

CHAPTER

29

THERIOGENOLOGY

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Theriogenology in mammals includes obstetrics, genital diseases and reproductive physiology. Theriogenology in birds includes these topics as well as egg anatomy, physiology and incubation. With the rising interest in captive propagation for avicultural and conservation purposes, modern avian theriogenology also includes veterinary and avicultural techniques designed to maintain optimal production. Many factors, including complex reproductive behaviors, affect avian reproduction. Avian clinicians can serve the avicultural community by developing a thorough understanding of the avicultural techniques, anatomy, physiology, nutrition and behavior necessary to maintain long-term reproductive health for individual pairs and the flock.

Reproductive disorders occur with surprising frequency but can be difficult to diagnose because the cloaca serves as the endpoint of the gastrointestinal, urinary and reproductive systems in birds. In 24 avian orders, an 8.9% prevalence of reproductive disease was described in necropsy specimens.⁸⁴ The most commonly affected companion bird species was the budgerigar, although this observation was probably skewed by biased sampling in this particular study. Domestic poultry that have been genetically selected for productivity traits are probably more susceptible to reproductive disorders than companion bird species. Poultry hens have been reported to have a reproductive disease prevalence of 27.5% and 53.3%^{155,159} with senility, malnutrition and infectious agents being the most often incriminated causes of disease. The most common infectious agent affecting the reproductive tract of laying hens appears to be *E. coli*.¹⁵⁹ Reproductive disease in hens is frequently multifactorial, complicating diagnosis and therapy. Additionally, more than one type of reproductive disorder is often present; however, because of the common pathogenesis of many of these disorders, preventive and therapeutic considerations are generally similar irrespective of the etiology.

Female Reproductive Anatomy and Egg Formation

Ovary

The normal reproductive tract of a mature hen consists of a left ovary and oviduct. The right ovary and oviduct are present in embryonic stages, but these tissues normally regress before hatching in most species (Color 29.10). In some species and individuals (raptors), these organs may be vestigial or functional post-hatching. The left ovary is located at the cranial end of the kidney and is attached to the abdominal wall by the mesovarian ligament. In young birds the ovary is flattened, in an inverted “L”-shape. It has nearly inappreciable folds and resembles a piece of fat (see Color 13). As birds mature and the gyri become more prominent, small primary oocytes give the ovary a cobblestone appearance (Color 29.24). This process occurs by about 25 weeks of age in Blue and Gold Macaws (see Color 13).¹⁷¹ Ovarian tissue can be more or less melanistic, especially in cockatoos, macaws and some conures. Gonadotropin secretion in maturing hens causes a hierarchy of follicles to develop, giving the ovary the appearance of a cluster of variably sized grapes (Color 29.20). As the breeding season approaches, the follicles undergo a period of rapid growth with the deposition of yolk proteins and lipid produced by the liver. At this point the yellow yolk is clearly visible through the highly vascularized follicular wall. The large follicle is suspended by a stalk. The normal post-ovulatory follicle becomes a thin-walled sac devoid of blood clots.⁸⁶ The hypertrophied granulosa cells are metabolically active for several days and may not be reabsorbed until eight to ten days post-ovulation in the chicken, and up to several months in the Mallard Duck.¹⁰⁰ The post-ovulatory follicle is thought to secrete non-steroidal hormones that are involved in oviposition and nesting behavior.⁸⁶

During the non-breeding season, the ovarian follicles normally collapse and exhibit atresia. Two kinds of atresia have been described.⁸⁶ Bursting atresia occurs when the follicle wall ruptures and yolk is harmlessly released into the peritoneal cavity where it is absorbed. Invasion atresia involves granulosa and theca cells invading the ovum with subsequent *in situ* yolk absorption. The earliest detectable indica-

tion that a large follicle is undergoing normal atresia is the appearance of a vesicular lesion. This vesicle formation continues until the entire follicle is covered. As the largest follicle is absorbed, the smaller follicles will progress similarly.³⁸ Small follicles can be covered by connective tissue, sometimes leaving a scar-like area (see Color 13). Large follicles may undergo cystic degeneration (Color 29.22).^{46,176} If ovulation ceases suddenly due to trauma or stress, then developing follicles may be hemorrhagic and result in regression of the developing follicle. Aging hens can exhibit permanent ovarian involution, which is believed to be a normal physiologic process. Aflatoxicosis can also cause follicular atresia.⁵²

Oviduct

Understanding the anatomic divisions of the oviduct and their associated functions is important when discerning pathologic changes in the reproductive tract. During active egg laying, the oviduct enlarges and occupies much of the left abdomen. During the non-breeding season, the oviduct shrinks considerably in length and width. The oviduct of a preovulatory hen is usually thin, straight and uniform in diameter, and can be confused with the laterally tracking ureters (Color 29.19). An ovary and oviduct can regress to a point where it is difficult to determine if a hen has ever been reproductively active.

The oviduct consists of five microscopically distinguishable regions: infundibulum, magnum, isthmus, uterus (shell gland) and vagina (Figure 29.1). Dorsal and ventral ligaments attach the oviduct in the peritoneal cavity.⁷⁹ In psittacine birds, the dorsal ligament is clearly visible crossing the cranial division of the kidney (see Color 13).¹⁷² Peristaltic activity in the cranial oviduct and stronger smooth muscle contractions in the uterus and vagina move the ovum down and the sperm up the reproductive tract. Oviduct transit time varies among species and is approximately 24 hours in the chicken. Similar transit times are discussed in companion and aviary birds, with the egg spending varying but proportional times in each section of the oviduct.

The cranial infundibulum consists of a thin, nearly transparent finger-like funnel that engulfs the ovum into an ovarian pouch (see Anatomy Overlay). More distally, the infundibular wall thickens as it becomes tubular. In domestic fowl, the infundibulum is about seven centimeters long, while in the Brown Kiwi it extends the width of the peritoneal cavity to receive oocytes from functional left and right ovaries.⁸⁶ Fer-

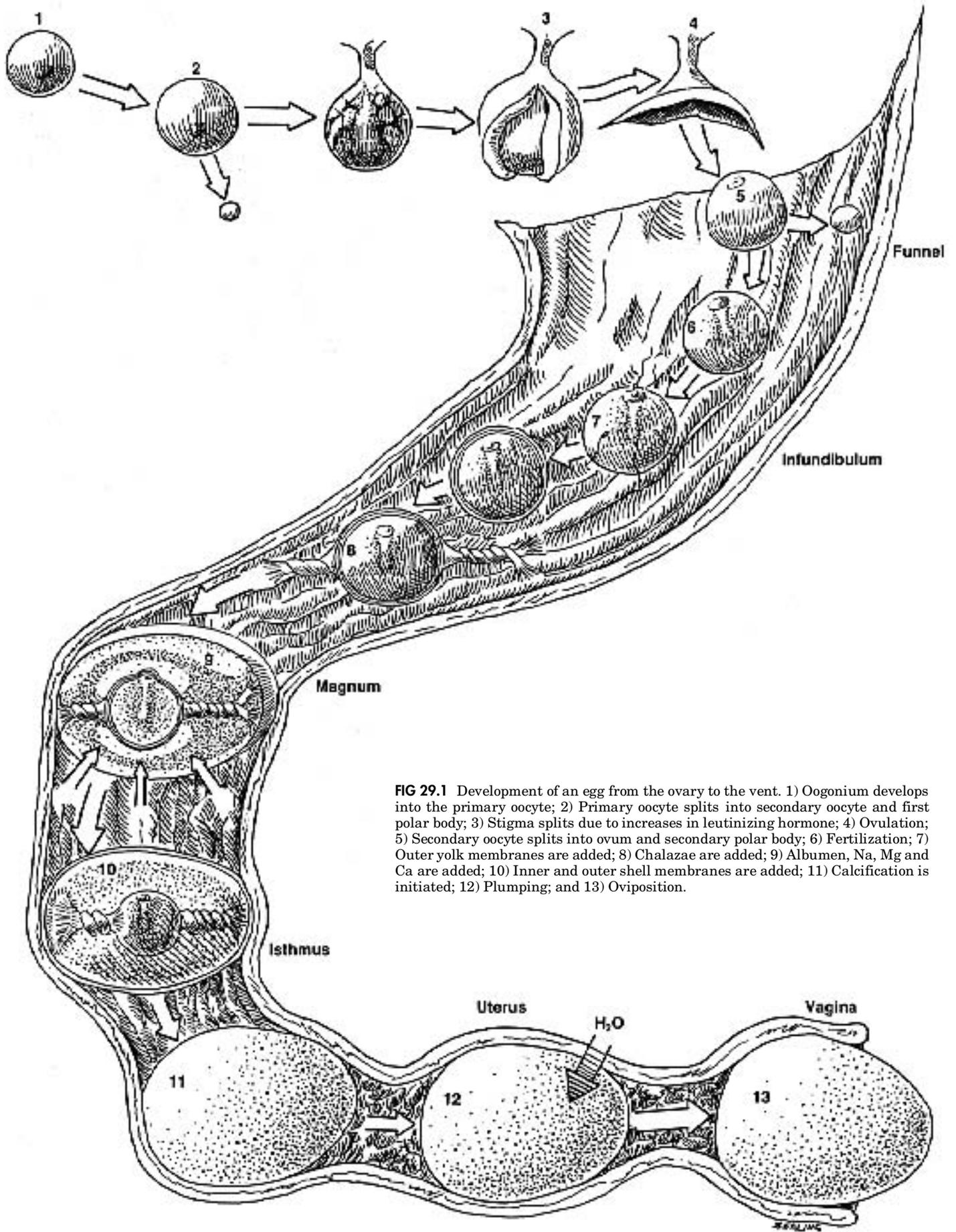


FIG 29.1 Development of an egg from the ovary to the vent. 1) Oogonium develops into the primary oocyte; 2) Primary oocyte splits into secondary oocyte and first polar body; 3) Stigma splits due to increases in leutinizing hormone; 4) Ovulation; 5) Secondary oocyte splits into ovum and secondary polar body; 6) Fertilization; 7) Outer yolk membranes are added; 8) Chalazae are added; 9) Albumen, Na, Mg and Ca are added; 10) Inner and outer shell membranes are added; 11) Calcification is initiated; 12) Plumping; and 13) Oviposition.

tilization occurs in the tubular portion of the infundibulum, where sperm may reside in glandular grooves awaiting the arrival of the ovum. Production of the chalaziferous layer of the albumen and the paired chalazae occurs also in the tubular portion of the infundibulum. Less than an hour later, the ovum exits the infundibulum and enters the highly glandular magnum that is differentiated from the infundibulum by its sudden enlargement in the mucosal folds. It is the largest and most coiled portion of the oviduct and deposits most of the albumen, sodium, magnesium and calcium used in egg development. The egg may remain in the magnum for three hours. Inner and outer shell membranes are added to the developing egg during its one to two hours in the isthmus. The isthmus has less well developed circular muscle and glandular tissue compared to the magnum.

The short uterus has numerous leaf-like lamellae composed of longitudinal folds, consisting of prominent longitudinal muscles and underlying tubular gland cells. This part of the uterus is ovoid in shape and holds the egg during shell deposition. The more cranial aspect of the uterus is difficult to differentiate from the isthmus. The egg remains in the uterus for 20 to 26 hours and receives salts, water, the shell and shell pigment. The uterus is highly vascularized during egg laying and must be carefully manipulated during any surgical procedure to prevent excessive hemorrhage (see Chapter 40 for considerations prior to performing uterine surgery). The egg is oriented in the uterus with its sharp end pointing caudally. In most species the egg is laid in this direction, although in some species the egg turns in the uterus just before oviposition.⁸⁶ The S-shaped vagina is the thickest walled portion of the oviduct; it begins at the uterovaginal sphincter muscle and terminates in the cloaca. The vagina is distinguishable by its numerous thin folds of mucosa. In juvenile chickens and some Anseriformes, the vagina is separated from the cloaca by a membrane that deteriorates at sexual maturity.⁸⁶ The vagina does not contribute to the formation of the egg, which passes through the vaginal lumen in seconds during normal oviposition. Along with the uterus, the vagina contributes to the muscular expulsion of the egg. Cranes that are disturbed during oviposition may retain an egg in the vagina for four days, followed by a normal egg delivery (Gee GF, unpublished).

Gallinaceous hens can store sperm in the spermatid fossulae (sperm host glands) at the uterovaginal junction and the glandular grooves and tubular

glands of the infundibulum. Other species may have only one set of these glands. Sperm located in these glands remain fertile for 7 to 14 days in the chicken and for 40 to 50 days in the turkey.⁷⁹ Anecdotal evidence suggests that sperm can remain viable for over a week in psittacine species.

Female Hormonal and Physiologic Factors

Primary oogenesis begins in the embryo when secondary oocytes are formed. Meiosis is arrested until adult life when follicles become active and grow in three phases, the first of which can last months to years.⁷⁵ This long initial stage represents the resting pre-nuptial period that occurs in all species. Stage two lasts about 60 days in the domestic hen, and during this period some yolk is deposited in vacuoles within the oocyte. This stage corresponds with ovarian regression that occurs in the non-breeding season in free-ranging species. Stage two growth in nondomestic species is influenced by luteinizing hormone (LH) and follicle stimulating hormone (FSH), which are both produced by the adenohypophysis under the control of the hypothalamus. Stage three involves rapid yolk deposition and normally occurs in free-ranging birds only when a mate is present. Courtship and nest-building activity seem to precede stage three follicular development, which terminates with either normal follicular atresia or ovulation.

Developing ovarian follicles consist of concentric layers of yolk, the oocyte, perivitelline lamina, granulosa cells, basal lamina and theca. Ovarian thecal and interstitial cells produce estrogen while the granulosa cells produce progesterone. Increasing concentrations of circulating estrogen stimulate an LH surge that is responsible for the continuation of meiosis two hours before ovulation. At this time, LH causes extrusion of the first polar body, the follicular wall ruptures and ovulation occurs. Extrusion of the second polar body occurs in the infundibulum, and the ovum is formed. The remaining granulosa cells of the ruptured follicle are under the control of LH and prolactin. These cells continue to produce progesterone, which inhibits further ovulation and induces behavioral and physiologic changes associated with incubation and brood care. During this time, the ovary and accessory reproductive tissues, such as the oviduct and comb, regress. Broody behavior is accompanied by the development of a brood patch in some species. Prolactin secreted by the anterior pituitary stimulates the production of "crop milk" in both genders of Columbiformes. In some avian species, pro-

lactin levels are usually higher in the parent providing the majority of the care.

In photoperiodic species, day length changes may terminate reproduction; however, in most species a photorefractory state develops that is controlled at the level of the hypothalamus.⁴² Long day length as a stimulus is blocked, and serum gonadotropin and gonadal steroid hormones decrease to minimum levels. Photorefractoriness is then terminated by shorter daylight periods. This change often corresponds to a surge in reproductive behavior in the fall in some species. Circadian rhythms are also involved in the photoperiodic control of synthesis and release of FSH and LH.

Domestic hens are continuous layers (indeterminate layers) and under optimum conditions are reproductively active year round. Hormonal mechanisms controlling continuous egg production have been artificially induced through selective breeding. In contrast, free-ranging birds lay one or more eggs in a clutch and then terminate egg production to begin incubation. The onset of reproductive activity is influenced by increasing photoperiod in temperate climates.⁷⁹ Photoperiod is less important in equatorial birds where day length is similar all year. Photoperiodic pathways are controlled by light passing through the eye via the optic nerve to the hypothalamus, and by light passing through the spongy calvarium stimulating the hypothalamus or pineal body. Photoperiod also affects the time of ovulation and oviposition. In poultry, the maximum effect of photostimulation occurs when birds are provided 12 to 14 hours of light; however, normal egg production can occur when hens receive 12 to 18 hours of light. Because reproductive activity continues even when hens are placed in continuous darkness, other factors also control egg laying.⁷⁹

Hypothalamic control of reproduction is influenced by environmental factors other than light, especially in periodic breeders of equatorial climates. In arid-dwelling species, such as the budgerigar and Zebra Finch, the rostral pituitary is constantly stimulated by the hypothalamus to release gonadotropins except when inhibited by negative external conditions such as drought. During these dry conditions when food would be scarce, the hypothalamic secretions suppress reproductive activity.

Ovulation in the domestic hen occurs shortly after oviposition allowing a 24 hour lay interval. In psittacine birds, the laying interval is generally two

days.⁴ In most Passeriformes, lay intervals are 24 hours, but they can extend up to four to five days in the Andean Condor and up to 44 days in the Brown Kiwi.⁸⁶ It is not clear whether transit time in the oviduct, a delay in ovulation or both are responsible for longer lay intervals.

Several hormones secreted by the follicle affect the oviduct. Progesterone in large doses may inhibit ovulation or, if given 36 hours before expected ovulation, will induce follicular atresia.^{75,79} If given 2 to 24 hours pre-ovulation, progesterone can induce a preovulatory surge of LH and premature ovulation. The dose and critical period for exogenous progesterone administration appear to vary among species and experimental designs. The extrapolation of any data collected in gallinaceous birds should be applied to companion bird species with caution. If progesterone is used to prevent egg laying, it should be administered when a complete clutch has been laid. Premature administration can cause an abnormal ovulatory process that may lead to soft-shelled eggs, mummification, peritonitis and death (Harrison GJ, unpublished). In general, the seasonal hypertrophy of the oviduct in free-ranging birds is dependent on estrogen secreted from the ovary. Progesterone and prolactin also interact with estrogen in stimulating the growth and secretory activity of the oviduct. In the oviduct, estrogens influence the synthesis of oviductal proteins, oviduct growth and the formation of tubular glands. Androgens in estrogen-primed birds influence the synthesis of proteins in the oviduct and in conjunction with estrogen initiate medullary ossification.

Ovum transport in the oviduct is primarily accomplished by contractions of the oviduct in response to a stretch stimulus. Prostaglandins, which contract the smooth muscle of the oviduct and vagina, may also influence egg transport and expulsion.⁷⁹ Arginine vasotocin released by the posterior pituitary stimulates uterus contractility *in vitro*, but it is not clear what role it plays in oviposition. The effect of oxytocin, also produced by the posterior pituitary, in inducing premature oviposition may be mediated *in vivo* by prostaglandins. It is likely that oviposition is a complicated process involving neurohypophyseal hormones, prostaglandins and hormones of the pre- and postovulatory follicles. Oviposition may last from seconds to hours depending on the species. Before expulsion of the egg can occur, the abdominal muscles and cervix must relax.

The thecal cells of the pre-ovulatory follicle and the ovarian stromal interstitial cells (homologous with the interstitial Leydig cells of the testis) produce androgens.⁷⁵ Sexual behavior and secondary sexual characteristics are influenced by androgens in the female and may become apparent depending on the stage of ovarian regression or growth. Examples of androgen-controlled characteristics include comb growth, bill growth and aggressive male-type behavior (territoriality).

LH levels increase in female cockatiels when they start nest-searching behavior and reach their highest levels during egg laying.¹¹⁶ LH levels decline during incubation, hatching and chick-raising, but increase again in both genders if a second clutch of eggs is laid. In male cockatiels, LH levels are highest during nest inspection and are lowest during egg laying. Prolactin levels increase in both genders during egg laying, peak during incubation and then decline to a resting level.

Calcium Metabolism

High levels of circulating calcium are needed for shell formation. Estrogen increases total plasma calcium by increasing the production of blood calcium-binding proteins.⁵ During the laying process in psittacine birds, the calcium levels can become extremely high, reaching levels of 30 mg/dl.⁸² However, increased blood calcium levels alone are insufficient for shell formation because hens can deplete the entire blood calcium in 8 to 18 minutes during the time the egg is in the uterus. Increased intestinal absorption and bone mobilization of calcium are needed to constantly replenish blood calcium. Laying hens will preferentially consume calcium-rich diets. This has also been observed in reproductively active psittacine hens. For psittacine birds, it is recommended that laying hens be offered at least 0.3% calcium (1:1 or 2:1 ratio with phosphorous) in their diet to prevent bone mobilization, but no more than 1% to ensure that egg shells are not excessively thick.¹⁴² For continuous layers, higher levels of dietary calcium (up to 3.34% in turkey hens) are necessary for maximum egg production and hatchability.¹⁰⁹ In domestic fowl, most of the egg shell calcium is obtained from the intestine, and bone calcium is used only when blood calcium levels are low.

Bone calcium does serve as a source of calcium for shell development in hens that lay eggs during morning hours when food intake and subsequent intestinal absorption of calcium is decreased.⁷⁹ Calcification of the medullary spaces of the long bones, particu-

larly the femur and tibia, occurs in female birds approximately ten days before egg formation (see Figure 15.16). In budgerigars, the primary sites of medullary calcification are the humerus and femur.¹⁴⁹ If insufficient calcium is consumed, cortical bone will be mobilized. At some point in the mobilization process, calcium deficiency causes a reduction in FSH secretion that stops the laying process. High-fat, low-calcium diets exacerbate a calcium deficiency by decreasing calcium absorption from the intestines. Increased activity and concentrations of renal and circulating Vitamin D₃ occur prior to and during egg laying. In budgerigars, it is theorized that polyostotic hyperostosis in non-laying females results from aberrant estrogen metabolism. Affected budgerigar hens have been shown to have normal ovaries and no evidence of hormone-secreting tumors or other endocrine diseases. The liver is responsible for inactivating estrogens, and it has been suggested that impaired liver function may be responsible for this disease.

Other Metabolic Changes

Hematogenic changes associated with egg laying include a slight increase in white blood cell count, packed cell volume, total serum solids and total protein. Total serum solids and total protein are increased because of a need for protein for calcium transport as well as from the estrogen-controlled liver synthesis of lipid and proteins produced during yolk formation (Table 29.1). Alkaline phosphatase levels may also increase due to estrogen stimulation. This has been shown to occur in budgerigars with estrogen implants.¹⁴⁹ Alkaline phosphatase levels were also shown to be elevated in egg-laying cockatiels and pigeons.⁵⁷

TABLE 29.1 Typical Hemogram of Normal Laying Cockatoo Hen

	Values	Change from Non-laying Status
WBC	15-18 x 10 ³ /mm ³	Increased
PCV	48-55%	High range of normal
Total protein	4 - 6.5 gm/dl	Increased
Calcium	15-20 mg/dl	Increased
AP	>500	Increased

Egg Structure and Physiology

The avian egg is made of concentric layers of tissue that originate from different portions of the oviduct (Figure 29.2). Each component of the egg is responsi-

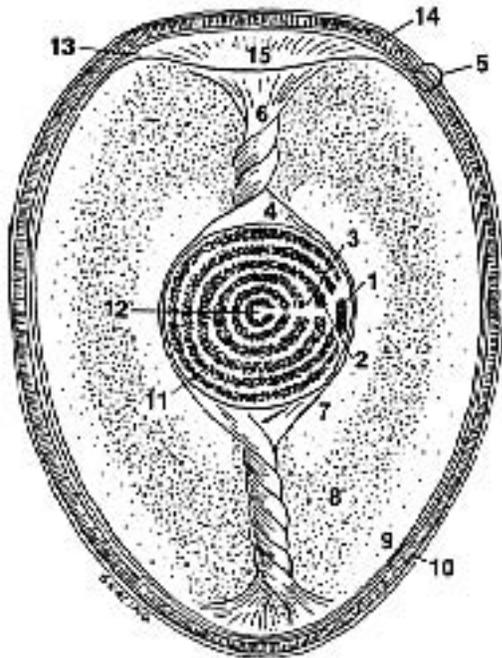


FIG 29.2 Components of an egg: 1) blastoderm before fertilization, 2) disc of latebra 3) neck of latebra 4) chalaziferous layer 5) shell (testa) 6) chalazae 7) internal layer of thin albumen 8) thick outer albumen 9) external layer of thin albumen 10) cuticle 11) vitellus aureus 12) center of latebra 13) internal shell membrane 14) outer shell membrane 15) air space (modified from Stoodley).¹⁶⁵

ble for various physiologic functions needed to support the growing embryo (Figure 29.3) (Table 29.2).^{79,86} Understanding normal egg anatomy allows the clinician to recognize abnormalities and instigate appropriate therapeutic or preventive measures to resolve embryonic death problems and female reproductive disorders.

The germinal disc is a small, circular, opaque white spot on the surface of the yolk that contains cytoplasm and the oocyte (Color 29.1). The yolk is classified as either “white” or “yellow” and is layered in strata that are visible when stained with potassium dichromate.⁴⁹ The yolk is 50% solids, 99% of which are proteins.⁸⁶ Maternal antibodies (IgG) are present in the yolk. These antibodies are absorbed by the chick and provide waning passive immunity until it becomes immunocompetent. The concentration and longevity of specific immunoglobulins in psittacine egg yolk are not known. It has been demonstrated that vaccinated hens pass anti-PBFD virus antibod-

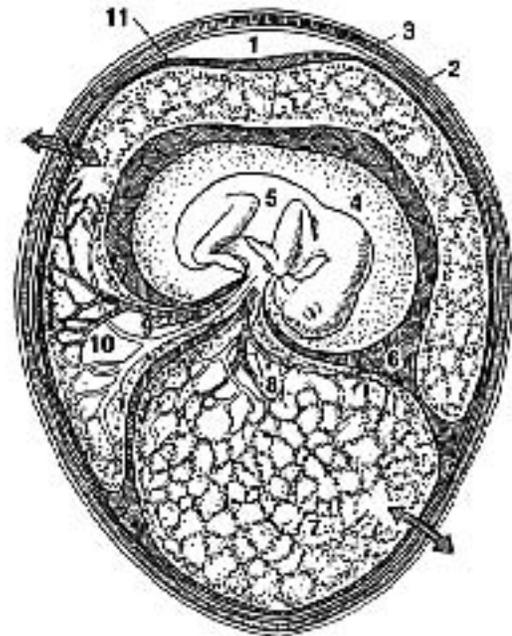


FIG 29.3 Compartments and components of developing egg: 1) air space 2) shell 3) outer shell membrane 4) amniotic sac 5) embryo 6) albumen sac 7) yolk sac 8) omphalomesenteric artery and vein 9) allantoic artery and vein 10) chorioallantois formed from the fusion of the chorion and the allantois 11) inner shell membrane (modified from Stoodley).¹⁶⁵

ies to their chicks and that these antibodies wane to undetectable levels between 30 and 45 days of age.¹³⁴

The yolk is surrounded by layers of membranes, collectively called the vitelline membrane. The normal yolk is various shades of yellow (depending on the diet and species of hen), firm, intact and separate from the albumen. The albumen is made of the chalaziferous layer, chalazae, and inner, middle and outer layers. The outer clear and inner layers are thinner than the middle layer, which can be macroscopically distinguished (see Figure 29.2). Although less viscous than the yolk, the middle layer of albumen is quite viscous, which makes it appear as a whitish-clear gel. It remains adhered to the yolk through the chalazae and albumen ligaments. At the blunt end of the egg the two shell membranes separate from each other, forming the air cell. The outer layer is adhered to the testa layer of the shell and the inner layer is attached to the dense portion of the albumen. The outermost surface of the egg is covered by a thin, sometimes waxy, cuticle. Microscopic pores in the egg shell allow for passive diffusion of oxygen,

TABLE 29.2 Embryonic Structures and Their Physiologic Function

Structure	Function
Shell	Physical protection, protects embryo from microorganisms, transpiration, regulate evaporation, source of calcium carbonate for bone formation.
Shell membrane (outer and inner)	Transpiration, protects embryo from microorganisms.
Air cell (between two shell membranes)	Transpiration.
Albumen	Nutrition, protects embryo from microorganisms
Chalazae (thick strands of albumen)	Stabilization and centralization of the yolk.
Yolk	Nutrition, maternal antibodies.
Vitelline membrane (yolk sac membrane)	Protects embryo from microorganisms.
Germinal disc	Infertile - blastodisc; Fertile - blastoderm.
Positive development	No embryo, only extra-embryonic structures.
Blastoderm without embryo	No embryo, only extra-embryonic structures, including blood islets and some vessels.
Chorioallantoic membrane	Transpiration, metabolism, waste collection, calcium transfer (shell to embryo).
Amnion	Structure around embryo, provides protective fluid environment, muscles gently rock embryo for five to twelve days of incubation, protein source (albumen drains into amnion, which embryo drinks late in incubation).
Cuticle	Regulates evaporation, protects embryo from microorganisms.

carbon dioxide and water during embryo development (see Color 48).

The shell thickness, size, shape and pigmentation vary depending on the species of bird, and a certain amount of individual variation occurs intraspecies (Figure 29.4). Hens that produce precocial young generally have larger, thicker-shelled eggs. Larger eggs usually have thicker shells. This can be clearly noted with conure eggs that have thinner shells than their somewhat close relatives, the macaws. Eggs may be conical, spherical, oval or cylindrical in shape. Cockatoos have more spherical-shaped eggs than macaws. In most species, the egg has a blunt end, which contains the air cell and is the point of exit for the emerging chick (Color 29.18). Other eggs, like those of ratites, are almost spherical. Psittacine eggs are generally smooth, but depending on the species, may be glossy (such as in macaws), greasy, chalky, powdery, ridged or pitted.

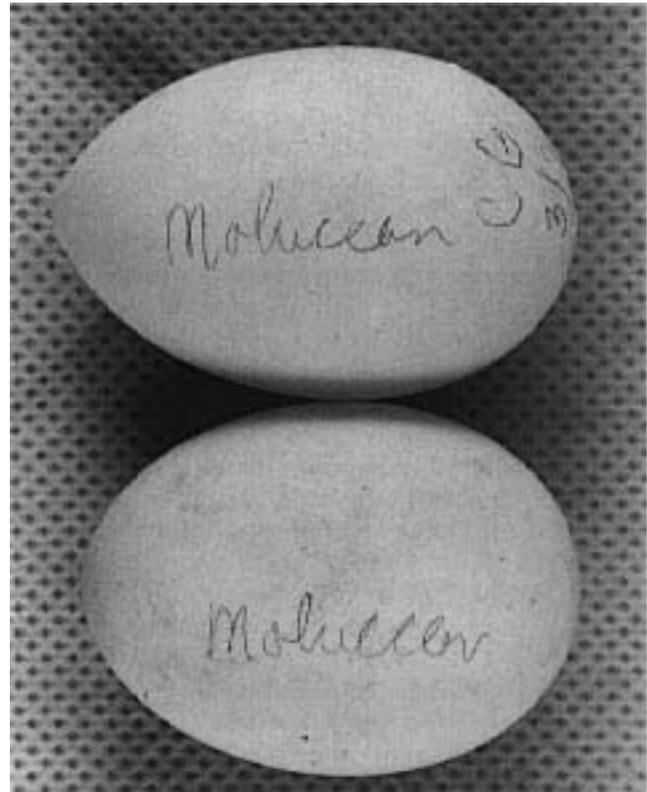


FIG 29.4 Egg morphology and size vary dramatically among species and individuals in the same species. Normal eggs from two different Moluccan Cockatoo hens illustrate the difference in egg shape. The egg is normally passed with the pointed end caudally. A soft pencil is nontoxic and can be used to mark eggs for record-keeping purposes.

Most avian embryology has been investigated in domestic species, but embryogenesis is thought to be similar for all species (Color 29.1 to 29.13).^{12,55}

Male Reproductive Anatomy

The paired testes are located within the body cavity ventral to and near the cranial border of the kidney and the abdominal air sac (see Anatomy Overlay). The testis is attached to the body wall by the mesorchium and is encapsulated by two fibrous coats. Occasionally, one testis may be larger but both should be functional in mature birds.⁸⁶ The author has noted testes of unequal size on numerous occasions in young and mature psittacines, the importance of which is unknown. The bilateral testes are not al-

ways symmetrically located within the body cavity. One can be located more caudally than the other. Dimensions, color and shape can vary, not only by age but also among species. During the resting stage, most testes are small, yellow-white and bean-shaped. In young birds, the testes can appear flattened and pointed when compared to the rounded shape of the mature testicle. Melanistic testes, like melanistic ovaries, can occur in some species of Psittaciformes (Golden Conure, Blue and Gold Macaw, some cockatoos), Passeriformes and Piciformes (Keel-billed Toucans). Under hormonal control the testes can increase in size by 300- to 500-fold (Figure 29.5).¹⁶⁸ The increase in size appears to be proportionally greater in finches and Columbiformes than in Psittaciformes (Harrison GJ, unpublished). Vascular supply increases during reproduction, resulting in a more prominent pattern of blood vessels on the testicular serosal surface. During the breeding season, yellowish testes may turn white, while melanistic testes may change from black-grey to grey-white.⁸⁶ The testes normally atrophy during periods of sexual inactivity; however, the testes never become as small as they were in the prenuptial stage.

Convulated seminiferous tubules comprised of germ (spermatogonia) and Sertoli cells make up the bulk of the testes and are responsible for spermatogenesis. Leydig cells, also called interstitial cells, produce male androgens and occupy the interstitial spaces between the tubules. Melanistic cells responsible for the color of the testicles are found in the same location. Mature spermatozoa exit via straight tubules into the rete testis, which connects the testis to the cranial aspect of the epididymis. The rete testis is not present in all birds. The epididymis, considered vestigial in birds, lies along the dorsomedial aspect of the testes and is concealed from view during laparoscopic examination, even during the breeding season when it enlarges considerably. In some species of birds, the epididymis is connected throughout its length by tubules to the rete testis.⁸⁶ Spermatozoa exit the epididymis and enter the ductus deferens, which forms a zigzag tubule running parallel with the ureter just medial to the kidneys. The ductus deferens is under hormonal control and is more convoluted during the breeding season. In the non-breeding season, it blends indistinguishably with the ureter and kidneys. The ductus deferens penetrates the dorsal wall of the urodeum, which functions as a receptacle for sperm. The last two to three millimeters of the ductus deferens project into the urodeum forming a papilla. In passerine birds and budgerigars, the caudal end of the ductus deferens forms the

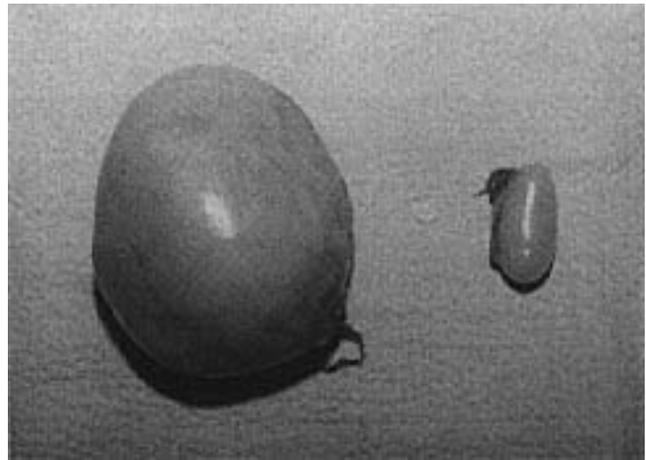


FIG 29.5 The testicle of a normal bird can increase substantially in size during the breeding season. In this Eclectus Parrot, the right testicle was of normal size and enlargement of the left testicle was caused by a seminoma (courtesy of Kim Joyner).

seminal glomus, which enlarges during the breeding season to form a prominent projection in the cloacal wall for the storage of sperm.⁸⁵ This prominence allows passerines to be easily sexed during the breeding season. Birds that do not have this structure have little sperm storage capacity. Proctodeal glands develop to varying degrees in birds and undergo hypertrophy in response to increases in steroid sex hormones.¹¹⁷

The ejaculatory papillae (terminal projectory papillae of the ductus deferens), paracloacal vascular bodies, cloacal folds and the phallus are involved with male copulation and are variably developed in avian species. The cloacal lymph folds and paracloacal vascular bodies contribute to the lymphatic erection of either cloacal or phallic tissue, and release a lymph-like transparent transudate when engorged.⁸⁰ Although not all avian species have been adequately studied, it is known that ratites, tinamous, Anseriformes, some members of the family Cracidae and one Passeriforme, the Black Buffalo Weaver, have phalli that are intromittent (inserted into the female).^{11,86} Other species have phalli that may become engorged during copulation, but semen transfer occurs by direct cloaca-to-cloaca contact without intromission.

The phallus, if present, is located ventrally in the proctodeum. Dysfunction or disease of the phallus can cause reproductive failure. Psittacine birds do not have a phallus, and copulation is accomplished by an eversion of the cloacal wall, which contains the slightly raised papilla that transfers semen to the everted orifice of the oviduct.¹⁰⁶ Determination of

gender can be accomplished by identifying the seminal papilla in the male of many cockatoos but not in Amazon parrots and macaws (Harrison GJ, unpublished).

Semen

In domestic fowl, spermatozoa undergo maturation and become fully fertile in the ductus deferens. The transient time required for sperm to pass from the testes to the distal ductus deferens is estimated to be from one to four days. Seminal plasma that is formed in the efferent and connecting ductules of the epididymis and ductus deferens accompanies the spermatozoa. Seminal plasma composition can vary among species but is similar in concentration and constituents in the budgerigar and domestic fowl.¹⁴⁵ In most birds, semen is stored in the ductus deferens. In Passeriformes, semen is also stored in the seminal glomus, which is the enlarged terminus of the ductus deferens. A lymph-like fluid, called “transparent fluid,” originates in the proctodeum and mixes with the semen during ejaculation. The function of this fluid is uncertain, but it does contain blood clotting agents that are deleterious to the spermatozoa.⁸⁶

In Passeriformes, spermatozoa are of the complex type, which can be differentiated from the simple type of sperm found in other birds by their predominantly spiral structure.

Semen can be collected from birds for artificial insemination, to evaluate its reproductive potential, to detect disease and to distinguish species or subspecies.¹⁵⁴ The consistency of normal semen ranges from that of water to that of heavy cream. Watery semen may indicate high volumes of transparent fluid in the sample. Normal semen is light white to milky, and brown, green or red discoloration may be due to fecal contamination or cloacal hemorrhage from over-exuberant semen collection. Production of fertile eggs is the best indicator of sperm viability, but determining sperm count and motility can be used to estimate function (Table 29.3).⁴⁵ One million sperm are required for optimal fertility in the domestic fowl.

Sperm concentration can be determined by mounting semen on a hanging drop slide, use of a spermatocrit or direct counting in a hemocytometer. In those species with higher sperm concentrations, dilution with artificial insemination semen extender may be necessary prior to evaluation. Motility is estimated as the percentage of spermatozoa moving in a forward motion as seen under high magnification. Live-dead counts using an eosin-nigrosin stain make it possible

TABLE 29.3 Volume and Concentration of Ejaculates from Selected Birds ^{21,69,80,106,147}

Bird	Sperm Concentration	Ejaculate Volume
Chicken	1.7-3.5 billion/ml	500-1000 μ l
Budgerigar	9.5-11.3 billion/ml	3.5-13 μ l
Pheasant	10 x 10 ⁹ / μ l	50-250 μ l
Large Psittaciformes	9-10 million/ml	50-100 μ l
Emu	4.4 billion/ml	1200 μ l

to evaluate the concentration of live sperm. Live-dead counts, computer-assisted measurement of spermatozoal swimming speed and metabolic rates of semen can also be used to determine semen quality.

Male Hormonal and Physiologic Factors

In the male, FSH and LH exert gonadotrophic properties similar to those described in the hen. FSH initiates the growth of seminiferous tubules and results in increased spermatogenesis. LH promotes development of the testosterone-producing cells of Leydig. Testicular growth is approximately logarithmic until half of the ultimate size is attained.⁴² Higher levels of testosterone are then responsible for male sexual behavior. Testosterone increases spermatogenesis and growth of accessory reproductive organs, such as the epididymis and cloacal gland. Testosterone also causes manifestation of secondary sexual characteristics such as comb growth, plumage and



FIG 29.6 Male cockatoos will frequently attack and sometimes kill their hens. This behavior can occur in pairs that have been stable and producing young for years. The precise cause of these attacks is unknown, but they are most common in the early part of the breeding season. Males generally become reproductively active earlier than the females, and a hen's failure to respond to a soliciting male may facilitate an attack. In this cockatoo hen, most of the beak and a part of the skull had been removed by the male.

bill color, structure of feathers, vocalizations and reproductive behavior. Specific reproductive behavior affected by testosterone, and probably mediated by other hormones, includes territorial aggression, courtship, copulation, nest building, incubation and care of the young. Testosterone levels are highest in many species at the time of establishment and defense of territory, courtship and nesting activity.⁴² Substantial field observations suggest that the testes become functional earlier than the ovaries, and that coordination of the reproductive effort is mediated by environmental and photoperiodic stimulation in the female.⁴² As in the hen, prolactin, progesterone, estrogen and androgens are all involved with incubation and brood care. This complex control of the reproduction cycle may account for breeding failures and mate aggression in captive Psittaciformes (Figure 29.6).



Female Reproductive Disorders

■ Egg Binding and Dystocia

Two of the most common clinically recognized reproductive disorders seen in avian species are egg binding and dystocia. Egg binding is defined as the failure of an egg to pass through the oviduct at a normal rate. Most companion bird species lay eggs at intervals greater than 24 hours, and individuals within a species may vary by more than one day from the normal oviposition rate.⁴ Variability in egg transit times makes it difficult to determine when a problem is occurring.

Dystocia defines a condition in which the developing egg is in the caudal oviduct and is either obstructing the cloaca or has caused oviduct tissue to prolapse through the oviduct-cloacal opening. Egg movement through the oviduct can stop at various locations. The most common anatomic areas for problems to occur are the caudal uterus, vagina and vaginal-cloacal junction.

The pathogenesis of egg binding in a particular case can be multifactorial. The pubic bones are not fused in birds, and pelvic deformities seldom play a role in dystocia. Common causes of dystocias are oviduct muscle dysfunction (calcium metabolic disease, selenium and vitamin E deficiencies), malformed eggs,

excessive egg production, previous oviduct damage or infection, nutritional insufficiencies, obesity, lack of exercise, heredity, senility and concurrent stress such as environmental temperature changes or systemic disease.^{62,66,140,160} Dystocia can also result from breeding birds out of season, egg production in virginal hens and a persistent cystic right oviduct (Color 29.20).⁷²

Abnormally prolonged presence of an egg in the oviduct causes a multitude of complications in the hen (Figure 29.7). The severity of these complications depends on the species, the bird's previous health, the cause of egg binding, the egg's location in the oviduct and the time elapsed since egg development began. An egg lodged in the pelvic canal may compress the pelvic vessels and kidneys, causing circulatory disorders and shock.¹⁴⁰ An impacted egg may cause metabolic disturbances by interfering with normal defecation and micturition, inducing ileus and renal dysfunction.⁶⁶ Pressure necrosis may occur to all three layers of the oviduct wall and lead to rupture.

Clinical Signs

Budgerigars, canaries, finches, cockatiels and lovebirds most frequently have problems with dystocia.¹⁴¹ This is probably because the presentation of a palpable egg for more than a few hours in small birds is generally more serious than it is in larger birds. The patient's clinical signs will depend on the severity of the complications. Generally, the hen appears depressed, has an abnormally wide stance, is reluctant to fly or perch and may show persistent wagging of the tail and straining movements of the abdomen. Canaries often exhibit drooped wings. Rear limb paresis or paralysis may occur. Egg-related peritonitis, septicemia, leg injuries and abdominal neoplasia show similar clinical signs (Color 29.27). Any depression can lead to anorexia, which further compromises the bird's condition.

Hens with dystocia frequently present with depression and secondary complications that require emergency therapy. A complete history including information of past breeding activity and the diet "consumed" will often suggest a pathogenesis. A thorough but rapid physical examination can also establish contributing factors such as obesity, concurrent disease or a malformed egg. Dystocias are most critical in passerines and other small birds, many of which can survive only a few hours without aggressive therapy.⁶⁶ Initially the therapeutic plan is to stabilize the patient (see Chapter 15) with an emphasis being placed on correcting the most likely etiology for the dystocia.

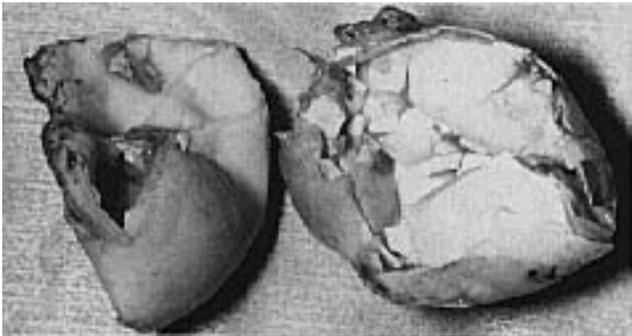
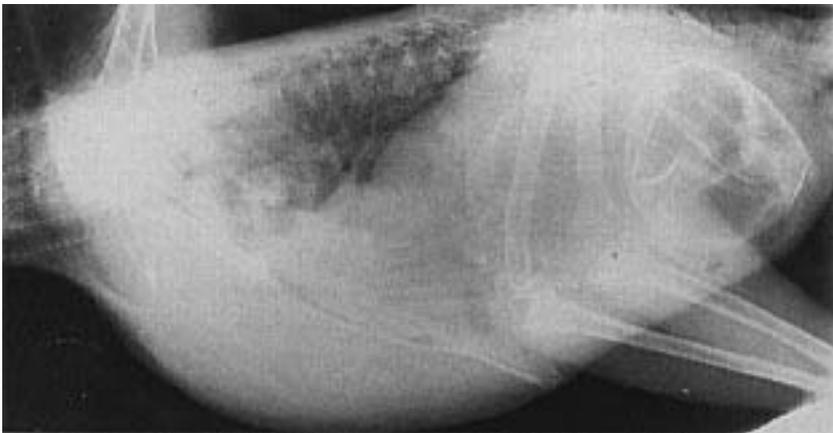
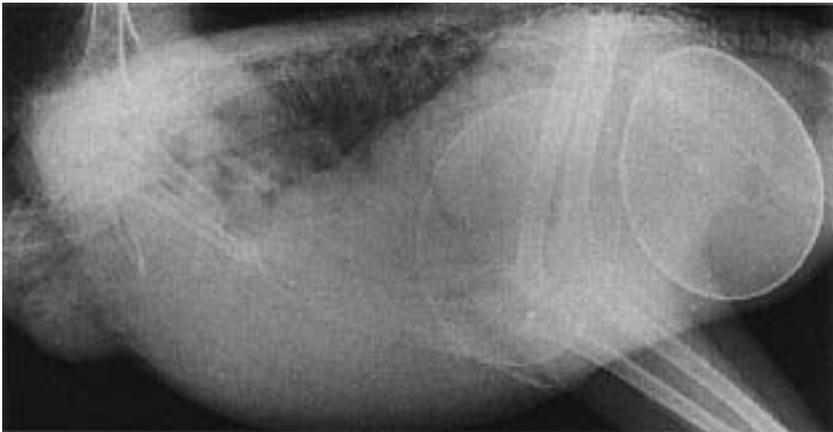


FIG 29.7 An Amazon parrot hen was presented for depression, a hunched stance and tenesmus. A firm palpable mass was present in the caudal abdomen. Radiographs revealed two calcified eggs. The caudal egg could not be located through the cloaca. The contents of the eggs were removed by oocentesis, and the eggs were collapsed. Radiographic appearance of the collapsed eggs. The hen's attitude improved immediately and she began eating normally. The fragmented egg shells were passed two days after they were collapsed.

The severity of the dystocia, and speed of correction that will be required, can be partially estimated by the level of depression. Careful abdominal palpation and a cloacal examination are required to determine

the egg's position in the reproductive tract. In smaller birds the displaced ventriculus may make palpation of an egg difficult. Soft-shelled eggs, shell-less eggs or eggs located cranial to the uterus can also be difficult to palpate. Suspected egg masses must be differentiated from palpable hernias, lipomas or ascites. Radiographs are a useful confirmatory tool but may not delineate a shell-less egg. Radiographically identifying more than one egg in various stages of development is common.

Therapy

The most important consideration in initiating therapy for dystocia is to establish a physiologic normal state. Attempts to remove the egg are secondary to stabilizing a patient in shock. In minimally depressed patients with few complications, the egg will usually pass if the hen is provided with supplemental heat, injectable calcium, selenium, vitamin E, vitamin D₃ and easy access to food and water.⁵⁶ Others require subcutaneous or IV fluids, rapidly acting steroids to combat shock, antibiotics to treat sepsis or peritonitis and injectable vitamins and minerals to address further nutritional deficiencies. Prolapsed oviductal or cloacal tissues should be moistened and cleaned with warm, sterile saline washes and water-based antiseptic ointments, such as chlorhexidine. Lubricating tissues surrounding the egg or the cloaca or vagina itself may be of some help to egg expulsion.

The bird should be placed in an incubator at 85 to 95°F with an inflow of heated, moisturized air. If the egg is not expelled within a few hours, then a prostaglandin injectable product (dinoprost tromethamine) can be administered IM or applied topically to the oviductal tissue. This compound appears to be superior to oxytocin because it has the combined effect of inducing uterine contraction while relaxing the uterovaginal sphincter (see Chapter 18).⁷³ Prostaglandin or oxytocin should be used only in cases where the uterus is thought to be intact and no adhesions to the oviduct are suspected. A hen receiving these agents must be able to withstand the increased contractions of the oviduct and abdomen that occur following the administration of oxytocin. Clinical signs

of oxytocin response include tail pumping, panting, abdominal contractions and elimination of the egg. Increasing and repeated doses of oxytocin can be given if initial injections have no effect. Experimental use of prostaglandins and arginine vasotocin in domestic species has shown that injections of either of these drugs may result in oviposition.^{144,157} Arginine vasotocin likely causes the release of prostaglandins from the uterus. Clinical use of vasotocin in reptiles suggests that this drug may be of some value in birds (0.01 to 1.0 mg/kg BW) (Lloyd M, personal communication). It has been shown that uteri are more sensitive to vasotocin than to oxytocin.⁹⁷ Complications of oxytocin or vasotocin use include oviduct rupture.

If medical therapies fail to elicit oviposition, then more aggressive approaches that require manual manipulation of the patient may be necessary. Massaging the abdomen or cervix may help stimulate egg passage or relax the cervix so that the egg can be passed. The egg itself can be digitally manipulated caudally for expulsion (Color 29.30). The use of warmed water-soluble solutions or ointments (saline with methyl cellulose) to lubricate the urodeum or vagina is equivocal. Gentle, persistent, caudally directed pressure on the egg may supplement weakened muscular contractions and loosen any recently formed adhesions. Only gentle traction should be used to prevent rupture of the oviduct. As long as the bird remains stable, repeated attempts at digital egg removal should continue. The cervix can be dilated by using a speculum to insert a blunt probe that is advanced in gentle, twirling motions (Figure 29.8). Eggs may be fertile and viable and should be incubated following expulsion. Digital manipulation and contracting therapy should not be used if one suspects ectopic eggs, uterine torsion, uterine rupture, or uterine constrictions due to adhesions (mucosal adhesions to the egg or the opposite uterine wall or serosal adhesions to other abdominal structures).

Ovocentesis

If the bird's condition is deteriorating or if an inappropriate period of time has passed since the dystocia was first noted, then more aggressive therapy such as ovocentesis must be considered. Ovocentesis is performed by aspirating the contents of the egg with a large needle (18 ga).¹⁴¹ Preferably the egg is manipulated with the use of a speculum so that it is observable and tapped through the cloaca.² If this is not possible, the egg is brought in juxtaposition to the abdominal wall so that other organs are not damaged during a transabdominal aspiration procedure. Fol-

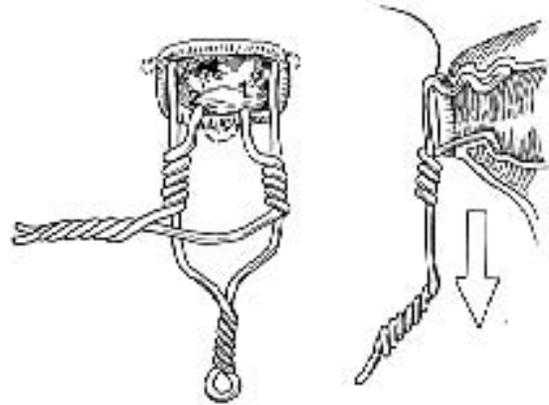


FIG 29.8 A speculum can be used to facilitate evaluation of the cloaca and removal of eggs. 1) opening of the ureter and 2) vaginal opening.

lowing aspiration of the egg contents, the egg can be gently collapsed (Figure 29.9). The risk of tearing the oviduct and producing peritonitis does exist but appears to be minor (see Chapter 48). The shell fragments and remaining contents of the egg should pass within several days (see Figure 29.7). Fragments that are visible through the cloaca can be gently removed. Some clinicians advocate flushing the uterus post-oviposition with an iodine, chlorhexidine or saline solution to help remove egg fragments and to decrease the incidence of metritis.⁵⁶ A Brunswick feeding catheter (3 to 5 Fr) can be placed through the cervix for this procedure (Figure 29.10). A course of broad-spectrum antibiotics, chosen based on the results of a Gram's stain collected from the uterus and confirmed as the correct choice by culture and sensitivity, is also recommended.

If the egg is lodged in the caudal oviduct or cloaca and the survivability of the egg is critical, then an episiotomy may be beneficial in delivering the egg.¹⁴¹ A laparotomy may be necessary to remove egg material or to perform a hysterectomy in cases where the uterus is ruptured or severe adhesions exist. Soft-shelled eggs located cranial to the uterus or ectopic eggs also require surgery.^{104,140}

Many hens with dystocia will attempt to lay another egg. Administration of medroxyprogesterone will stop ovulation, but there are side effects and its use is controversial. Following medroxyprogesterone administration, eggs already present in the proximal oviduct may continue to descend, complicating the

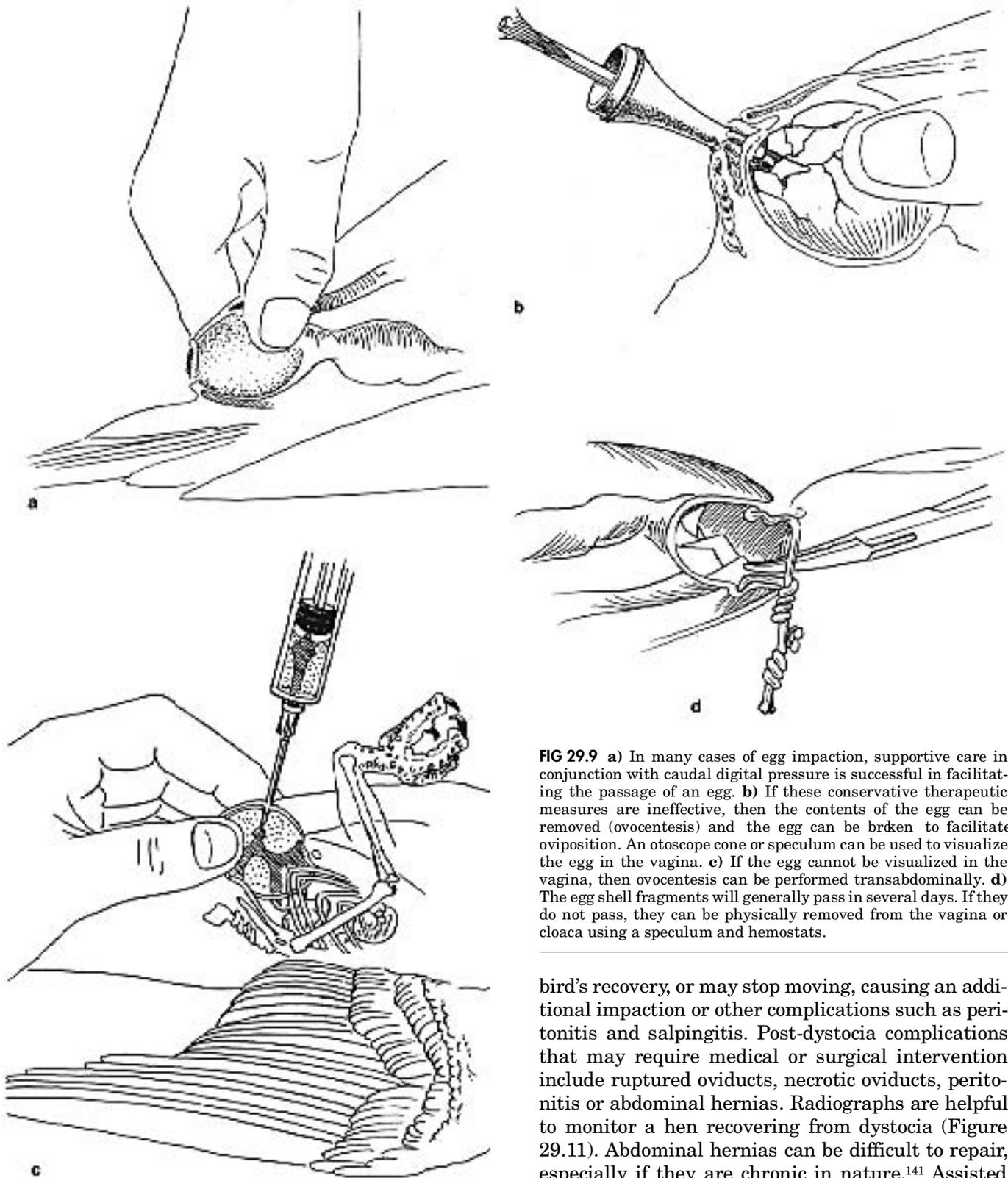


FIG 29.9 **a)** In many cases of egg impaction, supportive care in conjunction with caudal digital pressure is successful in facilitating the passage of an egg. **b)** If these conservative therapeutic measures are ineffective, then the contents of the egg can be removed (ovocentesis) and the egg can be broken to facilitate oviposition. An otoscope cone or speculum can be used to visualize the egg in the vagina. **c)** If the egg cannot be visualized in the vagina, then ovocentesis can be performed transabdominally. **d)** The egg shell fragments will generally pass in several days. If they do not pass, they can be physically removed from the vagina or cloaca using a speculum and hemostats.

bird's recovery, or may stop moving, causing an additional impaction or other complications such as peritonitis and salpingitis. Post-dystocia complications that may require medical or surgical intervention include ruptured oviducts, necrotic oviducts, peritonitis or abdominal hernias. Radiographs are helpful to monitor a hen recovering from dystocia (Figure 29.11). Abdominal hernias can be difficult to repair, especially if they are chronic in nature.¹⁴¹ Assisted oviposition may cause a flaccid cervix, allowing reflux of feces and urine into the uterus. Daily flushing and Gram's staining of the uterus to monitor pro-

gress following the removal of an egg appears to reduce the occurrence of metritis (Harrison GJ, unpublished).

■ Prolapsed Oviduct and Cloaca

Prolapse of the oviduct may occur secondary to normal physiologic hyperplasia and egg laying or as a sequela to dystocia (particularly in canaries and budgerigars) (Color 29.31 and 29.32).⁶ Excessive contraction of the abdominal muscles, perhaps exacerbated by poor physical condition and malnutrition, may cause these prolapses.¹⁴ Usually the uterus protrudes through the cloaca, often together with a partial prolapse of the vagina and cloaca. Distal portions of the oviduct may also prolapse, and frequently an egg is present (Color 29.30). Oviduct prolapses have been associated with deformed, soft-shelled and shell-less eggs.⁸⁴ Timely, aggressive therapy is needed to prevent devitalization of uterine tissues and secondary infections. All exposed tissue must be kept as moist as possible and cleaned thoroughly with sterile saline solution. Topical steroid preparations containing antibiotics or dimethyl sulfoxide gel can be used to reduce swelling so that prolapsed tissues can be replaced. If no egg is present, tissue replacement is accomplished by gently guiding the tissues through the cloaca with pressure from a lubricated swab or thermometer. Repeated replacement of tissues may be required, as prolapses often recur. Stay sutures placed in the cloaca or percutaneous retention sutures may prevent further prolapsing while uterine tissues regress in size, abdominal tissues regain structural integrity and the hen has a chance to regain normal muscle tone and strength.^{104,139} The prognosis for birds with uterine prolapses is good as long as they are treated immediately.⁶⁶

If an egg is present in the prolapsed tissue, it must be removed before the tissue is replaced in the abdomen (Color 29.31). Digital manipulation or implosion of the egg as discussed under dystocia may be effective. Chronically displaced tissue that contains eggs or egg material may require surgical debridement due to adhesions and shell abnormalities. In severe cases of uterine damage and necrosis, a partial or complete hysterectomy may be necessary, but is best delayed until the bird's condition is stable (see Chapter 41).¹⁴¹

■ Salpingitis and Metritis

Salpingitis may occur from air sacculitis, pneumonia, liver disease or retrograde infections of the lower

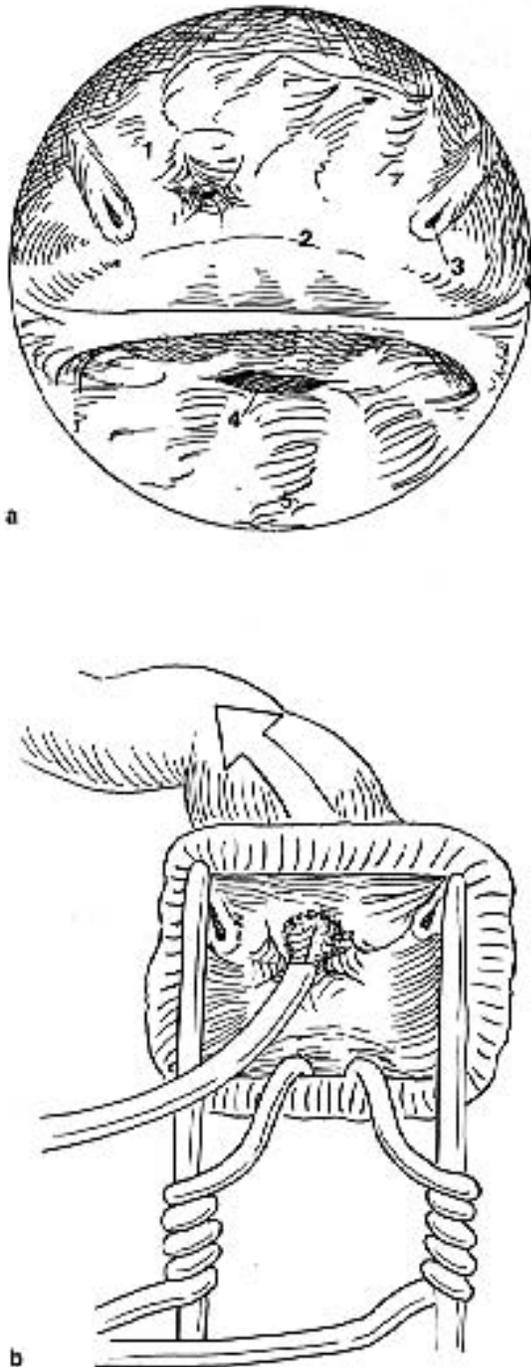


FIG 29.10 **a)** Caudal to cranial view of the normal cloacal anatomy of a hen: 1) vaginal opening, 2) urodeum, 3) opening of ureter, 4) rectal opening and 5) coprodeum. **b)** Post-dystocia flushing of the uterus with chlorhexidine or sterile saline may reduce the occurrence of salpingitis.



FIG 29.11 a) An adult Amazon parrot hen was presented for evaluation of a ventral abdominal mass. b,c) Radiographs indicated a herniated egg and a second abnormally formed egg in the dorsal abdomen. Surgical correction of the hernia and a salpingohysterectomy were recommended but refused. d) Additional radiographs taken eight weeks after the initial presentation indicated that the herniated egg was being resorbed, and that the second egg was increasing in density.

Theriogenology

Egg Incubation

From the poster of Cockatiel Embryonic Development, reprinted with permission. Posters are available from the University of California, ANR Publications, 6701 San Pablo Avenue, Oakland, CA 94608-1239.

Color 29.1

a) The unincubated fertile egg has a distinct ring consisting of a white peripheral region (area opaca) surrounding a clear central region (area pellucida). **b)** The same area in the unincubated infertile egg is small in size, lacks cellular organization and looks like a small piece of cotton on the surface of the yolk. The diameter of the fertile blastodisc is four to five times that of the same area in an infertile egg.

Color 29.2

Unincubated egg with a large blood spot indicative of bleeding from the follicle during ovulation.

Color 29.3

Incubated fertile egg showing two common types of first-day failures. Neither shows development of embryonic structures. **a)** Eggs with positive development fail before blood formation, while **b)** eggs with blastoderm without embryo continue cell division until blood-forming stages. Blastoderms without embryos can be seen during candling, when a blood ring is usually present. Positive developments and blastoderms without embryos may vary in size depending on the length of incubation.

Color 29.4

A profile shot of a yolk 48 hours after the onset of incubation showing a bleb that represents the embryonic area. This bleb can also be seen by candling.

Color 29.5

Cockatiel egg at approximately three days of incubation (embryo stage 17). The heart

and major blood vessels of the yolk sac are visible. The embryo has turned to its left side.

Color 29.6

Cockatiel egg at approximately 4.5 to 5 days of incubation (embryo stage 24). The subdivisions of the brain, the developing pigmented eye, increased yolk sac circulation, developing wing, leg buds and the allantois (arrow) are visible.

Color 29.7

Embryo removed from an egg at 4.5 to 5 days of incubation. The divisions of the brain are pronounced. The limb buds and tail fold are prominent.

Color 29.8

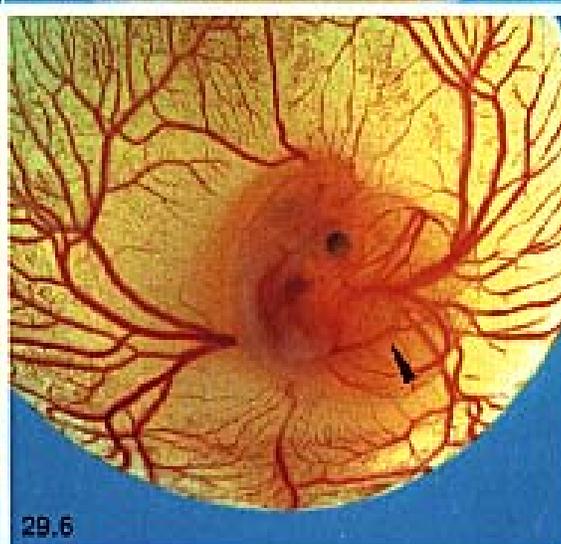
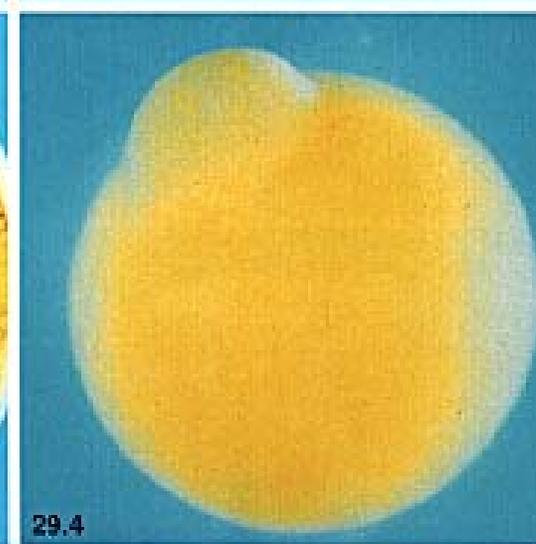
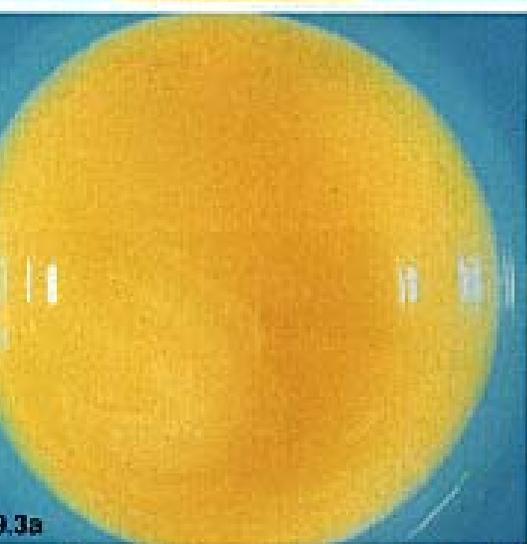
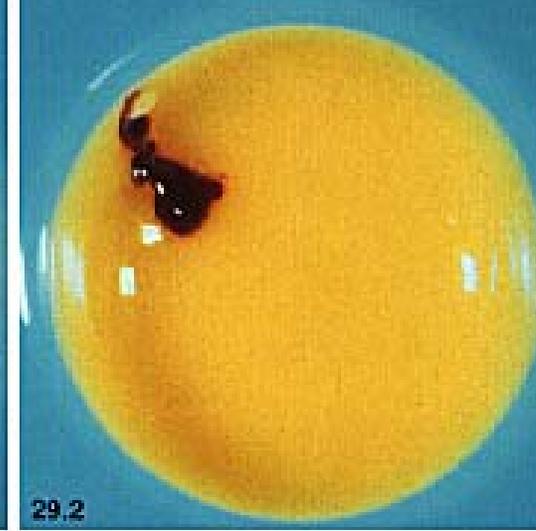
Normal cockatiel embryo approximately six days of incubation (embryo stage 28-29). The characteristic cockatiel-shaped head shows prominent mid-brain divisions. There is further development of the eye, including appearance of the choroid fissure (arrow). Outlines of the digits can be seen on the developing wings and legs.

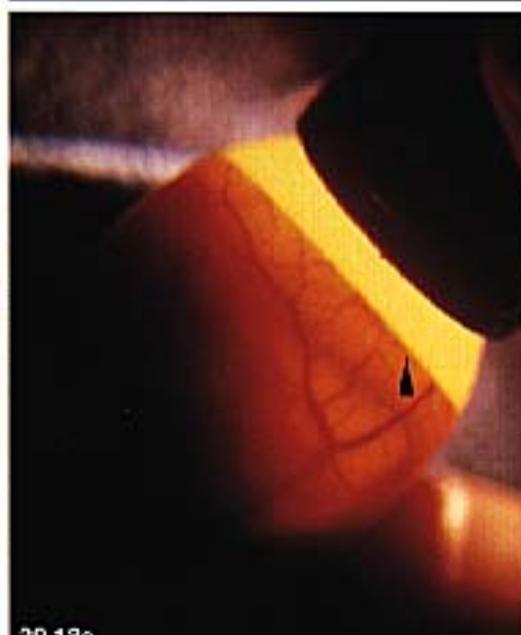
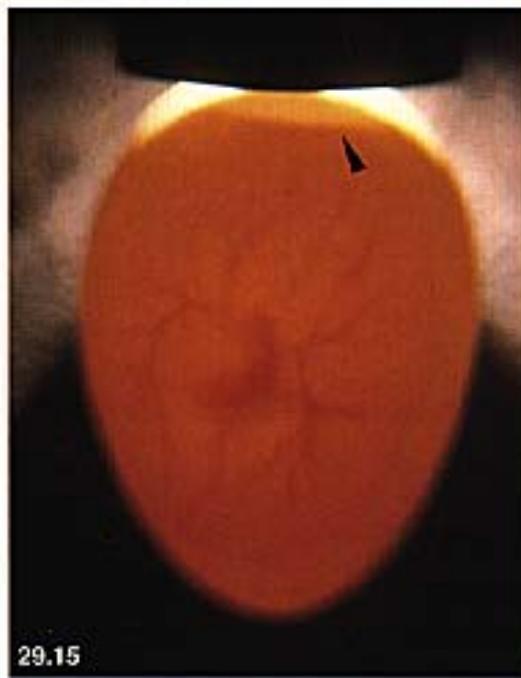
Color 29.9

Normal embryo at approximately 7 to 7.5 days of incubation (stage 31). The egg tooth, beak and scleral papillae of the eye are evident. The limbs are developing and the bones are beginning to calcify. The first feather follicles can be seen.

Color 29.10

Normal cockatiel embryo approximately nine days of incubation (embryo stage 35). A prominent egg tooth and early development of the upper beak are evident. Scleral papillae in the eye form a complete ring. The head shape is square with a less prominent mid-brain. Note the feather formation on the dorsal body surface and calcification in the long bones and toes.





Theriogenology

Color 29.11

Normal embryo at 12 to 13 days of incubation illustrates features unique to cockatiels. The lower beak is shorter than the large square upper beak. Note the serrations on the inner surface of both beaks. The scleral papillae can no longer be seen as the eyelid is now nearly closed. Calcification of the characteristic psittacine toes and growth of sparse feathers are evident.

Color 29.12

Normal cockatiel embryo at approximately 16 to 17 days of incubation (stage 42). The eyelids are closed, the beak is enlarged and the egg tooth and toenails are almost completely formed. The yellow pigmentation of the skin reflects the metabolism of fat. The extremities may appear pinker than illustrated.

Color 29.13

Normal cockatiel chick at hatching (18 days of incubation). The yolk sac is completely resorbed and the umbilicus is sealed. Hatchlings would not be expected to appear as hyperemic as illustrated.

Color 29.14

The appearance of a blood spot in this egg indicates that the egg was fertile. The de-

velopment of a blood ring is characteristic for early embryonic death (courtesy of Kim Joyner).

Color 29.15

Day ten of incubation in a Black Palm Cockatoo egg. Note the centrally located embryo with the developing blood vessels and the small air cell (arrow) (courtesy of Kim Joyner).

Color 29.16

Day 14 of incubation in an Umbrella Cockatoo egg (courtesy of Kim Joyner).

Color 29.17

Day 12 of incubation in an Eclectus Parrot egg (courtesy of Kim Joyner).

Color 29.18

a) Day 21 of incubation in a Military Macaw egg. Note the development of the air cell (arrow) and the still prominent blood vessels. **b)** Day 26 of incubation in the same egg. The shell is pipped and the blood vessels are collapsing as the bird switches from chorioallantoic to pulmonary respiration (courtesy of Kim Joyner).

uterus, vagina or cloaca. In Passeriformes, salpingitis has been associated with impaction of the oviduct and egg-related peritonitis.⁸⁵ Foreign bodies such as wheat grains located in the oviduct can cause metritis and salpingitis.¹³² Excessive abdominal fat has been associated with many cases of salpingitis in domestic fowl.¹²⁷ The etiologic agent most frequently isolated from birds with salpingitis is *E. coli*.¹³² Other bacteria such as *Mycoplasma gallisepticum*, *Salmonella* spp. and *Pasteurella multocida* can affect other organ systems simultaneously with the salpinx. In raptors, *E. coli* and *Streptococcus* spp. have been described as causes of salpingitis.³² Ascending infections from the cloaca induced by copulation, inappropriate treatment for egg binding or uterine prolapse may induce salpingitis.⁶⁶

While salpingitis is most common in adult hens, it can also occur in young birds.¹³² Salpingitis reportedly occurs less frequently than oophoropathies, obstruction of the oviduct and ectopic ovulation in a variety of avian species.⁸⁴ Depression, anorexia, weight loss and abdominal enlargement can occur with salpingitis. A discharge from the cloaca may also occur. Acute salpingitis in poultry is characterized by an enlarged, dark red oviduct with involvement of the infundibulum.¹⁵⁵ The lumen may contain cream-colored, slimy fluid or cheesy, yellowish fibrinous exudate. The oviduct may be thin-walled or decreased in length (common with Newcastle disease virus or infectious bronchitis virus).^{13,19} Congestion may be the only grossly observable change, although salpingitis may not be recognized macroscopically.¹¹⁹ Bacteriologic and histologic examinations are necessary to establish a diagnosis.

Cockatiel hens that have a history of egg laying followed by mild depression and weight loss may have a low grade salpingitis or focal egg-related peritonitis (Harrison GJ, unpublished).

Metritis is a localized problem within the uterine portion of the oviduct. It can be a sequela to dystocia, egg binding or chronic oviduct impaction. Bacterial metritis is often secondary to systemic infections.¹⁴¹ Metritis may affect shell formation or uterine contractility or cause infections in embryos (embryonic death) or neonates (weak chicks). Metritis can also cause egg binding, uterine rupture, peritonitis and septicemia. Coliforms, especially *E. coli*, are frequently implicated. Coliform metritis may be complicated by poor diet, and death rates are highest in hens during the ovulatory and egg-laying period.⁶²

In more advanced cases, birds may be depressed and have an enlarged abdomen and a palpable turgid uterus. Radiographs often reveal indistinct abdominal detail with a diffuse increase in soft tissue density. Ultrasound has been used in ostriches.⁶⁹ An affected ostrich hen may have a history of erratic production, malformed or odoriferous eggs or a sudden drop in production. An odoriferous cloacal discharge may occur, and the WBC may range from 20,000 to 100,000 mm³. Metritis and salpingitis are treated aggressively with parenteral antibiotics, supportive care and therapy for shock (see Chapters 15 and 18). In non-responsive cases, a laparotomy may be necessary to remove necrotic tissue, inflammatory exudates or egg material. The oviduct may be flushed directly with lactated Ringer's solution (with or without antibiotics) by placing an IV fluid tube or soft catheter into the vagina.⁶² Visualizing placement of the tube can be augmented by use of a cloacal protractor (see Figure 29.10).

Oviduct Impaction

Impaction of the oviduct is often a sequela to salpingitis (most frequently), metritis or egg binding. One study found that impactions were nearly always associated with obvious salpingitis in older birds.⁸⁴ Impactions may occur from excess secretion of mucin and albumen associated with cystic hyperplasia or inspissated egg material in the magnum. Soft-shelled, malformed or fully formed eggs can impact in the distal oviduct. Cockatiels, canaries and budgerigars are frequently affected, and the condition has been documented in raptors and an African Grey Parrot.^{6,14,35,66,83,85} Clinical changes are not always obvious and may include a cessation of egg production, progressive loss of condition and alternation between constipation and diarrhea. Chronic deterioration is particularly common if concurrent peritonitis or salpingitis is present. The abdomen may be diffusely or unilaterally (usually left side) enlarged, birds may be reluctant to fly or walk and periodic anorexia may occur.⁶⁶ Radiology can be helpful in some cases, but many oviduct impactions can be diagnosed only through endoscopy or exploratory laparotomy or at necropsy. Impacted oviducts may contain obvious egg material, gray or yellow purulent material, calcareous deposits or albumen. Diffuse peritonitis with adhesions can also occur with oviduct impactions. Treatment consists of parenteral antibiotics and in most cases, surgery to clean, repair or remove necrotic portions of the oviduct.^{3,66}

Oophoritis

The ovary can reflect the general health of a mature hen because many infectious diseases and physiologic abnormalities cause retrogressive changes in this organ.¹²⁷ Endoscopic evaluation of the female reproductive tract should include the ovum as well as the ovary. The normal ovary with mature follicles has yellow, turgid ova (Color 29.21). When diseased, the ovum can be wrinkled, black, enlarged, firm or hemorrhagic. In addition, abnormal yolk may appear coagulated or “cooked” and flake off onto the ovary or into the abdominal cavity.¹⁵⁵ Adhesions may exist between follicles and the follicles may be slightly stalked.¹⁵⁹ Pullorum disease of domestic fowl is characterized by discolored, pedunculated and inspissated ova. Other bacteremias may cause congestion, distortion and atresia of the follicles.¹³² Peritonitis commonly occurs with oophoritis.

Clinical signs of oophoritis include depression, anorexia, chronic wasting and sudden death. Therapy includes supportive care and parenteral antimicrobial agents as dictated by the etiologic agent.

Parasites

Eggs may contain adult ascarids that probably enter the oviduct from the cloaca due to reverse peristalsis.⁷⁴ Flukes (*Prosthogonimus ovatus* and related trematodes) inhabit the oviduct of Anseriformes and Galliformes. Heavy infections may cause soft-shelled or shell-less eggs, resulting in salpingitis.⁸⁷ Adult flukes less than 1 cm long may be passed in the eggs. Prevention involves the control of aquatic snails and dragonflies that serve as intermediate hosts.

Cloacal Problems

Cloacitis, cloacal strictures, cloacal liths and chronic prolapse of the cloaca can interfere with egg laying and copulation (Figure 29.12). These conditions may in turn result from traumatic egg laying. The cloaca may become chronically impacted with an egg, resulting in severe cloacitis and abdominal adhesions.⁸⁴ Feathers, fat and abdominal lipomas may occlude the vent, inhibiting reproductive ability.¹³⁹ Both medical and surgical approaches are helpful in treating cloacal problems (see Chapter 19). It is interesting to note that the cloaca prolapses normally in the Vasa Parrot during the breeding season.

In some cases, cloacal papillomas may interfere with copulation and semen transport (see Color 19). Pain-

ful lesions in the cloaca may also discourage individuals from mating; however, healthy chicks can be produced by breeding pairs of psittacine birds where one or both adults have mild to moderate cloacal papillomatosis.¹⁰⁷ The etiology of these lesions is unknown and it is recommended to exclude birds with this condition from a breeding aviary. Affected birds in a collection should be isolated from unaffected birds. Affected parents may or may not produce affected offspring, but regardless, chicks from affected parents should be hand-raised in isolation. Results from various treatment regimes for cloacal papillomatosis vary.^{139,180} A diet low in fat and high in fresh fruit and vegetables with high vitamin A or beta carotene was considered useful in resolving cloacal papillomatosis and cloacal adhesions in one case.¹⁷⁸

Cystic ova (reported in budgerigars, canaries and pheasants) may be single or multiple and may be noted during laparoscopy in apparently healthy psittacine hens (Color 29.22).^{66,84} Ovarian tumors and cystic hyperplasia of the oviduct can occur secondarily.^{14,66} The etiology of this condition and its clinical importance are unknown, but a primary endocrine disturbance is suspected because this lesion is frequently associated with hyperostosis.⁶ In affected birds, dyspnea, altered movement and diffuse distention (ascites) of the abdomen are common.¹³² Although not always palpable, abnormal ova may be firm, soft, fluctuating or pedunculated. Cysts may rupture, so palpation should proceed carefully. Radiographs may show a diffuse soft tissue density near the cranial lobe of the left kidney. Endoscopically, the ovary may be enlarged with many thin-walled cysts full of straw-colored fluid. Respiratory distress may be eliminated by transabdominally aspirating cystic

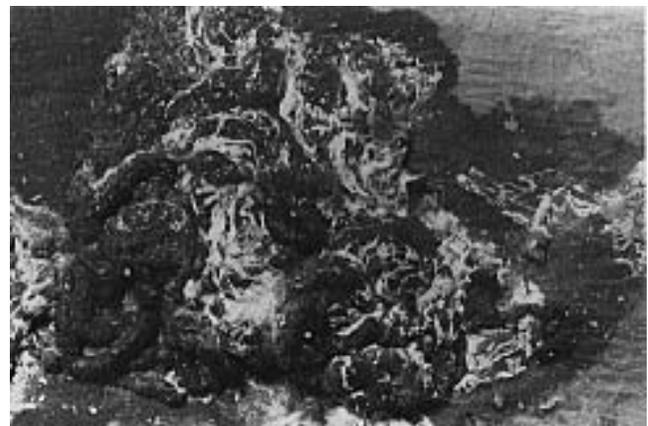


FIG 29.12 Voluminous stools are common in hens that are preparing to lay or that are incubating a clutch of eggs. Similar quantities of feces may be produced by birds with cloacal pathology that interferes with the normal passage of excrement.

fluid with a needle and syringe.¹³² Cystic ovaries were successfully treated in two budgerigars with oral testosterone.¹⁴ Removal of the ovaries, although technically difficult, may be the only long-term treatment.

■ Cystic Hyperplasia of the Oviduct

Most reports of cystic hyperplasia of the oviduct are in budgerigars and domestic fowl.¹⁴ The entire oviduct is dilated with a white or brown mucoid fluid, white or creamy masses or occasionally secondary cysts (Figure 29.13). Cysts also can occur secondary to improper formation of the left oviduct (possible degeneration during embryonic development) or from adhered lips of the infundibulum.¹²⁷ The ovary in affected hens may also have cystic changes suggesting an endocrine abnormality. Progressive abdominal distention, ascites and respiratory distress are the most common clinical changes. Palpation and radiographs may reveal the distended oviduct. Abdominal paracentesis may be attempted either for diagnosis or for relief of respiratory distress. Laparotomy will provide a conclusive diagnosis. Hormonal therapy with testosterone may prove effective in resolving the immediate problem, but a hysterectomy may be necessary to prevent future problems.

If a rudimentary right oviduct (or ovary) exists, it may also become cystic (Color 29.20).^{86,127} Cysts are of walnut size, contain watery or milky fluid and are situated near the cloaca in domestic fowl.¹¹⁹ Small cysts may go undetected, but large cysts may place pressure on abdominal organs. Egg binding has occurred secondary to a fully developed right oviduct in a budgerigar.⁷² The hen was depressed and thin and had a distended abdomen. Successful bilateral hysterectomies were performed to remove the egg-filled left oviduct and the right oviduct that contained a walnut-sized cyst with gelatinous fluid.

■ Neoplasia

In one study, neoplasia of the reproductive tract accounted for up to 4.3% of all reproductive disorders.⁸⁴ Budgerigars often have neoplasia in the ovary or oviduct.¹⁴ Ovarian neoplasia has been reported less frequently in other Psittaciformes. Ovarian and oviduct neoplasia occur more commonly in gallinaceous birds^{155,159} and occasionally in waterfowl and have been reported in a free-ranging Great Tit and a Mauritius Kestrel.^{31,120}

Presenting signs are similar to those seen with cystic ovaries or oviduct impactions.⁶⁶ In small birds, ascites

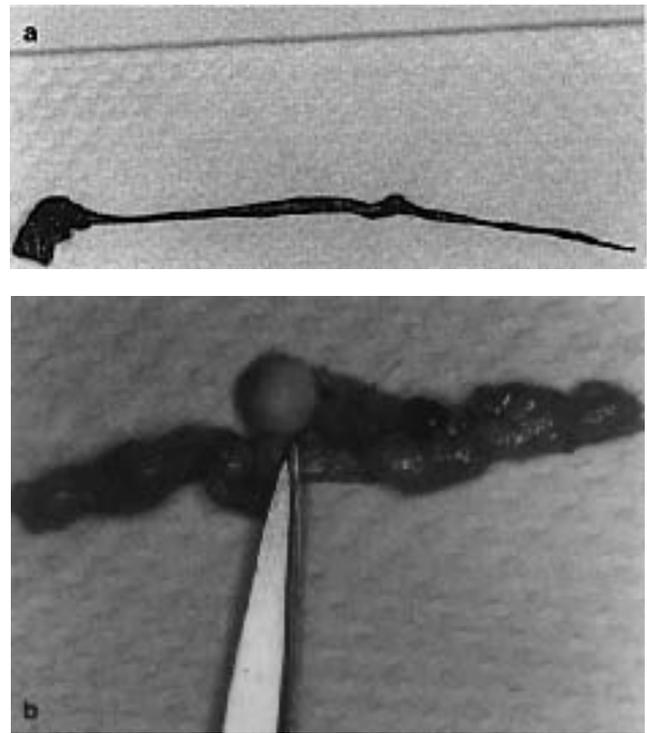


FIG 29.13 a) Normal oviduct from a cockatiel (cranial is to the left) b) and an oviduct from a hen filled with creamy masses and a mummified egg.

and peritonitis can produce clinical changes similar to those associated with reproductive neoplasias. Ovarian tumors can be very large and represent up to one-third of the body weight. Egg retention,¹⁴¹ concurrent cysts,⁶ ascites and herniation are common sequelae to reproductive tract neoplasias. Changes in secondary sex characteristics (cere color change in the budgerigar) may also occur. Radiographs can be helpful, although an enlarged ovary or oviduct creates an image similar to that seen when uncalcified eggs are present. A confirmatory diagnosis requires exploratory laparotomy and histopathologic examination of biopsy samples. Lymphomatosis is suggested by cauliflower-like growths of the ovary in domestic fowl.¹¹⁹ A variety of other tumor types have been reported including adenocarcinomas, leiomyomas, leiomyosarcomas, adenomas¹¹⁹ and granulosa cell tumors. Excisional surgery is the traditional therapy, although prognosis for long-term recovery is poor.

■ Ectopic Eggs and Non-septic Peritonitis

Egg material may gain access to the abdomen through ectopic ovulation and discontinuous or ruptured oviducts (Color 29.25). Ectopic ovulation occurs when the infundibulum fails to engulf an ovum. It

may be caused by reverse peristalsis of the oviduct, which occurs during normal egg laying, or by trauma to the oviduct that interferes with normal function.¹⁴¹ Restraining or stressing a hen during egg laying has been incriminated as a cause of ectopic ovulation.¹³² Ectopic ovulation is thought to occur frequently, and in one study it was the most common reproductive disorder (28.6%) described in necropsy specimens from nine avian orders.⁸⁴ Peritonitis may or may not develop from ectopic ovulation. If present, it can occur in either a septic or non-septic form. Yolk itself only causes a mild histiocytic response and if free of pathogens will gradually be reabsorbed by the peritoneum.¹⁰²

Depending on the location of rents in the oviduct, completely or partially shelled eggs may be deposited in the abdomen. Ruptured oviducts can result from acute and chronic oviduct impaction, including egg binding, cystic hyperplasia, neoplasia and salpingitis. Large, misshapen eggs may cause uterine disintegration and rupture resulting in ectopic eggs.¹⁸²

Ectopic eggs have been reported in Passeriformes and Psittaciformes.¹⁴¹ Uncomplicated ectopic ovulation may go unnoticed for a protracted period of time. Abdominal distension, a penguin-like stance and weight loss may be the only clinical changes. Free yolk in the abdomen may be absorbed and systemic antibiotics may be needed until the abdomen clears itself of yolk. The condition may recur if predisposing factors are still present. Excessive accumulations of egg material or fully formed eggs should be removed surgically. Damaged oviduct tissue should be repaired or removed.

Cockatiel hens with a history of egg laying frequently present with gradual weight loss, intermittent depression and ascites. If the abdominal fluid is sterile (rules out septic peritonitis), these birds will frequently respond to therapy that includes dexamethasone and medroxyprogesterone acetate.¹⁴⁰ The dose of medroxyprogesterone varies with the size of the bird (150 g [0.05 mg/g]; 150 to 300 g [0.04 mg/g]; 300-700 g [0.03 mg/g]; 700 g [0.025 mg/g]; Umbrella Cockatoo [0.018 mg/g]). Some hens may not cease egg laying activity with the administration of medroxyprogesterone alone and require administration of testosterone as well (Harrison GJ, unpublished). Scientific investigations are necessary to determine the pathogenesis of ascites in these hens and what role the empirically derived therapeutic regime plays in resolving this problem.

■ Egg-related Septic Peritonitis

Peritonitis is the most frequent cause of death associated with reproductive disorders.⁸⁴ It may not be a single disease but part of several syndromes, including ectopic ovulation, ruptured oviducts and salpingitis. It is theorized that it may be the cause instead of the result of a ruptured oviduct.¹⁴ It is uncertain which component of the egg is most important in inducing peritonitis but it is likely to be the yolk that is secondarily contaminated with bacteria. Frequently, hens that have been hysterectomized behave as if they have ovulated but do not develop egg-related peritonitis. Experimentally, egg yolks from other hens can be placed near the infundibulum of a laying hen and the yolk will be delivered normally. Peritonitis was never induced by the yolk (Ringer RK, unpublished). In another study, 87% of hens with ectopic ovulation also had egg-related peritonitis (Color 29.28).⁸⁴ In domestic fowl, fatal peritonitis can occur alone.¹²⁹ The peritoneum is commonly congested and edematous and appears lusterless; adhesions may be present. Peritonitis appears to be described most frequently in cockatiels, budgerigars, lovebirds, ducks and macaws.¹⁴¹

Presenting clinical signs include sudden death, abdominal swelling, respiratory distress, depression, anorexia and cessation of reproduction. The hemogram may show a severe inflammatory response. Radiology, abdominocentesis and laparotomy are helpful diagnostic aids. Septic peritonitis leading to severe debilitation, sepsis and death can occur if the yolk is contaminated with bacteria. Egg yolk in the peritoneal cavity is thought to be a predisposing factor to septic peritonitis.¹⁵⁵ Turbid yellow, green or brown yolky fluid or cheese-like yellowish masses of inspissated yolk material in the abdomen are indications of ectopic ovulation or a ruptured oviduct. Peritonitis may lead to secondary infection of other abdominal organs, and in advanced cases, extensive adhesions may form in the abdomen. Egg-related pancreatitis may cause temporary diabetes mellitus, especially in cockatiels.¹⁴¹ A temporary stroke-like syndrome has been described in cockatiels with yolk peritonitis, possibly due to yolk emboli. Aspirin may be used as an anticoagulant in cases where yolk emboli are suspected (1 tablet/30 cc water, 0.5 cc/kg PO TID). The etiologic agent of egg-related peritonitis is often coliforms, especially *E. coli*.¹²⁹ In cockatiels, *Yersinia pseudotuberculosis* and *Staphylococcus* spp. have also been reported in association with egg-related peritonitis.⁸⁴

Treatment consists of antibiotics, steroids to reduce inflammation and supportive care (heat, fluids, nutritional supplements). Long-term antibiotic therapy may be necessary and diet correction is advised. Most cases resolve with medical therapy alone, but early diagnosis is essential. If surgery is required to remove egg material or perform abdominal irrigation, the patient should be stabilized first with supportive care and antibiotics.

■ Chronic Egg Laying

Chronic egg laying occurs when a hen lays eggs beyond the normal clutch size or has repeated clutches regardless of the existence of a suitable mate or breeding season. Humans, inanimate objects (stuffed animals, enclosure toys) or birds of another species may act as substitute mates and stimulate excessive egg laying. This problem is particularly common in hand-raised hens that are imprinted on humans. The chronically reproductively active female may exhibit weight loss from constant regurgitation and feather loss or mild dermatitis around the cloaca in association with masturbatory behavior. Removing eggs from the hen effectively induces a form of double clutching and can facilitate the problem. The continuation of egg laying is ultimately hormonally controlled. The most domesticated psittacine birds, cockatiels, lovebirds and budgerigars, are notorious chronic egg layers. Perhaps the high incidence of problems in these species indicates a lack of hormonal balance in controlling egg laying that has occurred due to selective pressures designed to make birds produce continually in a variety of environmental situations.

Hens on a completely nutritious diet can continuously lay eggs for years without deleterious effects. In most cases, however, malnutrition and the progressive stress and physiologic demands of egg laying ultimately will compromise the hen. Calcium deficits lead to abnormal egg production, reduced oviduct inertia and generalized muscular weakness. Egg binding is common in hens that chronically lay eggs. Behavior modification can be attempted to stop the laying cycle (see Chapter 4). The stronger the environmental stimulus to cease egg laying activity, the better. Diminishing exposure to light to only eight to ten hours a day should interrupt the hormonal cycle, and egg laying should cease. Objects stimulating masturbatory behavior or sexually oriented regurgitation should be removed, although many birds will continue reproductive behavior despite this environmental change. Nest boxes and possibly

enclosure mates should be removed. Changing the location of the enclosure may also be helpful. Owners may discourage reproductive behavior by decreasing the amount of time spent with a hen until egg laying ceases.

Medical therapy is designed to correct any nutritional imbalances or reproductive tract abnormalities. Mineral and vitamin supplements should be given parenterally and added to the diet. Caloric intake with adequate protein levels should be increased. Medroxyprogesterone injections can be used to interrupt the ovulatory cycle. Depression, polyuria, weight gain, liver damage, immunosuppression and occasionally diabetes mellitus (especially in cockatiels) can occur with use of medroxyprogesterone. Egg laying may be stopped from two weeks to several months following therapy and repeat injections are often necessary. Some patients experience no problems, while others experience continual or permanent side effects. Dangerous spikes in drug concentrations can be prevented by implanting a progesterone pellet that allows for continual drug delivery.⁶¹ Despite behavioral and medical therapy, affected hens may continue to lay eggs. The long-term solution in these cases is a salpingohysterectomy (see Chapter 41).

■ Over-production

Maximal safe levels of egg production and chick care have not been determined for companion bird species. Dietary and environmental conditions in different aviaries would be a factor in determining safe production levels. Free-ranging psittacine hens may produce only one, at the most two, clutches per year. Egg production in excess of two clutches a year would thus be considered unnatural. Many captive psittacine birds (particularly Blue and Gold Macaws, cockatoos and Eclectus Parrots) routinely produce four clutches of eggs per year with no apparent side effects; however, continued levels of unnatural clutch production may lead to reproductive tract disease or other disorders precipitated by poor body condition. Over-producing hens may be thin and in poor feather condition, have poor muscular tone and be unable to quickly involute the uterus after egg laying has stopped. To ensure the long-term health of a reproductively active hen, egg production should be limited to two clutches a year in birds exhibiting medical problems secondary to excessive egg production.

■ Anatomic Abnormalities

Congenital atresia of the oviduct has been described in domestic fowl and is one of the causes of egg-related peritonitis. Oviduct discontinuity can occur due to degeneration of part of the Muellerian duct during embryonic development.

Normally, only the left ovary is present in birds, except in Falconiformes and Kiwis,⁸⁶ where both ovaries are frequently present.⁸⁴ Persistence of the right ovary has been reported in corvidae, ducks, swans, a Funereal Cockatoo, owls and grouse without mention of the presence of a right oviduct.^{84,86} The author has repeatedly seen right ovaries in young macaws and cockatoos and once in a mature Golden Conure, all without evidence of a functional right oviduct. In these birds, the right ovary appeared vestigial as has been reported in other cockatoos and owls.⁸⁴ A right ovary and oviduct were present at necropsy in a mature Scarlet Macaw that died from complications of egg impaction in the left oviduct (Color 29.20) (Ritchie BW, unpublished). In domestic fowl, about 90% of enlarged right ovaries are the result of damage to the left ovary. Occasionally, fertile eggs can result from ovulatory activity from the right ovary.⁸⁶

All early embryos have bilateral oviducts. After the first trimester of incubation, growth of the right oviduct appears to be inhibited. Persistent right oviducts without right ovaries (although they could have been rudimentary and overlooked) have been described in penguins and budgerigars.^{72,84} Right oviducts, occasionally paired with right ovaries and oviductal orifices, have been reported in domestic fowl.¹⁵¹ The incidence is highest in inbred strains, probably due to hormonal imbalances or abnormal genes that affect growth and differentiation of the Muellerian duct.¹¹³ Functional right oviducts have been reported in the domestic fowl, occasionally associated with cystic changes.^{86,119,127,151} A fully developed cystic right oviduct has also been observed in a budgerigar. Some domestic fowl with bilateral reproductive tracts can lay two eggs a day, and this unusual condition has also been reported in a duck. A functional-appearing right oviduct that is not altering the health of a companion bird can remain intact.

■ Abnormal Eggs

Dietary problems, environmental factors and reproductive tract abnormalities can all result in the production of abnormal eggs. Soft-shelled eggs may be an incidental occurrence or may indicate an underlying

ing nutritional or medical disorder. Nutritional deficiencies of calcium and vitamins A and D₃ have been associated with soft-shelled eggs. Therapy consists of both parenteral and oral nutritional supplements.

Oviduct pathology may also cause abnormal egg production. Suggestive abnormalities include thin-shelled eggs, irregular external calcium deposits, or overly thick-shelled eggs. Uterine infections may cause rough-shelled eggs, which can be corrected by flushing the uterus with appropriate antibiotics (Harrison GJ, unpublished). Organochloride pesticides (DDE) cause egg shell thinning by inhibiting deposition of calcium by the shell gland.^{30,126,131} Contaminated free-ranging birds in captive breeding projects may have reproductive abnormalities for many years due to the residual activity of these poisons and the long-term storage of these chemicals in body fat.

Domestically bred birds may be exposed to chemical toxins that may cause abnormal eggs through contaminated feed and agricultural spraying. In fact, it is legal to divert contaminated feeds from human food production into food used for animals, directly or by dilution, if the process of manufacturing or cooking will deplete the pesticide to "safe" levels.⁵⁰

Metritis, ectopic ovulation and ovarian disease may cause yolkless, small or sterile eggs that appear grossly normal. Inconsistent transient times of the egg passing through the oviduct may cause abnormally sized eggs due to deposition of differing amounts of albumen. A slow passage time of a preceding egg may allow for double ovulation to occur and result in a double-yolked egg.⁶² This occurs with some frequency in domestic fowl but has also been observed in psittacine birds. The problem is usually self-limiting. Hatchability is decreased in moderately abnormal eggs, but these eggs may still produce normal chicks.

Blood clots (meat spots) are described in poultry eggs in association with hemorrhage at ovulation or at other locations in the reproductive tract (Color 29.2). Shell color, yolk color and the odor of an egg can be influenced by diet, hereditary factors and microbial contamination. Some drugs and environmental toxins may cause abnormal egg production, resulting in early embryonic death or weak chicks. Examples include crude oil, exhaust fumes, nicotine, chlorinated hydrocarbons and certain antibiotics (furazolidone).¹²¹

Male Reproductive Disorders

Toxins

Numerous toxins can affect spermatogenesis in mammals. Reduced spermatogenesis has been reported in Japanese quail exposed to mercury.¹⁷⁴ Copper fungicides in feed have been found to suppress spermatogenesis and induce testicular atrophy.¹⁴⁸ Cystic testicular degeneration occurs in ducklings given feed with furazolidone.¹⁸⁵

Anatomical Abnormalities

Cyclic seasonal testicular atrophy occurs in many species (up to 500-fold in Passeriformes). Following the correct environmental stimuli, the testicles can undergo hypertrophy in preparation for breeding. Because of the seasonal change in testicular size, pathologic cases of atrophy can be difficult to diagnose (see Color 25). Serial laparotomies may be indicated to evaluate changes in testicular size. Testicular atrophy can be caused by orchitis as a result of trauma or genital infection or can be due to progressive infertility. Malnutrition, toxicity or bacteremia may also cause testicular degeneration. Affected birds may demonstrate a lack of libido or be infertile. Therapy is limited to addressing infectious or behavioral problems. If fibrotic or infiltrative changes have occurred, spermatogenesis may be permanently altered.

Testes can be abnormally joined at their anterior ends, which may not prevent spermatogenesis.¹¹⁹ Testicular hypoplasia may be attributable to congenital or hereditary conditions. Similarly, true agenesis may occur, causing parts of the genital tract to be absent (monorchidism).

Orchitis

A variety of bacteria can cause orchitis in birds, including *E. coli*, *Salmonella* spp. and *Pasteurella multocida*.¹³² Infections may originate from prolapsed or ulcerated phalli, renal obstruction, cloacitis and septicemia. Clinical signs are similar to those expected for any generalized infection. Antibiotics may be helpful in resolving the active infection but may not prevent or reverse infertility.

Neoplasia

Testicular tumors commonly occur in older budgerigars but can also be found in larger Psittaciformes and other birds.¹⁴ A seminoma of the testes was reported in a Collared Turtle Dove¹³³ with progressive emaciation for several months before death. The author diagnosed a seminoma in an Eclectus Parrot that died suddenly with an enlarged left testicle that occupied much of the left peritoneal cavity (see Figure 29.5). The surface of the testicle was smooth, which is typical of primary tumors. This bird had a long history of aggressive behavior (atypical for the species) toward numerous hens. It was theorized that hormonal imbalances associated with this tumor were responsible for the behavioral changes.

Sertoli and interstitial cell tumors have been described in birds (see Chapter 25).⁴⁷ Lymphoproliferative diseases, such as leukosis, can also affect the testes resulting in infertility.⁹⁹ Regardless of the tumor type, testicular neoplasias can involve one or both testes. Unilateral paresis, progressive weight loss and abdominal enlargement are typical clinical signs. Affected birds may have reduced secondary sex characteristics and become more feminine in nature (cere of the male budgerigar turning from a blue to brown color). Metastasis from testicular tumors usually affects the liver.¹⁴ Surgery may be successful if the tumor is easily approached and unilateral and the cock is in good health. Long-term prognosis is guarded due to the possibility of metastasis.

Phallic Prolapse

Birds with large phalli may develop partial or complete prolapses, which are frequently secondary to trauma, infection or extreme weather fluctuations.⁶⁹ Infections may be secondary to mucosal irritation (over-exuberant mating or vent sexing) or fecal contamination. The phallus may become enlarged and ulcerated. In geese, *Neisseria* spp. have been isolated from erosions of the phallus, oviduct and cloaca and are believed to be sexually transmitted.¹¹⁸ Occasionally, a prolapsed phallus will revert preventing evaluation of secondary infections. In severe cases of phallic prolapse, the birds may be severely depressed, anorectic and disinterested in copulation. Permanent infertility is a common sequela. In ostriches, frostbite and necrotizing dermatitis may occur secondary to a prolapse.

Exposed tissue should be thoroughly cleaned with a sterile saline solution, carefully debrided and cov-



FIG 29.14 Environmental factors that would make a pair of birds feel insecure should always be considered when evaluating a lack of breeding success. The design and location of the nest box, enclosure and aviary are all critical factors to consider. This male Sulphur-crested Cockatoo with fertile eggs was eliciting proper nest box defense behavior in response to an intruder.

ered with antibiotic cream. Topical DMSO may help reduce swelling, making replacement of the phallus easier and more permanent. Daily therapy and cloacal mattress suture may be necessary to prevent recurring prolapses (see Chapter 41). Systemic antibiotics should be considered due to the possibility of ascending urogenital infections. If large areas of necrosis are present, then surgery is necessary to debride the wound. Nodular ejaculatory papillae have been described as abnormal in pigeons.¹¹

Behavioral Abnormalities

Reproductively active males and females (particularly budgerigars and cockatiels) may exhibit masturbatory behavior or excessive regurgitation. These are normal reproductive behaviors that may become pathologic in birds that are isolated. Cockatiel cocks incubate the eggs, and a single male may spend much of the time on the enclosure floor mimicking incubation activities. Removing the bird from its enclosure for long periods of time (with available food and water sources) or changing the enclosure or enclosure location may stop this behavior. In other cases, displaced reproductive behavior may occur in males

housed with females. In these cases, the male is often “imprinted” on humans and cannot complete the reproductive cycle with its own species. Exchanging mates may prove helpful, but usually these males should be removed from the breeding program. Human imprinting can also occur in females, and in both genders behavioral abnormalities due to improper imprinting may not be obvious. Indeed, lack of pair-bonding, lack of egg production or infertility may be the only signs associated with the use of hand-raised imprinted birds in a breeding program. The interaction of a chick with its parents and nesting conditions may be critical for successful reproduction in some species (see Chapter 4).

Under-production

Establishing the existing level of production is the first step to managing a breeding pair. This includes calculating levels of fertility, hatchability and chick fledging rates. Average clutch sizes have been reported for many species of birds;^{4,43} unfortunately, clutch size can vary among individuals and within genera. Under-production is particularly important with endangered species where maximum production is critical to ensure the survival of the species.

If production from a breeding pair does not approach the average, then medical, physical or behavior problems should be addressed. Correction of any medical or physical abnormalities, such as clipping overgrown feathers near the cloaca, dieting overweight birds or treating birds for localized infections can be instituted. The diet should be carefully analyzed and any deficiencies should be corrected. Environmental deterrents to breeding may be determined by using a video camera to observe the pair’s daily behavioral patterns. Some changes that may be necessary include re-pairing birds, improved enclosures, different nest boxes, varied diet, altered climate, different lighting or reducing aviary disturbances induced by humans, other birds or vermin (Figure 29.14).

Endocrine manipulation for improving reproductive success in birds has been studied with marginal success. The dependence of the avian endocrine system on environmental stimuli makes clinical manipulation of the avian reproductive system difficult. Specific behavioral manifestations of endocrine abnormalities have been treated medically, such as testosterone injections in timid male *Electus* Parrots, but such therapy is experimental in nature and long-term use can cause testicular atrophy.

Testosterone has been suggested to induce singing in male canaries but can cause serious side effects. Male canaries that do not sing are usually sick or malnourished. PMSG (gonadotrophin serum) administered to canary hens induced defeathering and vascularity of the brood patch and accelerated reproductive tract development with accompanying increased incidences of egg binding and oviposition on the enclosure floor.¹⁶⁴ In another study, PMSG administration stimulated reproductive activity in birds maintained in both long and short daylight cycles.¹⁶³

Birds that consistently fail to produce should be removed from the breeding program.

Artificial Insemination

Artificial insemination (AI) has been used successfully in cranes, cassowaries, raptors, gamebirds, Anseriformes, Columbiformes, Galliformes and some psittacine species, including budgerigars, a cockatiel and Hispaniolan Amazon Parrots.^{45,60,146} This technique may be beneficial in endangered species, especially when a limited number of individuals of a species is available. Problems with incompatible pairs, poor fertility due to physical or behavioral difficulties and large distances between individuals housed at different breeding facilities may be resolved using AI.

Collection Technique

Successful collection techniques include cooperation, massage and electroejaculation. Cooperative AI requires human-imprinted males that are encouraged to copulate and deposit semen in or on a suitable receptacle.¹⁶ This technique is extensively used in raptor breeding.^{173,184} Using imprinted birds requires a tremendous amount of time and effort and may not be possible in all species.¹⁷ Semen may also be collected during or after natural copulation or through the use of dummy mounting devices and artificial vaginas.¹¹ Semen collected with these techniques is usually free from contamination.

The massage collection technique requires two people, one to restrain the bird and the other to collect the semen. The inner thigh, ventral abdomen, tail, vent and synsacral area are stroked. The tail is posi-

tioned dorsally and with continued stroking pressure is placed laterally on the cloaca to encourage ejaculation. Even without ejaculation, semen can be expressed from the cloacal region with this technique. Contaminated semen samples often contain feces and urine and should be discarded. Contamination can be reduced by fasting the birds and evacuating the cloaca before semen collection. Visualization of semen volumes in small Psittaciformes can be augmented using a strong light source. Massage has been used successfully in poultry, Casuariiformes, Anseriformes, Columbiformes, Falconiformes, Gruiformes, Passeriformes and Psittaciformes including budgerigars, Hispaniolan Amazon Parrots, Maroon-bellied Conures and Monk Parakeets.^{11,21,45,78,147}

Five to fifteen μ l of semen could be artificially collected from budgerigars twice weekly using a modified massage technique where the semen is collected by simply applying pressure on both sides of the cloaca to empty the contents of the seminal glomus. Hispaniolan Amazon Parrot males were collected three times a week during the months corresponding with the natural breeding season although semen yields remained high in one individual for several months after the normal breeding season. In Hispaniolan Amazon Parrots, paired and unpaired males yielded statistically similar semen volumes, sperm motility and sperm concentrations. Collection of semen from males paired with reproductively active hens did not affect their breeding performance.

Electroejaculation under anesthesia is not a common method for collecting avian semen although it has been successfully used with a variety of birds including waterfowl, domestic fowl,⁹¹ psittacine birds⁵⁸ and pigeons.¹¹ Electroejaculation may cause semen samples to be expelled with feces and urine.¹¹ This technique, however, can be attempted if massage and cooperative techniques fail. Electroejaculation was successful in 95% of mature pigeons when used in conjunction with a cloacal retractor.¹⁰ A combination of massage and electrostimulation was used to collect semen from macaws, cockatiels and Amazon parrots.⁵⁸

The volume and sperm concentration of semen varies among species (see Table 29.3).⁴⁵ Semen volume can be increased by multiple collections in a week. Excessively frequent collection can cause cloacal irritation, swelling and hyperemia of the vent. Adding diluent to the semen may help preserve sperm viability in low-volume samples.⁴⁵ Semen should be evaluated before and after any storage or preservation and before insemination. The phallus, anal gland secre-

tions, urine, feces and transparent fluid can all interfere with semen collection and viability.

Higher fertility levels are achieved when semen is used for insemination immediately following collection. Uncontaminated semen in chickens can be stored for 1.5 days and still result in 37% fertility.⁹⁵ Short-term storage requires temperatures near freezing and protection from drying and contamination. Diluents should be used when semen is stored for more than one hour. Commercially available poultry diluents have been used successfully in non-domestic species.^{21,78} Long-term semen storage requires cryopreservation using either glycerol or dimethylsulfoxide (DMSO) as a cryoprotectant. Glycerol must be removed prior to insemination using dialysis. Frozen-thawed semen has proven to be viable in Sandhill Cranes, American Kestrels, Peregrine Falcons and budgerigars.^{20,124,146,152} Modifications in cryopreservation methods and fluids may be necessary when handling semen of other avian species.

Insemination Technique

Cooperative techniques may be used to inseminate females that are encouraged to respond to handlers allowing semen to be deposited in the cloaca or oviduct.¹⁸⁴ Massage techniques involve manual stimulation, eversion of the cloaca and placement of the semen via a tube, straw or catheter through the cloaca into the vagina. Anatomical structures can be better visualized using speculums and specially designed cloacal retractors.¹⁰ Deep vaginal insemination results in the best fertility levels; however, with frequent and timely inseminations fertile eggs can occur when semen is deposited in the cloaca.

Fertile eggs were produced by budgerigar hens using five to ten μ l of fresh semen or 40 μ l of frozen-thawed dialyzed semen placed by cannula into the oviduct two hours post-oviposition. Determining when to inseminate is crucial for fertilization to occur. It is best to inseminate every other day after the first egg in a clutch is laid or after each egg is laid.¹⁸⁴ The frequency of insemination is governed by the species, sperm concentration, durability of the sperm, method of insemination and ovulation patterns. Hispaniolan Amazon Parrots were successfully inseminated before egg laying started but fertile eggs were not laid until insemination procedures were discontinued.

Initiating egg laying usually requires behavioral stimulation provided by the presence of a mate. Female budgerigars were successfully inseminated only

after they began laying when stimulated by the presence of a vasectomized male.¹⁴⁶ Vasectomized cocks show normal male sexual behavior, including courtship and copulation. Hispaniolan Amazon Parrot hens begin egg laying when housed separately but adjacent to males.

Non-disease Factors Affecting Reproduction

Gender

The most common cause of reproductive failure in companion birds is pairing of two birds of the same gender. Several techniques for determining the gender of birds have been described. The appropriate method to use depends on the species, age of the bird and the information to be derived from the procedure.

Physical Characteristics

Many species of birds are sexually dimorphic, with visual characteristics that distinguish males from females. The degree of dimorphism varies with species and may not always be obvious. Even with monomorphic species, subtle differences may exist that allow determination of gender. With most monomorphic species, definitive gender differentiation requires laboratory or laparoscopic procedures.

With birds of prey, the female is generally 30% larger than the male, although some size overlap occurs in the intermediate weight ranges.¹⁸⁸ In other groups of birds, the male is generally heavier and has a larger frame than the female. Head size as well as bill breadth, length and depth are often greater in males. Differences in beak size are usually obvious in toucans but may require calipers to appreciate in smaller species. The majority of psittacine birds are monomorphic although there are many exceptions (Table 29.4).

In dimorphic birds, feather color, iris color and bill characteristics typically differentiate hens from cocks.^{43,64,101,158,167} These secondary sexual characteristics become more obvious as birds reach reproductive maturity. Immature birds typically have color patterns similar to adult females. Feather shape and length may also be different. For instance, male Racket-tailed Parrots have much longer central

retrices than females, and male Princess Parrots have a spatula extension at the end of the ninth primary feather. In general, red to brown iris color is more common in female cockatoos; however, this technique is not always reliable, especially in Moluccan, Rose-breasted, Bare-eyed, Goffin and immature cockatoos. At maturity, wild-type (green) male budgerigars have lavender to dark-blue ceres while females have light-blue to tan or brown ceres. Gender determination based on cere color may not be effective in inbred color mutations. The White-fronted Amazon is clearly sexually dimorphic. Males have numerous red secondary wing coverts while females have few to none.

Vent Sexing

Gender can be determined in most Galliformes, Anseriformes, some game birds, ratites and some species of Cracidae by looking for the phallus on the wall of the cloaca. In Columbiformes and Passeriformes, which have prominent papillae of the ductus deferens, gender can be determined if these structures can be visualized using general anesthesia and a cloacal protractor.¹⁰ The clitoris is located at the same location as the phallus, and differentiating the phallus from the clitoris can be difficult in chicks and immature cocks (see Chapters 46 and 48).

Determining the distance between the pelvic bones (gapped in females, close together in males) has been discussed as a method of gender determination. The distance between the pelvic bones increases in post-ovipositional females but may narrow considerably in the months following oviposition. In larger psittacine birds, this is an unreliable method of gender determination. Some practitioners feel that this is a reliable method for gender determination in mature lovebirds.

Behavioral Characteristics

Behavioral characteristics generally vary with gender; however, birds can develop homosexual pair bonds with one bird behaving more like a hen than the other. Males are generally more aggressive and are responsible for territorial defense. The songs of the male finch, canary and cockatiel differentiate them from females. With some free-ranging psittacine species, the hen incubates while the male is perched nearby as a sentry. Depending on the cycle of incubation and the age of chicks, the hen may join the male in inter- and intraspecific territorial confrontations. In cockatiels and some other species of psittacines, the male shares in incubation duties.

Observing copulation in species in which the male completely mounts the female may indicate a successful pair bonding. Complete mounting is typical of raptors, waterfowl and Passeriformes. In New World Psittaciformes, copulation occurs side by side, and homosexual pairs have been observed precisely mimicking this procedure. The male usually places one foot on the caudal tail region of the female and has the more dorsally placed tail during cloacal contact. It is theorized that females exhibiting masturbatory or courtship behavior with inanimate objects, other species of birds or humans will often lay eggs. If no eggs are produced, it may indicate that the bird is a cock, but some masturbating hens have been known to wait twelve years after the onset of this behavior before laying an egg.

Laparoscopic Sexing

Although subject to error when used in young birds with undifferentiated gonads, laparoscopic examination is a definitive method of gender determination when performed by an experienced practitioner (see Chapter 13). Its major advantage over other gender determination techniques is that it allows for direct inspection of abdominal structures, especially reproductive organs, for evidence of disease or dysfunction. Its disadvantage is that it is an invasive procedure that requires anesthesia.

Laboratory Methods

Genetic determination of gender in birds is considered the most reliable of the available noninvasive techniques. One method employs feather pulp as a source of chromosomes. After culturing, staining and careful examination, the gender chromosomes can be identified in most species. The disadvantage of this technique is the difficulty of collecting an adequate number of growing feathers that will produce a viable culture that is not contaminated. Other problems include overnight mailing and a lag time in obtaining results, as only one laboratory^a offers this service commercially in the United States.

Determination of gender can also be accomplished by evaluating differences in the DNA composition between males and females. A small volume of red blood cells is necessary for this procedure, and advantages include easy and relatively non-traumatic sample collection and a long sample shelf life without refrigeration. Lag time in obtaining results is a disadvantage, as only one laboratory^b offers this service in the United States.

TABLE 29.4 Examples of Sexual Dimorphic Differences in Psittacines*

Species	Male	Female
Budgerigar	Cere lavender to dark-blue	Cere pink-brown to light-blue
Cockatiel	Lacks barring, bright yellow face and cheek patch	Tail and flight feathers have barring on underside
Malee Ring-necked Parrot, Adelaide Rosella, Red-rumped Parrot, Bourke's Parrot	Lacks barring when mature	Barring on underside of wings
Red-rumped Parrot	Red rump patch	Green rump patch, paler
Scarlet-chested Parrot	Scarlet breast	Green breast
Regent Parrot, Superb Parrot	Yellow feather patches	Green throughout
Princess Parrot	Spatular extension on the end of the ninth primary flight feather, coral beak	Lacks spatular extension, brownish beak
Red-bellied Parrot	Red belly	Scattered red feathers on green belly
King Parrot	Red head feathers	Green head feathers
Red-winged Parrot	Blue crown feathers, bright yellow-green underparts, black shoulder mantle, coral bill, orange-red iris	Duller green feathers, pale-brown iris
Duchess Lorikeet	Red side rump patch	Yellow side rump patch
Fairy Lorikeet	Blue rump patch	Yellow side rump patch
Josephine's Lory	Red lower back	Green lower back
Whiskered Lory	Red face-crown and crown	Green face-crown and crown
<i>Cacatua</i> spp.	Brown to black iris	Red to reddish-brown iris
Eclectus Parrot	Green	Red and purple
Pesquet's Parrot	Red spot behind eye	No red spot
Grey-headed Lovebird	Grey head, neck and breast	Green head, neck and breast
Ring-necked Parakeet	Colored ring around the neck	No ring
Mueller's Parrot	Red beak	White beak
White-fronted Amazon Parrot	Red alula and primary covert feathers	Slight to no red in primary covert feathers
Mountain Parakeet	Yellow forehead, lores, cheeks and throat	Green forehead, lores, cheeks and throat
Mexican Parrotlet	Blue lower rump and underwing coverts	Yellow-green lower rump and underwing coverts
Pileated Parrot	Red forehead	Green forehead
African Grey Parrot	Red vent and rump feathers	Grey tips on red feathers or mostly grey

* Not all members within a genus will portray the listed sexual differences. One representative species is listed for each genus. Differences are usually observable only in mature species.)

Fecal steroid assays have been used to determine gender in birds.⁸ Steroid hormone levels including estrogens and androgens (mostly testosterone) are measured by radioimmunoassay, and an estrogen/testosterone ratio is used to estimate the sex of the bird. An individual bird's production of steroid hormones varies with age and sexual activity and leads to some overlap in the estrogen/testosterone ratios. Commercial laboratories^c claim high accuracy, although no blind studies have been performed to validate the test. One recent report suggests that fecal sex steroid determination in most parrots is not effective, especially in small birds.⁷ This technique of gender determination requires that a bird be sexually mature.

Sexual Maturity

In birds, the female is the heterogametic gender (ZW), and the gender of the embryo is determined prior to ovulation and not at fertilization as occurs in mammals. Males are homogametic (ZZ). Sex differentiation occurs in the developing embryo during the first trimester of development. Secondary sexual characteristics under hormonal control may be obvious before functional sexual maturity is achieved.

The age of sexual maturity varies greatly between species. For example, Zebra Finches and captive Japanese Quail are sexually active by two months of age. By comparison, the Fumar begins breeding at about eight years of age.⁸⁶ Many smaller Passeriformes begin breeding in the first or second spring after hatching. In larger Psittaciformes (Amazon parrots, African Grey Parrots, large cockatoos and macaws),

viable eggs can be produced when the birds reach three to six years of age. Pionus and smaller cockatoos and macaws may be sexually mature by two to four years. Lories and lorikeets produce young at two to three years, conures at one and one-half to two years, and budgerigars, lovebirds and cockatiels at six months to one year of age.¹⁰⁷ Initial clutches of eggs may be infertile in young birds due to immaturity of reproductive tissues or reproductive inexperience.

Environment

Light

The most important factor for reproductive stimulation of free-ranging birds in mid to high latitudes is day length. Neuroendocrine systems control the annual development of the reproductive system with sufficient precision to assure that young will be produced when trophic resources are optimal. Lengthening photoperiods elevates LH secretion, which is the primary reproductive hormone. The effect of photoperiod on certain species of captive companion birds has been partially studied, and this information may be applicable to other related species.

Female budgerigars have been found to lay eggs when exposed to male vocalizations even when kept in continuous darkness.⁷¹ Male song appears to be a strong factor for successful oviposition, but a dark nest box is also important. Photoperiod can have a direct effect on male vocalizations, which in turn stimulates the female reproductive cycle. In poultry, the onset of sexual maturity is affected by lighting.⁴⁰

Climate

High environmental temperature combined with high humidity increases the physiological stress on birds and can decrease reproductive activity. An ambient temperature greater than 27°C induces a state of thermal stress in pullets⁶⁷ and decreases egg production, feed consumption and feed efficiency. Increases in humidity appear to have less effect on reproductive activity than increases in dry bulb temperature. Temperature extremes have been shown to decrease semen production and cause thinner egg shells. The use of misters in hot weather can alleviate this problem.

Rainfall triggers courtship behavior in male Zebra Finches. Juvenile finches may form pair bonds before the rainy season so that they are ready for breeding when rain occurs. Finches are thought to maintain testicular function throughout the year, making

rapid reproduction post-rainfall possible.¹⁷⁹ Cold temperatures inhibit Zebra Finch reproduction.

Season

Effects of environmental conditions on reproduction may be similar in captive birds and free-ranging birds; however, heavily domesticated species like canaries, budgerigars and finches react differently than their free-ranging conspecifics.¹⁷⁹ For larger Psittaciformes, insufficient information has been collected to make comparisons between the reproductive characteristics of free-ranging and captive birds. In Central America, free-ranging Yellow-naped Amazon Parrots have been found to produce eggs as early as December and can have eggs hatch as late as April. Conures in the same area have roughly the same cycle although in both species, the majority of eggs hatch in January (dry, cooler weather). Budgerigars in Australia breed throughout the year, but in each geological or ecological area only produce young for a set number of months each year.¹⁸⁹

Individual pairs of a particular species have the capacity to re-nest if necessary to account for yearly changes in environmental conditions as well as to adjust to geographic or ecologic changes in a region. This same adaptability may be expressed in captivity.

Egg production was monitored for one year at a large Psittaciforme breeding facility in California.¹⁶⁹ Amazon parrots were found to breed during a 17-week period from early March to early July. Blue and Gold Macaws had the longest breeding season, with eggs being produced year round, although egg numbers decreased in September and November. Rose-breasted Cockatoos had the shortest season, which lasted nine weeks from late winter to early spring. Amazon parrots bred at a time when the mean high temperature and humidity was highest (87.9°F and 44.4% RH), as opposed to the cockatoos, which bred with a mean high temperature of 79°F and RH of 27%. Blue and Gold Macaws bred during the broadest range of temperature and humidity.

The Amazon parrots quit producing when day length began to decrease and did not breed again until day length had increased considerably. Blue and Gold Macaws could breed in both increasing and decreasing light. Cockatoos produced only during periods of increasing day length. It could not be determined from this study if temperature, humidity, day length, a combination of all three or other unrecorded factors controlled reproduction. All three of the evaluated species originate from temperate climates and are

probably affected by other factors as well as day length. In Hyacinth Macaws in a Florida aviary, eggs are produced between May and October.

Mate Presence

In cockatiels, mate access is essential to ensure nesting behavior.¹⁵⁶ Increases in LH levels necessary for oviposition occurred only in females given full mate and nest box access. Visual, but not auditory, isolation of mates did not negatively affect cockatiel reproduction in one study.¹⁹⁰ Testicular development was found to be greater in starlings when males were housed with females. In other species, sufficient photoperiods will induce testicular development regardless of whether a female is present.¹⁷⁹

Separating a pair during the non-breeding season may not affect reproductive success in a subsequent season, and mates will usually reunite, especially if they have previously been reproductively successful. Mate retention has been found to be associated with greater reproductive success than mate replacement in a variety of species that naturally separate in the non-breeding season.

Orange-winged Amazon Parrots in one study were stimulated to breed by separating them from their self-selected mates for three months and then placing them into enclosures with nest boxes that permitted them to “chew” an entrance hole into the nest box.¹¹¹ In addition, these “enriched pairs” were given fruit instead of a complete crumble diet and were exposed to water misters on alternate days. The reproductive success in these birds was better than in a control group, but no one factor could be identified as being responsible for the increased egg production.

Mate Selection

In some monogamous birds, such as California Quail and Turtle Doves, forced pairing of mates can result in successful breeding. In other species like cockatiels, forced pairing was found to result in decreased reproductive activity.¹⁹⁰ In some species, force-pairing may result in increased mate aggression.

Specific mate characteristics may affect mate acceptance and the strength of the pair bond. Rather startling is the fact that leg band color is important in determining mate preference in finches. This is just one indication that color of feathers and beaks, which often changes seasonally, can be an important reproductive stimulant. External physical characteristics of birds can be dependent on health and nutrition as well as environmental cues that influence the pro-

duction of hormones responsible for secondary sex characteristics.

Aggressive mates can inhibit reproduction by preventing the opposite sex from eating or through direct physical abuse. Aggressive behavior is most noteworthy in cockatoos and is seen occasionally in Eclectus Parrots. Male cockatoos, even in long-term successful pairs, may suddenly attack and sometimes kill their mates. The beak, eyes, skull, feet and cloaca are most commonly traumatized.

Old World Psittaciforme males completely mount the female during copulation, and male aggression may occur from failures in proper copulatory behavior. Evidence suggests that males become sexually active before hens, which may precipitate the aggressive behavior. If a free-ranging male cockatoo becomes aggressive, the hen is able to escape to prevent serious injury.

Lighting, food and the presence of other birds may induce aggressive behavior. As a solution, male cockatoo flight feathers are often clipped, and the nest box is provided with a lower “escape” hatch so that the female cannot be trapped within the nest box. Males should be introduced into the hen’s enclosure by being placed within a smaller enclosure. After mate acceptance, the male can be released into the female’s enclosure. In the case of Eclectus Parrots, females are more aggressive than males, although they rarely seriously injure a mate. Excessively aggressive males or timid females should be removed from the breeding program.

Mate Pair-bonding

Pair-bonding refers to the behavioral acceptance that exists between a compatible hen and cock and is evident in all successful pairs, although considerable species and individual variation exists. Strong territorial defense coordinated between the male and female, such as lunging at the front of the cage with upraised wings in macaws and tail-fanning with crown and nape feather ruffing in Amazon parrots, are examples of proper pair-bonding (Figure 29.15). Other behaviors include mutual preening (see Chapter 4), feeding, nest box inspection and copulation (see Color 8). Homosexual pairs may also bond and exhibit these same behaviors.

Evaluating a breeding pair through the aid of video recorders will help identify causes of behavior-induced infertility. Some copulatory efforts may be handicapped by physical, medical or behavioral abnormalities. Abbreviated copulatory efforts may nor-



FIG 29.15 Using a video camera to observe a pair of birds' daily behavior can help determine if an effective pair bond exists and may help identify environmental factors that are inhibiting breeding. These strongly pair-bonded Green-winged Macaws are displaying proper territorial defense behavior in response to an intruder.

mally precede successful copulation. Birds should be allowed to choose their own mates to increase the likelihood of pair-bonding.

Mate Vocalization

Male auditory signals stimulate female reproduction in several species. In turn, males will maintain spermatogenesis longer when paired with sexually active females. Budgerigars may be the only avian species in which an auditory stimulus promotes ovarian development and ovulation.²² Isolation of budgerigar pairs both visually and auditorially from other budgerigars will cause reduced reproductive behavior.²² A similar effect has been hypothesized in macaws, Amazon parrots, cockatoos and African Grey Parrots (Harrison GJ, unpublished).

Social Interaction

The presence of other breeding birds is a reproductive stimulus in highly social and colony breeding species. Social birds such as budgerigars should be housed within hearing, if not visual, range of the same species to stimulate successful reproduction.

Social behavior varies during the breeding season in some species. For example, some parrots will feed only in pairs during the breeding season while cocka-

toos will interact daily with other pairs and feed in larger groups. Free-ranging populations of Zebra Finches are highly social until courtship behavior begins.

In captivity, housing similar species near each other may reinforce the pair bond and strengthen endocrine controls by eliciting territorial defense behavior. In contrast, excessive territorial defense may waste energy and interfere with pair interactions that are critical for reproductive success. Monitoring of a pair's behavior and analysis of enclosure diagrams in multiple-pair and multiple-species aviaries will help define proper housing for each species and individual pair.

Human Interaction

Human interference can affect reproduction. Circulating levels of prolactin are easily altered by stressful events; stress is one method used to discourage incubation in egg-producing turkeys.³⁸ Aviary disturbances and handling birds near the breeding season may disrupt endocrine control of the reproductive cycle or disturb the birds so that mating is not initiated or completed, incubation is interrupted, eggs are damaged or chicks are cared for improperly. Successful territorial defense appears to have a positive effect on reproduction, and males that feel they have defended their nest from humans may be more reproductively active. Although evidence is rather anecdotal, barren pairs have been induced to breed by disturbing, handling or relocating the pairs. Some species of birds are withdrawn and display fear as opposed to aggression when approached by humans, indicating improper territorial defense. These pairs may be more productive once conditioned to human activity or if placed in an isolated, protected area. Annual physical exams can be performed on properly conditioned birds and do not appear to negatively affect reproduction.

Nests

Availability and acquisition of a proper nest site and nesting material may be a strong environmental stimulus for breeding particularly in male cockatiels and finches.^{77,110,179} In fact, LH surges that stimulate reproduction and spermatogenesis may be induced

because of male defense behavior and not because of the presence of a female. Starling males that did not defend their nest boxes were found to have similar LH levels to males without a nest box.⁵¹ The relationship of the perch to the nest box hole (perch ten cm below the hole) played a significant role in reproductive success in budgerigars.²³ This phenomenon may be applicable to considerations of nest box design in other species as well.

Birds are generally classified as being either determinate or indeterminate layers. Determinate layers will lay only a set number of eggs in a clutch regardless of whether any egg is removed or destroyed. Indeterminate layers will continue to lay until they “recognize” the correct number of eggs. Prolactin is released from the pituitary gland in response to the incubating bird’s physical contact with eggs in the nest. The concentration of prolactin, which is responsible for regression of the ovary and incubation behavior, was found to increase gradually in cockatiels that were incubating eggs.¹¹² These cockatiels were also found to be able to continue to lay additional eggs if previous eggs were removed from the nest. In turkeys, follicular atresia occurs when egg incubation starts.³⁸

Applying these principles to companion birds, it is logical that if birds are thought to be indeterminate layers, eggs should be removed before incubation starts if production of more eggs is desired. The longer incubation is allowed to proceed the more complete the ovarian regression would be, which would make a hen less likely to lay another clutch. Budgerigars are believed to be determinant layers.

Territory

In finches, males with a breeding territory had larger testes with a longer functional period than males that were photo-stimulated without a breeding territory.⁴² The presence of sexually active females may also affect the influence of breeding territory in stimulating male reproductive activity. The male’s reproductive condition appears to be more easily synchronized by environmental cues than does that of the female. Female reproductive performance appears to be more affected by captivity than does that of males. The presence of a sexually active male probably has a positive effect on females in many species, even though the effect may not be recognizable because of the lack of stimulation in the female from other cues.

Enclosures

Enclosure design can affect reproduction. Some species of birds appear to breed better in flights as opposed to flight enclosures. The actual dimensions may be important, but longer, wider and higher enclosures may not always be better, as a larger enclosure may represent a territory that a pair feels it cannot adequately defend. Enclosure design in general and nest position, including whether it is within the enclosure, how high it is and whether it is open or obscured by walls, may all influence a pair’s feeling of security (see Chapter 2).

Reproductive Experience

Previous reproductive activity may decrease the requirement for environmental cues to stimulate breeding. This has been shown to occur in budgerigars and is suspected in Orange-winged Amazon Parrots.¹⁸ Mate familiarity increases reproductive success in cockatiels,¹⁹⁰ and mate retention throughout successive breeding seasons has been correlated with greater reproductive success in monogamous birds. Mate familiarity may improve pair coordination, decrease aggression between mates and increase male reproductive behavior.³⁹

Hand-raising neonates may result in imprinting on humans or a lack of early environmental “learning,” which may affect future reproductive success. Imprinting often appears stronger in males than in females. Hand-raised cockatiel hens were more likely to lay eggs (and more of them) than parent-raised birds; however, eggs were often laid on the floor of the enclosure. Pairs with hand-raised male cockatiels were less likely to inspect nest boxes or produce fertile eggs.¹¹⁶ The behavioral deficits of hand-raising can be attenuated by successful and repeated breeding experience. Imprinting on the wrong species is common in birds and has been reported to occur when Rose-breasted Cockatoo chicks are foster-raised by Major Mitchell’s Cockatoos.¹⁴³ During fledging, chicks are thought to imprint on habitat, which will later control nest site selection.

Nutrition

Low dietary calcium levels (0.056% to 0.3%) have been shown to cause a complete cessation of egg laying in gallinaceous birds. Decreased energy intake causes decreased LH levels followed by ovarian atresia. Low-sodium diets also result in cessation of egg laying. Zinc-rich diets decrease feed intake and

may directly decrease egg laying by altering metabolism and endocrine functions.

Psittacine birds being fed largely seed diets should be expected to consume low levels of vitamin A, D₃ and E as well as other nutrients (see Chapters 3 and 31). Vitamin E deficiency can cause reduced spermatogenesis in domestic fowl and Coturnix Quail. Vitamins A and D₃ are needed for proper reproductive gland secretions and calcium metabolism, respectively. Over-nutrition may precipitate infertility by either mechanically blocking the cloaca or reducing successful ovulation. Abdominal fat and lack of condition may contribute to oviduct inertia and egg-laying problems. Amazon parrots, Scarlet Macaws and Rose-breasted Cockatoos are commonly obese and should be carefully monitored to prevent weight-related infertility. Fat can accumulate in pendulous folds near the cloaca and crop, often differentiating into lipomas (see Color 8). Subcutaneous fat deposits over the coxofemoral and flank regions are more subtle indications of excessive energy intake.

The availability of certain food items and not simply energy consumption may be one of the many stimulants needed to begin or strengthen reproductive activity. Aviculturists can mimic naturally occurring variations in food availability by reducing food intake and variability in the non-breeding season and then dramatically increasing the quality, quantity and variety of foods before the breeding season. The success of this method is equivocal but suggests the need for further study.

In White-crowned Sparrows, consumption of green wheat enhanced photo-stimulated ovarian growth. Testicular growth was not affected.⁴¹ It was not shown if the ovarian stimulatory effects of green leaves were ecologically significant or if specific substances in the green plants induced the change.

Aflatoxins and mycotoxins in feed have been shown to reduce egg production and cause infertility. Thiotepea, although mutagenic, teratogenic and carcinogenic in large doses, proved in one experiment to be a safe, effective temporary chemosterilant when fed to free-ranging male Red-winged Blackbirds.¹²⁸

Physical and Medical Characteristics

Adequate exercise is important to reproductive success and decreases the likelihood of reproductive disorders, such as egg-binding. Any physical abnormality or medical condition affecting mobility, balance,

the cloacal region or the reproductive tract can cause infertility or decreased reproductive success. Heavy cloacal feathering, such as in Rose-breasted Cockatoos and fancy pigeon breeds, may prevent copulation resulting in infertility. A female Golden Conure with an abscessed preen gland repeatedly attacked her mate when he attempted copulation. The pair laid fertile eggs three weeks after the abscess was resolved. Medications, especially certain antibiotics, can cause infertility or decreased or abnormal egg production. For example, testosterone injections in males can cause infertility, and an entire season of reproduction can be interrupted after the use of injectable doxycycline therapy.¹⁶² These changes may have been due to the stress of restraint and injection; however, similar cessations of reproductive activity have been noted following the administration of doxycycline in other psittacine species (Harrison GJ, unpublished).

Inbreeding

Males inherit fertility and semen quality characteristics. Some mating behavior is learned and some is inherited. Inbreeding may lead to infertility or decreased production due to genetically controlled physical or behavioral deficits. Lethal and sub-lethal genes that are more frequently expressed during inbreeding can cause decreased hatching rates. Genetic selection for large body types (budgerigars and turkeys) may cause a physical inability to breed.

General

Frequently, the positive environmental factors that stimulate breeding and the negative factors that prevent it cannot be discerned. Successful captive breeding depends on establishing environmentally enriching conditions that stimulate reproductive activity.

In environmentally stimulated cockatiels, the period from ovarian development to first oviposition was independent of environmental conditions.¹¹⁰ In pair-bonded cockatiels, a combination of diet, photoperiod, light intensity, ambient temperature and misters increased breeding activity. It was found that changing from a low-quality to a high-quality diet was not necessary to elicit a strong reproductive response. Of the remaining four factors it could not be determined if a non-stimulatory period was necessary or if the presence of any or all of the four factors was necessary to induce breeding.

Free-ranging populations of budgerigars were found to have minimal requirements for reproduction, including correct ambient temperature, day length and

water and food availability.¹⁷⁹ Captive budgerigars have similar requirements, which include sufficient food and water, a nest box with an opening at least ten cm below the nest box hole, combination of loud and soft warbles from the males and, in some cases, exposure to short photoperiods before the breeding season. Budgerigars are strongly stimulated by the vocalizations of other pairs.

Canaries respond principally to photoperiods, with low ambient temperatures causing a delay in egg laying.¹⁷⁹ Inducing reproductive activity in canaries requires exposure to a sequence of short photoperiods to abolish photorefractoriness followed by increasing day length to 14 hours of light. Providing a nest pan is not a prerequisite for ovulation and egg-laying in canaries but improves egg production.¹⁸² In reproductive success in canaries, the presence of a male or male vocalizations plays only a modifying role compared to photoperiod.

Natural Incubation

Natural incubation is a behavior under hormonal control that can be externally affected by many factors. Improper parental incubation can lead to a complete lack of egg development, arrestment of embryo development, late embryo death or abnormal or weak chicks at hatch. Additionally, many species such as macaws tend to be rather nervous in captivity and are notorious for breaking eggs. Minor punctures and hairline cracks can cause the death of a developing embryo. Foster parents or artificial incubation can be used in pairs with incubation problems. Failures in incubation can also originate from embryo-related problems, diet or environmental factors. In a group of Hyacinth Macaws, natural incubation of the eggs and chick-raising did not occur until the diet contained 15% fat and 2.5% fiber with limited seeds and nuts (Harrison GJ, unpublished). Studying pertinent egg information and performing thorough diagnostic procedures can help determine the cause of some of these incubation failures.

Information Collection

The attending veterinarian should review existing records concerning the parent's reproductive and medical history and fate of any eggs or chicks. Developing an accurate and consistent record-keeping system and regularly scheduling on-site visits will help identify factors that could explain incubation failures (see Chapter 2).

Reproductive information from each pair including numbers of eggs per clutch, number of clutches per

year, time of day eggs are laid, previous fertility and hatchability statistics, causes of egg failure and chick survivability will all help in evaluating a collection. To get a true fertility rate for a pair, one must necropsy all eggs as soon as possible after they are determined to be dead.

Fertility

Documenting if an egg is infertile or was fertile and died in early incubation is the first step in investigating egg problems. Eggs that are fertile but were not incubated or that failed to develop past two to five days of incubation will generally appear infertile when candled. These eggs should be opened to determine if they were fertile. Fresh, infertile eggs have a well organized small blastodisc, which in domestic species can be easily differentiated from the large, sometimes cottony or doughnut-shaped fertile blastoderm (Color 29.1). Old, addled or infected eggs in which fertility cannot be determined should not be included in fertility calculations. Additionally, any misshapen, mis-sized or otherwise abnormal eggs that are discarded should not be used in calculating fertility rates. The preferred method would be to include these eggs, as they can be fertile, or to calculate a separate fertility rate for abnormal eggs. Hybrid eggs should also be discounted, as they may have decreased fertility. Fertility rates can be calculated by finding what percentage of the total number of eggs laid were fertile. Undetermined eggs should not be included.

Fertility rates can be useful for discerning problems within a flock or individual pair. Infertility can be a result of behavioral, environmental, nutritional and medical problems (Table 29.5). Factors that should be considered include age of the birds, time the pair has been together, time the pair has been in the aviary, enclosure type, enclosure location, production of eggs in the past, past fertility and hatchability, hybrids, inbreeding, date of lay, environmental parameters (temperature, humidity, day length, rainfall) and behavioral characteristics of the pair. Fertility within an aviary should be evaluated on an individual pair and species basis within an aviary. Fertility rates of free-ranging birds may vary among species due to natural physiological processes. Fertility is normally reduced in older birds, in younger birds and at the beginning and end of a breeding season. Infertility in these cases may be a natural occurrence and not an indication of disease.

Domestic poultry have been genetically selected to produce high fertility rates of approximately 95%.

TABLE 29.5 Causes of Infertility

- **Behavioral:**

Immaturity, pair incompatibility, normal species differences, normal occurrence as part of clutch, sexual inexperience, lack of early learning, aviary disturbances, lack of social interaction, excess social interaction, homosexual pairs, lack of pair-bonding, asynchronous breeding condition, improper imprinting, infrequent matings.

- **Environmental:**

Incorrect photoperiod, incorrect nest box design or nesting materials, incorrect enclosure design, lack of visual barriers, excessive rain, insufficient rain, temperature, humidity, availability of correct foods, loose or incorrect perches.

- **Medical:**

Obesity, age (young or old breeders), inbreeding, vent feathers, drug therapy (causing vitamin deficiency or direct, decreased fertility), previous hormonal therapy (testosterone injections), musculoskeletal, neuromuscular or other disease (causing pain, paresis, ataxia, weakness, decreased muscle tone or incoordination), neurologic disease (causing paresis, ataxia, lack of muscle control), reproductive tract disease, nutritional deficiencies or excesses, systemic disease, parasitic disease leading to malnutrition, cloacal abnormalities, abnormal cloacal pH, possible thyroid deficiency, toxins (pesticides, chemicals, mycotoxins).

The fertility rates of most free-ranging companion birds have not been determined, although in some species studies have indicated that fertility can be quite high. Captive companion and aviary birds have the potential for similar fertility rates but more commonly the rates are lower, probably due to a combination of environmental and dietary factors.⁹² Free-ranging macaws do not necessarily nest and produce offspring each year. This cyclic production is probably related to environmental factors and not due to disease-related infertility. Aviculturists should establish their own fertility rates and standardize data so that comparisons can be made among similar aviaries.

Hatchability

Hatchability rates are determined from eggs that were known to be fertile. Including infertile eggs in hatchability statistics will artificially lower hatchability rates and confuse diagnostic efforts. Hatchability rates are calculated by finding the percentage of fertile eggs that successfully hatched.

“Successfully hatched” may or may not include chicks that were weak and died soon after hatching from pre-nursery-associated problems. Hatchability rates can be calculated for individual pairs, separate clutches, different species, eggs incubated naturally, eggs incubated artificially and eggs that had various kinds of physical problems or that were manipulated during incubation or hatching. The more precise the hatchability statistic, the more diagnostic the information that is provided (Figure 29.16).

In domestic fowl, the hatchability of naturally and artificially incubated fertile eggs approaches 85 to 90%. With companion and aviary birds, this figure may be much lower, and ranges from 8% to 100% have been discussed.^{29,92} Lower hatchability rates are probably due mostly to improper parent or artificial incubation techniques. Aviculturists should be encouraged to develop their own standards for hatchability and then strive annually to improve their level of success.

The number of lethal or chromosomal abnormalities reported in companion bird species is low when compared to domestic species. Evaluating fertility and hatchability statistics from parents and sisters of breeding males may help identify lethal or semi-lethal genes in some family trees. Breeding tests may

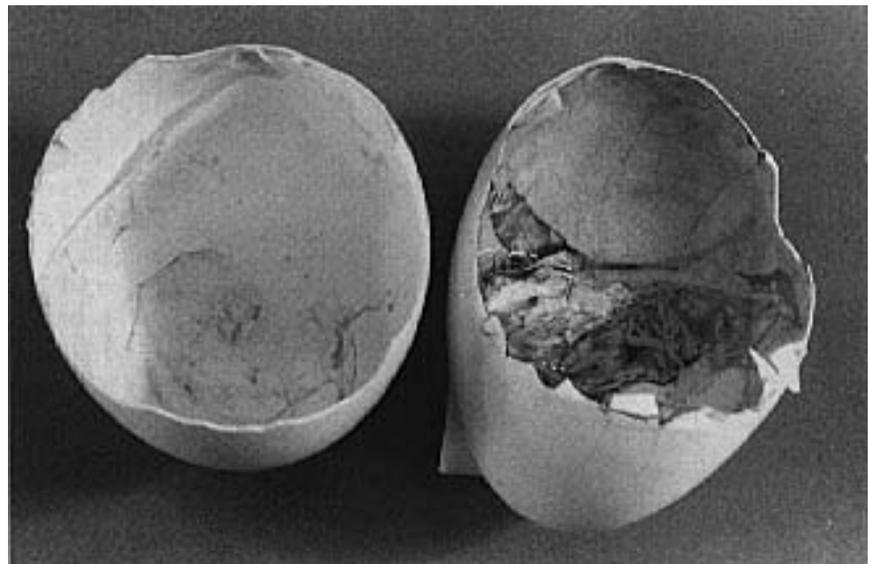


FIG 29.16 Complete and accurate hatching records can be indispensable in helping a veterinarian identify reproductive or neonatal problems. The egg on the left was from a normal unassisted hatch, and the chick from this egg was strong and developed normally post-hatching. The egg on the right was from a difficult hatch that required assistance. Note the retained blood within the membranes and excrement in the egg. The chick from this egg had intermittent problems with depression and slowed gastric emptying times for the first week of life.

TABLE 29.6 Causes of Death or Abnormalities in Embryos⁹²**FIRST TRIMESTER**

- Egg handling
 - Eggs stored too long
 - Eggs stored under incorrect conditions
 - Incorrect egg fumigation or sanitation (dirty hands)
 - Excessive vibrations (jarring)
 - Rapid temperature change
- High temperature in early incubation
- Incubation faults
 - Temperature, humidity, turning
 - Cooling after development has begun
 - Suffocation due to incorrect ventilation
- Inbreeding
- Chromosome abnormalities
- Egg-transmitted infectious diseases
- Parenteral nutritional deficiencies
- Abnormal or aged sperm
- Idiopathic developmental abnormalities
- Drugs, toxins, pesticides
- Cracked eggs
- Small holes in eggs

SECOND TRIMESTER

- Parenteral nutritional deficiencies
 - Riboflavin, vitamin B₁₂, folic acid, biotin, manganese, pyridoxine, pantothenic acid, phosphorous, boron, linoleic acid, vitamin K, vitamin D
- Secondary vitamin deficiencies
 - Antibiotic therapy destroying vitamin-producing flora
 - Diet imbalances, inadequate food intake
- Viral diseases
- Bacterial infections
- Fungal infections
- Egg jarring or shaking in the first trimester
- Incubator faults
 - Incorrect turning, temperature, humidity and ventilation
- Inbreeding resulting in lethal genes

THIRD TRIMESTER

- Malpositions
 - Inadequate or incorrect turning
 - Abnormal egg size or shape
 - Incorrect incubator temperature
- Incubator faults
 - Poor incubator ventilation
 - Egg cooling early in incubation
 - Inadequate or incorrect turning
 - Incorrect temperature
 - Incorrect humidity
- Incorrect hatcher temperature or humidity
- Long storage time pre-incubation
- Infectious disease
- Nutritional deficiencies
 - Vitamin A, D, E, K, pantothenic acid, folic acid
- Lethal genes
- Chromosomal abnormalities
- Idiopathic developmental abnormalities

be required to establish whether such genes are sex-linked or autosomal, dominant or recessive.

Parental Factors

The medical history of each parent should be examined to identify factors that may affect fertility and hatchability. Table 29.6 lists factors associated with

embryonic death according to the stage of incubation (first to third trimester). A pair with persistent fertility or hatchability problems should be completely evaluated by performing physical examinations, complete cloacal examinations, cloacal and choanal cultures, Gram's staining and culturing of uterine samples (many uterine problems are anaerobic), complete blood counts, serum chemistries, radiographs, exploratory laparoscopies and evaluation of sperm. Exposure to toxic compounds, either directly or in the food or water, should be considered. Behavioral problems including lack of pair-bonding, inconsistent parental incubation and egg trauma in the nest may also cause hatchability problems.

Diet

Diets should be analyzed for adequate levels of protein, fat, carbohydrates, calorie content, minerals, fiber, calcium and vitamins and for the presence of aflatoxins. Total caloric intake and food selection behavior for each individual bird should be evaluated. Nutritionally deficient hens can produce eggs, but the low level of nutrients may prevent the eggs from hatching. The age of embryonic mortality will usually depend on the degree and type of deficiency or toxicity.

Severe hypovitaminosis A causes a complete cessation of egg production. Partial hypovitaminosis A may cause circulatory collapse and embryo death and has been suggested as a cause of egg binding. Vitamin E deficiencies can cause lethal rings in which the embryo is seen surrounded by a ring of separated tissue. Vitamin D₃ deficiencies can cause small eggs with poorly calcified shells. Ultraviolet light exposure may improve hatchability in these cases while excess D₃ may lead to a complete cessation of egg production. Embryonic hemorrhage is common with deficiencies in vitamins E and K. Vitamin K is also involved with calcium transport, and vitamin K deficiencies can mimic the clinical signs associated with hypocalcemia.

Calcium-deficient eggs exhibit reduced hatchability, poor shell calcification, embryos with rickets and excessive loss of water and weight during incubation. The calcium/manganese ratio regulates the rate of hatching, and imbalances of these minerals may cause early or late hatching. Given the wide variability in the types of food (and thus the composition of these foods) consumed by free-ranging birds of different species, it is not surprising that a single commercially available diet cannot meet the needs of all captive companion birds. For example, free-ranging

Hyacinth Macaws that feed on high-fat nuts may require nutrients contained in these foods for successful reproduction and embryo health. It is speculated that breeding third and fourth generations of companion bird species will result in higher fertility and hatchability rates in birds fed commonly available commercial diets (see Chapters 3 and 31).

Environmental Factors

Perches should be stable enough for breeding, and nest boxes of suitable size should be easily accessible (see Chapter 2). Nest box size and shape and bedding material should be evaluated. The microclimate of the nesting area, including temperature and humidity, is important for proper incubation and is adversely affected by soiled bedding and improper nest box design. Cultures from bedding material may help identify infectious agents. Ambient temperature, humidity and to a lesser degree rainfall, wind and barometric pressure may affect the success of parental incubation.

Pre-incubation Factors

Non-incubated, fertile eggs will not develop if held at 55°F to 75°F. Cockatiel eggs stored at 55°F and 60% relative humidity did not show decreased hatchability until after three to four days of storage.³³ Eggs can be incubated for two days, removed and placed at 55°F, and placed back in an incubator without a decrease in hatchability. These temperature manipulations are convenient for shipping eggs and for synchronizing hatching times.

Parents may not initiate incubation until more than one egg is laid. Under natural conditions, the failure of a parent to incubate the first egg when temperatures are not within safe preincubation ranges can result in the death of the egg. Exposure of eggs to temperatures that are higher than 55 to 75°F but below optimal incubation temperatures can cause death of the embryo. Parent behavior, climate and nest box characteristics may be responsible for lack of development or deaths in embryos during the first and last third of development.

Artificial Incubation

Hatchability of artificially incubated eggs is frequently lower than naturally incubated eggs.³⁶ Eggs

that have been naturally incubated for the first five to ten days may have higher hatchability levels than eggs that are artificially incubated for the entire developmental period (Figure 29.17). The fact that different hatchability rates exist between natural and artificially incubated eggs highlights the need for a wider dissemination of information on successful incubation protocols.^{1,24,33,53,65,81,122,165,166,184}

Many aviculturists prefer to use foster parents rather than artificial incubators, particularly during the first week to ten days of incubation. Foster parents must exhibit broodiness and be accepting of the shape, size and color of the foster eggs (see Chapter 6). Bantam and Silkie chickens have been used successfully to foster eggs from many psittacine species. The number of eggs under each foster parent should not exceed the number that the hen can adequately incubate.

Incubation Requirements

Important incubation factors include temperature, humidity, air flow in the incubator and hatcher, egg position during incubation, the angle for egg turning and the number of times per day the egg is turned. Incubator temperature and humidity affect the incubation period, and published incubation periods may vary with different incubation parameters (Table 29.7). Substantial research is necessary to establish the optimal incubator parameters for companion bird species. Most psittacine eggs are incubated at 99.1° to 99.5°F (37.3°

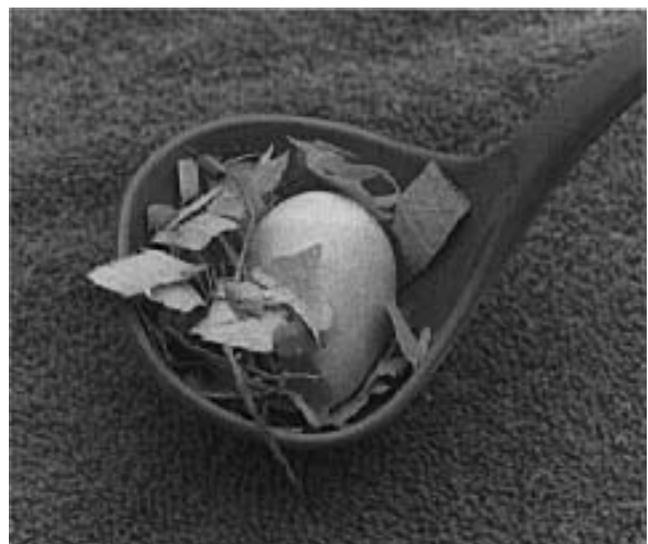


FIG 29.17 A plastic soup ladle makes an ideal tool for removing eggs from the nest. It has smooth edges and a long handle and can be easily sterilized between uses (courtesy of Apalachee River Aviary).

to 37.5°C) and 80° to 82°F (26.7° to 27.8°C) wet bulb, and hatched at 98.5°F (36.9°C) and 88° to 90°F (31.1° to 32.2°C) degrees wet bulb. Lower incubator humidities and higher hatcher humidities have also been described (Jordan R, unpublished). Research involving fertile cockatiel eggs determined that 99.5°F (37.5°C) with 56% relative humidity and 98.4°F (36.9°C) with 67% relative humidity were optimal settings for incubation and hatching, respectively.³³

Temporary shifts in temperature (as long as not excessively hot or cold) probably have no effect on hatchability. Such fluctuations are common when the incubator door is opened and the eggs are candled. It is best to turn off the fan when the incubator door is opened. Daily temperature and humidity charts should be maintained for each incubator. Individual incubators may have hot or cold spots that affect hatchability, and placing numerous thermometers at different locations within an incubator can help to identify these areas. Thermometers and hygrometers should be calibrated frequently to make certain that they are accurate.

A 2°F excess in temperature during the first few critical days of incubation can result in embryonic death.¹²² Increasing or decreasing the incubation temperature by 1.4°C caused poor hatchability and increased the incidence of abnormalities in cockatiel chicks.³³ Chicks produced by higher than optimal incubation temperatures were small, weak and dehydrated and frequently had umbilical openings and exposed yolk sacs. Scissor beaks, curled toes and wry necks were also common. Slightly higher temperatures will further increase mortality, and temperatures approaching 104°F (40°C) will kill all embryos.

Marginally lower-than-optimum temperatures may cause a delay in hatching. Temperatures that are constantly a degree or so lower than optimum have been shown to cause an increased number of “late dead” embryos, and if hatching occurs, chicks are weak with large, soft bodies and unabsorbed yolk sacs. Some chicks may be ataxic post-hatching. Hatching may occur several days later than expected. Low humidity results in lower egg weight, larger air cell size and small dehydrated chicks, possibly due to inadequate calcium mobilization for bone development.

Chicks from eggs incubated at high humidities may have excessive amounts of fluid, including residual albumen, that may obstruct the nostrils causing asphyxiation. Eggs should be turned at least five to eight times a day for at least two-thirds of the incu-

TABLE 29.7 Incubation Periods for Psittacine Species Common in Aviculture

Species	Incubation Period (days)	Pip to Hatch Interval (hours)
African Grey Parrot	26-28	24-72
African parrots (small)	24-26	24-48
Amboina King Parrot	20	24-48
<i>Aratinga</i> conures	24	24-48
Blue-fronted Amazon Parrot	26	24-48
<i>Brotogeris</i> parakeets	22	24-36
Budgerigars	18	12-24
Caiques	25	24-48
Cockatiels	21	24-48
Cockatoos (large)	26-29	24-72
Cockatoos (small)	24-25	24-72
Eclectus Parrot	28	24-72
Goldie's Lorikeet	24	24-48
Grass Parakeet	18	24-48
Green-cheeked Amazon Parrot	24	24-48
Hyacinth Macaw	26-28	24-72
King Parrot	20-21	24-36
Lories	26-27	24-36
Lovebirds	22	24-48
Macaws (medium)	24-28	24-72
Macaws (miniature)	23-27	24-60
<i>Amazona ochrocephala</i> parrots	26-28	24-48
Parrotlets	19	24-36
<i>Pionus</i> parrots	25-26	24-48
<i>Psittacula</i> parakeets	24-26	24-48
<i>Pyrrhura</i> conures	23	24-48
Quaker Parakeet	23	24-48
Red-lored Amazon Parrot	24	24-48
Rose-breasted Cockatoo	22-24	24-72
Palm Cockatoo	28-30	24-72
White-fronted Amazon Parrot	24	24-48

Compiled by Susan Clubb and Keven Flammer

bation period. More frequent turning, up to 24 times a day, may improve hatchability in Psittaciformes or with embryos suspected to have a lack of vigor or delayed development. Eggs should be positioned on their sides with the round or air-cell end slightly elevated. Poultry eggs tilted or placed in a horizontal position were found to have an increased incidence of malposition of embryos.⁷⁶

Still-air incubator temperature requirements are usually higher than forced-draft incubators. Placing incubators in a room that maintains a relatively cool (70-80°F; 21-26°C), dry (50-60% relative humidity), environment is ideal. Extreme temperature and hu-

midity fluctuations (5°F or 5% relative humidity) in the incubation room should be avoided. Incubator ventilation, sanitation, abnormal vibrations, improper mechanized egg turning, inaccurate thermometers, inaccurate hygrometers and placement of incubators near walls and windows can all affect incubator function.⁸⁹ Incubators with horizontal grill-type turners may be too rough for sensitive embryos. Excessive jarring and shaking, particularly during the early stages of development, can result in embryo death or malformation. Improper egg position and faulty egg turning during development may result in malpositions and incomplete closure of the ventral body wall.

Hatchers should be evaluated in a manner similar to incubators (Figure 29.18). The success of the sanitation program and the presence of microbial contamination can be estimated with cultures of the incubator surfaces, water trays, egg trays, and incubator room floor, shelves and instruments.

Incubation Preventive Techniques

Prevention of most incubation problems involves correcting the three most common causes for decreased hatchability in artificially incubated eggs: improper temperature, humidity and egg turning.⁸⁹ Accurate record-keeping is mandatory for identifying hatching-related problems. A protocol for carefully evaluating incubator performance and stability should be followed.⁸⁹ Pre-conditioning incubators a month prior to breeding season and evaluating daily fluctuations in temperature and humidity in the incubator room and in the incubators may help identify problem areas. The egg-turning mechanism should be checked periodically to confirm that it functions at the correct time interval, maintains the necessary egg angle and does not excessively vibrate the eggs. Excessive vibrations have been associated with reduced hatchability.¹⁸⁴

Bacterial and fungal agents infrequently cause problems in psittacine eggs, but occasionally a contaminated incubator, hatcher or incubator room can cause high egg losses. Water trays should be removed and disinfected daily and should be filled with distilled water. In the non-breeding season incubators should be dismantled, and non-sensitive parts should be thoroughly cleaned with a glutaraldehyde solution followed by a long period of air drying. Disinfecting incubators with formalin and potassium permanganate is extremely dangerous and cannot be recommended.



FIG 29.18 Plastic strawberry baskets with a section of small plastic mesh in the bottom make ideal hatching containers. They are safe, have no sharp edges and can be easily sterilized between uses. This hatching basket contains the perfectly opened egg from a Greater Sulphur-crested Cockatoo chick (courtesy of Apalachee River Aviary).

Periodic culturing of newly hatched chicks, eggs and incubator surfaces will indicate if bacterial contamination is occurring. A sterile contact tape can be used to culture nursery surfaces and eggs. An open microbiological agar plate can be placed in an incubator to determine what bacteria are present in the air. Incubators can be tested for the presence of PBFD virus or polyomavirus by taking swabs for DNA probing.

The incubator room should be kept scrupulously clean to prevent particulate matter from contaminating egg shell surfaces. Clothes and shoes worn around other birds should be removed or covered before entering the incubation area. Hands should be thoroughly washed with a disinfectant or gloved before handling eggs. Problems associated with incubation are listed in Table 29.8.

Eggs are relatively resistant to bacterial invasion, but eggs that may have been contaminated with infectious agents should be incubated separately from non-contaminated eggs. Feces and dried particulate matter can be gently sanded off the egg surface, although over-exuberant sanding should be avoided.

Chlorine dioxide foam may be a safe sanitizing agent for contaminated eggs,¹²⁵ or eggs can be washed with a warmed iodine solution (104°F) or immersed in warm water baths (110°F) for up to five minutes. Bacterially contaminated eggs can be dipped into a cold water antibiotic solution by warming the eggs to 37°C, and placing them in cold water (4°C) containing 1000 µg/ml of gentamicin solution.¹⁶¹ Psittacine eggs should not be incubated with eggs from other species.

Monitoring the Embryo

Candling

A candling program allows one to follow developmental progression of an embryo and detect any abnormalities that may occur. Chicken eggs can be used for practice. Periodic candling will ensure the removal of non-developing eggs as soon as they are identified. This increases the likelihood that the cause of an embryo's death can be determined and reduces the possibility of an infected egg contaminating the incubator. Daily candling will improve an individual's ability to recognize developmental stages of the various species.

Initial candling of psittacine eggs should occur no later than six to seven days into incubation. The eggs should be handled with care to prevent sudden jarring or chilling from inducing embryonic death or malformation.¹⁷⁵ Shortly after oviposition, an egg may not have an air cell; this develops as the egg cools and the internal volume is reduced. It is during this initial cooling process that surface contaminants can be drawn into the egg. Eggs can be marked for identification using a #2 pencil (see Figure 29.4). Candling between the seventh and tenth day of incubation will indicate if an egg is fertile and whether it is developing normally. After the initial candling, eggs can be evaluated every two or three days if desired, and should be examined at least once just before transfer to the hatcher.

Candling naturally incubated eggs should be considered; however, the disadvantages of disturbing the adults and eggs must be weighed against the possible advantages of identifying eggs that need manipulation or intervention for hatching to occur. Candling to determine if the egg is fertile (five to seven days post-laying) followed by evaluation just prior to the expected date of pipping will usually be sufficient for evaluating parent-incubated eggs.

Extended flashlight type candler may not satisfactorily illuminate eggs when ambient light is present, and more specialized tools may be required (Figure

TABLE 29.8 Chick Abnormalities Caused by Incubation Problems

Abnormalities	Possible Causes
Early hatch, thin, excessive vocalizations	Small eggs, species differences, high incubator temperature, low incubator humidity, high incubator or hatcher temperatures (bloody navels).
Late hatch	Large eggs, old parents, eggs stored too long pre-incubation, low incubator temperature, inbreeding.
Sticky chicks, albumen present	Low incubation temperature, high incubation humidity, incorrect turning, very large eggs.
Dry chicks, stuck to shell	Low humidity during egg storage, incubation or hatching, incorrect turning, cracked eggs, poor shell quality, high incubator temperatures.
Small chicks	Small eggs, low humidity during egg storage or incubation, high incubator temperature, high altitude, thin or porous shell, tetracycline used in hen.
Weak chicks	Variety of causes including incorrect humidity and parental malnutrition.
Umbilicus fails to close with varying degrees of unretracted yolk sac	Incorrect incubation temperature, low hatcher temperature, high hatcher humidity, parental malnutrition, omphalitis (can be caused by contamination or incorrect incubation temperature), inadequate ventilation.
Short, wiry down (species dependent)	Nutritional deficiencies, toxins (eg, mycotoxins), high incubation temperature first two trimesters.
Dwarf embryos, stunting in growing chicks	Egg contamination, heredity, parental malnutrition, possible hypothyroidism.
Short beak, missing beak, face, eye or head abnormalities	High incubator temperatures early first trimester, lethal genes, idiopathic developmental abnormalities, parental nutritional deficiencies (eg, niacin), low oxygen early first trimester, sulfa drug use in hen, insecticides, herbicides, excess dietary selenium, nicotine, viral.
Red hocks	Prolonged pushing on shell during pipping and hatching, parental vitamin deficiencies, thick shells, high incubator humidity, low incubator temperature.
Musculoskeletal or neurologic abnormalities	Incorrect incubation temperature (curled or crooked toes, splayed legs), low incubation temperature (bent necks, nervous disorders), high incubation temperature (ataxia, star gazing), low humidity, unsuitable hatching substrate, sulfa drug use in hen, insecticides (scoliosis, lordosis).

29.19).¹⁰³ High intensity lights (heat) may injure embryos during early development.

- Candling Data:** Candling helps identify the degree of egg shell thinning, egg shell cracks, blood rings, meat spots, membrane and blood vessel integrity, heart rate, stage of development, development progression, air cell size and shape, and yolk size, color and shape (Colors 29.14 to 29.16). Candling later in incubation helps to evaluate malpositions, chick movement, size, shape and location, and internal pip-to-hatch interval (Colors 29.17, 29.18). Lack of embryo vitality can be recognized by poor vessel integrity, decreased movement and retarded development. Embryo death in early incubation results in cessation of development, blood rings and loss of membrane and vessel integrity. Late embryo death is somewhat harder to recognize due to the natural opaqueness of the developing embryo, but lack of

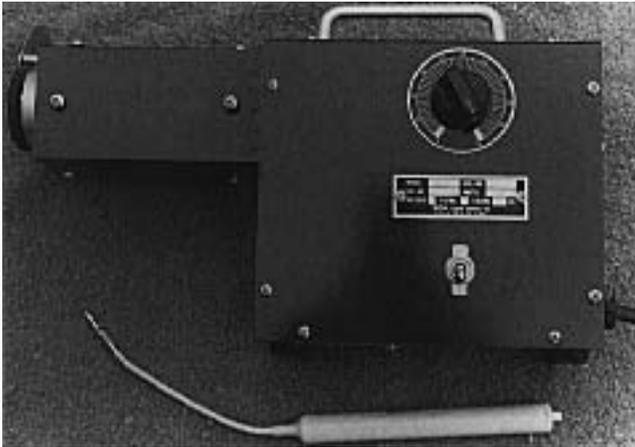


FIG 29.19 Small utility lights (below) can be used to perform basic candling tasks in dark areas, but have limited application for performing in-depth diagnostic evaluation of an egg. It is best to use a commercial candler (above) when detailed information on the developmental state of an egg is required (courtesy of Apache River Aviary).

vessel integrity and movement are indicative of late incubation deaths.

Egg Weights

Eggs should be weighed when they are candled, and weight loss rates can be recalculated throughout the incubation cycle. Air cell dimensions can be evaluated to provide an estimate of weight loss. Eggs from most Psittaciformes should lose an average of 12% to 13% of their weight from the beginning of incubation to the point of transfer to a hatcher. An additional 3% of weight should be lost during pipping.^{24,81,130} Desired egg weight loss can be determined using a variety of mathematical formulas.^{24,68,81,130} Egg weight loss is affected by egg shell porosity, air circulation, altitude, temperature and humidity.^{24,26} Egg weight loss rates can be used to detect incubation problems and can also be used to manipulate the humidity or egg to ensure proper weight loss.

Egg Necropsy

Every egg that fails to develop or that dies should be necropsied. (“Breakout” refers to opening eggs for diagnostic purposes.) Candling cannot distinguish very early embryonic deaths from infertile eggs, and the presence of fertility is an important criteria when proceeding with a diagnostic program in avian reproduction. All data relating to the egg should be reviewed prior to necropsy. The veterinarian should be able to identify various embryologic structures and understand the physiological purpose of each structure that might be related to embryo death (see Table 29.2, Figures 29.2, 29.3).^{1,44,55,135,137}

Necropsy should be performed on eggs or chicks as soon after death as possible to prevent rapid autolysis from destroying valuable information; however, one should always make certain that an embryo has stopped developing before initiating a necropsy. The majority of eggs for necropsy will fall into two distinct age groups: embryonic death at three to five days of incubation and death perihatching.¹³⁵ Early embryonic mortality is common with improper incubation temperature, jarring, inbreeding and chromosomal abnormalities. Deaths at the end of incubation are usually associated with hatching, and the stressful period of switching from allantoic to pulmonary respiration. Factors including improper incubation humidity, temperature and turning are thought to be the leading causes of late embryonic death in psittacines. Mid-incubation deaths occur in poultry embryos when the hen is fed a diet deficient in proteins, minerals or vitamins.

- **Technique:** All eggs should be candled before necropsy to determine the best point for entering the egg. This also permits the correlation of candling with necropsy findings. Eggs should be weighed and measured, and external shell characteristics (egg shape, egg size, external calcium deposits, cracks or thinning) should be noted. Pip marks should be evaluated for turning direction, location and size. Chicks normally pip counter-clockwise from the round end of the egg. The egg necropsy should be performed under sterile conditions until cultures have been taken of embryonic fluids and tissues.

The egg is opened over the air cell with sharp-blunt scissors. Shell membranes are examined for abnormalities and then carefully peeled back to expose the internal egg contents (Color 29.36). When exposed, the albumen and amnion can be cultured and visible microbial growth may be noted (Color 29.37). The chorioallantoic membrane (CAM) normally adheres to the inner shell membranes after the first trimester

CLINICAL APPLICATIONS

Egg Weight Formulas — Weight loss trend to pip:

$$([\text{laid weight}] - [\text{current weight}]) \div (\# \text{ days incubated}) = (\text{average daily weight loss})$$

$$(\text{average daily weight loss}) \times (\text{total incubation period prior to pip}) \div (\text{laid weight}) = (\text{decimal percentage})$$

$$(\text{decimal percentage}) \times 100 = (\text{weight loss trend to pip})$$

$$(\text{laid weight}) = (\text{species specific coefficient}^{\text{B}^1}) \times (\text{length egg in mm} \times \text{breadth}^{\text{2}})$$

of development. Adhesions of the embryo to the CAM are abnormal. Eggs that are not necropsied shortly after death may develop adhesions or sticky membranes. Late-term embryos have anatomical differences from adults, but are similar to young chicks.¹¹⁴

Internal egg membranes and structures should be evaluated before being altered by manipulation or culture techniques. A ruptured yolk sac can obscure the necropsy field. Color, size and location of the albumen, yolk and allantois is recorded. Presence and characteristics of the circulatory tree are observed. Abnormal odors should be noted.

Enlarging the hole over the round end of the egg is performed by careful removal of egg shell fragments with thumb forceps. If a small chick or no chick is identified, the contents of the egg can be carefully poured into a sterile container (Color 29.38). If a well developed chick is present, the position of the air cell with respect to the egg, orientation of the embryo as a whole within the egg, position of the head, beak and neck in relation to the body and position of the beak in relation to the air cell should be evaluated (Color 29.34).

Different malpositions have various success rates for hatching in domestic species, and many can hatch unassisted (Table 29.9). Although only six malposition classifications are traditionally discussed, there is an almost endless variety of malpositions that can occur, some of which are very subtle and require close inspection.

Psittaciformes appear to have different malpositions than domestic species.^{28,29} Frequently the head appears on an even plane with the right wing, and the entire body may be rotated such that the spine is on a horizontal plane with the short axis of the egg. The significance of this malposition is not known. Common causes of malpositions include turning problems, incorrect position in the incubator, oxygen deprivation, excess CO₂, lack of embryo vigor and delayed development.³⁷

If a chick can be seen in the egg, it should be weighed, measured and staged according to standards.⁵⁵ Remaining albumen, chorioallantoic membrane blood vessels and ruptured yolk or allantoic contents may adhere to the shell once the chick is removed. If size permits, the chick should have a full necropsy performed, being careful to keep the yolk sac membrane intact. Special attention should be given to the hatching muscle for size, edema and hemorrhage. Other gross evaluations, including skin color and hemor-

rhage, musculoskeletal deviations, internal hemorrhages and contents of the mouth, nares, crop and esophagus, should be made. The liver may be hemorrhagic from exuberant kicking, especially if a chick was malpositioned (Color 29.39).

The hatching muscle (*Muscularis complexus*) is a primary storage site for lymph in the embryo and is normally enlarged perihatching. Lungs are evaluated for evidence of air intake or for the presence of fluid, although this differentiation usually requires histopathology. Tissues should be fixed in formalin for histopathologic evaluation. Eggs can be frozen for future analysis of toxic substances.⁹⁶

At hatching, a chick's weight, activity, vocalizations, body measurements and degree of yolk sac retraction and the presence of any abnormalities should be recorded. Monitoring chick growth rates, food intake, behavior and development progression can help detect any subtle problems that may occur.

Microbiology of Eggs

External egg structures prevent but do not stop microorganisms from entering the egg. Bacteria located in an egg could suggest environmental contamination that occurred after embryonic death. If the ne-

TABLE 29.9 Classic Malpositions of Chick Embryos

Malposition 1:	Head between the thighs. Failure of the chick to lift and turn its head to the right in the middle of the last trimester. Completely lethal. Incidence increased by high incubation temperature.
Malposition 2:	Head in the small end of the egg. Chick is upside down in the egg. Hatchability reduced by 50% in domestic species. Incidence increased by incubator egg position and low temperature.
Malposition 3:	Head is under the left wing. Chick rotates its head to the left as opposed to the right. Usually lethal. Incidence increased by incubator egg position, temperature and parental malnutrition.
Malposition 4:	Beak is away from the air cell. Upward turned aspect of maxilla and egg tooth is not near the air cell; however, the rest of the embryo is normally positioned. Slightly reduced hatchability. Incidence increased by incubator egg position.
Malposition 5:	Feet over head. Usually lethal.
Malposition 6:	Head is over the right wing. Normally the head is under the right wing in domestic species. Psittacines may normally hold the head in the same plane as the wing. Reduces hatchability slightly in domestic species. Incidence may be increased by parental malnutrition.

cropsy is performed immediately following embryonic death, finding bacteria in the embryo can indicate bacteremia or an infected ovary or oviduct in the hen. Bacterial contamination of an egg usually originates from the atmosphere, the nesting area or the surface of the cloaca.²⁵

In poultry, reducing microbial loads in nest shavings does not increase hatchability, although it does decrease bacterial contamination in dead or dying eggs. With duck eggs, however, hatchability and chick hatch weight could be increased by having clean eggs (not visibly soiled).^{82a} Generally, egg infection is not caused by environmental contamination although the type and concentration of bacteria in the nest box can definitely increase the likelihood of an egg infection. Eggs with hairline cracks, pinpoint punctures and thin shells are more prone to colonization by microorganisms. Reduced cuticle deposition, which occurs in older birds, does not affect hatchability except in extreme instances, but will affect the number of dead or dying eggs that are bacterially contaminated.

Egg structures are affected by bacteria in different ways. The source of persistent egg infections may be identified by culturing the hen's cloaca, nest box contents, the exterior egg shell, albumen, yolk and embryonic tissues. Gram-positive bacteria occur mostly on the surface of the egg, and the insides of contaminated eggs contain mostly gram-negative bacteria.¹⁵ Chicks that are infected in the egg usually die with macroscopic yolk lesions (coagulated yolk and yolk sac hemorrhages) (Color 29.39). In other cases, embryos may die before the production of macroscopic changes in the yolk and a histological examination is necessary to establish the presence of an infection.⁶³

■ Egg Therapeutics

Treatment of bacterially infected eggs is possible although preventing infections is more effective. Medical intervention should be attempted only in those cases where the embryo is at risk of dying. Perpetuation of weak genetic lines in companion birds may be exacerbated by assisting in the hatching of troubled eggs.

Pre-incubation

Defects in egg shells can be repaired by using sparing amounts of Elmer's glue, surgical glue or paraffin. Thick or excessively large applications of these substances can retard air exchange or create difficulty

for the chick during hatching. It has been suggested that large defects can be covered with egg shell remnants from other eggs, although the prognosis for these eggs should be considered poor.¹⁶⁵ Tremulous air cells resulting from trauma or weak shell membranes may indicate blastoderm shock and ruptured shell membranes. Eggs with tremulous air cells usually have reduced hatchability but should not be discarded because embryos may develop and hatch normally.⁹⁰ These eggs should be hand-turned as should all eggs with suspected shell or membrane defects.

Incubation

Manipulation of eggs before the point of hatching should be limited. The most frequently used techniques are designed to change the weight loss of an egg. Eggs can be moved to higher or lower humidity incubators based on weight loss. Eggs can also be gently sanded or have small holes placed in them over the air cell to increase weight loss. Paraffin can be used to partially cover the egg to reduce weight loss although no more than 60% of the area above the air cell or total air cell surface should be coated.²⁶ Sealing a large portion of the air cell may decrease oxygen intake and cause the embryo to invert within the egg. Eggs that have had their shell altered should be hand turned to keep the sealant intact and to reduce chances of damaging the shell. Irregular or weak vascular patterns may be corrected by increasing the turning frequency (Thormahlen M, unpublished).

Injecting sterile lactated Ringer's solution into severely dehydrated eggs has proven successful in some cases.²⁶ Injection of sterile water into the small end of the egg (albumen) during the first half of incubation led to addled eggs in one study¹⁵⁰ but was of benefit if given later in incubation. Replacement volumes to be given are calculated from egg weight deficits. Injecting antibiotics (piperacillin 200 mg/ml, 0.02 ml for macaw eggs, 0.01 ml for cockatoo eggs) into bacterially contaminated eggs has been attempted with some success.¹⁰⁸ If done properly, injecting gentamicin into the albumen was not found to lower hatchability.²⁷

Small dental drills or needle puncture holes can be used to make a pathway for delivering injections into either the small end of the egg or over the air cell. Holes should be resealed with paraffin or glue. Pre-incubatory egg injections with 2.4 mg of tylosin and 0.6 mg of gentamicin was successful in eliminating *Mycoplasma meleagridis* from turkey poults.¹⁰⁵ Ap-

appropriate drugs, dosages, site of inoculation and timing of inoculation for companion bird species have not been determined, and egg injections should be avoided except in special cases.

Late Incubation

As the expected hatch date approaches, the egg should be candled frequently to monitor changes in the configuration of the air cell. As the chick develops, the head comes to lie under the right wing, with the tip of its beak directed towards the air cell.¹²² When the air cell drops and enlarges, the hatching process has begun and a chick begins the transition from chorioallantoic circulatory respiration to pulmonary respiration. As the circulation to the allantois no longer has the capacity to meet the embryo's needs for gas exchange, the chick begins to move its head to the air-filled end of the egg. This stage of hatching can be observed only by candling. Some chicks will begin to vocalize during this period. The CO₂ level in the embryo rises causing the neck muscles to twitch, and the embryo will penetrate the membrane into the air cell. At this point, the embryo begins to breathe air, and the patent right-to-left cardiovascular shunts close.¹²² The muscle twitching also occurs in the abdominal musculature initiating retraction of the yolk sac into the coelomic cavity.¹²² As the chick becomes more active and depletes the oxygen in the air cell, its carbon dioxide level increases to 10%, producing even stronger muscle contractions of the neck until the beak creates a puncture in the shell.¹²² At this point the chick is breathing room air and vocalizations can be heard. External embryo structures such as the yolk sac or enlarged allantois can be accidentally ruptured during the pipping process.

Once eggs have started to pip and are transferred to the hatcher, they should be left undisturbed. As it hatches, the chick alternates between jerking head movements, which continue to chip the shell, and prolonged muscle contractions of the neck and back, which straighten the neck and force the body to rotate slightly counterclockwise. When these muscles relax, the head is in a new position, and additional jerking movements chip a different portion of the shell. During this process, called cutting out, the chick rotates within the shell 360°, cracking the shell circumferentially (see Figure 29.16). Eventually, the chick will push off the top of the shell and emerge. In most cases the process proceeds normally and a healthy chick emerges.

The average incubation period and pip-to-hatch interval of each species varies (see Table 29.7) and eggs

that deviate from these average values can successfully hatch without assistance. Premature intervention in the hatching process can cause embryonic death. Proper intervention at the correct time can definitely result in a hatched chick that would have otherwise died. The amount of assistance required is difficult to determine but it is generally best not to rush the hatching process, but to gently assist each stage as necessary. Pip-to-hatch intervals are 36 to 48 hours in most species and hatching times of less than 24 hours and greater than 80 hours usually indicate a problem.⁵⁴ Chicks that pip one-fourth to one-half of the egg and then stop for an extended period of time, or that reverse direction and return to the pip site, usually require assistance.⁶⁸

Eggs that lose an abnormal amount of weight or contain a malpositioned chick can hatch, although hatchability is generally reduced. The timing and degree of intervention is dictated by the recession of active blood vessels, yolk sac retraction and extent of delayed hatching. A weak chick may also require assistance with hatching. Weak chicks will emit faint and infrequent vocalizations. These chicks may be normally positioned and may have appropriately entered the air cell; however, if the hatching process is delayed, the embryo may be in jeopardy.

Chicks can be safely removed from their eggs if the yolk sac and blood vessels have retracted. In general, chicks that have made one quarter of a turn during pipping can usually be safely removed from the egg. Chicks can bleed to death or rupture their yolk sacs if removed prematurely, although in some cases minor bleeding and a partially unabsorbed yolk sac must be accepted to remove a chick before death occurs. Candling or dampening the inner shell membrane with sterile water will help elucidate the position of unretracted blood vessels. Once chicks pip internally, it is important that they have an unoccluded path for air intake. Malpositioned chicks or chicks with delayed albumen ingestion may need egg shell fragments removed and fluid cleared from their nares.

If drawdown fails to occur, little can be done to assist the chick. The transition from allantoic respiration to breathing air is delicate and timely. Forcing the process will result in the death of the chick. If internal pip has occurred but external pip does not, a small hole can be safely created in the air cell to provide a source of fresh air. If there are no signs of external pip after 36 hours from drawdown, a breathing hole should be created. The breathing hole need only be a few milli-

meters in diameter and can be created using a bur with or without magnification depending on the size of the egg.

To perform an ovotomy, the egg is candled and the air cell identified and marked with a soft pencil. The shell over the air cell is cleaned with dilute chlorhexidine or povidone iodine. It is important to keep the ovotomy site totally over the air cell where there are relatively few blood vessels in the outer shell membrane. If the shell is opened over any other area of the egg, severe, life-threatening hemorrhage may occur. Vessels that are regressing take on a ghostlike appearance and are often only partially filled with blood.

If, after providing a breathing hole, external pip still does not occur, the shell should be removed over the air cell. This procedure is usually performed 48 hours after drawdown or early on the scheduled hatch date if it is accurately known.⁸¹ Using a bur, the shell is removed without disrupting the outer shell membrane, which lies directly below and attached to the shell. A circular area of shell 0.5 to 1.5 cm in diameter should be removed, depending on the size of the egg. Once the shell has been removed, the outer shell membrane should be moistened with saline on a cotton-tipped applicator. Once moistened, the membrane becomes translucent, making it easy to identify any vessels that might need to be coagulated using bipolar radiosurgical forceps. After the vessels are coagulated, the membrane can be opened with the bipolar forceps revealing the chick within the air cell.

If the chick has entered the air cell, there will be a small nick in the inner membrane through which the beak has penetrated allowing respiration. The inner membrane is generally moister and more translucent than the outer shell membrane, except in the area where the beak has penetrated. In this region, the vessels retract and the membrane will usually appear dry and white. The inner shell membrane is delicate and highly vascular. The membrane should be carefully manipulated to prevent tearing. The entire exposed inner shell membrane will rapidly desiccate and should be kept moist by adding drops of warm, sterile saline or lactated Ringer's solution. Small quantities of fluids should be used to keep the chick from drowning. If the membrane is opaque, it is not properly hydrated.

At this point the position and status of the chick may be assessed. A major cause of late embryonal death is suffocation caused by occlusion of the nares by the

inner shell membrane. The bipolar forceps may be used to coagulate the vessels around the site of membrane penetration followed by the creation of a small circular defect in the membranes. Removing this tissue allows the chick to breathe and prevents the membranes from occluding the nares.

For a successful hatch to occur, there must be an increase in CO₂ within the air cell to stimulate the chick to struggle, which ensures retraction of the yolk sac and the break out from the egg. The hole created in the shell can be partially sealed to allow this increase in CO₂ to occur. A stretchable, wax type test tube sealant can be used to effectively seal the hole created in the shell. The edges should be smoothed out and the egg returned to the incubator. An alternative solution is to place the egg in a small plastic bag with moistened sterile gauze. The bag can be partially sealed to allow an increase in CO₂ to develop, and the moistened gauze will ensure adequate humidity.

Chicks that are slightly malpositioned may create an external pip below the air cell. Strong chicks will continue to move their head toward the air cell creating additional external pips along the way. Appearance of more than one external pip may be an indication that the chick is malpositioned and may need assistance. If the pip is properly located, the egg should be returned to the incubator with the pip up. If the pip is on the opposite end of the egg from the air cell, it is likely that the chick is inverted and will need major assistance. If the pip is close to, but not within the air cell, intervention is indicated (see Chapter 41).

Embryo Extraction

The time period between external pip and hatching varies with species, shell thickness, incubation regimen, genetics and the strength of the chick. These factors make it difficult to determine when intervention is indicated. Embryos that enter the air cell prematurely may defecate inside the shell causing a compromise in the normal metabolic management of waste (see Figure 29.16).

When sufficient time has passed that the risk of fecal contamination is high or if the chick appears to be weakening based on decreased vocalizations and movements, the hole in the shell and shell membranes should be enlarged to allow gentle extraction of the chick's head and neck. The chick should be grasped by the beak and gently pulled out of the shell to allow visual inspection of the yolk sac (Figure

29.20). If the inner shell membranes have not adequately retracted, the yolk sac will still be visible, (incompletely absorbed). If no feces are found, the chick is gently replaced and the egg is sealed to allow hatching to proceed. The chick should be re-evaluated every one to three hours for the presence of feces. If the chick appears weak, oral administration of 5% dextrose solution may be beneficial. This can be alternated with lactated Ringer's solution to provide additional electrolytes. Since embryos are very susceptible to drowning, it is best if the solution can be placed into the esophagus or ingluvies using a 1 mm diameter silicone catheter or metal feeding tube. Excessive quantities of fluid should be avoided to prevent the accumulation of fluids in the allantois, which may increase the potential for membrane ruptures or delayed yolk sac absorption.

Once feces are observed within the shell, the chick should be removed. The chick is gently extracted with care taken to control hemorrhage from any unretracted vessels. The major attachment of the chick to the shell is in the area of the umbilicus where the vessels of the inner shell membrane attach to the yolk sac and umbilicus. The chick is extracted to a point where these vessels are visible and a vascular clip can be easily applied. The vessels are transected

using radiosurgery and the chick is completely removed from the shell.

Aggressive hatching assistance is indicated for inverted chicks to prevent their dying of hypoxia or drowning. The earliest indication that a chick is inverted is an external pip at the small end of the egg. In approximately one of three inverted chicks, the air cell will have drawn down far enough to supply the chick with air. This is beneficial as the key to saving inverted embryos is providing air and enough time to allow retraction of the yolk sac. A breathing hole should be created over the air cell, which will change the pressures within the egg and allow the embryo to slide down into the large end of the egg and the air cell to migrate to the small end of the shell.⁸¹ The original pip site should be enlarged, with care taken not to damage the vessels within the membranes. If bleeding occurs it should stop in ten seconds. Sustained bleeding of chorioallantoic membranes can be stopped by applying pressure with sterile swabs or with the careful and specific application of a chemical coagulant such as silver nitrate. Experimentally, excessive bleeding can be controlled by placing injectable vitamin K sparingly on the bleeding CAM. Dehydrated chicks can be given fluids orally or subcutaneously while still in the egg. A small amount

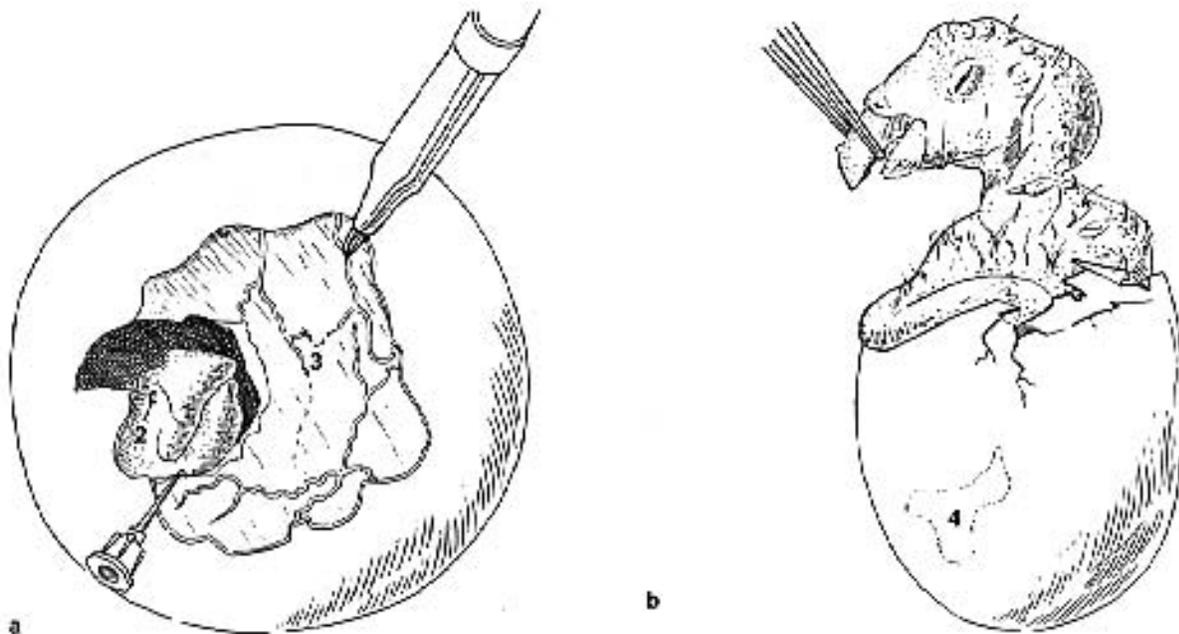


FIG 29.20 a) Assisting with the hatching procedure can be a life-saving procedure in some situations. The most important factor is to ensure that the chick's nostrils are clear of the shell membranes so that it can breathe. The avascular membranes can be gently teased away from the nostrils using a hooked needle. Radiocautery is used as needed to control bleeding. b) Smooth, plain pick-ups can be used to grasp the beak across the egg-tooth to elevate the head and evaluate the progression of yolk sac absorption. If the yolk sac is not absorbed, the head should be replaced, the end of the egg should be partially covered with parafilm and the egg should be returned to the hatcher. If the yolk sac is absorbed, the chick can be removed. 1) egg tooth, 2) nare, 3) receding vessels and 4) ghosted yolk sac (modified from Stoodley).¹⁶⁵

Theriogenology

Color 29.19

Normal left melanistic ovary in a mature Umbrella Cockatoo hen (open arrow). The cranial, middle and caudal divisions of the kidney are clearly visible. The inactive ovary is found in its normal location at the cranial medial border of the cranial division of the kidney. The right and left adrenal glands (arrows), the ischium (i), the pubis (p) and the ureter (u) are also visible.

Color 29.20

A 25-year-old Scarlet Macaw hen was referred with a history of egg binding that had supposedly been resolved with a hysterectomy. The hen was losing weight, regurgitating and had a distended, painful abdomen. An exploratory laparotomy indicated peritonitis and a fibrous constriction of the bowel. A side-by-side intestinal anastomosis was performed, but the bird did not recover. Necropsy findings included an abdominal egg yolk (arrow) and a fully developed left oviduct (open arrow). The abnormal development of a right ovary (also present here) can predispose a hen to reproductive problems.

Color 29.21

Infectious salpingitis with secondary infection of the ovary. A normal follicle (arrow) is seen adjacent to degenerating, hemorrhagic follicles (open arrows). The lungs (l) are also visible (courtesy of R. Korbel).

Color 29.22

A 23-year-old Amazon parrot hen was presented with a history of progressive abdominal swelling and weight loss. Cytology of fluid collected by abdominocentesis revealed a modified transudate. Radiographs indicated a diffuse soft tissue opacity in the intestinal peritoneal cavity that was pushing the ventriculus cranially. The bird did not respond to supportive care. The ovary (arrow) was reddish-brown, enlarged, firm and contained numerous hemorrhagic follicles. Histopathology indicated cystic follicular degeneration and bacterial hepatitis.

Color 29.23

Grayish-yellow, nodular follicles in a gallinaceous hen with a Marek's disease virus-induced ovarian neoplasm (courtesy of R. Korbel).

Color 29.24

a) Ovary and oviduct from a normal 22-month-old ostrich hen. Note the size of the oviduct and suspensory ligament. **b)** Close-up view of the ovary showing several follicles that are beginning to mature (courtesy of Brett Hopkins).

Color 29.25

A reproductively active Sun Conure was found dead in the nest box. The bird was in excellent overall condition and had delivered a normal fertile egg two days prior to presentation. The liver was enlarged, friable and congested. Histopathology indicated acute gram-negative bacterial hepatitis. Several active ovarian follicles (arrow) and the size of the oviduct (open arrows) in a reproductively active hen are evident.

Color 29.26

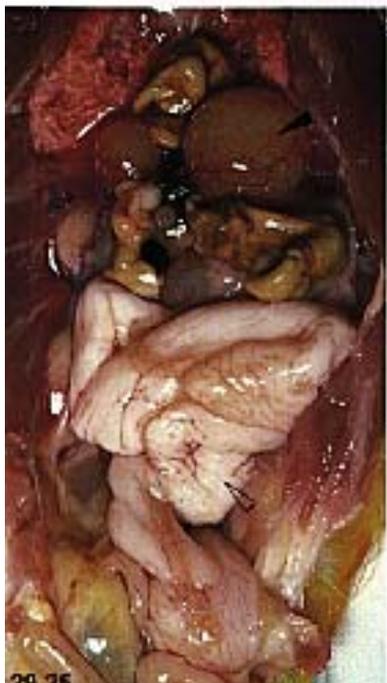
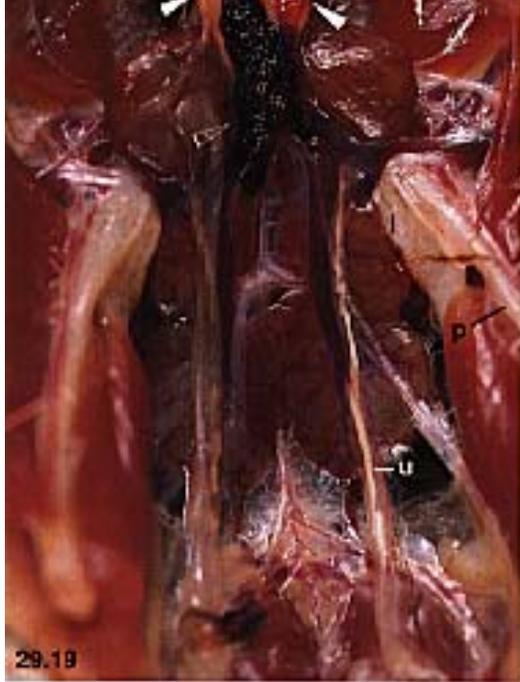
An Amazon parrot hen was presented for necropsy following several days of anorexia, depression and straining to defecate. A firm mass was present in the caudal abdomen. Necropsy indicated the retention of an egg (open arrow) in the caudal portion of the uterus. The degree of dilatation and hyperemia of the uterus (arrow) are evident. Histopathology indicated bacterial salpingitis.

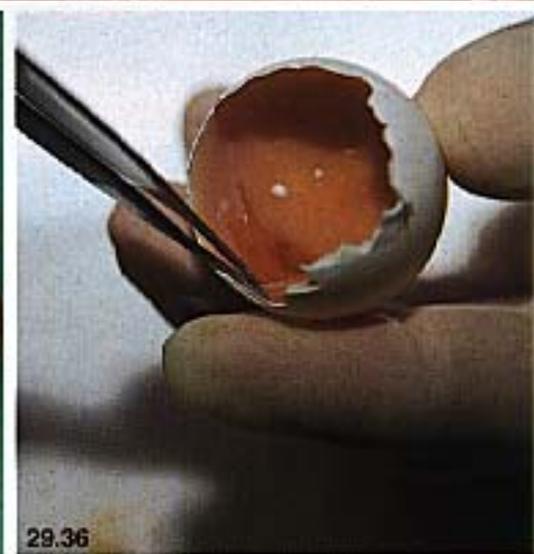
Color 29.27

An Umbrella Cockatoo hen was presented with a three-day history of depression following the delivery of its first egg of the season. Radiographs indicated a granular, soft tissue opacity in the intestinal peritoneal cavity that was pushing the proventriculus and ventriculus cranially. Clinicopathologic changes included WBC=35,000 (toxic heterophils), AST=1500, LDH=1200 and calcium=8 mg/dl. An exploratory laparotomy indicated diffuse peritonitis with adhesions throughout most of the abdominal cavity. The bird was euthanized. At necropsy, necrotic, brown, fibrous, peritonitis-related material was located on most of the abdominal organs.

Color 29.28

Egg-related peritonitis in an Amazon parrot hen with chlamydiosis. The bacterial peritonitis was considered to have occurred secondary to the chlamydial infection.





Theriogenology

Color 29.29

Typical presentation of an impacted egg. In this conure, the excessively large egg was lodged in the caudal uterus and vagina. The egg was removed by performing ovocentesis through the cloaca and collapsing the egg.

Color 29.30

a) An impacted egg in the cloaca of a budgerigar. Note the vagina (arrow). **b)** The egg could be seen through the vent and was removed by performing ovocentesis and gently removing the fractured portions of the eggshell.

Color 29.31

A mature cockatiel hen was presented with a one-day history of depression and tenesmus. The client became extremely concerned when blood was noted in association with a mass protruding from the cloaca. The hen had a prolapsed uterus that contained an egg. The egg was removed and the uterus was coated with a steroid-containing antibiotic ointment and was gently replaced in the cloaca with a moistened cotton-tipped applicator. Retention sutures were placed on both sides of the cloaca. These sutures were removed two days later, and the hen had no further problems.

Color 29.32

Mild prolapse of the vagina in the immediate post-oviposition period (courtesy of Kim Joyner).

Color 29.33

a) A necropsy should be performed on every egg that fails to hatch. In this case, a Moluccan Cockatoo embryo pipped three days earlier than expected. The embryo was properly positioned, but the excessively large embryo was preventing the development of a normal air cell. **b)** Hyperemia in newly hatched chicks is characteristic of dehydration or septicemia. In this exces-

sively large (32 g) Moluccan Cockatoo embryo, the hyperemia was believed to have been caused by struggling in the egg and anoxia.

Color 29.34

Malposition 2 in an Umbrella Cockatoo chick. Note that the head is positioned at the pointed end of the egg opposite the air cell (courtesy of Kim Joyner).

Color 29.35

Soft-shelled eggs with depigmentation from a gallinaceous hen with salpingitis (courtesy of R. Korbelt).

Color 29.36

During an egg necropsy, the membranes should be gently peeled away using fine-tipped forceps to ensure that all underlying structures are examined (courtesy of Kim Joyner).

Color 29.37

Fungal infection in a macaw egg. Note the proliferative growths on the eggshell membranes (courtesy of Kim Joyner).

Color 29.38

Examination of a dead-in-shell embryo should be performed sterily to allow the collection of diagnostic culture samples. In this case, the egg has been opened and its contents have been placed in a sterile petri dish for further evaluation. The partially autolyzed, 20-day-old embryo and the yolk sac are easily distinguishable. Note the blood-tinged fluid, indicative of hemolysis.

Color 29.39

A dead-in-shell Eclectus Parrot embryo with hemorrhage of the liver and a ruptured yolk sac. These are common findings in embryos from bacterially contaminated eggs. Note the well developed pipping muscle that is a major storage site of lymph in the developing embryo.

of air (depending upon the size of the egg) may be injected into the egg through the original pip site to infiltrate under the membrane and expand it in any areas not trapped by the shell.

The egg should then be returned to the incubator with the pip site elevated at a 45° angle. Air should be injected through the pip site every two hours for the first day. On the second day, the pip site should be enlarged. The membranes should be left dry allowing the shell to separate from the membranes more easily. During the second and third days, the membrane should be gently and very gradually torn around the pip site allowing vessels to retract between manipulations. Eventually, as the shell is removed from the small end of the egg, the yolk sac should be visualized to determine if it has retracted. Once the end of the shell and its associated membranes are removed and the yolk has retracted, the chick will usually emerge without further assistance.

Altricial birds have a relatively small yolk sac at hatching because the parent birds begin to feed the hatchlings almost immediately. Conversely, precocial birds have a relatively large internal yolk sac because they leave the nest soon after hatching. Over the subsequent several days they learn to select food items by observing the parent birds. During this time period, they maintain their nitrogen balance with the aid of the residual internalized yolk sack. The internalized yolk sac of altricial birds comprises five to ten percent of their total body weight and of precocial birds is 12 to 25%. Additionally, altricial birds use their internalized yolk sac faster than precocial birds.

Unabsorbed yolk sacs are best left unattended and allowed to fully retract. This may require leaving a chick in the egg for several hours longer than normal so that the shell protects this fragile sac. Small umbilical protuberances can generally be ignored although the chick should be handled carefully until the umbilicus is sealed. Frequent application of disinfectants such as iodine solutions will prevent infec-

tions of the umbilicus and yolk sac. Larger protuberances can be carefully placed into the abdomen with the aid of a swab dipped in a water-based sterile ointment. The umbilicus is then sutured or surgically sealed with glue.

Surgical ligation and removal of the yolk sac may be needed in cases with a persistent or very large external yolk sac (see Chapter 48). Chicks that require amputation of the yolk sac can survive but have higher mortality levels. The chick is anesthetized with isoflurane to prevent traumatic injuries to the yolk sac and a hemostatic clip is applied to the umbilicus between the chick and the yolk sac. Two sutures (8-0 to 10-0) are placed to aid in closure of the umbilical opening with care taken to place them shallow enough to avoid penetrating umbilical vessels. The hemostatic clip is outside the body and an occlusive dressing is applied to protect the umbilicus. Occasionally, herniation of intestinal contents can occur through the umbilical opening while the chick is still within the egg. The prognosis is poor in these cases, although surgical resolution of the hernia should be attempted. Exteriorized tissues should be adequately cleaned with sterile saline and kept moist with the application of ointments if necessary. Umbilical openings can be surgically enlarged if necessary to replace herniated intestines (see Chapter 41).⁶⁸

Appreciation is extended to G. J. Harrison and R. Avery Bennett for detailing the surgical aspects of assisted hatches.

■ **Laboratories Mentioned in the Text**

- a. Avian Genetic Sexing Laboratory, Barlette, TN
- b. Zoogen Inc., Davis, CA
- c. A.U.D. Laboratory, Aztec, NM

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