Macrosopic and microscopic findings of infant lung in case of live or still birth

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In the case of a newly born infant, either stillborn or found dead, the state of the lungs is of particular forensic interest. The aim of this study was to determine whether lungs are adequate for pathological comment about stillbirth or live birth of an infant. In order to evaluate the relevance of 171 autopsy reports of infants, macroscopic and microscopic findings, and body measures at autopsy, microscopic investigations on the degree of alveolar expansion performed on the lung specimens were obtained. Results revealed on the one hand, that the lung filled the thoracic cavity and covered the anterior surface of the heart in 45 (26.37%) cases; on the other hand did not fill the thoracic cavity and presented at the back side of the thoracic cavity in 56 (32.7%) of the cases. In a total of 40 histological sections, 19 (47.5%) uniform, 10 (25%) semi-collapsed, 11 (27.5%) collapsed alveoli sections in shape, 11 (28.2%) amnion aspiration, 9 (23.1%) mild amnion aspiration, 9 (23.1%) moderate amnion aspiration, 10 (25.6%) evident amnion aspiration and 22 (57.9%) pulmonary emphysema were determined. The paper thus revealed macroscopical findings of infant lungs with combination of histological findings were more formative than their histological findings alone in the assesement of still or live birth of an infant.

Key words: Infant, stillbirth, lung, live birth, autopsy.

INTRODUCTION

The body of a newborn is very often disposed of in drains, rivers, or rubbish dumps etc (Hausmann et al., 2004; Ong and Green, 2003; Lavezzi et al., 2003). Some births especially in rural area of Turkey occur at home without medical assistance. If any infant is found dead in Turkey, the body is sent to the autopsy center by the public prosecutor. Most parents claim that their child was dead before birth suspecting infanticide. Some parents also claim about medical malpractice. Determining live birth in these instances may make a difference in subsequent criminal proceedings. To investigate a dead infant about live or stillbirth is important in relation with both penalty code and civil law. Civil law in Turkey order that if any infant was live birth, infant would have all civil rights since the time of fetus fertilization (Soysal and Cakalir, 1999). The contract law also stipulates that healthy and executed birth is in advance of fetus rights (Code of Obligations, 2010 – www.mevzuat.adalet.gov.tr/html/407.html).

The determination of live birth is one of the most important aspects of the autopsy of an infant whose death has been found suspected. A careful evaluation of clinical data and family diseases, investigation of death scene area, accurate postmortem examination is very
important in differential diagnosis of live birth or stillbirth. Postmortem examination includes external examination, weight of organs, macroscopic and microscopic evaluation of lungs, hydrostatic tests of lungs, evaluation of umbilical cords, stomach contents and placenta. Natural diseases, congenital anomalies, trauma and birth injuries that could have caused or contributed to death have naturally been included in medicolegal investigations of infant autopsies (Janssen, 1984).

In the case of a newly born infant, either stillborn or found dead, the state of the lungs is of particular forensic interest (Janssen, 1984; Weibel et al., 2007). The critical evaluation of the histological and macroscopic examinations may aid in solving the case (Lavezzi et al., 2004). Besides histopathological examination, macroscopic appearance, the texture, border features of lungs and their filling of chest cavity are valuable parameters in diagnosis of live or stillbirth. Some authors state that pulmonary interstitial emphysema is also an important parameter in differentiation of live or stillbirth. Pulmonary interstitial emphysema is defined as interstitial dissociation of the lung with the air due to degradation of alveolar structure. Pulmonary interstitial emphysema is accepted as a certain live birth criteria by some authors. There is also some discussion that pulmonary interstitial emphysema may occur by artificial respiration or putrefaction (Lavezzi et al., 2003; Marchetti et al., 2007; Busuttil and Keeling, 2009).

The aim of this study was to determine whether lung samples together with other macroscopic and microscopic autopsy findings are adequate for pathological comment about stillbirth or live birth of an infant. We compared our findings which had an impact on the reports’ conclusions about live or still birth of infants and reviewed difficulties and deficiencies encountered during the evaluation of these cases.

**MATERIALS AND METHODS**

In this study, retrospective data in the period between 1999 and 2006 and investigating vitality of cases during birth by prosecutor was obtained from the infant autopsy database of the 1st Specialization Board of the Council of Forensic Medicine in Turkey.

The Council of Forensic Medicine is the official organ of the Ministry of Justice and the only official expert institution in Turkey. The duties and responsibilities of the Council of Forensic Medicine are considered by the law. Each year approximately 85000 reports are written about scientific and technical subjects related to forensic sciences asked by the courts and the district attorneys. The Council of Forensic Medicine includes specialized departments such as the Morgue and the Toxicology Department, specialization boards such as the First Specialization Board which deals with autopsy, toxicological analysis and medical and legal records.

Details of autopsies that have been performed in the cities of Turkey since 2001 have been recorded in a database in the first board which deals with the cause of death and medical malpractice. The first board includes a general surgeon, cardiovascular surgeon, neurosurgeon, gynecologist, internist, cardiologist, hematologist, immunologist, pediatrician, pathologist and forensic medicine specialists.

A total of 171 autopsy reports of infant cases in which infanticide or malpractice during birth was suspected were included in the study comprising year of death, gender, any witness statements, any medical records, report conclusion about live or stillbirth, body measures and weight, any traumatic changes, any congenital anomaly and any disease, umbilical cord examination, macroscopic and microscopic findings of the lung (sharpness of edge and shape, texture of lungs, their expansion in the chest cavity, degree of alveolar expansion, pulmonary interstitial emphysema and amnion aspiration).

In order to evaluate the relevance of reports, the conclusion about live or still birth, medical records, macroscopic and microscopic findings, and body measures at autopsy, microscopic investigations on the degree of alveolar expansion, were obtained from autopsy reports of infant deaths. Affected data that resulted to cases of live or still birth were compared with statistical analysis.

The findings of the study were evaluated by utilizing Statistical Package for Social Sciences 13.0 (SPSS 13.0) program. Descriptive analysis, chi square test between groups comparison of numerical data was used. P value of < 0.05 was accepted as statistically significant in all these comparisons.

**RESULTS**

The entire 171 infant autopsy reports on cases of live or stillbirth requested by prosecutors were concluded at the 1st Specialization Board of the Council of Forensic Medicine in Turkey between the years 1999 and 2006 (Figure 1).

99 of these cases (57.9%) were female and 70 (40.9%) were male. In two cases no information was given about gender because of advanced putrefaction.

Reports of infant autopsy cases were finalized as 47 stillbirths (27.5%) and 34 live births (19.9%). In 42 cases, conclusion about live or still birth could not be obtained due to putrification of body (24.5%) and insufficient autopsy information. Any comment was not provided in 48 (28.1%) cases.

According to the statement of witnesses in medical records, there were 56 (32.7%) stillbirth and 36 (21.1%) live birth cases. 79 (46.2%) cases revealed death at the death scene and/or did not have any eye-witnesses. There was a statistically significant correlation between witness statement and conclusion about live or still birth at reports of cases (p<0.000).

As maturation of infant has been compared in height, weight, body measures (foot length, crown-heel, crown-rump length, head circumference), weight of organs, development of ossification centers in the lower end of the femur, calcaneus and talus, presence of hair, lanugo, ear arcade, breast nodule, morphological appearance of genital organs, 71 (41.5%) cases were immature and 100 (58.5%) cases were mature. There was no statistical correlation between maturation of the infant and the conclusion about live or still birth at reports.

The weight of a termed infant has been estimated normal as equal or heavier than 2500 gr; small for the gestational age as lighter than 2500 gr; very small for gestational age as lighter than 1500 gr; and incompatible with life as lighter than 800 gr compared with newborn...
standard charts. It was established that 112 (65.5%) infants are normal, 26 (15.2%) small for gestational age, 21 (12.3%) very small for gestational age and 2 (1.2%) incompatible with life. There was no statistical correlation between birth weight of infants and the conclusion about live or still birth.

No obvious congenital abnormality and/or traumatic change was detected in autopsy reports. Umbilical cord of 71 (41.5%) cases was examined during autopsy and sampled for histopathological examination. No inflammation was detected in the samples.

The lung filled the thoracic cavity and covered the anterior surface of the heart in 45 (26.37%) cases (Figure 2), whereas in 10 (5.8%) cases the lung partially filled the thoracic cavity and partially covered the anterior surface of the heart. In 56 (32.7%) of the cases, the lung did not fill the thoracic cavity and presented at the back side of the thoracic cavity. In 60 (35.1%) of the cases, no information about macroscopical appearance of lungs in the thoracic cavity was given in the reports. There was a statistically significant correlation between medical record data about live or still birth and macroscopical appearance of infant lungs at the autopsy (p≤0.000) (Figure 3).

Hydrostatic test of lungs showed 25.7% sinking, 29.2% flotation, and 0.6% partially flotation. In 44.4% of the cases, no hydrostatic test was performed. Hydrostatic test of lungs significantly correlated with the conclusion about live or still birth in the reports (p≤0.000) (Figure 3). Live birth infants according to the medical record were found to have a high percentage of sinking in water. Correlation of cases with medical data was non-significant with the hydrostatic test (p: 0.008).

Unaerated lungs, because of intrauterine death, were shown before autopsy with radiological examination in one case (Figure 4).

The histological sections of 40 cases were obtained. These 40 preparates were re-evaluated microscopically by an experienced pathologist. 19 (47.5%) uniform, 10 (25%) semi-collapsed, 11 (27.5%) collapsed alveoli sections in shape were determined. When the existence and the diffuseness of amnion aspiration in alveoli was searched, 11 (28.2%) amnion aspiration, 9 (23.1%) mild amnion aspiration, 9 (23.1%) moderate amnion aspiration, 10 (25.6%) evident amnion aspiration was determined in 39 sections (Figure 5). 22 (57.9%) cases had pulmonary emphysema in lung sections; 16 cases did not express any pulmonary emphysema while we could not evaluate pulmonary emphysema in 2 cases because of technical causes.

There was no statistical correlation between medical record data about live or still birth and macroscopical findings (alveolar shape, amnion aspiration and pulmonary interstitial emphysema). Prosecutions were initiated in 34 live birth cases (19.9%) of alleged abandonment.

DISCUSSION

The number of suspected infant death cases reported by the 1st Specialization Board of the Council of Forensic Medicine decreased between the years 2005 and 2006, although there was an increase in the number of these cases from 1999 to 2004. Among factors which may have influenced the change are improved birth control, adaptation facilities and welfare payments (Ong and Green, 2003).

One of the most important medico-legal questions is whether an infant found abandoned had been born alive or not. The mothers in abandoned cases often claim that their infants were stillborn (Lavezzi et al., 2003; Marchetti et al., 2007; Knight and Saukko, 2004). In this study, registered medical data was found to be more valuable than any oral data. The mother's statement especially with the claim of stillbirth was found to be less valuable than other witnesses (medical staff, or eye-witness of scene not related with the case) statements with respect to the conclusion about live or still birth at reports (Knight and Saukko, 2004).
Figure 2. Macroscopical assessment of respired lungs during autopsy. The lung filled the thoracic cavity and covered the anterior surface of the heart in the shown case which also had an anamnesis of live birth by medical staff.

Figure 3. Significant correlation of live birth (in blue line) and stillbirth (in red line) conclusion of cases with autopsy findings (Floating lung: Floating lung at the hydrostatic test; Expansion macros: The lung covered the thoracic cavity and anterior surface of the heart; Witness with LB: Witness with live birth comment; Expansion micros: Uniform expansion in alveoli microscopically; Collapse micros: Collapse in alveoli microscopically; Witness with SB: Witness with still birth comment; Collapsed lung macros: The lung uncovered the thoracic cavity and anterior surface of the heart; Sinking lung: Sinking lung at the hydrostatic test).
Figure 4. Radiology of unaerated lungs because of intrauterine death.

Figure 5. Insufficient alveolar expansion in histopathological section of a case.
As the maturation of the infant has been compared in height, weight, body measures, weight of organs, the development of ossification centers in the lower end of the femur, calcaneus and talus, morphometric measurements were normal in relation with the gestational age and consistent with standard infant development charts. Since not every mature infant is assumed to be born alive, clinical information, macroscopic and microscopic lung findings were found to be more valuable information to estimate of newborn as having been live or stillbirth.

Live birth can be established with certainty if there is an inflammatory reaction at the site of umbilical cord division area (Ong and Green, 2003; Lavezzi et al., 2003; Knight and Saukko, 2004). Histopathological inspection should be made in relation with umbilical cord including umbilical skin and intra-abdominal vessels parallel to vertical axis (Jannsen, 1984; Busuttil and Keeling, 2009). But the absence of this finding does not indicate stillbirth. Histologically, inflammation around umbilical cord can be observed as early as 2 to 3 h after birth (Janssen, 1984; Busuttil and Keeling, 2009; Graham and Hanzlick, 1997). However, in most cases, infanticide usually occurs just after birth (Ong and Green, 2003; Knight and Saukko, 2004). In our study, the identification of the cord alone was not sufficient as inflammation in samples at a site of division of the cord would be more helpful. Umbilical inflammation in itself cannot be taken as the only certain evidence of stillborn or live born in the absence of other macroscopic and microscopic findings, and information regarding the duration of delivery and other circumstances. We did not have any record from where the umbilical cord had been sampled during autopsy. In the absence of the knowledge of birth circumstances, any inflammation of umbilical cord samples would be a more valuable finding than the lack of inflammation would be.

The death of the infant may be a result of natural causes which are incompatible with life (e.g. congenital abnormalities) (deRoux and Prendergast, 2006). Maceration is a definite proof of stillbirth (Knight and Saukko, 2004). Curiously, there was no evidence of intrauterine maceration, congenital anomaly or traumatic findings in the 171 cases. Nor was there injury on the skin around the nose and mouth and froth in the gastrointestinal tract indicating the possibility of smothering (Knight and Saukko, 2004; Tabata and Morita, 2000).

The presence of food in the stomach would have been a reliable indicator of live birth if we had found any in our cases. Some traces of air in the gastrointestinal tract or the middle ear were mentioned in reports with suspicion of decomposition occurring before the autopsy (Lavezzi et al., 2003; Marchetti et al., 2007; Knight and Saukko, 2004).

At birth, reflex inspiration will aerate the lung, and depending on the period of survival, the lungs will gradually become aerated (Weibel, 2008). Unaerated lungs, because of intrauterine death, were successfully shown with radiology before the autopsy in one case. However aerated lungs do not mean in every case that the infant was alive during birth. Just as was stated by numerous literature, under various conditions the aerated lungs may turn into unaerated ones and, on the other hand, a stillborn's lungs may seem to be aerated (e.g. mouth-to-mouth revival, external cardiac massage, the administration of oxygen). Unfortunately, there is no consensus as to the use of the hydrostatic test for diagnosis of live birth at textbooks. Gross examination of the lungs was considered to be more valuable than the hydrostatic test (Ong and Green, 2003; Knight and Saukko, 2004). Macroscopic evaluation of the lung can contribute to a differentiation between stillborn infants and infants born alive but dying shortly after birth in the absence of resuscitative efforts and putrefacation (Hausmann et al., 2004; deRoux and Prendergast, 2006).

Both the histological and macroscopic examination of lungs is needed to solve the problem (Lavezzi et al., 2004).

We classified lungs with their macroscopic findings as respired lung (pink or light red color, soft texture, round edges with filled chest cavity and covered front surface of the heart) and as unrespired lung (dark red color, solid texture, sharp edges without filling chest cavity and without covering front surface of the heart). In unrespired infants, lung resembles the liver and they do not have any crepitation with squeezing if artificial respiration has not been applied or any putrefaction has not occurred. Partial aeration may occur during intravaginal parturition and resuscitation. So we especially state that radiological examination of lungs and gastrointestinal system should be performed before the autopsy in the absence of putrification and artificial respiration (Stocker and Dehner, 2002).

In our study, we did not find any significant correlation related with live or stillbirth between the hydrostatic test and macroscopic findings of the lung in cases with clinical data. The hydrostatic test of live birth infants with premature, hyalen membrane disease, exposed to dense stress or 100% O₂ respired may turn to be false negative (Busuttil and Keeling, 2009). As has also been known, infection or edema in lung of live birth infants may result in false negative finding, hence the hydrostatic test is not a diagnostic test to differentiate live or stillbirth (Soysal and Cakalar, 1999).

In many perinatal infant autopsies, a frequent microscopic finding was advanced autolysis of many internal organs limiting a pathologic comment. In antepartum fetal death, histological abnormalities are most commonly found in the liver or lungs, for example, if showing evidence of fetal bacterial or viral infection (Breeze et al., 2008). In our cases, amnion aspiration was microscopically considered as similar to a lung of stillbirth which also expressed the collapse in alveoli. Also, microscopical alveolar aeration of lungs was related with live birth in medical records in deficiency of putrification and artificial respiration.
A frequent cause of infant death at birth is the aspiration of amniotic fluid. The assessment of a fluid infiltration in the alveoli can pose forensic-medical problems in newborns if there is no detailed knowledge of the circumstances of birth (Janssen, 1984). At least two samples should be taken from each lobe of the lungs in order to determine the diffusion of amniotic fluid.

The findings of uneven aeration and pulmonary interstitial emphysema (PIE) are supportive of live birth (Ong and Green, 2003; Lavezzi et al., 2003; Lavezzi et al., 2004; Marchetti et al., 2007; Tabata and Morita, 2000). In the vast majority of cases, PIE affects premature, often low birth weight infants, who have primary surfactant deficiency and who are receiving mechanical ventilation (Graham and Hanzlick, 1997; Breeze et al., 2008; Pursnani et al., 2006). In our study, cases with PIE were less in number to evaluate statistically. Studies including both PIE and birth medical data have only been case presentation until now.

Hyaline membranes in the lungs of infants who lived without resuscitative efforts is an important clue to predict live or still birth although our cases were not explored for hyaline membrane in the lungs (Janssen, 1984; Tabata and Morita, 2000; Sakaihara et al., 2001; de la Monte et al., 1986). Moreover, stereology of lung morphometry for these cases would have a great potential in the evaluation of respiratory physiology and pathology (Weibel et al., 2007; Dembinski et al., 2002).

Conclusions

At literature, studies about infant death regarding live or still birth usually include a few parameters or case presentations. In our study, a series of infant autopsy could be investigated. The information including medical records, correct sampling at umbilical cord was essential in evaluation of these cases. The degree of maturati on, the weight of infant and hydrostatic tests of lungs was useless to comment about stillbirth or live birth of an infant. Macroscopic examination of lungs was especially valuable in evaluation of these cases. Also, microscopically alveolar aeration of lungs was related with live birth in deficiency of putrification and artificial respiration. From the histological consideration of lungs alone, the manner of death could not be proven. In such cases, a critical and reserved macroscopic and microscopic assessment of lung samples in conjunction with a careful ascertainment of case history and medical records is advisable. When cases were evaluated to reach a conclusion of stillbirth or live birth, more than one finding had been in a scale of assessment.

Further investigations with prospective studies are needed to take measures in the comment of infant live or stillbirth. Definition and classification of infant autopsy findings as major and minor criteria will facilitate the standardization of infant autopsies as well as their results to estimate live or still birth.

LIMITATIONS

We could re-evaluate microscopically histological sections of only 40 in total of 171 cases because some autopsies were performed in different cities of Turkey and histological sections of these autopsies could not be obtained. Also, we had to exclude some prepares from the study as a result of autolysis which is common in putrified bodied that had been found left.

There is no clinical research paper investigating live or still birth of infants in Turkey. So we could not compare our findings with any Turkish literature data (www.ncbi.nlm.nih.gov/pubmed).