Introduction

The incidence of sepsis is higher in neonates than in adult patients, and the risk of mortality is higher, in particular, neonates with low birth weight show relatively high morbidity and mortality ***(Jordan et al., 2006).***

Furthermore, neonatal sepsis is difficult to diagnose, as clinical signs are often obscure and laboratory parameters are unspecific. No clear-cut definition of neonatal sepsis has been agreed to, although several studies have tried to identify one. Clinicians thus permit over-treatment based on the high risk of mortality if sepsis is left untreated ***(Chan et al., 2009).***

Currently, there are no reliable biochemical markers for confirmation of sepsis. Blood culture tests are still considered the gold standard for the diagnosis of sepsis although results are not available until at least 48 hours. There is a need for an effective and accurate biochemical marker to support or exclude the diagnosis of infection ***(Mishra et al., 2006)***.

Recent epidemiologic studies have shown increasing numbers of invasive fungal infections (IFIs), main pathogens responsible for these infections are yeast-like fungi from *Candida* genus and *Aspergillus* spp. moulds ***(Pagano et al., 2006) (Enoch et al., 2006)***.

Candida species are an increasingly important cause of invasive candidiasis (IC) in patients hospitalized in intensive care units (ICUs). Studies have shown that the incidence of IC in ICUs peaks on the 8th to 10th day of stay in the ICU. Prolonged hospitalization in the ICU is the single most important risk factor for IC development, other important risk factors for IC include central venous catheterization, post operative, acute renal failure, diabetes, haemodialysis, and broad-spectrum antibiotics. Risk of IC is also high among patients who are neutropenic ***(Martino and Subirá, 2002)***.

IC is associated with high mortality rates reaching up to 40–50%. Early and precise diagnosis of deep mycosis is difficult because of lack of specific symptoms. In numerous comparison studies it has been shown that blood cultures, despite monitoring on a daily basis, are positive in only 50% of cases ***(Yera et al., 2001) (Ostrosky-Zeichner and Pappas et al., 2006)***.

Classic mycological examinations, including histo-pathological assessment, microscopic methods and cultures of clinical samples, also have a limited diagnostic value ***(Marr et al., 2002)***.

Serological methods used in fungal diagnostics are generally based on detection of cell-wall components of selected pathogenic fungal species, i.e. mannoproteins, functioning as soluble antigenic markers. During the course of a systemic fungal infection these antigens are present in the

bloodstream only transiently, and they are eliminated by forming immune complexes as well as via endocytosis by Kupffer cells in the liver ***(Yeo and Wong, 2002)***.

Immunodiagnostics of IC rely on the detection of circulating mannan antigen in the serum, being the main component of the fungal cell wall of *Candida* spp. Mannan is a highly immunogenic polysaccharide antigen with immuno-modulating properties, and its oligomannose epitopes may induce humoral response ***(Sendid et al., 2003)***.

Aim of the Work

This study aims to evaluate the use of serum *Candida* mannan antigen in newborn infants as a marker of neonatal fungal sepsis.

Neonatal Sepsis

Definition:

The term neonatal sepsis describes a disease of infants who are younger than 1 month of age, are clinically ill, and have positive cultures ***(Schelonka et al., 2005)***.

Classification of Neonatal Sepsis:

1. Early-Onset Sepsis (EOS):

Neonatal EOS is defined by the Centers for Disease Control (CDC) as blood or CSF culture-proven infection occurring in a newborn who is younger than 7 days of age ***(Phares et al., 2008).***

2. Late-Onset Sepsis (LOS):

Neonatal LOS is defined as occurring from 8 to 90 days of life. The risk factors for LOS in premature infants are related to the necessities of their care and the bacteria that cause LOS are often acquired in the NICU ***(Puopolo, 2008)***.

3. Very Late-Onset Sepsis:

Very late-onset sepsis occurs after 3 months of life. It affects premature infants who are of very low birth weight (VLBW) in the neonatal intensive care unit. This infection has usually been associated with prolonged instrumentation.

Overall mortality was 18% of infected infants versus 7% of uninfected infants. And the neonatal mortality rate in Egypt was 25 per 1000 live births ***(Edwards, 2011).***

Risk Factors for Neonatal Sepsis

1- Maternal Risk Factors:

Maternal factors may influence the development of systemic bacterial infection in the neonate. Maternal factors such as malnutrition and recently acquired sexually transmitted diseases can also increase the risk of infection ***(Schrag et al., 2000)***.

This risk is increased if colonization is associated with prematurity, maternal fever, or prolonged rupture of membrane (ROM). Asymptomatic bacteriuria has been associated with premature birth ***(Edwards, 2011).***

2- Peri-partum Risk Factors:

Some of the peripartum factors associated with an increased risk of neonatal infection are untreated or incompletely treated focal infections of the mother (including urinary tract, vaginal, or cervical infections), as well as systemic infections, such as maternal septicemia or maternal fever without a focus. and in the presence of prolonged ROM has been associated with an increased incidence of sepsis ***(Edwards, 2011)***.

3- Neonatal Risk Factors:

Although no significant sex difference has been documented for infections acquired in utero, it was noted in the 1960s that male infants had a higher incidence of neonatal

sepsis than female infants. Possibly, this is related to X-linked immune-regulatory genes ***(Edwards, 2011)***.

Factors that may affect late-onset neonatal infection include prior antimicrobial use; prematurity; a high infant-to-nurse ratio in the neonatal intensive care unit; and the presence of foreign materials such as endotracheal tubes. Contaminated parenteral fluids, including lipid emulsions, also have been associated with systemic infections ***(Edwards, 2011)***.

4- Other Risk Factors:

It has been suggested that bottle feeding may predispose to infection. Prepared formula does not contain several important biologic factors found in colostrum, which have a local gastrointestinal protective effect against gram negative enteric bacilli. Breast milk also contains immunoglobulins, macrophages, and lymphocytes, all of which play a role in immunologic defense ***(Edwards, 2011)***.

Diagnosis of Neonatal Sepsis

The diagnosis of sepsis is challenging due to the nonspecific nature of clinical presentation,

1. Obtain a Focused History:

Infants have a predisposition to infection. The pathogenesis of this predilection includes a complex interaction between obstetric and newborn factors; therefore, both the mother's and the infant's history are critical to a comprehensive risk assessment ***(Short, 2004)***.

- Obstetrical History

Obstetrical history should include maternal health before pregnancy. Prenatal care as well as antepartum and intrapartum events, any risk factor associated with preterm labor is a risk factor for neonatal sepsis ***(Klein et al., 2006).***

- Neonatal History

Events surrounding the labor and the infant's need for resuscitation as well as measures used in the early stabilization should be reviewed. Being cared for in a NICU is a risk factor for LOS due to invasive medical procedures and environmental issues ***(Klein et al., 2006)***. **Table 1** outlines additional neonatal risk factors for infection and/or sepsis.

Table (1): Neonatal Risk Factors for Infection/Sepsis

Immaturity of Immune System: ➢ Immature reticuloendothelial system. ➢ Lack of adequate humoral immune responses. ➢ Low immunoglobulin and complement levels and function. ➢ Impaired cellular and phagocytic activity. ➢ Prematurity (heightens immaturity of immune system already presented) ***(Wolach, 1997)***.
Anatomic Defects (late onset >3-5 days): ➢ Obstructive uropathy. ➢ Gastroschisis ***(Klein et al., 2006).***
NICU Care/Environmental Issues (late onset >3-5 days): ➢ Frequent invasive procedures - Poor skin integrity. ➢ Repeated courses of antibiotic therapy. ➢ Indwelling vascular catheters and long-term mechanical ventilation. ➢ NICU outbreaks, understaffing and impact on handwashing. ➢ Long-term glucocorticoid therapy ***(Klein et al., 2006)***.
Poor Nutrition: ➢ Delayed initiation of enteral feedings. ➢ Prolonged period to reach full enteral feedings. ➢ Prolonged period of time to regain birth weight ***(Stoll et al., 2002)***.
Complications of Prematurity: ➢ Patent ductus arteriosus. ➢ Bronchopulmonary dysplasia. ➢ Necrotizing enterocolitis **(Stoll et al., 2002)**.

2. Physical Assessment

Non-Specific Features of Sepsis:

Signs of fetal distress can be the earliest indication of infection in neonates with sepsis. Fetal tachycardia in the second stage of labor was evaluated as a sign of infection. A low Apgar score, suggesting distress at or before delivery, also has been correlated with sepsis in the newborn period ***(Klein et al., 2006)***.

Neonatal sepsis is often subtle and need a high index of suspicion for early diagnosis. Babies with sepsis may present with one or more of the following symptoms and signs:

a. Hypothermia or fever (former is more common in low birth weight babies).
b. Lethargy, poor cry or refusal to suck.
c. Poor perfusion or prolonged capillary refill time.
d. Hypotonia or absent neonatal reflexes.
e. Bradycardia or tachycardia.
f. Respiratory distress, apnea or gasping respiration.
g. Hypoglycemia or hyperglycemia.
h. Metabolic acidosis.

(Rajiv et al., 2001).

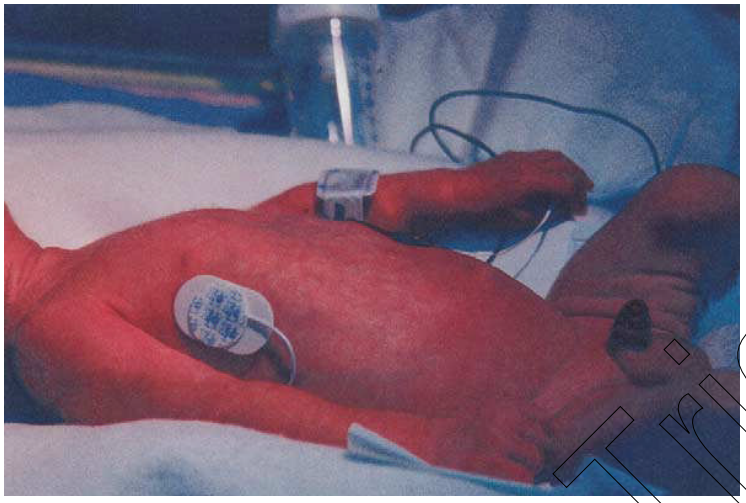

Figure (1): Generalized symmetrical hypotonia in suspected sepsis ***(Short, 2004)***

Specific Features Related to Various Systems *(Short, 2004):*

a. Neurological Manifestations:

- Bulging anterior fontanelle.
- High-pitched cry.
- Excess irritability.
- Not arousable.
- Comatose.
- Seizures.
- Neck retraction.

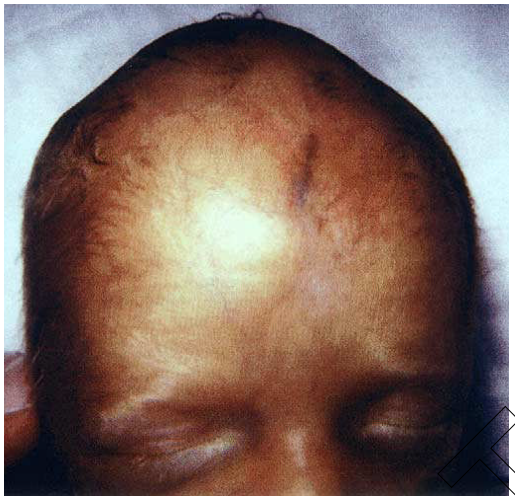

Figure (2): A bulging fontanel may be associated with meningitis ***(Short, 2004)***

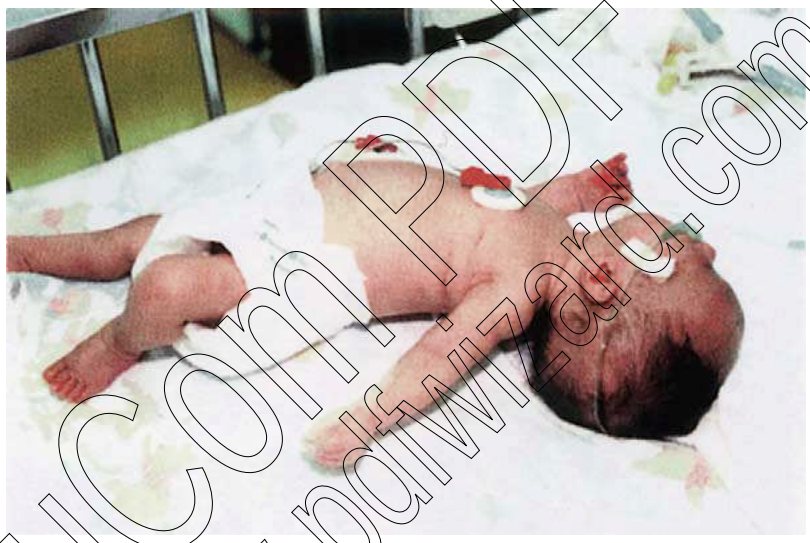

Figure (3): Opisthotonic positioning may be a sign of meningitis, or alternatively, may be seen in an infant who is attempting to compensate for airway edema or stridor ***(Short, 2004).***

b. Respiratory Manifestations (Ohlin et al., 2010):

- **Tachypnea:** Persisting breathing rate > 60 breaths per minute and not responding to standard nursing procedures.

- **Apnea:** onset of repeated episodes of apnea (lack of breathing movements >20 sec) leading to active intervention.

- **Increased oxygen need:** any increase in the amount of oxygen needed to obtain oxygen saturation of 88–92% persisting > 60 min. Rapid fluctuation in oxygen need around a stable baseline was not considered as increased oxygen need.

c. Cardiac Manifestations:

- Hypotension.
- Poor perfusion.
- Shock.

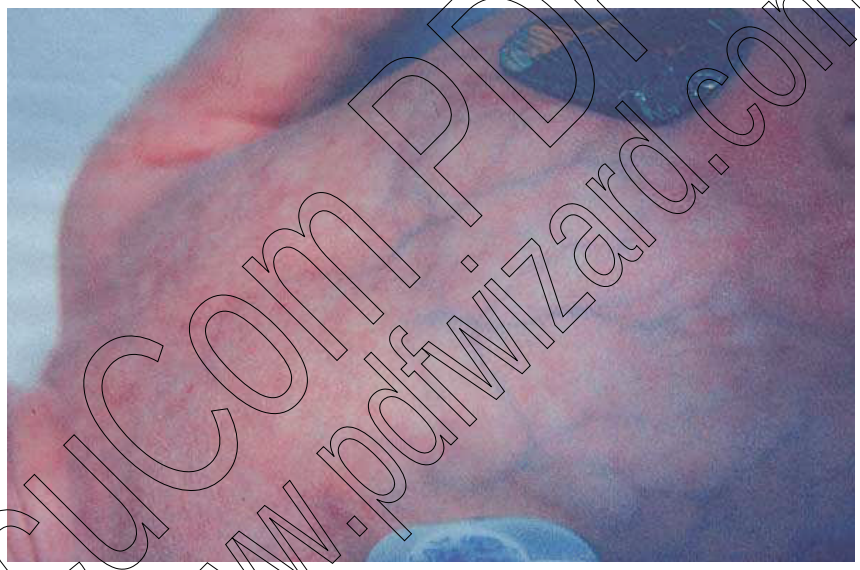

Figure (4): Diffuse mottled, bluish-gray appearance of this infant's skin suggestive of systemic poor perfusion ***(Short, 2004).***

d. Gastrointestinal Manifestations:

- **Feed intolerance:** difficulty in accepting enteral feedings presenting as repeated vomiting or increased gastric

retention, leading to a decrease in the frequency of enteral feeds ***(Ohlin et al., 2010)***.

- Diarrhea.
- Abdominal distension.
- Paralytic ileus.
- Necrotizing enterocolitis (NEC).

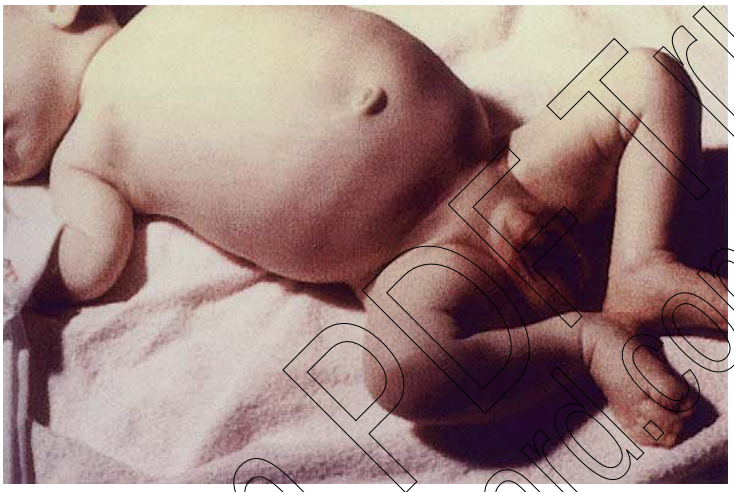

Figure (5): Abdominal distension is a nonspecific sign of infection and sepsis. Bowel loops are visible through the abdominal wall (Short, 2004).

e. <u>Hepatic Manifestations:</u>

- Hepatomegaly.
- Direct hyperbilirubinemia (especially with Urinary Tract Infection (UTI)).

f. <u>Renal Manifestations:</u>

- Acute renal failure occurred in 26% neonates with sepsis. Although ARF in neonates has been reported to be predominantly oliguric, it was observed that ARF secondary to neonatal sepsis was predominantly non oliguric. Low birth

weight was an important risk factor for the development of ARF ***(Mathur et al., 2006)***.

g. Hematological Manifestations:

- Bleeding.
- Petechiae.
- Purpura.

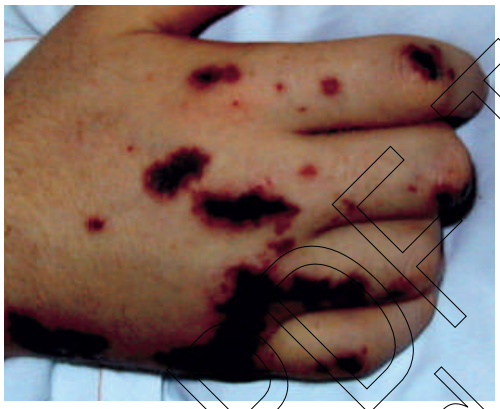

Figure (6): Purpura fulminans ***(Short, 2004)***

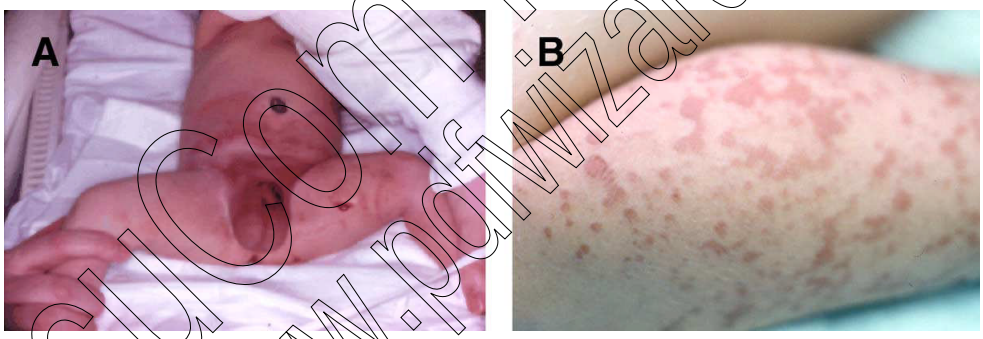

Figure (7): Petechiae and Purpura

(A) Petechiae. Infant with an infected circumcision; the responsible organism was Group B streptococcus. Scattered petechiae are surrounding the penis and extending upward to the periumbilical area.

(B) Purpura. Area of small hemorrhages on the right leg of the infant compared to the petechiae (minute, pinpoint-sized hemorrhages) in Figure 7A. Purpura is often associated with decreased platelet counts ***(Short, 2004)***.

h. Skin changes:

- Multiple pustules.
- Abscess.
- Sclerema.
- Mottling.
- Umbilical redness and discharge.

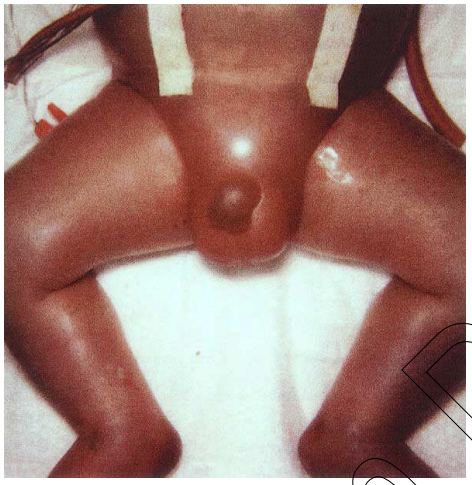

Figure (8): Swollen subcutaneous tissue and shiny edematous skin, often called sclerema; is a late sign in sepsis, often associated with positive fluid balance ***(Short, 2004).***

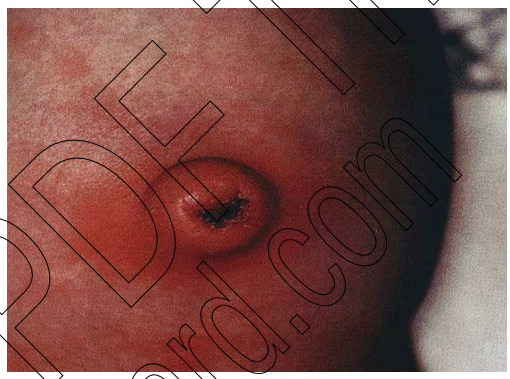

Figure (9): Omphalitis can present as periumbilical erythema, warmth, or drainage; the infection may be localized or may be a source for a systemic infection ***(Short, 2004).***

Diagnostic Markers of Infection in Neonates

A wide variety of hematological and biochemical markers have been investigated for the evaluation of clinical sepsis.