REVIEW OF LITERATURE
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Sepsis is a greek word meaning 'putrefaction'. The concepts of sepsis arose from separate sources at about the same time in mid nineteenth century. In America, Oliver Wendell Holmes and in Vienna, Ignaz Philipp Semmelweis independently made the observation of high mortality among women hospitalized with puerperal fever. Both observed the death of a fellow physician who had been infected during participation in the autopsy of an infected subject. This experience brought attention to the fact that infection was being transmitted directly and both urged washing of hands and changing of clothing. These policies caused reduction in maternal mortality from 11.4 percent in 1846 to 1.3 percent in 1848. Subsequently, Pasteur's discovery of bacteria prompted Lister to publish his "antisepsis theory" in mid nineteenth century. Later progress by German physician enabled Kocher to report 2.3 percent infection in clean wound.

Definitions:

Bacteremia:

The term bacteremia as suggested by Ritchie(1990) denotes that bacteria are present in the blood but gives
no indication as to whether they are harmless or likely to cause injury.

**Septicemia:**

It implies the presence of organism and their toxin in the blood. The term is used when a patient shows severe toxemia, often with a high swinging temperature, severe prostration and all constituent sign of infection according to Richie (1990).

**Neonatal Septicemia:**

Gottoff & Behrman (1970) described neonatal septicemia as generalised bacterial infection documented by positive blood culture in the first four weeks of life.

**INCIDENCE:**

Septicemia remains the most important cause of morbidity and mortality in the newborn more so in the developing countries due to delivery and postnatal followup conducted in unclean environment having more chance of contamination with infective organism (Khatua et al 1981).

Gluck et al (1966) reported the incidence of neonatal septicemia as 1.8 per 1000 live birth. According to Gottoff and Behrman (1970) the incidence of neonatal septicemia has remained fairly constant over the past
40 years and varies from 1 per 500 to 1 per 1600 live birth. Wilson and Reichenwald (1974) reported the annual rate of sepsis neonatorum in a Modern North American Hospital as approximately 1 per 1000 live birth.

The incidence in India varies widely. Chaudhary et al (1975) reported the incidence of septicemia as 11.2 percent. Clinical and bacterial proven septicemia was found as 10.97 and 6.55 percent per 1000 live birth by Khatua et al (1986). Recently, a study by Mondal et al (1991) reported the incidence of neonatal septicemia as 15.5 per 1000 live birth.

**Early onset versus late onset neonatal septicemia:**

Neonatal sepsis can be divided into two main groups depending upon whether the onset is in the first 48 hours of life or later.

There are important differences between the two as has been suggested by Paul & Singh (1988). Early onset disease occurs in 51% of all cases according to data available from All India Institute of Medical Sciences. According to Freedman et al (1981), it is caused by organism prevalent in maternal genital tract and infection occur either due to ascending infection following rupture of membrane or during passage of baby through
birth canal or at the time of resuscitation in the labour room whereas late onset sepsis is nosocomial in origin.

High mortality is reported in early onset septicemia in various studies. Battisi et al (1981) reported a mortality of 64% for septicemia during first 48 hours and 29% for late onset septicemia. In the same hospital another study by Placzek et al (1983) also reported a high mortality of 70% in early onset septicemia as compared to mortality rate of only 12.5 percent for the late onset. Bhakoo et al (1974) and Chugh et al (1986) also reported a rate of 83.3% and 63.3% for early onset septicemia.

ETIOLOGY:

Frequency of infection by various organisms varies from one institution to another and even from year to year in the same institution. However, most of the Indian studies indicate that occurrence of Gram negative septicemia is approximately 1.5-3 times more than that of Gram positive septicemia. Most of the Indian studies (Nellian et al 1981, Sharma et al 1987, Mishra et al 1985, Khatua et al 1986) suggested that predominant aetiological agents of neonatal sepsis are Escherichia coli, staphylococcus aureus and
Klebsiella, other organism like pseudomonas proteus, enterobacter and salmonella has been identified to a lesser extent.

Starr (1985) reported that in last decade staphylococcus albus and streptococcus viridans have emerged as important pathogen in neonatal intensive care unit, where indwelling catheter is routinely applied. In the study Sinha et al (1986) pure Gram negative organism were causitive agent in 86.6% cases. Of these pseudomonas aeuroginosa was most common followed by klebsiella and E coli.

The incidence of various agent in septicemia is shown in table - I
### Aerobic Bacterial Isolates (%) from Cases of Neonatal Septicemia

<table>
<thead>
<tr>
<th></th>
<th>Bhakoo et al</th>
<th>Singh et al</th>
<th>Guha et al</th>
<th>Mishra et al</th>
<th>Monga et al</th>
<th>Mondal et al</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n=132 n=113</td>
<td>n=113</td>
<td>n=100 n=124</td>
<td>n=92 n=150</td>
<td>n=33</td>
<td></td>
</tr>
<tr>
<td>Klebsiella</td>
<td>08.3 06.0 18.6</td>
<td>14.0 76.1 58.6</td>
<td>26.2 01.6 19.5</td>
<td>15.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Escherichia coli</td>
<td>17.4 22.1 12.4</td>
<td>13.0 07.2 09.8</td>
<td>40.0 25.0 10.6</td>
<td>-</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pseudomonas</td>
<td>12.8 09.7 03.5</td>
<td>24.0 07.2 07.6</td>
<td>06.2 20.8 13.3</td>
<td>12.1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Strepto. faecalis</td>
<td>05.3 06.8 12.4</td>
<td>- - -</td>
<td>- - -</td>
<td>-</td>
<td></td>
<td></td>
</tr>
<tr>
<td>\textsuperscript{ß} Strepto. haemolyticus</td>
<td>00.7 - - 03.0</td>
<td>- - 03.4</td>
<td>03.7 - -</td>
<td>-</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Staph. aureus</td>
<td>12.0 35.4 30.0</td>
<td>21.0 04.8 01.1</td>
<td>22.5 34.1 21.2</td>
<td>12.1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Staph. albus</td>
<td>- - -</td>
<td>19.0 04.8 11.9</td>
<td>- - -</td>
<td>-</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Others</td>
<td>42.7 16.0 23.1</td>
<td>06.0 - 10.0</td>
<td>01.25 15.2 03.6</td>
<td>30.4</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Changing pattern of micro-organism causing neonatal septicemia:

The type of bacterial isolates from cases of neonatal sepsis have been varying with time to a remarkable extent the world over. In the west prior to 1940 predominant organism was group A beta haemolyticus streptococcus, which was replaced by staphylococcus aureus in 1950's according to Harris and Folin (1983).

Wilson & Eichenwald (1974) opined that predominant role of staphylococcus lasted only for a short time with the organism being replaced inturn by Gram negative enterobacteria. Since then many studies confirmed that group B streptococcus is most important cause of neonatal septicemia in Europe and America. However, a study from Finland by Veskari et al (1985) reported that incidence of group B streptococcus accounted for 53% in less than twenty hours, 14% in 24 hours to seven days and 27% in 8-28 days of onset, whereas E coli was found in 14%, 24% and 10% of the cases in three groups respectively.

Changing trend is evident from data available from various institution in our country also. Bhakoo et al (1974) and Guha et al (1979) reported E coli as the commonest pathogen. Reports by Khatua et al (1986)
and Chuyn et al (1986), showed significant increase in incidence of klebsiella. Notwithstanding individual studies, it can be concluded that in our country, the aerobic bacterial agents - Escherischa coli, klebsiella, staphylococcus aureus and pseudomonas are the important cause of neonatal sepsis according to Paul and Singh (1986).

The antibiotic sensitivity pattern have also been showing an alarming trend, with most of the organism showing increasing resistance to commonly used antibiotics like penicillines and aminoglycosides, employed as the first line of therapy. The most significant finding in a study from Bombay by Ronga et al (1986) is a sharp fall in percentage sensitivity of klebsiella, E coli and pseudomonas to gentamicin from 80-90 percent in 1981-82 to 20-57 percent in 1984. The sensitivity pattern staphylococcus to gentamicin has fortunately remained unchanged. All the Gram negative isolate from the Intensive care Unit in above mentioned study showed very poor sensitivity to ampicillin. Similar findings have also been reported by Guha et al (1978) and Gurmeet et al (1990).

Pre disposing factors for neonatal septicemia:

HOST FACTORS:

1. Sex:

  Neonatal septicemia is more common in males.

Males have an approximately 2 fold higher incidence
of sepsis, meningitis and UTI than females according to Vaughan and Behrman (1987).

The probable explanation is that factors regulating the synthesis of gamma globulin are probably situated on the X chromosome in the male infant confer less protection compared to female counterpart (Schaffer 1977).

In a study by Khatua et al (1986) 70.7 percent of total cases were male and mortality rate in male was also higher as 63% compared to female as 44.4 percent.

2. **Prematurity**:

Preterm babies are more vulnerable to catch infection and the incidence of sepsis as well as mortality is very high in premature babies. Khatua et al (1986) found that 63 percent of cases in their study were premature babies. They suggested that preterm babies has low level of IgG at birth because its maternal transfer is a function of gestational maturity. Inherent defensive machinism as well as both cellular and humoral immunity lack in preterm. Their complement system and mucosal defence are also poorer as compared to term infant according to Paul & Singh (1988). They also described low storage reserve in neutrophil is a contributory factor for development of sepsis in preterm babies.
Mc Craken and Shinefield (1966) reported mortality of 60 percent in premature babies as compared to 31% in term babies. Similar reports of high incidence and mortality were also obtained by various Western as well as Indian workers (Buetlow et al 1965, Bavkatte et al 1979, Bhskoo et al 1974, Guha et al 1978, Khatua et al 1986).

**Intrauterine Growth Retardation :**

According to Paul and Singh (1988) a neonate with IUGR suffers significant deficiency of cell mediated immune response. Their thymus is atrophied and relative as well as absolute number of T cells is remarkably diminished.

Meneriker et al (1976) opined that there is evidence to suggest that maternal transfer to immunglobulin is affected as a result of placental changes associated with IUGR.

Also small for date babies exhibit poorer neutrophil function in term of chemotaxis, phagocytosis and intra cellular killing as has been described by Chandra (1984).

**Agent Factors :**

**Inoculum size :**

Small neonate when exposed to a large inoculum of pathogens present in the genital tract and
environment have increased chance of infection (Paul & Singh 1988).

**Bacterial capsular antigen:**

The importance of capsular antigens in imparting virulence to group B streptococci and E Coli has been partially elucidated recently according to McCracken (1981).

Singh and Paul (1988) described that presence of K1 antigen on E coli is associated with invasive disease specially meningitis. K1 antigen consist of sialic acid moieties which block the alternate pathway of complement system enabling the organism to evade opsonization and phagocytosis.

**Bacterial toxin production:**

Neonate with gram negative septicemia has fulminant clinical course because bacteria produce endotoxin which mediate the reaction responsible for manifestation like shock, fever, intravascular coagulation and changes like thrombocytopenia, leucocytosis and complement activation according to Singh & Paul (1988).
**Tissue Tropism:**

According to Edwards & Baker (1984), the variable adherence of bacteria to epithelial cell surfaces is determinant of organ susceptibility to infection. *E. coli* and group B streptococcus have tendency to infect meninges, whereas *neisseria gonorrhoeae* have tissue tropism for eyes.

**ENVIRONMENT FACTORS:**

*Early rupture of membrane:


Blanc (1959) described a comprehensive account of progress of events leading to what he termed as "Amniotic infection syndrome". Fryles et al (1963) studied 138 infants born to mother who had history of leaking P/v more than 6 hours and his finding revealed that clinical sepsis was present in 31 percent of such babies.
In a study by Khatua et al (1965), the total of 65 percent cases of neonatal septicemia were associated with prolonged rupture of membranes.

**Maternal Genital Tract Flora:**

Throughout the pregnancy and until the rupture of membrane the environment of infant is normally sterile. Human birth canal is colonised by large number of bacteria, mycoplasma, chlamydiae, fungi, yeast and viruses. Kishore et al (1987) confirmed that 90% of pregnant women harbor potentially pathogenic organism of which Gram-ve organism were isolated in his study from 48.6 percent of pregnant women.

In the west where group B streptococcal infection is the leading cause of neonatal septicemia 5-30 percent of women population harbor it as has been suggested by Starr (1983).

**Nursery Flora:**

Hands of nursery personnel are a potent source of infection unless due precaution are taken. This fact has been nicely described by Davies (1982) in his article "Please wash your hand". High risk neonates are exposed to variety of fomites which are potential source of infection such as incubators, cots, linen, suction or

**Clinical Manifestation:**

Clinical features of neonatal septicemia is quite vague, nonspecific and subtle. According to Nyhan & Fousek (1958), the first manifestation may be failure to thrive, or the baby not doing well or may be an alteration in established feeding pattern.

The baby who had been sucken normally gradually or suddenly become inactive and irresponsible. He or She appears sick, pale with grayish blue circumoral cyanosis and vacant stare. These symptoms suggest the diagnosis of septicemia as has been described by Singh (1985).

Gotoff and Behrman (1970) tabulated the symptoms and signs of neonatal septicemia as below:

<table>
<thead>
<tr>
<th>CNS</th>
<th>Respiratory system</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lethargy</td>
<td>Tachypnoea</td>
</tr>
<tr>
<td>Hyporeflexia</td>
<td>Dyspnoea</td>
</tr>
<tr>
<td>Irregular respiration</td>
<td>Cynosis</td>
</tr>
<tr>
<td>Irritability</td>
<td>Apnoea</td>
</tr>
<tr>
<td>Full fontanelle</td>
<td></td>
</tr>
<tr>
<td>Aponea</td>
<td></td>
</tr>
<tr>
<td>GIT</td>
<td>CIRCULATORY CHANGES</td>
</tr>
<tr>
<td>-------------------------</td>
<td>------------------------------</td>
</tr>
<tr>
<td>Abdominal distension</td>
<td>Pallor/cyanosis/mottling</td>
</tr>
<tr>
<td>Hepatomegaly</td>
<td>Hypothermia</td>
</tr>
<tr>
<td>Vomiting</td>
<td>Abnormal respiration (Apnoea, Tachypnoea etc)</td>
</tr>
<tr>
<td>Diarrhoea</td>
<td>Cold clammy skin</td>
</tr>
<tr>
<td>Decreased stool</td>
<td>Hypotension</td>
</tr>
</tbody>
</table>

**General**

**Fever**

**Hypothermia**

**Sclerema Neonatorum**

**Laboratory Diagnosis:**

The clinical diagnosis of neonatal septicemia remains a difficult task as it is mimicked by various non infective conditions like hypoglycemia, hypothermia, intracranial haemorrhage etc or many a times, septicemia may be associated with any of the above mentioned conditions according to Chandra et al (1988). In such conditions we will have to depend on laboratory parameters for diagnosing neonatal sepsis. Laboratory tests which can help in the diagnosis of neonatal sepsis as have been suggested by various workers from time to time are -

Gastric aspirate cytology, examination of external ear
canal debris (Scalon 1971), leucocytosis more than 10,000/ cmm, leucopenia \( \leq 5000/ \text{cmm} \), B-band cell/Neutrophil ratio. Toxic granulation in neutrophil, thrombocytopenia (Williams et al 1970) and Buffy coat smear examination. Micro ESR (Evans et al 1970), acute phase reactant in serum, like CRP (Aimbender et al 1981) and leucocyte alkaline phosphatase activity (Donato et al 1979) are some of the other tests which have been done for the diagnosis of neonatal septicemia.

**ERYTHROCYTE SEDIMENTATION RATE (ESR):**

A laboratory test which measures the distance in mm that erythrocyte fall per unit time (usually one hour) is called erythrocyte sedimentation rate according to Wintrobe (1967).

The term ESR was first introduced by German Scholar Robin Fahraeus in 1918. He found out and confirmed that sedimentation depends upon ability of plasma to lower electrostatic charge on surface of RBC so that agglutination can occur.

According to Fishel (1967) the erythrocyte sedimentation rate depends upon the formation of rouleaux and aggregation of RBC's. The characteristics of RBC's and the shear forces of macromolecules are responsible for rouleaux formation.
Red cell sedimentation has three phases. Phase of aggregation, phase of rapid fall and lastly the phase of collection. In first phase vander wall forces weakly bind red blood cell together. The second phase of rapid fall takes place when red cell aggregate with macromolecules and forms clumps, which fall more rapidly than single cells. In the last phase of packing red cell mass collected on the bottom of the tube (Hintrope 1967).

ESR IN CLINICAL PRACTICE:

Fahraeus published his observations on this test in pregnancy in 1918. It was then that ESR was first adopted in clinical practice.

The erythrocyte sedimentation rate is a diagnostic test and it is simple, non-specific, inexpensive and widely acceptable test for tissue damage. A normal, low or elevated rate has been of great help in diagnosis and therapy of a wide range of disease. The ESR when elevated is useful first to determine the presence or absence of disease. Secondly to monitor the progression or improvement of an already recognised disease and finally to measure the response to therapy as has been described by Dacie & Lewis (1977).
**Micro ESR:**

Micro ESR method requiring only a few drops of capillary blood was described 60 years ago by Landau in 1933. Micro ESR value correlates closely with the Westergren level at 15 mm per hour and below. Value over 15 mm per hour correlate progressively with higher Westergren level. Micro ESR level of 25 and 30 mm per hour are equivalent to Westergren value of 40 and 70 mm per hour respectively according to Landau (1933).

**Normal values:**

Landau (1933) watched this parameter in 32 newborn over one week of age and reported a level of 1 - 2 mm per hour. However, he did not analyse his data in term of sex and birth weight.

Smith (1936) evaluated the micro ESR in infant aged 12 day to 6 weeks and observe the micro ESR level of 4 - 6 mm per hour in first 30 minutes.

Evans et al (1970) studied 100 normal male and females low birth weight infants of 3 days or less and 30 normal low birth weight babies in late neonatal period. His findings revealed that 95th percentile Micro ESR levels for both the sexes were 6 mm per hour in first 3 days of age and 11 mm in late neonatal period.
No correlation was noted between ESR, gestational age, birth weight in either premature or the full term group.

Adler & Denton (1975) studied 71 normal newborn including 26 low birth weight and 45 full term infant. The test was performed by the same person daily at same time for first two weeks of life. He found that the value for 95th percentile of micro ESR rises from 1 mm per hour at 24 hours of age to 17 mm per hour at 14 days of age. Majority of patients showed small daily fluctuations.

In a study by Karsten et al (1980) 125 newborns without infection were studied. They found that the value of micro ESR rose slowly from 2 mm per hour at birth to 4 mm per hour at 8 days of age.

Farida et al (1980) in their work described normal value for 75 healthy neonates and found that values of 8 mm and less indicated normality with 75% certainty.

Mishra et al (1981) considered 0-8 mm value during first hour for first week of life as normal.

Singh et al (1987) described that nearly 81% of normal newborn infants have micro ESR value less than 14 mm per hour.
Micro ESR in neonatal infections:

Landau (1933) and Smith (1936) were the first workers to describe a micro ESR method requiring few drop of capillary blood. However, their data is this parameter for neonates were quite limited.

Evans et al (1970) observed an elevated value of micro ESR in 8 of 9 newborn infants with serious infection they studied. In addition normal values of ESR observed in idiopathic respiratory distress syndrome may be useful in differentiating this entity from infectious process.

Adler and Denton (1975) observed that all the infected patients had elevated micro ESR at sometime in the course disease except two patient with DIC. They observed that in most of the patients with in twenty four hours of onset of infection the micro ESR values were elevated. In majority of cases the values returned to normal with clinical improvement. The workers opined that this test is very useful in the evaluation of the patient whose culture results are affected by concurrent antibiotic therapy. Most of the noninfected neonates with moderate to serve respiratory distress syndrome or without any serious illness had value well with the normal range.
Boyle et al (1978) performed micro ESR test on 60 neonates. They observed that rise in micro ESR values takes place 24 to 48 hours after development of infection.

Parida et al (1980) observed that 74.4% of definitely infected and 24% of probably infected babies had elevated values of micro ESR. However, three cases of DIC had low values of micro ESR. These low values were thought to be due to lowered fibrogen levels as has been suggested by Edson et al (1967) and Beiger et al (1971).

Namdeo et al (1985) studied micro ESR with other haematological parameter for diagnosis of sepsis. They found an elevated values i.e. more than 8 mm in 24 out of 25 culture positive and 8 out of 25 culture negative infants.

Singh et al (1987) evaluated different parameters of sepsis screen for diagnosis of neonatal sepsis and found that 55% cases of definite sepsis and 62% cases of probable sepsis micro ESR was more than 14 mm per hour.
Mishra et al (1989) performed simple haematological tests for diagnosis of neonatal sepsis and observed that elevated micro ESR i.e. more than 8 mm during first hour was found in 73% cases of proved sepsis and 22% cases of probable sepsis.

Sharma et al (1993) reported mean micro ESR value on first day as 15.3 mm first hour in clinically suspected septicemia and 21.1 mm first hour in clinically suspected septicemia newborn who in addition had obvious focus of infection like pyodlerma, pneumonitic etc. These workers also reported that there was no significant variation in value of micro ESR in culture positive and culture negative cases. They also ruled out any prognostic significance of micro ESR.

C-reactive protein:

Pepys (1982) described C-reactive protein as acute phase reactant. Acute phase reactants are group of plasma proteins, concentration of which increases following most forms of tissue injury or inflammation or association of malignant neoplasia.

While a number of acute phase reactants have been described for early diagnosis of neonatal septicemia such as fibrinogen, C-reactive protein, orosmucoid and pre-albumin, majority of them have low sensitivity. CRP,
however, is a notable exception and therefore, been found clinically applicable as reported by Sann et al (1984).

The discovery of C-reactive protein was reported in 1930 by Tillet and Francis. They were investigating serological reactions is pneumonia with various extracts of pneumococci. The workers observed that a non type specific somatic polysaccharide - CPS fraction which, they designated as fraction C and was precipitated by sera of acutely ill patient. After the illness the capacity of the patients sera to precipitate C poly saccharide rapidly disappeared.

Abrenethy and Avery (1941) characterised C-reactive protein as a protein which required calcium ion for its reaction with CPS and introduced the term acute phase to refer to serum from patients acutely ill with infectious disease and containing the C-reactive protein.

Lofstrom (1944) independently described a non specific capsular swelling reaction of some strains of pneumococci when mixed with acute phase sera and subsequently showed that substance responsible was CRP. He detected CRP in infectious as well as non infectious conditions.
Structure & Synthesis:

Osmond et al (1977) described structure of CRP and proposed that molecule of human CRP (mole weight 105,500) is composed of five identical non glycosylated polypeptide unit which are non covalently associated in a disc like configuration with cyclic pentameric symmetry.

Hurliman et al (1966) reported that CRP is synthesized by hepatocyte and no evidence is obtained of its production by any other cell type.

Kushner et al (1978) stated that early after inflammatory stimulus CRP production was increased in periportal hepatocytes and with time, recruitment of cells to CRP production proceeded centripetally in liver lobule until most of the cells were involved.

Properties:

Claus et al (1970) reported that there was a rapid rise in C-reactive protein concentration from normal level by as much as 3000 fold in response to acute tissue injury, inflammation or infection.

Klindmark (1971) reported that CRP enhance phagocytosis of various bacterial species by peripheral blood leucocytes in a serum free medium.
CRP is a potent activator of complement system when reacting with capsular polysaccharide and choline phosphatidite. According to Seigel et al (1975) CRP has the power to combine with T lymphocyte and inhibit certain of their functions. It can also bind with Fe receptor of macrophages to bring about phagocytosis by acting as an opsonin as has been suggested by Mortenson et al (1977).

Pepys's (1981) reported that CRP may take part in pathogenesis of many inflammatory conditions in which its circulating concentration is elevated, it seem unlikely that this is its major role. He opined that increased concentration of CRP in plasma only indicates that potentially toxic antigenous material is released from damaged tissue.

Normal values:

Nudelman et al (1984) reported the CRP level in normal individual as follows:

<table>
<thead>
<tr>
<th>Age group</th>
<th>Normal maximum level (Mg/Lit)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cord blood</td>
<td>10</td>
</tr>
<tr>
<td>Neonate &lt; 1 week</td>
<td>30</td>
</tr>
<tr>
<td>Neonate 1 week-1 month</td>
<td>10</td>
</tr>
<tr>
<td>Infant</td>
<td>10</td>
</tr>
<tr>
<td>Children</td>
<td>02.2</td>
</tr>
<tr>
<td>Adult</td>
<td>03.0</td>
</tr>
</tbody>
</table>
Peltola (1984) reported that value upto 20 mg/lit may be considered as normal. Among Indian workers Chandana et al (1988) considered value of \( \geq 76 \) ug/ml as abnormal, while Sharma et al (1993) considered the value of \( \geq 6 \) ug/ml as abnormal.

**CRP in clinical practice:**

In various studies efforts have been made to establish relationship between CRP concentration and infectious, non infectious and inflammatory conditions. Johansson et al (1972) and Pepy (1982) in myocardial infraction, Jodal et al (1975) in kidney infections, Fischer et al (1976) in post operative infections in surgical patients reported importance in CRP estimation.


CRP as Screening Test for neonatal sepsis:

In an effort to evaluate comparative role of different test used to diagnose neonatal sepsis, Desai et al (1982) observed high sensitivity (67.5%) and specificity (83.3%) for CRP test among four parameter viz, Band/total neutrophil, leucocytes ≥5000/cm, micro ESR and C-reactive protein, they used.

Singh et al (1987) in their study found CRP as most sensitive (80%) and specific (91%) screening test for neonatal sepsis.

Chandana et al (1988) also documented that CRP was the most useful simple test with high degree of sensitivity (83%), specificity (43%) and positive accuracy (57%) while comparing with TLC, Band cell/Neutrophil ratio, buffy coat smear and gastric aspirate cytology test for diagnosis of neonatal sepsis.

CRP in neonatal sepsis:

Ainbender et al (1982) studied infants who were prone to develop septicemia due to H/o prolonged rupture of membranes and maternal fever. They observed that the raised CRP value is obtained in significant number of cases and maximum value is obtained during first three days.
Kindocho et al (1984) reported that eleven out of twelve infants of proven sepsis had raised serum CRP which decreased significantly after the successful treatment.

Vyas et al (1985) evaluated the role of CRP in neonatal infections. They worked with hundred neonates of suspected septicemia of which some were definitely infected and others were probably infected. They found that CRP test was better tool to distinguish between definitely infected from probably infected as compared to routine haematological investigations.

Kalra et al (1985) estimated CRP in various neonatal infections. They found that CRP is equally effective diagnostic tool as blood culture in neonatal septicemia, while is superficial infection, it is not of much importance.

Sethi et al (1990) evaluated role of CRP in superficial and deep infections of neonates. They reported that CRP was raised in 100% cases of deep and 50% cases of superficial infection. Rate of detection and mean percentage were also significantly higher in deep infections because of greater tissue destruction.
Suri et al (1991) compared the diagnostic and
prognostic utility of CRP with other acute phase
reactant viz, alpha-1 antitrypsin, and alpha-2 macro-
globulin and reported that CRP is better parameter for
early diagnosis and predicting outcome.

Sharma et al (1993) reported diagnostic and
prognostic utility of CRP in neonatal septicemia. They
estimated CRP in septicemic neonates on 1,7,14 days and
found that on first day CRP was raised in all cases of
septicemic however, on fourteenth day it touched zero
among those babies who had improved however, in babies
who deteriorated or died the values showed an increasing
tendency.

Leucocyte alkaline Phosphatase :

First idea of leucocyte alkaline phosphatase
was proposed by Kay in 1929, who on working with
plasma phosphatase in osteitis deformans and other
diseases of bone suggested, that neutrophil contain a
phosphatase enzyme, the optimal activity of which was
around 9 and phosphomonoesterase in nature.
However, the worker reported that specially there is
no correlation between leucocyte alkaline phosphatase
and serum alkaline phosphatase.
Wachstein and Middle Town (1946) reported a series of cases consisting of 28 normal subjects and some cases of infection and leukemia. They found that some leucocytes were weakly phosphatase positive in normal subjects and also in patients of leukemia, while in infections, staining was considerably more frequent. These workers also reported that phosphatase concentration was higher in bone marrow precursor than in peripheral leucocytes.

Plum (1950) reported that one third of leucocytes of human blood contain alkaline phosphatase. In his study alkaline phosphatase activity was normal in hypochromic anemia higher in polycytemia vera and decreased in myeloid leukemia.

Kerppola (1951) demonstrated high concentration of leucocyte alkaline phosphatase in cases of panmyelopenia, myeloma, Hodgkin's disease, certain leukemias, bone tumours, malignant liver and pernicious anemia after the use of antianemic substance. In the same year Valentine et al. (1951) made several studies in leucocyte alkaline phosphatase. According to them the phosphatase concentration was increased in leucocytes after operations, trauma and in polycytemia vera,
normal in rheumatoid arthritis, rheumatic fever and disseminated lupus erythematosus and low in chronic myeloid leukemia. These workers also reported increased LAP activity after administration of cortisone and ACTH.

Sirola & Sirola (1957) studied LAP in blood and bone marrow. They observed that presence of alkaline phosphatase is not a characteristic of any specific disease. However, it was absent in hypochromic anemia, pernicious anemia, allergic disease collagen disease, endocrinal disease and disease of nervous system. LAP activity was higher in acute infections, panmyelopenia, Hodgekin's disease and cirrhosis of liver.

Trubowitz et al (1961) made an attempt to isolate purify and describe some properties of human leucocyte alkaline phosphatase. They reported that 250 fold purification could be attained with best preparation having a specific activity of 4075 ug of phosphorus per mg nitrogen per hour. The purified enzyme contained 44.1 ug of zinc per milligram of nitrogen. The least purified preparations were capable of liberating inorganic phosphorus from mono or disphosphorylated nucleotide.
King et al (1962) studied leucocyte alkaline phosphatase activity in mongolism and reported significantly high value in both males and females. They suggested that LAP activity is controlled by gene situated on chromosome 21 and in mongolism, there is trisomy of this chromosome, thus extra gene available to give high values of LAP activity.

**Cell types showing LAP activity:**

Wachstein (1946), Plum (1950) demonstrated that LAP activity is confined to the cytoplasm of only mature neutrophils while all other blood cells failed to show any enzyme activity, but contrary to this Vercauteren (1951) reported presence of this activity in eosinophils also. Four year later Kaplow (1955) could not demonstrate LAP activity even in a single eosinophil in a smear of absolute neutrophil count of 34,000 per cu/mm and contradicted the earlier report.

Hayhoe and Wonglino (1958) reported alkaline phosphatase activity in the cytoplasm of limited number of neutrophils.
Miller et al (1983) reported that alkaline phosphatase activity is confined to the tertiary granules of neutrophils.

Trubowitz et al (1958) also reported that LAP activity is confined chiefly in polymorphs one year later same workers reported that the bone marrow polymorphs contained 50 percent less activity than that of peripheral smear. They postulated that polymorphs synthesise additional alkaline phosphatase after their release from marrow and during their life span in blood and tissue.

**LAP activity in neonatal period:**

LAP score may differ from laboratory to laboratory and in addition to an objective criterion of assessment. The result are liable to altered by room temperature, reagent used delay in examination of slides, thickness of smear and circadian changes according to Sharma et al (1983). However, normal adult value described by Corberand et al (1973) were 105.5 ± 52.5.

Okell and Axon (1965) reported that LAP scores of cord blood of 25 newborns ranged from 163 to 303 with mean of 236, whereas, normal adult range in their
study was 60 ± 20. They suggested that higher values in neonates at time of birth were due to free passage of progesterone through placenta, a hormone, which is also responsible for high LAP scores in pregnant ladies as has been reported by an earlier worker Pritchard (1957).

Kommissarova et al (1971) reported LAP score in 3–5 days old neonates as 168 ± 16.4 and 5–30 days old neonates as 135 ± 9.8 respectively.

Halbrecht and Shabtay (1972) compared LAP scores of preterm, full term and post term neonate with that of adults. They observed that term and post term neonate of either appropriate or low birth weight had LAII activity higher than normal adult however, premature babies had a significantly low levels. They suggested that LAII activity may be used as an extra criterion for prematurity. They also examined LAP activity with respect to the influence of maternal toxemia, effect of sex and possible relationship to ABO blood group but no statistically significant difference were found.
Corberand et al (1973) also compared LAP activity of newborns and adult along with four other cytochemical leucocytes reactions and found that newborn had distinctly higher values than adults.

**LAP activity in neonatal infections:**

LAP activity determined by cytochemical method is known to be increased in bacterial infection of adults, however, in majority of studies in neonatal infections, it is found to be decreased.

Donato et al (1979) determined LAP activity in 36 healthy newborns and 13 neonates with severe bacterial infections like septicemia, pneumonitis and meningitis. They observed significant low scores in infected neonates. In their study all the healthy newborn had LAP scores ranging from 163-270, whereas, all infected newborn had LAP score below 160. They also denied any significant difference in LAP activity between neonates who survived or those who died. LAP activity was found to be a better indicator than other cytochemical diagnostic test like Nitroblue tetrazolium test.

Sharma (1980) studied LAP activity in 32 healthy newborn and equal number of infants with infections like septicemia, pneumonitis and meningitis etc. He observed
that healthy neonates had significantly higher LAP scores than those of infected ones. In his study all the healthy neonate had LAP score between 144 to 244, whereas, among all infected neonates scores were below 132. In his study there was also no correlation between LAP score and mortality. He suggested that low LAP activity could be explained by rapid release of functionally immature neutrophil by bone marrow which had low LAP enzyme content.

Sharma et al (1983) compared LAP activity of healthy neonates below one month and babies under one year of age to babies with bacterial as well as non-bacterial infections. They reported that mean LAP scores were 142 for healthy babies, 115.9 for bacterial infection and 120.9 for non-bacterial infection below one month age group. Among infants upto one year age group, mean LAP score for healthy babies was 100.5 for bacterial infection, 147.7 and for non-bacterial infection 95.8 score was obtained. They observed that the value of LAP score was much lower in severe infections like pyogenic meningitis as compared to minor, illness like gastroenteritis and boil. In their study, fall in LAP activity was reported in
neonates having bacterial as well as non-bacterial infection but due to high standard deviation, and narrow margin between mean value of both groups, these two cannot be differentiated on basis of LAP score.

Paul and Kumar (1984) reported 'high' LAP activity in neonatal infections in contrast to other studies. They determined LAP scores of 15 newborn with bacterial infection, 15 of viral illness and 10 controls. All the cases in their study were full term. The mean score obtained was 87.5 for control group, 160.3 for viral illness and 232.7 for bacterial infections. They explained that low scores obtained in earlier studies were due to inclusion of premature babies in study group, who distinctly had LAP score significantly low as has been suggested by Halbrecht and Shabtay (1972).

Sharma et al (1985) studied LAP activity in 29 preterm and 45 term neonates. The cases were of healthy neonate; neonates with suspected septicemia and superficial infections. They reported that 91.7% term neonates and 92.8% preterm neonates with confirmed bacterial infections had LAP score significantly below the control group, while all the term and 81.1% of preterm babies with clinically suspected septicemia
had LAP score below the range of control value. Although no significant difference was found in LAP activity of healthy control and those with superficial infections. There were also not significant variation of scores of fullterm and premature babies of all the study groups.

Sharma et al (1989) evaluated LAP score and other haematological indices for early diagnosis of neonatal sepsis and reported that LAP activity was significantly low (P<0.001) in both preterm and term neonates with confirmed bacterial infection and clinically suspected septicemia as compared to healthy neonates but the procedure is time consuming and requires high degree of precision.