blood culture are needed, this is not possible in many cases. A large no of variables determines the success rate of isolation from blood culture, Many aspects may influence the result such as timing of specimen, culture media and significance of isolates.[2]

In view of these problems we thought of substitute of blood culture which may give higher positivity than blood culture, easy to perform that is splenic aspirate culture through FNAC. There still is considerable danger in any splenic puncture technique in which a piece of splenic tissue is removed for histologic examination. On the other hand there is little if any danger in the type of puncture which yield material for cytologic study.[3]

There were many studies performed in the past. In nineteenth century Widal has used splenic puncture for the purpose of culture and showed 90% positive results in cases of typhoid. Interestingly, for first time salmonella typhi was isolated in post mortem tissue of spleen only. Many authors have done splenic puncture for same purpose.[4]

But all the studies done were on splenic biopsy where tissue is removed however in this study we have taken splenic aspirate through FNAC so there are no any chances of complications. In India little work has been done on FNAC spleen and too has been mainly for cytological purposes. No study on FNAC spleen used for culture purpose could be found.[5,6]

Keeping this view in mind the present study has been undertaken to evaluate the results of splenic aspirate culture through FNAC in cases of septicemia and bacteremia and its comparison with blood culture.

METHODOLOGY

One year prospective study.

Subjects: - 100 patients with symptoms of fever, with or without signs of 'septicemia' were taken. The cases are divided into three groups

Group I consist of mainly cases in which there are signs and symptoms septicemia. These symptoms and signs consists of fever 101 degree F or more than 101 degree F. with chills and rigors, vomiting, altered sensations, hypotension, tachycardia, leucocytosis or leucopenia in an appropriate clinical set up.

Group II Consist of cases of fever but without the clinical signs & symptoms of septicemia which are listed in upper paragraph.

Group III Consists of fever which are not of bacterial origin like malaria and fever due to Neoplasm.

- Clinical diagnosis of septicemia was accepted when in an appropriate clinical set-up high fever with chills, tachycardia & tachypnoea was seen.
- In 50 cases splenic aspirate & blood culture were performed simultaneously.
- (17 cases) of septicemia, 25 patients without sign of septicemia, 8 cases were fever of known etiology but without septicemia.)

Main Outcome Measure:

Early and superior results obtained by splenic aspirate culture as compared to conventional peripheral venous blood culture.

OBSERVATION TABLES

TABLE NO.1
DISTRIBUTION OF CASES ACCORDING TO CLINICAL CONDITION.

S. NO.	Groups	Blood culture	Splenic Aspirate Culture
Group 1	Fever with Clinical diagnosis of septicemia	35	17
Group 2	Fever without clinical diagnosis of septicemia	57	25
Group 3	Fever of known Etiology without Septicemia	8	8
Total	100		50

Comparison of percent positivity in blood culture and splenic aspirate culture

Groups	Blood Culture	Splenic Aspirate Culture
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	No. of cases	Positive cases	%	No. of cases	Positive cases	%
I	35	15	41.93%	17	15	88.21%
II	57	01	1.96%	25	01	5.26%
III	8	0	0	08	0	0

Group I comprises of the cases of frank septicemia. In this group the percentage positivity was 41.93% and 88.21% in blood and splenic culture respectively. Group 2 Fever without clinical signs of Septicemia, Blood & Splenic aspirate culture are positive in 1.96% & 5.26% respectively. Group III is having cases of Malaria, Tuberculosis and other fever of known etiology. In this group no organism was grown from blood & aspirate culture.

TABLE 2
INCIDENCE OF GRAM POSITIVE & GRAM NEGATIVE SEPTICEMIA

	Positive cases	Gram Positive	Gram Negative
Blood Culture	16/100	8	8
Splenic Aspirate Culture	17/500	9	8

Table shows that incidence of gram positive & gram negative septicemia is approximately 51% & 49% respectively.

TABLE 3
RELATION OF DURATION OF FEVER WITH POSITIVE CULTURES.

Days of fever	No. of cases	Blood culture Positive/Negative	Splenic culture Positive/Negative
1 to 7 days (I st week)	14	3/11	3/11
8 to 14 days (II nd week)	13	0/13	4/9
15 to 21 days (III nd week)	16	1/15	9/7
22 to 28 days (IV th week)	1	0/1	0/1
29 days onwards (V th week)	6	0/6	1/5

The table shows maximum blood culture positively in first week & after first week hardly any blood culture is positive because the patients were on high antibiotics before coming to Hospital. While splenic culture were positive in first (3/14) week, second week (4/13) & in third week (9/16) so splenic aspirate positively is gradually increasing from 1st to third week. The 3rd week splenic aspirate culture were positive in those cases also who had taken antibiotics prior to collection of splenic aspirate.

TABLE 4
RELATION OF POSITIVE CULTURE WITH PALPABLE SPLEEN.

	Total Cases	Splenomegaly	Spleen not palpable
	50	17	33
Positive cases	17	6	11
Percent positivity	34%	35.29%	33.33%

In this table the positivity of Positive of splenic aspirate culture was 35.29% in the cases of splenomegaly and 33.33% positivity where spleen was not palpable so as such there is no exact correlation of splenomegaly with the positivity of culture.

TABLE 5
RELATION OF TIME OF INCUBATION IN BLOOD CULTURE & SPLENIC ASPIRATE CULTURE.

Time of incubation	Splenic aspirate culture (Total cases 17/50)	Blood Culture (Total cases 17/50)
24 hr. (1 day)	15	02

8 hr. (2days)	01	10
72 hr. (3 days)	01	03
4 days and more	-	01

The table clearly shows that in splenic aspirate culture after 24 hrs. of incubation in 15 cases micro organism can be grown. While only in 1 case three days incubation was required. While in blood culture only 2 cases were positive in 24 hr. while in 10 cases 2 days incubation was required and in 3 cases 3 days incubation was needed. This indicate that incubation period required is more in blood culture as compared to splenic aspirate culture.

Table 6
SHOWS RELATIONSHIP OF THERAPY RECEIVED PRIOR TO CULTURE WITH POSITIVITY OF CULTURE

Antibiotics received prior to culture	Total	Positive culture	Negative Culture	Positive Culture	Negative Culture
	38	13	25	3	35
Antibiotics not received prior to culture	12	04	08	01	01

RESULTS

In septicemic patients the positivity of splenic & blood culture were 88.23% (15/17) & 42.85% (15/35) respectively. In non septicemic group also the positivity of splenic aspirate was three times to blood culture 4% & 1.7% respectively. 26 patients were neonates, 6 were infants, in the age group of 5-12 years, -7, 36 adolescent & 26 adults. In neonatal & infantile period the offending organism were streptococcus pyogenes & E. Coli. While in adult the gram negative bacilli & coagulase positive staphylococcus were common isolates.

Cases with Splenomegaly were 17 & without splenomegaly 33.35.29% positive culture in splenomegaly case 33.33% in cases of where spleen was not palpable. Time of incubation required was 24 hours in 15/17 cases of splenic aspirate. Whereas in blood culture 2/16 24 hours, 10/66 -48 hours & 3/16-72 hours were needed. Time of incubation was less in splenic aspirate culture.

In 36 cases patients had already received antibiotics & out of these 13 & 3 had shown positive results in splenic aspirate & blood culture respectively. Sensitivity of splenic aspirate is 88.2% & specificity is 96% with a positive prediction value of 98%. There was no single complication seen in this series after splenic puncture.

STATISTICAL ANALYSIS-

(A) Sensitivity of splenic culture in cases of Septicemia = True Septicemia

$$\text{Cases of septicemia} = 15 = 88.2\%$$

17

(B) Significance of splenic aspirate culture (Specificity) = True negative results = 24 = 96%
all non septicemia cases 15

(C) False negative rate of splenic aspirate culture = False negative = 2 = 11.7%
all septicemia patients 17

(D) False positive rate of splenic aspirate culture = False positive = 1 = 4%
all non septicemia patients 25

(E) Positive predictive value of splenic aspirate = True positive = 15 = 98%
all Positive 16

(F) Negative predictive value = True negative = 24 = 96.5%
all negative 26

DISCUSSION

The life threatening syndrome of gram negative or gram positive bacterial septicemia is now a common problem in all acute care hospitals. Despite the availability of new broad spectrum antibiotics, the incidence and outcome of sepsis and septic shock have not decreased significantly in the last two decades. This is due in part to the failure of early recognition and specific therapy of underlying infection and also because of the emergence of antibiotics resistant hospital strains. The incidence has also risen because of the aging of the populations and prolonged survival of persons with chronic illness. For the confirmatory diagnosis of bacteremia and septicemia the isolation of organism is mandatory. The commonest method which is used for isolation of microorganism is blood culture.

But routine and conventional blood culture suffer from many limitations: Blood culture is positive only 50-60% of cases, If the concentration of bacteria or microorganism /ml of blood is less, then the positivity is markedly reduced and large amount of blood (may be 50 ml) is required

1. If the bacteremia is intermittent then three or more blood cultures are required for the isolation of microorganism. Belli J Waisbren. A. 1956 [5]
2. Blood sample should be collected before the start of antibiotic therapy and when fever starts rising. Adam Finn 1986.[6]
3. If the peripheral veins are involved in thrombophlebitis then it is very difficult to get blood sample for culture purposes.
4. In severely anaemic patients and in neonates and children where the blood volume is less taking out even 10-15ml. of blood is associated with a risks.[7]

So looking to all these limitations of blood culture, a substitute for blood culture was looked for in literature. Some writers have done a very fruitful work over splenic puncture. These studies were done on the splenic biopsy tissue received by von silverman needle. Block and Jacobson (1950) had done splenic puncture in 55 cases. Complications which took place were haemorrhage from spleen (in 4 cases) and fatalities in two cases. Because of the risk of haemorrhage the procedure was banned in U. S. A. But these are many more studies done by Introzzi (1950), Witt RL, Sukumar V, Gerges F in various age group from neonates to old age and for various purpose like diagnosis sarcoidosis, Haemorrhagic disorders, tuberculosis, lymphomas and for isolation of microorganism without any major complication or mortality.[6,7,8]

After the advent of fine needle aspiration biopsy splenic aspirate through fine needle become more easy and without the risk of complications. In India also in last few years FNAC of spleen has started at number of institution. Verma & Singh had diagnosed a case of hairy cell leukemia by FNAC of spleen. (1990). Similarly Sharan and Sinha (1990) has used FNAC for the demonstration of L. D. bodies.[9]

However because the number of studies done in this subject were not many. Therefore this subject was chosen that is fine needle aspirate culture as a substitute of blood culture in cases of bacteremia and septicemia. Simultaneously blood splenic aspirate cultures were performed in 50 cases and results were compared.

In Indian subcontinent because of prevalent habits and social taboos, adolescent may not get sufficient nutrition and may be exposed to increased risk of infections. Blood culture positivity was claimed to be 50-60% in frank septicemia and bacteremia, that too when samples were collected before starting antibiotic therapy and at peak of fever.[10]

Widal in nineteenth century was first to culture salmonella typhae in splenic culture, when blood culture was negative. Introzzi (1950) supported this finding by demonstrating typhi bacilli in splenic tissue when all other method of investigation were negative. The finding in this series were similar to above findings. 85% positivity of

splenic aspirate culture was found in cases of septicemia and bacteremia inspite of antibiotic received prior to collection of sample.[8]

In the remaining three group where the signs and symptoms of bacteremia and septicemia were not present, the positivity was three times than blood culture. If we take the overall positivity of splenic aspirate culture (that is by combining all the groups). If comes to 34% our this finding is supported by authors who worked on Pyrexia of unknown origin and had concluded clearly that in only 36% of cases infections cause can be determined, 51% were of non infections origin and 13% are of unknown origin.[11]

The incidence of gram negative septicemia has been undoubtedly increasing because of the antibiotic resistance which is a common finding in gram negative rods due to plasmids. But incidence of infections due to resistance strains of staphylococcus aureus is also increasing in the hospital. In this series all patients have been indoor patients and out of all positive cases higher number has been that of staphylococcus. From our observations it is clear that the maximum positivity of blood culture was in the first week while the splenic culture were reported upto positive third week of fever also. Therefore when bacteremia cannot be demonstrated by blood culture in second or third week, splenic aspirate culture grow in bacteremia even after 3rd week, this is because microorganism take longer time to get clear off from splenic tissue.

RELATION OF PALPABLE SPLEEN WITH POSITIVE CULTURE

There is no specific relation of palpable spleen with the percent positivity of cultures. As it is clear from table that in 17 cases palpable spleen were present, out of this only 6 were positive (35.29%) whereas in 33 cases spleen was not palpable and 11 cases were giving positive culture (33.33%). Though in all septicemia there is enlargement of spleen but the spleen will be only palpable when it is enlarged to double its size. So it is not necessary that in all cases of septicemia spleen should be palpable and also vice versa all the splenomegaly are not due to infections. Relation of positive culture with widal test.[12]

Table clearly indicate that out of 40 widal seven show a significant rise in titre of 'O' & 'H' and six show titre less than this. Positive widal is taken as H1/160 and O:1/160, levels below this are taken as anemnesic reaction. Four out of seven cases salmonella paratyphi were cultures while three came to be negative out of six remaining cases which has show low titre, E. Coli and Proteus were grown in two cases respectively. Most of our cases had already received either Ampicillin, chloramphenicol or ciprofloxacin.[13]

In this series only one blood culture was positive for salmonella out of seven significantly widal sera while four splenic aspirate were positive. So we can conclude that widal test is still necessary in cases of suspected typhoid and for confirmation or early diagnosis splenic aspirate culture is necessary that the blood culture and more so when chloramphenicol and ciprofloxacin had been started already. Because blood culture may not reveal microorganism even when only two doses of antimicrobial therapy has been given.

It is clear from the table in 901% of cases one days incubation was required in splenic aspirate while in blood culture only 12% showed organism in one day period while 59% were positive after 2 days, 20% after 3 days and 5% after 4 days. So it is clear from above finding that time required for incubation period is comparatively less with splenic aspirate than the blood culture. More so the longer the time of incubation greater are the chances of contamination and delay in report and delay in specific antimicrobial therapy. In most of our cases of frank septicemia the splenic smear pattern was inflammatory with predominately neutrophilic response.[14]

There was no complication seen in the study. Though some patients complained of slight pain in the region of puncture site of 2-3 hrs. No complications like haemorrhage or fatality was recorded in FNAC spleen in the studies of many peoples, Block and Jacobson (1951)

have described 2 facilities in 55 cases in his series but they done splenic biopsy by vim silverman needle. As such there is no complications of FNAB spleen even in children & the patients of haemetological disorders.^[7,15]

Septic Splenomegaly unconscious patient and children must never undergo splenic puncture. We have done FNAB spleen in 17 frankly septicemia patients without any complications. Jacobson & Block have performed FNAB spleen in children without any complications. Unconscious patients is a relative contraindication because in case any complication occur during procedure patient may not able to complaint immediately.^[7,14,15]

The only contraindication to splenic aspirate is a decided haemorrhagic tendency. According to Block and Jacobson (1951) non palpable spleen is the absolute contraindication for the splenic biopsy. FNAB spleen can easily be done even in nonpalpable spleen. We had difficulty only in one case where the patient had severe emphysema and splenic dullness can not be elicited.^[7,16]

The present study has been conducted on the hospital patients, which may or may not represent a true cross section of the normal population. However it has served some useful purpose of highlighting the performance of splenic aspirate culture over blood culture in cases of septicemia and bacteremia. Further studies of splenic aspirate culture by FNAE are needed in children & adult in cases of septicemia.

CONCLUSION:

Spenic aspirate culture is superior and gives more positive results as compared to blood culture.

Spenic aspirate culture is most suitable investigation in case of Septicemia. Septicemia patients who had already received antibiotics. Patients is anaemic or have thrombophlebitis in peripheral veins. If the single and random sample has to be collected.

REFERENCES

1. Chatterjea JB, Arrau CM, Dameshek W. Splenic puncture. British medical journal. 1952 May 10;1(4766):987.
2. DEWARD O. FERRIS, M.D.; MALCOLM M. HARGRAVES, M.D. SPLENIC PUNCTURE AMA Arch Surg. 1953;67(3):402-407.
3. Morrison M, Samwick AA, Rubinstein J, Morrison H, Loewe L. SPLENIC ASPIRATION: CLINICAL AND HEMATOLOGICAL CONSIDERATIONS BASED ON OBSERVATIONS IN ONE HUNDRED FIVE CASES. Journal of the American Medical Association. 1951 Aug 25;146(17):1575-80.
4. Ferraioli G, Brunetti E, Gulizia R, Mariani G, Marone P, Filice C. Management of splenic abscess: report on 16 cases from a single center. International Journal of Infectious Diseases. 2009 Jul 31;13(4):524-30.
5. Belli J, Waisbren BA. The number of blood cultures necessary to diagnose most cases of bacterial endocarditis. The American journal of the medical sciences. 1956 Sep;232(3):284.
6. Finn A, Louviere J. Shopping-center patronage models: Fashioning a consideration set segmentation solution. Journal of Business Research. 1990 Nov 30;21(3):259-75.
7. Block M, JACOBSON LO. Myeloid metaplasia. Journal of the American Medical Association. 1950 Aug 19;143(16):1390-6.
8. Introzzi P. La punctura della milza. Ferrata, A.: Le emopatie. Milan, Societa Editrice Libreria. 1935;2(pt 3).
9. Witt RL, Sukumar V, Gerges F. Surgeon-performed ultrasound-guided FNAC with on-site cytopathology improves adequacy and accuracy. The Laryngoscope. 2015 Jul 1;125(7):1633-6.
10. Sharan K, Sinha RK. Fine needle aspiration biopsy of spleen in kala-azar. Indian pediatrics. 1990 Dec;27(12):1287-9.
11. Nagy GV. Über die Technik der Milzpunktion und ihren diagnostischen Wert. Klin. Wschr. 1924;3:274.
12. Donald E. Fry, MD; Leslie Pearlstein, MD; Robert L. Fulton, MD; et al Multiple System Organ Failure The Role of Uncontrolled Infection
13. Block M. Sarcoid diagnosed by needle biopsy of the spleen: report of a case. Journal of the American Medical Association. 1952 Jun 21;149(8):748-50.
14. Soderstrom N. Anatomy and histology of the spleen. Aspiration biopsy cytology. Monographs in clinical cytology. 1979;7(Part 2).
15. Moriarty AT, Schwenk Jr GR, Chua G. Splenic fine needle aspiration biopsy in the diagnosis of lymphoreticular diseases. A report of four cases. Acta cytologica. 1992 Dec;37(2):191-6.
16. Shields JW, Hargraves MM. An evaluation of splenic puncture. In Proceedings of the staff meetings. Mayo Clinic 1956 Aug (Vol. 31, No. 16, pp. 440-453).