Examination of the Placenta

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Introduction

The placental examination is an important component of the postmortem investigation of fetal and perinatal deaths.[1-3] The pathologist can determine the cause of many of these deaths if the results of a placental examination are integrated with clinical, laboratory, and autopsy findings (Table 18-1 [located at end of chapter]). In one study, placental abnormalities were found in 85% of stillbirths, the majority of these lesions contributing to fetal death.[4] In many cases, the placenta contains the only etiology of fetal death, without which the cause of death will remain undetermined. Autopsy findings often can answer questions about the results of medical therapy and the nature of terminal events, whereas identifying the initiating disorder commonly requires the placental examination.[5,6] The placenta is also valuable in the forensic arena, often being the key in assigning the manner of death, and in medical malpractice litigation, revealing conditions outside the control of health care professionals.[5,7,8] The placental examination begins with a thorough gross inspection, adequate sampling for histology and ancillary studies, microscopic examination, and careful correlation with the clinical history.

The quality of hospital placental pathology reports is variable. Usually at the time of the initial placental examination, very little clinical information is available to the pathologist. We recommend that the placenta be grossly reexamined and in most cases new sections be submitted.[9]

Scope of Chapter

- Role of the placenta in autopsy pathology
- Transport, storage, equipment, and ancillary studies
- Gross and microscopic examination: placental membranes, placental disc, umbilical cord, maternal vessels, multiple gestations and twin-twin transfusion

Gross Examination

Transport

Controversy exists as to whether the placenta should be refrigerated (not frozen), unfixed, or fixed in 10% formalin. If refrigerated, the placenta can be stored at 4°C for 3 to 7 days without loss of histologic information. The placenta ideally should be placed in a large flat container so as not to distort the shape. The placenta should not be frozen because freezing results in lysis of the red blood cells and marked distortion of the histology. Keeping the placenta fresh (unfixed) allows for tissue procurement and the use of certain ancillary studies, such as bacterial and viral cultures, DNA and cytogenetic studies, metabolic studies, electron microscopy, and infusion studies. If the specimen is fixed in 10% formalin, at least 10 times as much formalin as the placental tissue volume should be used to completely surround and immerse the placenta.[10,11] Fixing the placenta permits easier handling, facilitates cutting of the tissue, and allows for better recognition of gross lesions.[12] Adequate formalin fixation before processing into paraffin is essential to good histology. The blood within the placenta alters the formalin pH. Better fixation is achieved by quickly rinsing the cassettes after grossing, changing formalin when it becomes blood tinged, and agitating the cassettes during fixation. Inadequately fixed tissue will result in significant artifact of the villi, which may be misinterpreted as infarct or accelerated maturation.

Equipment and Ancillary Studies

Tools and equipment for examination include a scale, ruler, long sharp knife, scalpel, fine-toothed forceps, a pin, and scissors. A large container is needed for the placental disc. When examining placentas in the fresh state, materials for ancillary studies should be readily available. These include microbiologic/bacterial (aerobic and anaerobic) culture media, transport media for cytogenetics and virology, glutaraldehyde fixative for electron microscopy, rapid-freezing capability for metabolic studies, and colored saline or radio-opaque dye and a syringe for twin placenta injection studies.

Tissue for bacterial and viral cultures should be taken in a sterile fashion (using sterile scalpel blades) from the subamniotic chorionic plate. The membranes also may be swabbed with a culturette for bacterial cultures. The dual fetal and maternal surface culture routinely performed at the time of delivery is of questionable value.[13] Use of predelivery antibiotics and fastidious organisms that need specialized culture techniques (mycoplasma, ureaplasma, anaerobes) may prevent successful recovery of organisms. It is not uncommon to have polymicrobial chorioamnionitis; therefore, a mixed culture should not be dismissed as contamination.

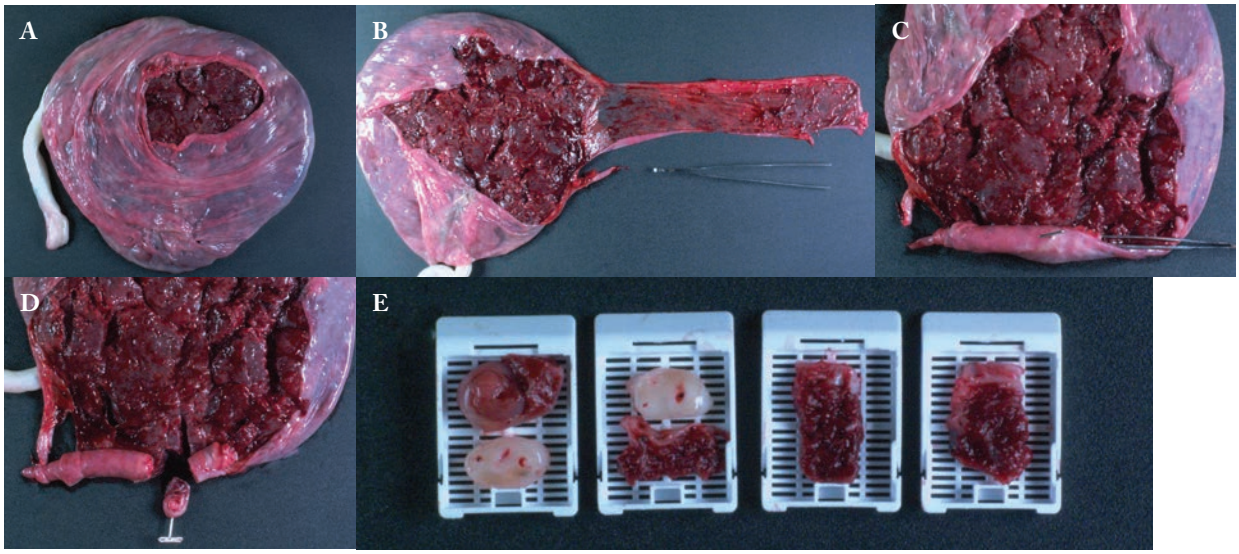


Figure 18-1. In a vaginal delivery, the zone of membrane rupture is closest to the cervix and has a higher yield for inflammation. A. Asymmetry of the zone of rupture is an indication of a placenta implantation other than in the fundus. B. A roll of the membranes done using fine-toothed forceps is usually effective, even in the fresh state. C. A pin is placed through the jaws of the forceps, which are then slowly removed. D. The section is taken with a sawing motion on either side of the pin. E. Recommended sections include two sections of umbilical cord from different areas; a membrane roll, including the zone of membrane rupture; and two full-thickness, nonmarginal sections of normal appearing placenta. Sections should include large chorionic plate vessels and maternal surface. Sections of gross abnormalities should be submitted in additional cassettes.

Submission of Sections for Microscopic Examination

The College of American Pathologists (CAP) has suggested that adequate sectioning includes two sections of umbilical cord, a membrane roll that includes the zone of membrane rupture, and two full-thickness sections of normal appearing, nonmarginal placenta.[11] Sections should include large chorionic plate vessels and maternal basal vessels. Additional sections should be submitted from areas of gross abnormalities (Figure 18-1).

Weight

The placenta may be weighed in the delivery room. The full-term placental weight has been estimated at 430 g to 650 g[10,12,14,15]; however, it must be noted that although some weigh the placenta with the membranes, cord, and any blood clots attached, others remove these components before weighing (Table 18-2). The latter is the preferred method and is what the normal-value tables are based on. Regardless of technique, the report should reflect how the weight was obtained. The umbilical cord weighs approximately 1 g/cm, and the membranes weigh approximately 50 g. Another variable is the amount of maternal and, even more so, fetal blood "trapped" within the placenta, accounting for as much as 370 g.[12] This additional weight can also vary. For example, the time of umbilical cord clamping will affect the amount of fetal blood within the placenta. If the placenta is not weighed after delivery, the pathologist is faced with other problems concerning weight. It has been shown that the stored, unfixed placenta loses 4% of its weight within 12 hours, 6% within 24 hours, and 10% within 48 hours.[12,15,16] This loss is especially marked in the edematous placenta. On the other hand, the formalin-fixed placenta gains weight—close to 10%.[15] One wonders: does the weight correlate with fetal/neonatal complications? Most studies report that the placental weight is a poor indicator or correlative of the condition of the child.[12] Possibly more valuable is the fetal:placental weight ratio. A fetal:placental weight ratio of greater than 10:1 is often associated with nonreassuring fetal heart tones. However, the small placenta has a tremendous reserve capacity, and the large placenta may be the result of incremental growth secondary to maternal or environmental factors.[17] Reserve capacity of a normal placenta is estimated to be approximately 30%, whereas an abnormally developed placenta may have as little as 10% reserve. In summary, the placental weight is most likely not a limiting factor in the status of the fetus/neonate but should still be included in the overall gross examination.[18-20] Most placentas associated with poor outcome have multiple abnormalities, which may be more than just additive.

Placental Membranes

The extraplacental membranes should be examined for their normal translucency and "glossiness." The mode of membrane attachment should be noted: marginal, circummarginate, or circumvallate (Figure 18-2). The following features should be noted: meconium (slimy, green) staining (Figure 18-3), opacity (suggesting infection/chorioamnionitis), dull nodules anywhere on the amnion (amnion nodosum, oligohydramnios; Figure 18-4), and/or brown discolorations (previous hemorrhage, amniocentesis; Figure 18-5). Thickened extraplacental membranes may be secondary to increased amount of

Table 18-2. Placenta and Umbilical Cord Weight and Measurement Based on Gestational Age					
Gestation Weeks	Mean Placental Weight (g)	Mean Placental Diameter (cm)	Cord Length (cm)	Cord Diameter (cm)	Fetus-to-Placenta Weight Ratio
8	13	NA	6	NA	NA
9	15	NA	NA	NA	NA
10	29	NA	10	0.4	NA
11	42	5.0	NA	NA	NA
12	56	5.6	13	0.5	NA
13	70	6.2	NA	NA	NA
14	83	6.9	16	0.4	NA
15	97	7.5	NA	NA	NA
16	110	8.1	19	0.8	1:1
17	124	8.7	NA	NA	NA
18	138	9.4	22	0.9	NA
19	150	10.0	NA	NA	NA
20	163	10.6	NA	1.5	2.7:1
21	176	11.2	NA	NA	NA
22	189	11.9	23	1.6	2.8:1
23	190	12.5	NA	NA	3.3:1
24	216	13.1	28-40	1.7	3.4:1
25	234	13.7	NA	NA	4.0:1
26	252	14.4	28-48	1.8	4.1:1
27	270	15.0	NA	NA	4.5:1
28	288	15.6	28-45	2.1	4.8:1
29	396	16.2	NA	NA	5.2:1
30	324	16.9	48	2.1	5.2:1
31	342	17.5	NA	NA	5.5:1
32	360	18.1	42-50	2.4	5.9:1
33	378	18.7	NA	NA	6.0:1
34	396	19.4	53	2.6	6.2:1
35	411	20.0	NA	NA	6.4:1
36	447	20.6	46-56	2.3	6.6:1
37	467	21.3	NA	NA	6.8:1
38	493	22.0	57	1.8	6.9:1
39	500	NA	NA	NA	7.1:1
40	510	NA	35-60	1.4	7.2:1
41	524	NA	NA	NA	7.2:1
42	532	NA	61	NA	7.1:1

Data extrapolated from Benirschke K and Kaufman P,[15] Manci EA et al,[50] and Heifetz.[51] NA = data not available.

decidua parietalis, which may show bland necrosis. This finding is thought by some to be a marker for decreased uteroplacental blood flow.[21] Subamniotic hematomas may be seen at the site of membrane rupture or near the umbilical cord insertion site. At the location of cord insertion, the membranes may have small patches of squamous metaplasia, a feature of a mature placenta (Figure 18-6).[15] Subchorionic fibrin plaques are normal at term and have been associated with an active fetus.

It is advocated that a section be taken from the site of membrane rupture and also from the placental margin.[12] In a vaginal delivery, the site of membrane rupture is the

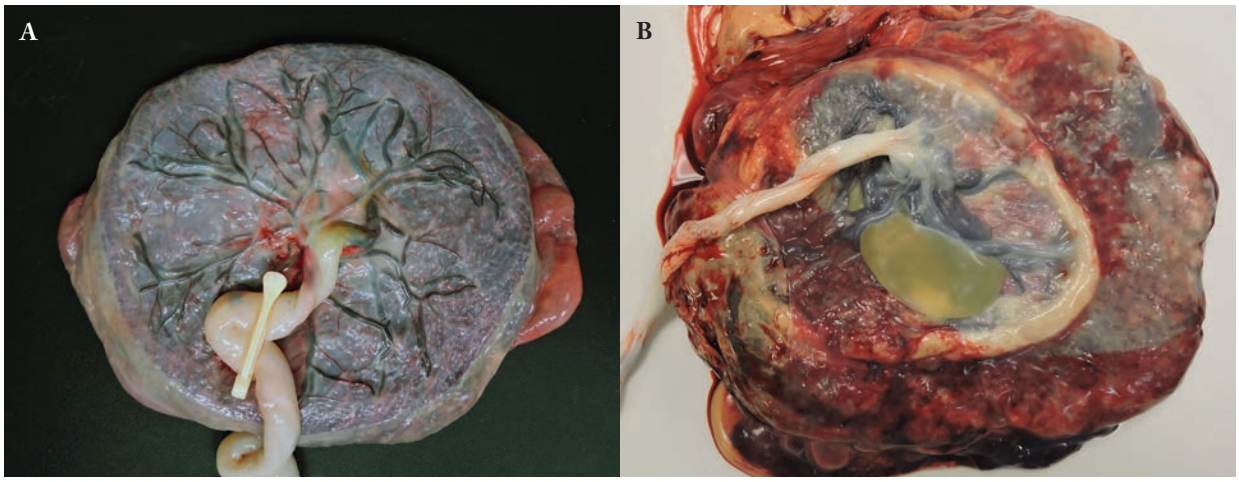


Figure 18-2. A. Normal membrane attachment to the disc may be at the margin, as shown in the inferior half of this placenta, or circummarginate, as shown in the superior half of this placenta. B. Circumvallate membrane attachment is considered pathologic only if it involves the entire membrane attachment site. The chorionic plate is folded back on itself forming a distinct ridge, often several centimeters away from the actual margin. This placenta also has a large chorionic cyst filled with yellow fluid.

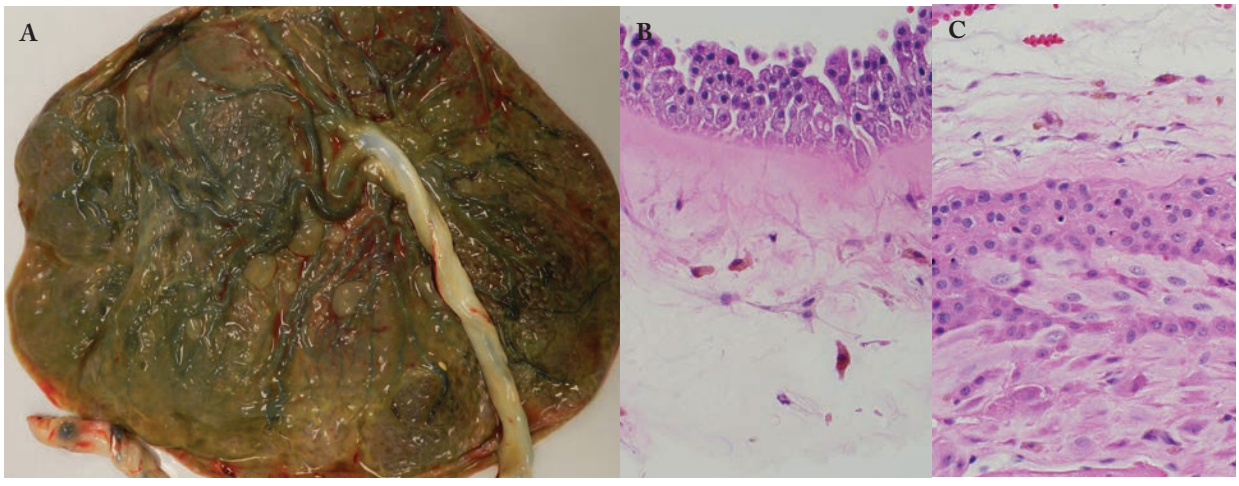


Figure 18-3. A. At term, meconium-stained membranes are a common finding. Thick, viscid meconium may be problematic from the standpoint of meconium aspiration syndrome or the underlying stressor that causes meconium to be passed. Initially, the meconium is dark green but with time will become browner to yellow. B. Meconium will cause vacuolization and hyperplasia of the amnion epithelium and be taken up by macrophages in the amnion in approximately 1 hour. C. Meconium will be taken into the chorion within 3 hours and into the decidua approximately 6 hours from the time of passage to delivery.

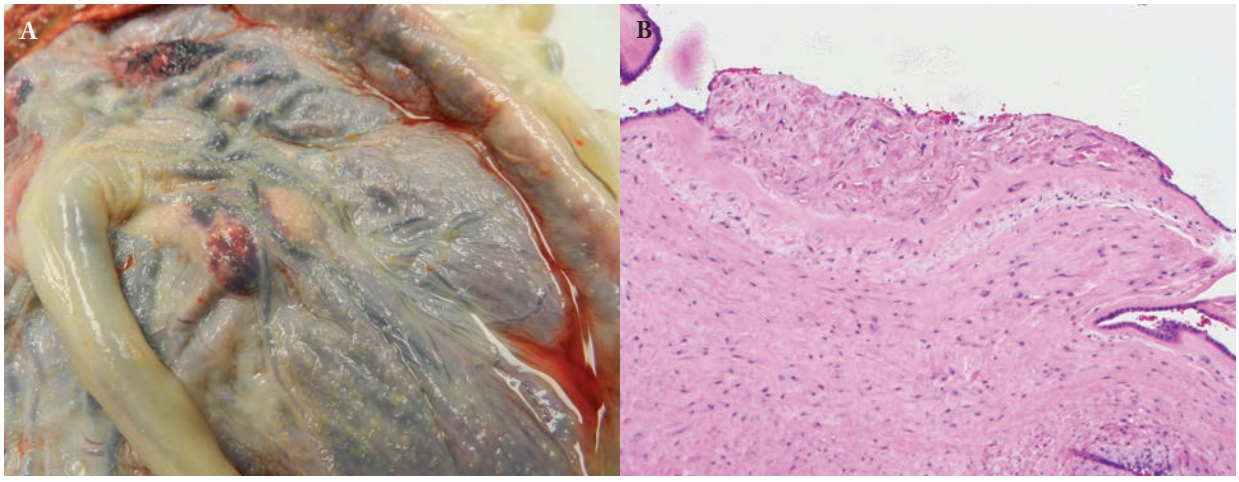


Figure 18-4. Amnion nodosum is evidence of severe and prolonged oligohydramnios. A. The small yellow-white plaques will be present on all amnion surfaces and can be scraped off. B. There are nodules of amniotic fluid debris attached to degenerated amnion.

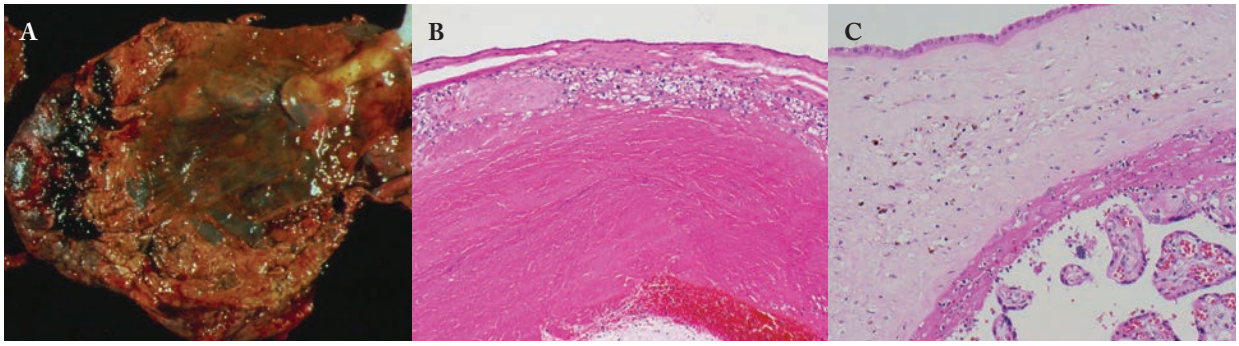


Figure 18-5. Hemosiderin staining of the membranes is most often secondary to chronic abruption or remote retromembranous hemorrhage, but may be evidence of fetal bleeding. A. The brown staining is often mistaken for remote meconium. There is tan granular debris at the periphery of the disc. B. The pigment is generally more granular than meconium and easily distinguished by an iron stain (C).

portion that was overlying the cervix and is the most likely site of inflammation. The site of rupture can also tell you something about the location of the placental implantation within the uterus. A low-lying placenta will have asymmetric membranes or site of rupture at the placental margin. The site of membrane rupture will have a slightly rolled margin, whereas tearing of the membranes that may occur during delivery of the placenta yields a sharp or ragged edge. The membrane roll taken for histology should have the site of rupture situated centrally, and the membranes should be rolled in a "jelly-roll" fashion using the fine-toothed forceps (Figure 18-1, A-D). The edge of the placenta can be used as a place to anchor a pin through the membranes and also serves as orientation. When the forceps are carefully removed, a sawing motion to cut on either side of the pin will prevent the center of the roll from being displaced. Other techniques, such as fixing briefly in formalin or Bouin's solution or use of special devices, may aid in obtaining membrane rolls.[22]

A single, full-thickness roll is submitted for histology. Two membrane rolls have been suggested as a way to increase the identification of acute chorioamnionitis and maternal vascular abnormalities.[23] It is important to make sure that both the amnion and chorion/decidua of the extraplacental membranes are represented in the sections—the amnion frequently becomes detached when there is meconium, edema, or chorioamnionitis. The decidua parietalis contains maternal vessels, which may show vasculopathy associated with poor fetal outcome.

Special stains may be used, such as iron stains for hemosiderin and bacterial stains. Of note, meconium macrophages are visible at 1 hour within the amnion and at 3 hours within the chorionic membrane.[15] Although meconium contains bile, none of the routinely used bile stains are reliable. Immunohistochemistry for zinc coproporphyrin (ZnCP-1; DAKO Japan, not available in the US) has been shown to be specific for meconium-laden macrophages.[24] It is very uncommon for meconium to be passed prior to 32 weeks' gestation. Iron-negative, pigmented macrophages may contain hemoglobin pigments.

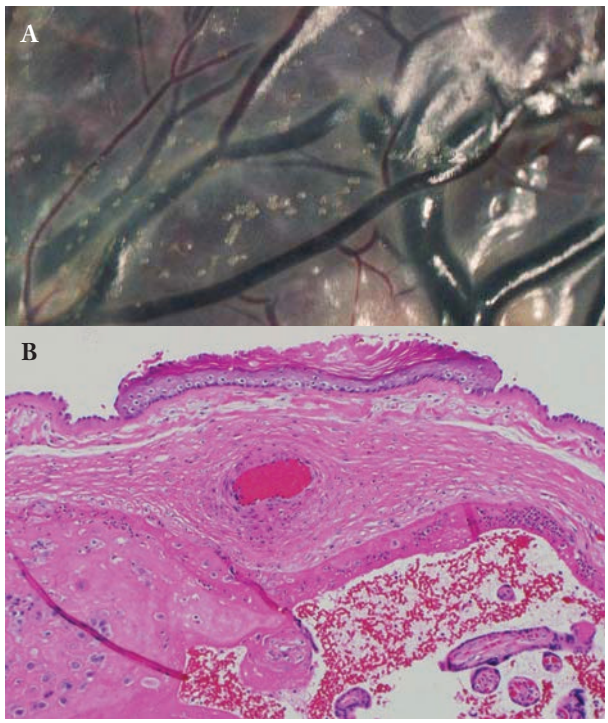


Figure 18-6. Squamous metaplasia is a normal finding at term. A. Small white deposits are found on the amnion of the chorionic plate near the cord insertion, often over the large chorionic plate vessels, and are not easily removed. B. Keratinization may occur.

Acute chorioamnionitis, a cause of 10% to 25% of fetal deaths, is due to an ascending bacterial infection.[25] Chorioamnionitis is subtle on gross examination, unless very severe (Figure 18-7). In most cases, inflammation can only be seen microscopically. The Perinatal Section of the Society for Pediatric Pathology has suggested terminology for both maternal and fetal inflammatory response.[26] Of note, intact membranes offer no true barrier to the spread of bacteria.[12] The bacteria can spread through the intact membranes to the fetus. The initial inflammatory response is maternal—maternal neutrophils within the decidua parietalis of the free membranes. Maternal neutrophils migrate into the intervillous space and

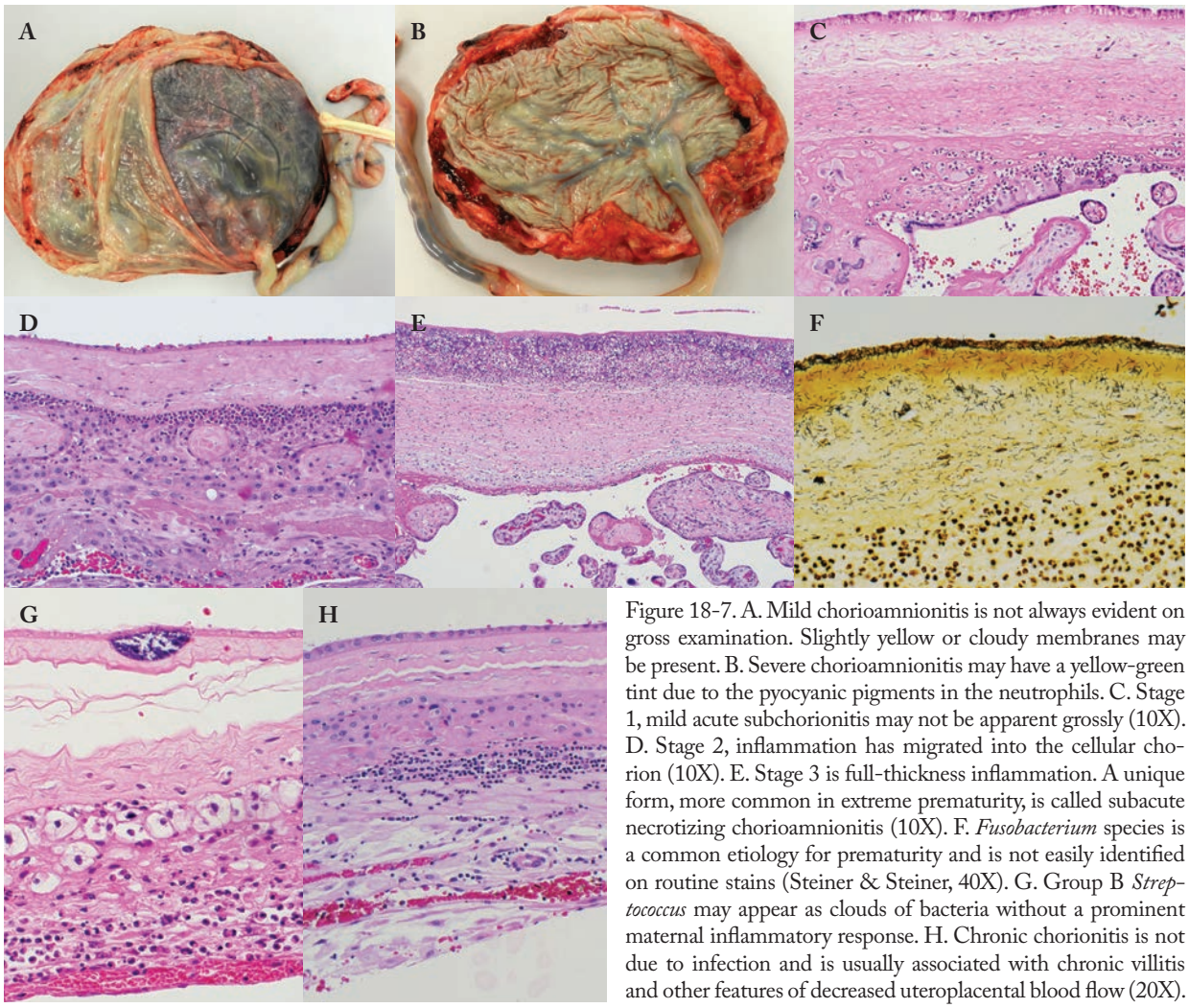


Figure 18-7. A. Mild chorioamnionitis is not always evident on gross examination. Slightly yellow or cloudy membranes may be present. B. Severe chorioamnionitis may have a yellow-green tint due to the pyocyanic pigments in the neutrophils. C. Stage 1, mild acute subchorionitis may not be apparent grossly (10X). D. Stage 2, inflammation has migrated into the cellular chorion (10X). E. Stage 3 is full-thickness inflammation. A unique form, more common in extreme prematurity, is called subacute necrotizing chorioamnionitis (10X). F. *Fusobacterium* species is a common etiology for prematurity and is not easily identified on routine stains (Steiner & Steiner, 40X). G. Group B *Streptococcus* may appear as clouds of bacteria without a prominent maternal inflammatory response. H. Chronic chorionitis is not due to infection and is usually associated with chronic villitis and other features of decreased uteroplacental blood flow (20X).

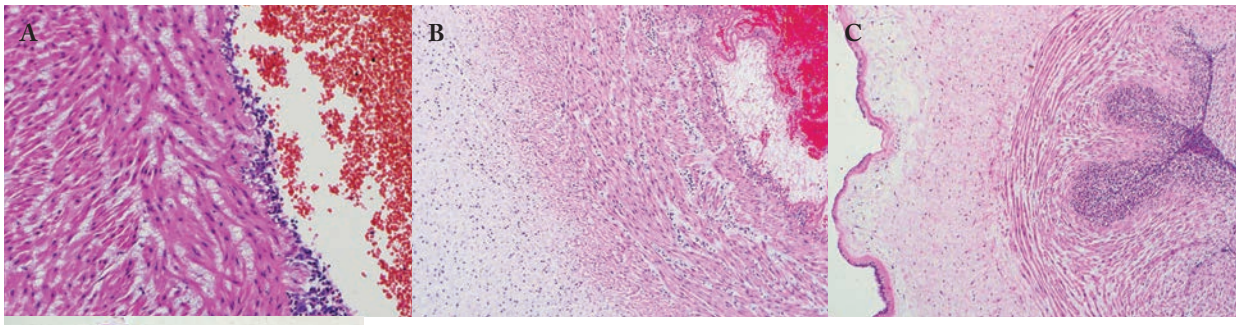
marginate in the subchorionic fibrinoid. At this phase, the neutrophils are purely maternal in origin.

For mild chorioamnionitis with few neutrophils, the maternal neutrophils migrate first beneath the chorionic plate (6-72 hours), into the chorionic plate (1-6 days), and to the amnion (>36 hours).[27,28] For cases of severe chorioamnionitis with many neutrophils, the migration can occur within as little as 24 hours[27]; however, there are a number of factors that can affect this: the virulence of the bacteria, the amount of bacteria, and the immunocompetence of both mother and baby, primarily related to gestational age. It should be emphasized that there is no clear correlation between the intensity of the histologic inflammation and the clinical severity of the fetal or maternal infection[12]; however, the more severe the fetal inflammatory reaction, the more likely the fetus is to be infected. Chorioamnionitis may result in premature rupture of membranes, premature labor, fetal defecation with meconium toxicity, villous edema with hypoxia and acidosis, abruption, umbilical vein thrombosis, congenital pneumonia, meningitis, sepsis, and fetal death.[12,27,29,30]

Generally, a fetal inflammatory response is delayed until after the maternal response is well established. The fetal neutrophils will migrate from fetal vessels in the chorionic plate and umbilical cord (Figure 18-8). Fetal inflammatory cells marginate beneath the vascular endothelium, then through the vessel wall, and finally out into the surrounding connective tissue. Inflammation begins first in the veins, then in the arteries. Fetal-derived inflammatory cells will be present only if the fetus was alive during the infection. After fetal demise, the vascular smooth muscle will autolyze, and care must be taken to distinguish between the nuclear changes of autolysis and those of inflammation (Figure 18-9). If the fetus is septic, there may be acute villitis and bacteria within the villous capillaries (Figure 18-10). There will be neutrophils within the fetal lung and stomach contents before sepsis. This is also evidence of viability during the infection.

Placental Disc

The placental disc should be weighed, minus the cord and membranes, and inspected for shape and consistency. Most placentas are round to oval. Approximately 10% of placentas will have an abnormal shape, such as having two nearly equal–sized lobes (bilobate or a smaller accessory lobe (succenturiate; Figure 18-11). The average dimensions of the placental disc are 22 cm in diameter



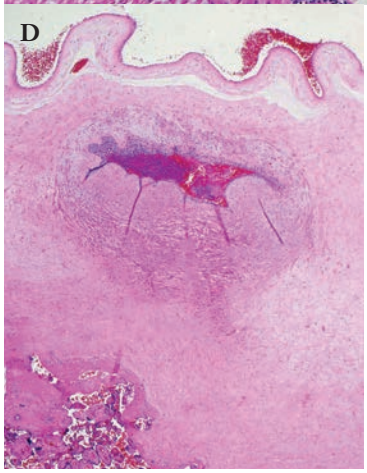


Figure 18-8. The fetal inflammatory response occurs in the umbilical cord and chorionic plate vessels. A. Inflammation begins in the vein with margination, then extends into the muscular wall and ultimately out into Wharton jelly (B), here with a developing thrombus. C. The arteries rarely are inflamed without venous inflammation. D. Inflammation of the chorionic plate vessels.

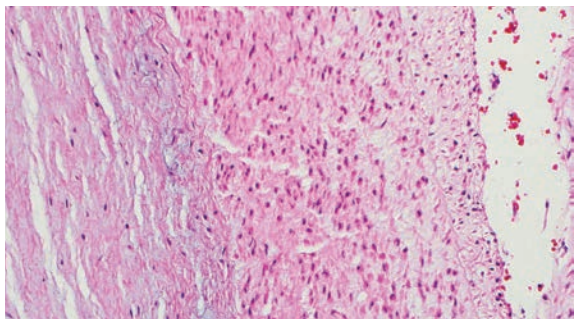


Figure 18-9. Maceration of the umbilical cord occurs from the fetal abdomen towards the placenta. The smooth muscle cells undergo degeneration and should not be mistaken for multilobated neutrophils.

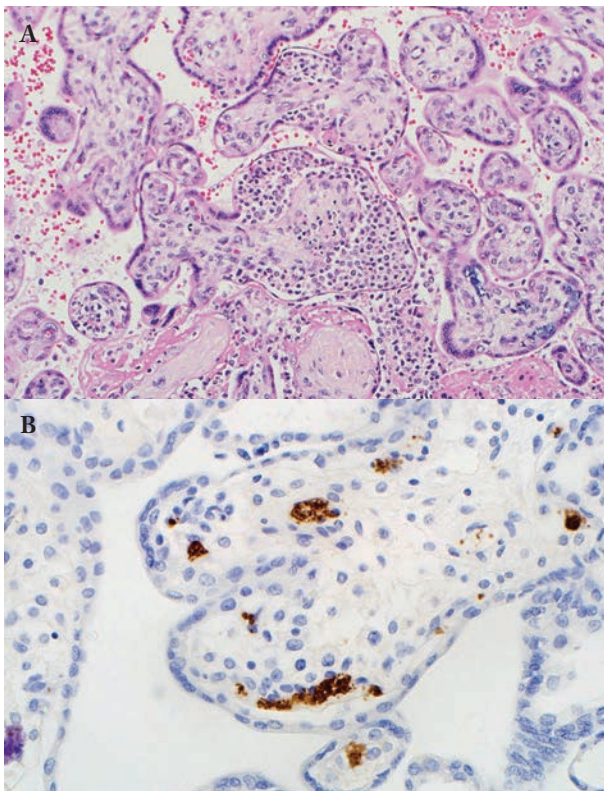


Figure 18-10. A. Fetal sepsis may occur via maternal sepsis and may show acute villitis, rarely with villous vessels containing bacteria. Maternal sepsis may be suspected by the presence of neutrophils and fibrin in the maternal intervillous space. B. Case of fetal sepsis with positive immunohistochemistry for Group B *Streptococcus* in fetal vessels.

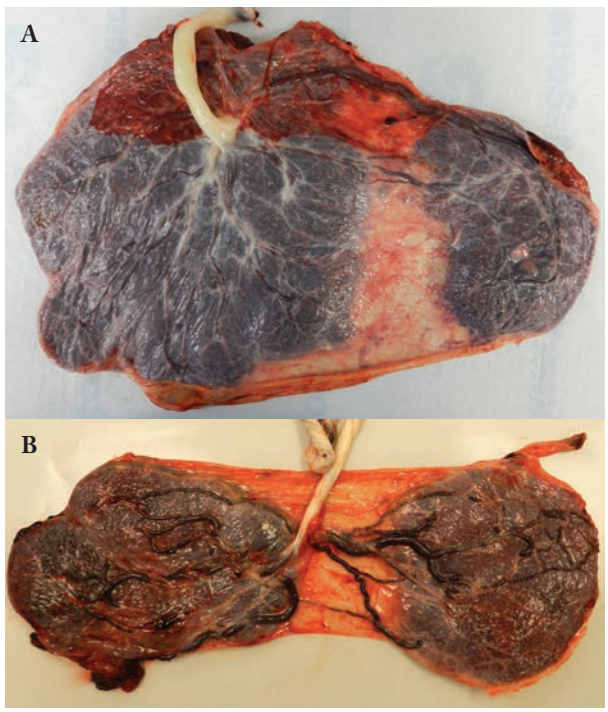


Figure 18-11. A. Accessory placental lobes may abut the main disc or be separated by several centimeters. The integrity of the vessels between the lobes should be documented. The subamniotic hemorrhage noted here may be an indication of a disrupted fetal vessel. B. If the lobes are relatively equal in size, the term bilobate is used. The umbilical cord inserts, unprotected between the two lobes, in the majority of bilobate placentas.