

Rhabdomyosarcoma

Skeletal muscle differentiation in a malignant mesenchymal tumor

Histopathology • Molecular pathogenesis • Clinicopathologic correlation

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Learning objectives

- Identify the four rhabdomyosarcoma subtypes and their typical age distributions.
- Recognize subtype-defining architecture, cytology, and skeletal-muscle differentiation.
- Connect PAX::FOXO1 fusions, RAS-pathway activation, and MYOD1 alterations to phenotype.
- Explain why subtype, site, and fusion status matter for clinical interpretation.

Definition: Malignant skeletal muscle differentiation

Rhabdomyosarcoma is a malignant mesenchymal neoplasm that exhibits skeletal-muscle differentiation.

- “Rhabdo-” refers to striated / skeletal muscle lineage.
- Tumor location need not contain mature skeletal muscle.
- Lineage assignment depends on morphology and, when needed, myogenic immunophenotype.

Skeletal muscle tumors: Malignant predominance

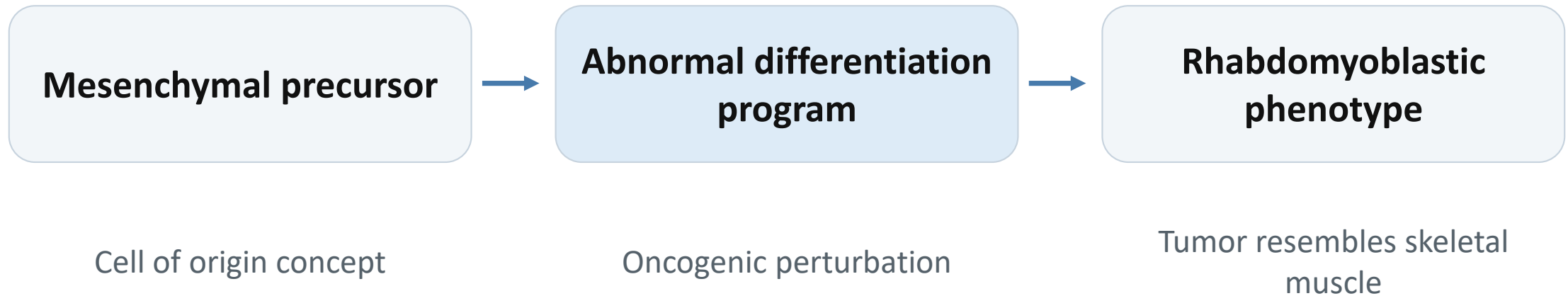
Rhabdomyosarcoma

- Malignant mesenchymal tumor
- Multiple histologic and molecular classes
- Important pediatric soft-tissue sarcoma

Rhabdomyoma

- Benign skeletal-muscle tumor
- May arise in heart or soft tissues
- Frequently associated with tuberous sclerosis

Lineage does not imply mature tissue of origin



Characteristic pediatric sites

Sinuses

Head and neck

Genitourinary tract

Rhabdomyosarcoma can arise where normal mature skeletal muscle is sparse.

The pediatric–adult subtype divide

Common in younger patients

- Embryonal rhabdomyosarcoma
- Alveolar rhabdomyosarcoma
- Usually present before age 20

Older-age pattern

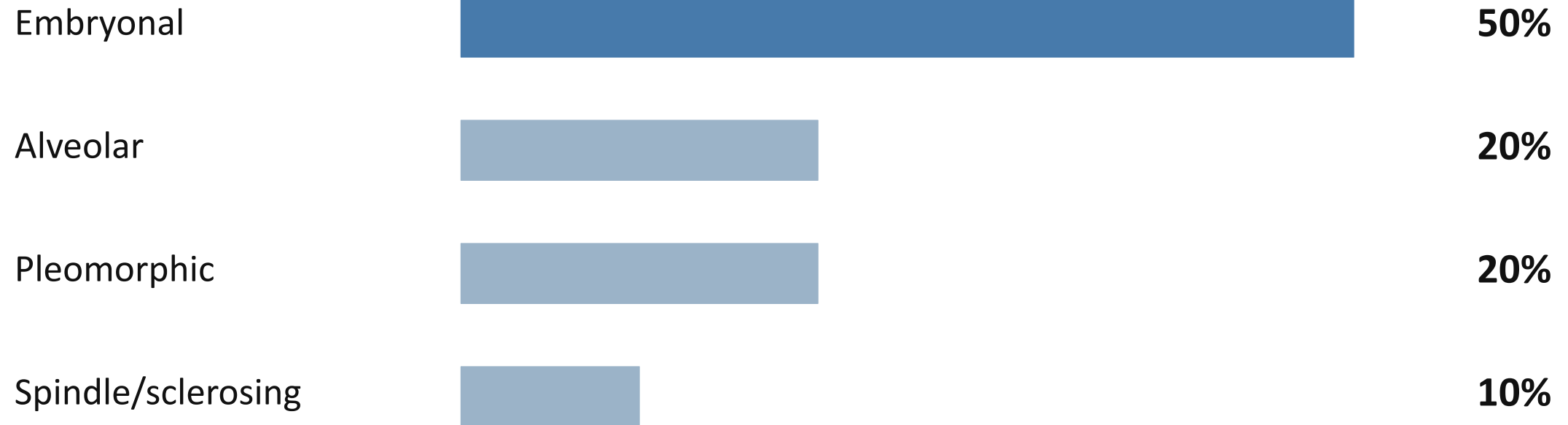
- Pleomorphic rhabdomyosarcoma: Adults
- Spindle cell/sclerosing: Across age groups
- Subtype changes the clinical context

Four major histologic categories

Subtype	Distinctive finding / teaching clue
Embryonal	Primitive round/spindle cells in myxoid stroma
Alveolar	Fibrous septa; Discohesive central cells
Pleomorphic	Bizarre, sometimes multinucleated cells
Spindle cell/sclerosing	Fascicles / storiform cells $\pm$ dense collagen

Subtype proportions

Percent of cases in the Robbins four-subtype overview



These are the approximate proportions stated for the four-category textbook overview; They are not pediatric-only proportions.

Do not transfer proportions between cohorts

Textbook overview

- Embryonal 50%; Alveolar 20%
- Pleomorphic 20%; Spindle/sclerosing 10%
- General subtype summary

Pediatric-specific NCI framing

- Childhood subtype mix differs
- Embryonal forms a larger pediatric share
- Cohort, age range, and classification affect proportions

Subtype-specific molecular mechanisms

Alveolar

PAX3/7::FOXO1

Embryonal

RAS pathway

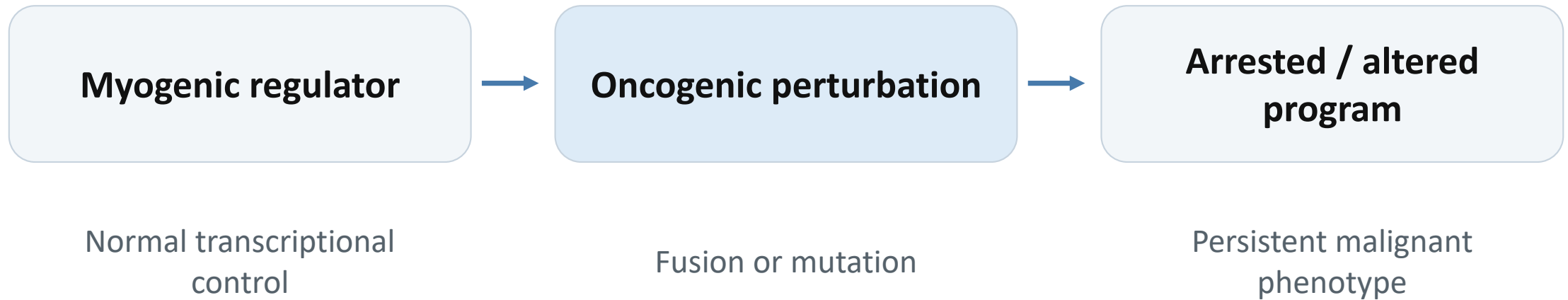
Spindle/sclerosing

MYOD1 in subset

Pleomorphic

Heterogeneous aberrations

Why molecular differentiation matters



Alveolar rhabdomyosarcoma: FOXO1 fusions

Recurrent rearrangement

PAX3 or PAX7

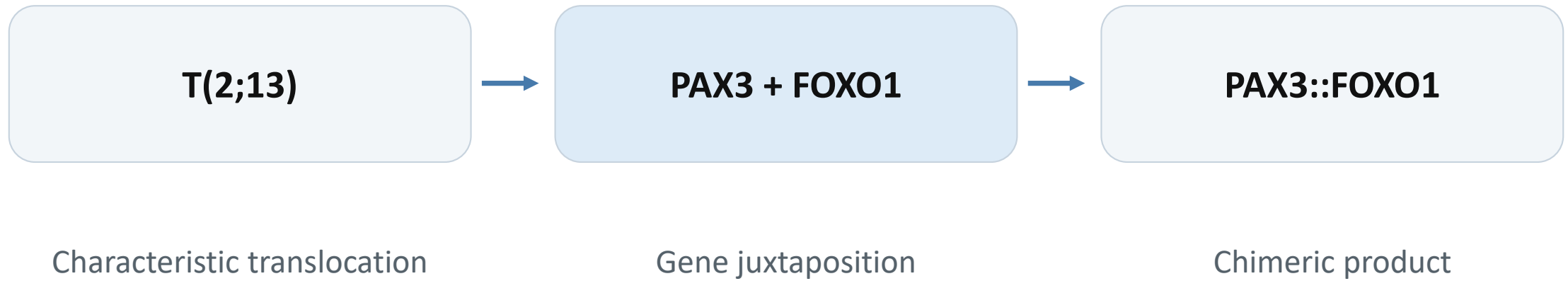


FOXO1

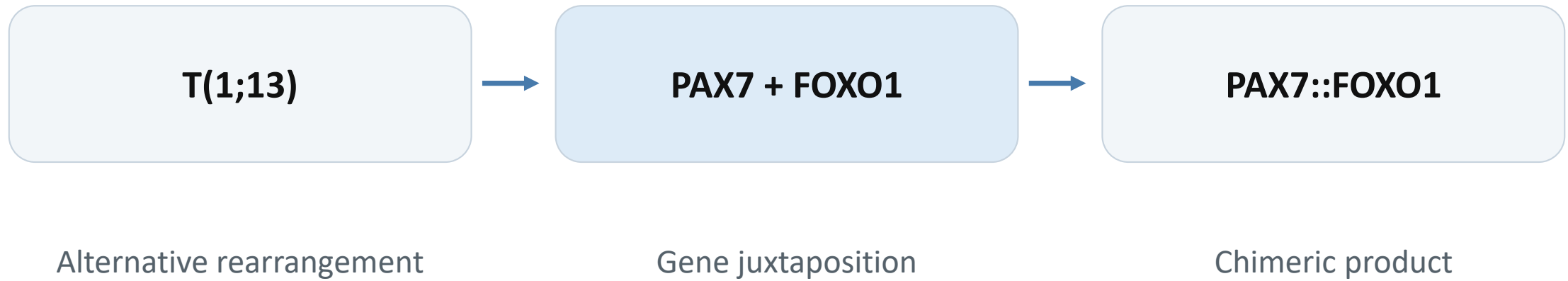
PAX3::FOXO1 / PAX7::FOXO1

Chimeric transcriptional regulator; Interference with myogenic differentiation

PAX3::FOXO1: Chromosomal origin



PAX7::FOXO1: Chromosomal origin



PAX3 and normal skeletal muscle programming

PAX3 is a transcription factor involved in initiating skeletal muscle differentiation.

- A transcription factor regulates gene-expression programs.
- Fusion to FOXO1 changes the regulatory context.
- The textbook links the chimeric product to impaired differentiation.

Fusion-driven differentiation arrest

PAX::FOXO1 fusion

Genomic rearrangement



**Abnormal transcriptional
control**

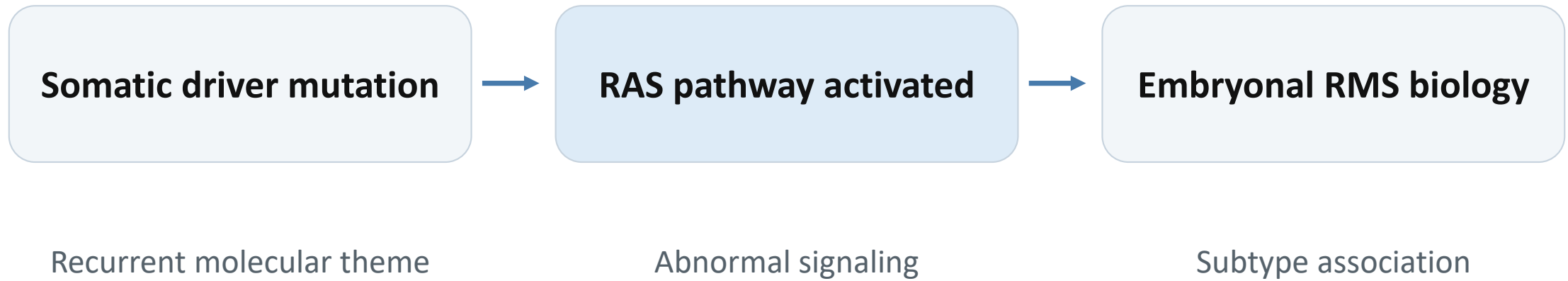
Chimeric regulator



**Impaired myogenic
maturation**

Persistence of immature
tumor phenotype

Embryonal: RAS-pathway activation



MYOD1 and spindle/sclerosing tumors

Mutation in a subset

MYOD1



Myogenic program

Altered myogenic regulation

The association is subtype-specific but not universal: Not every spindle/sclerosing RMS has a MYOD1 mutation.

Pleomorphic RMS lacks a single defining fusion

Pleomorphic rhabdomyosarcoma is genetically heterogeneous.

- Multiple genomic abnormalities may coexist.
- The textbook distinguishes its molecular landscape from the other three types.
- Do not infer a recurrent PAX::FOXO1 fusion merely from pleomorphic cytology.

Molecular associations are not absolutes

Specific association

- Alveolar: FOXO1 fusion often present
- Spindle/sclerosing: MYOD1 in a subset
- Embryonal: Frequent RAS activation

Diagnostic caution

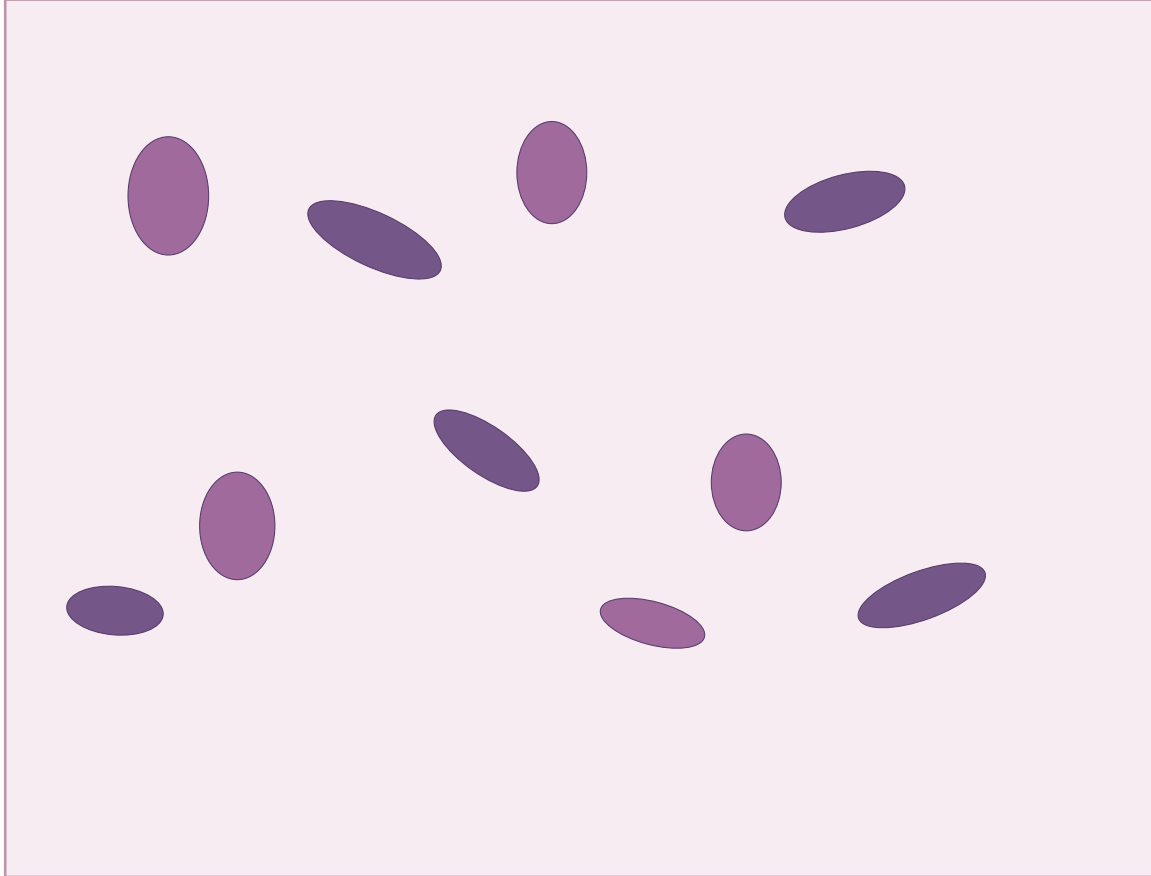
- “Frequently” does not mean invariably
- Histology and molecular status may diverge
- Integrate morphology, phenotype, and molecular findings

Embryonal RMS: Gross appearance

Soft, gray, infiltrative mass.

- The macroscopic phenotype is nonspecific.
- Infiltration illustrates the malignant behavior of the lesion.
- Microscopy is required to establish subtype and differentiation.

Embryonal RMS: Primitive cell population



- Round primitive cells
- Spindled primitive cells
- Myxoid background

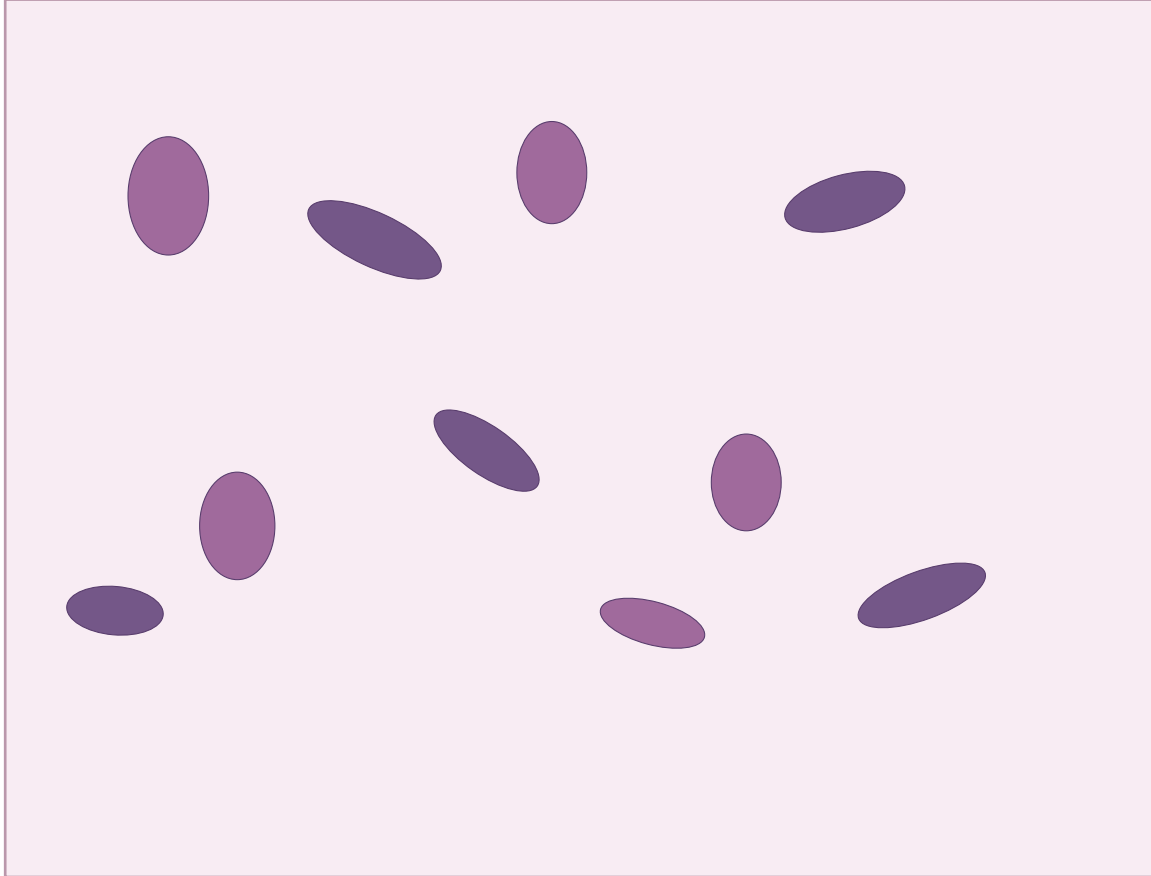
Schematic representation of the mixed round/spindle pattern.

Why “embryonal” is morphologically appropriate

Tumor cells recapitulate multiple stages of skeletal muscle differentiation.

- Primitive immature elements coexist with more differentiated cells.
- Round and spindle morphologies occur in the same tumor.
- Recognizable rhabdomyoblasts may appear within the population.

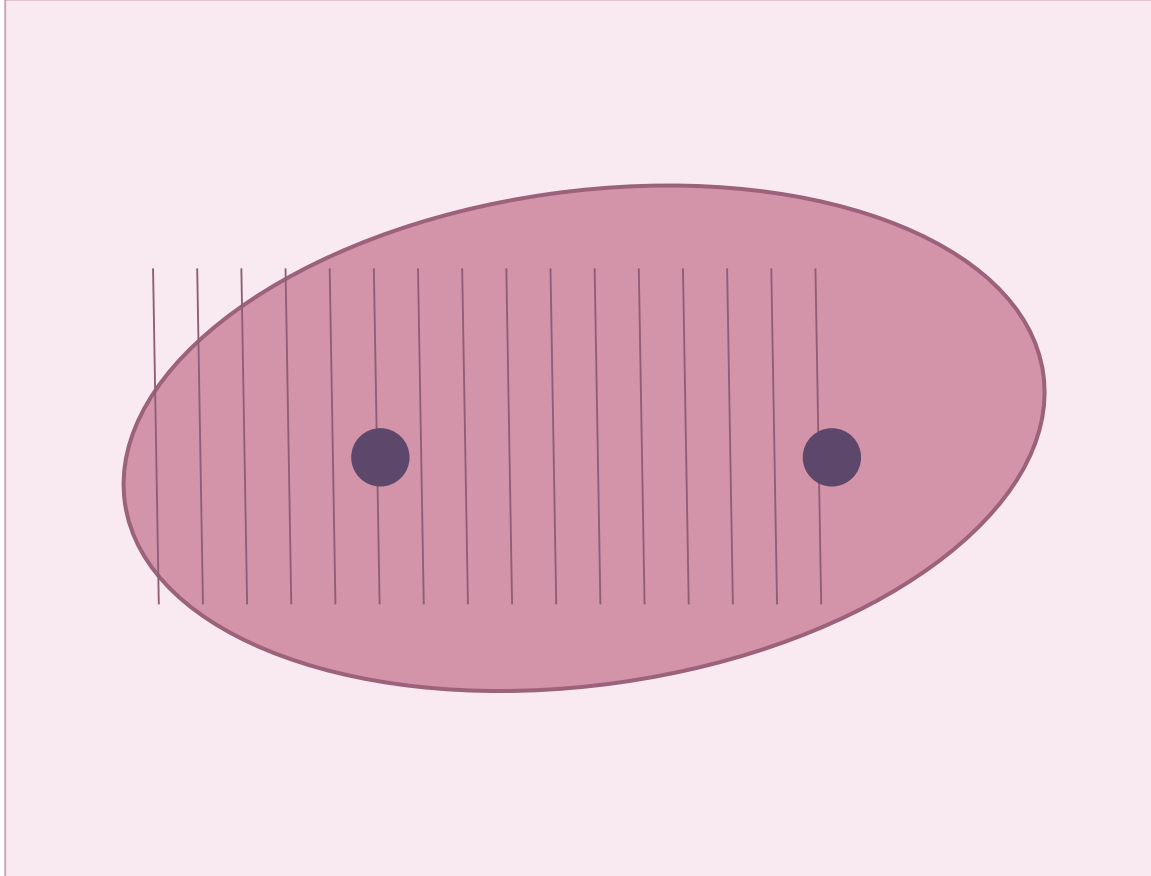
The myxoid stroma in embryonal RMS



- Loose myxoid matrix
- Scattered tumor cells
- Variable cellular maturity

Observe tissue context, not just isolated cell shapes.

Rhabdomyoblasts: A differentiation clue



- Eosinophilic cytoplasm
- Developing skeletal muscle phenotype
- Cross-striations may be visible

Cross-striations are supportive when identified; They are not required in every section.

Cross-striations are not always demonstrable

When seen

- Direct morphologic evidence of skeletal-muscle differentiation
- Can be decisive in the correct context

When absent

- Does not by itself exclude RMS
- Seek architecture and myogenic immunophenotype

Embryonal RMS: Integrated morphology

- Soft, gray, infiltrative gross mass.
- Variable primitive round and spindle cells.
- Myxoid stromal background.
- Variable rhabdomyoblastic maturation; Occasional visible cross-striations.

Sarcoma botryoides is an embryonal variant

Embryonal RMS

Shared differentiation



Hollow viscus wall

Bladder or vagina



Botryoid growth pattern

Grapelike polypoid masses

Botryoid RMS: Classic anatomical settings

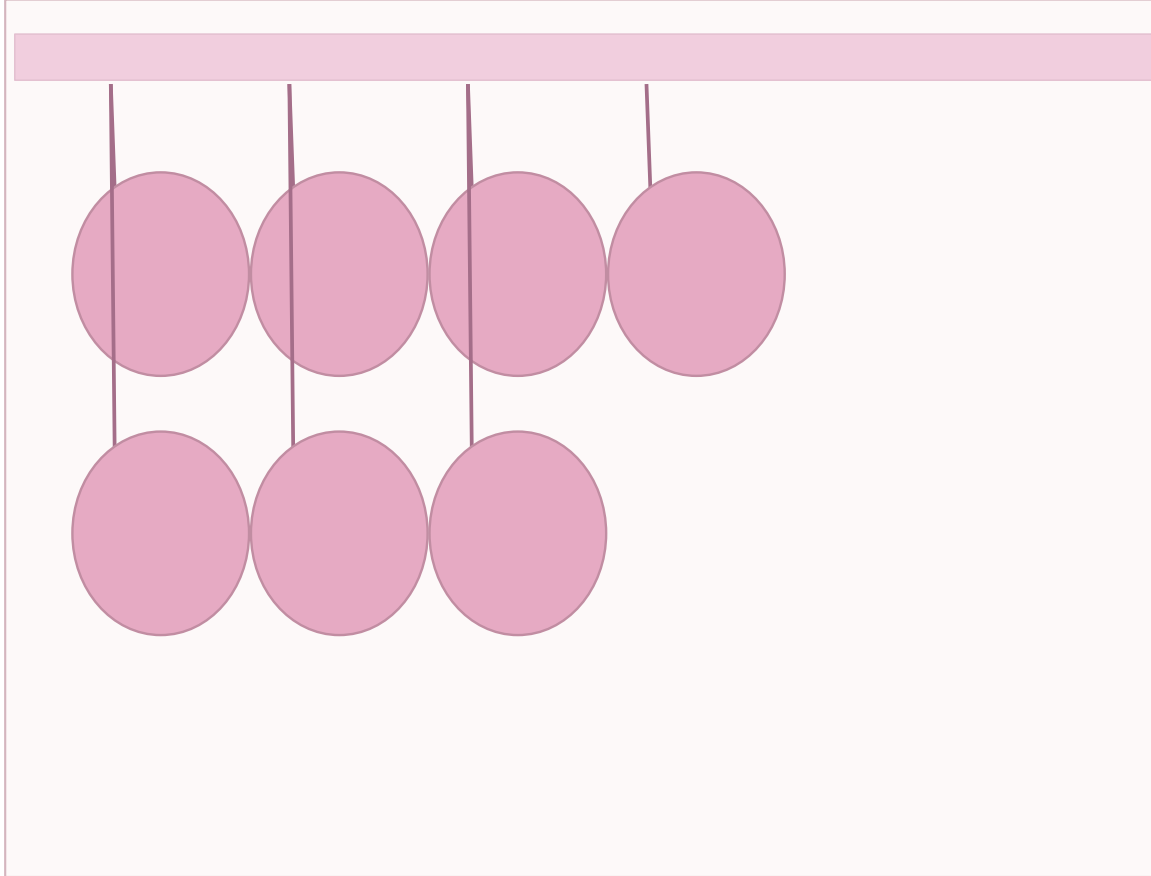
Genitourinary locations

- Vagina in infants / young children
- Urinary bladder wall
- Other hollow-viscus settings

Why location helps

- Subepithelial or wall-based tumor
- Lumen permits a polypoid mass
- Gross pattern suggests the variant

Botryoid RMS: Polypoid architecture



- Surface epithelium
- Grapelike polypoid lobules
- Underlying tumor-bearing wall

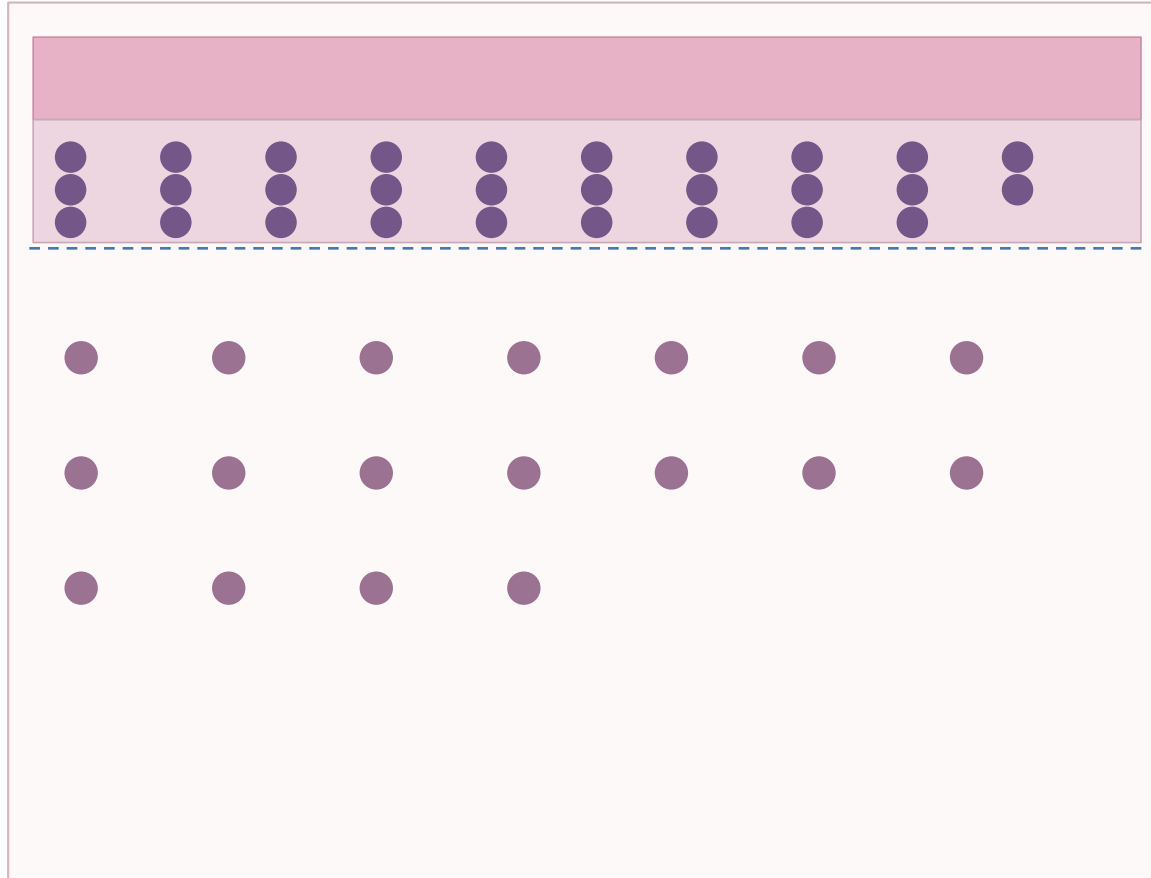
Schematic anatomy; Not a gross specimen.

Vaginal botryoid RMS: Age and gross pattern

The Chapter 22 vaginal example most often occurs in infants and children younger than 5 years.

- Often a bulky polypoid / rounded mass.
- Grapelike clustering explains the historical name “botryoides.”
- Appearance is not sufficient to establish the diagnosis.

Botryoid RMS: The cambium layer



- Surface epithelium
- Subepithelial cellular condensation
- Deeper edematous stroma

The cambium layer is a concentrated subepithelial tumor-cell zone.

Botryoid RMS: Cells in a loose deep stroma

Deeper tumor cells may lie in an edematous fibromyxoid matrix.

- The subepithelial zone is more cellular (cambium layer).
- An inflammatory infiltrate may accompany the loose stroma.
- This variation creates a potential benign-polyp mimic.

Botryoid RMS can resemble an inflammatory polyp

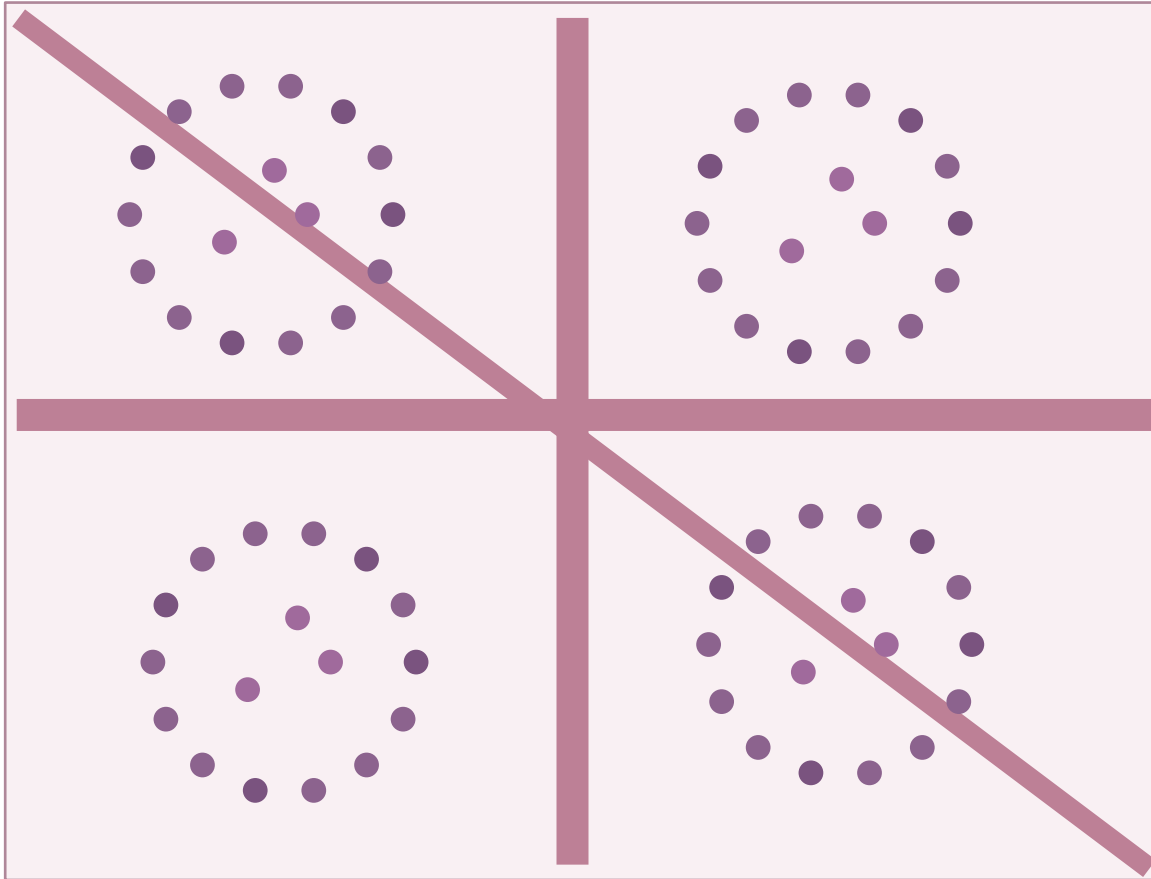
Potential pitfall

- Polypoid vaginal lesion
- Edematous matrix
- Inflammatory cells

Features that demand attention

- Subepithelial primitive-cell condensation
- Malignant rhabdomyoblasts
- Integrated tissue sampling and interpretation

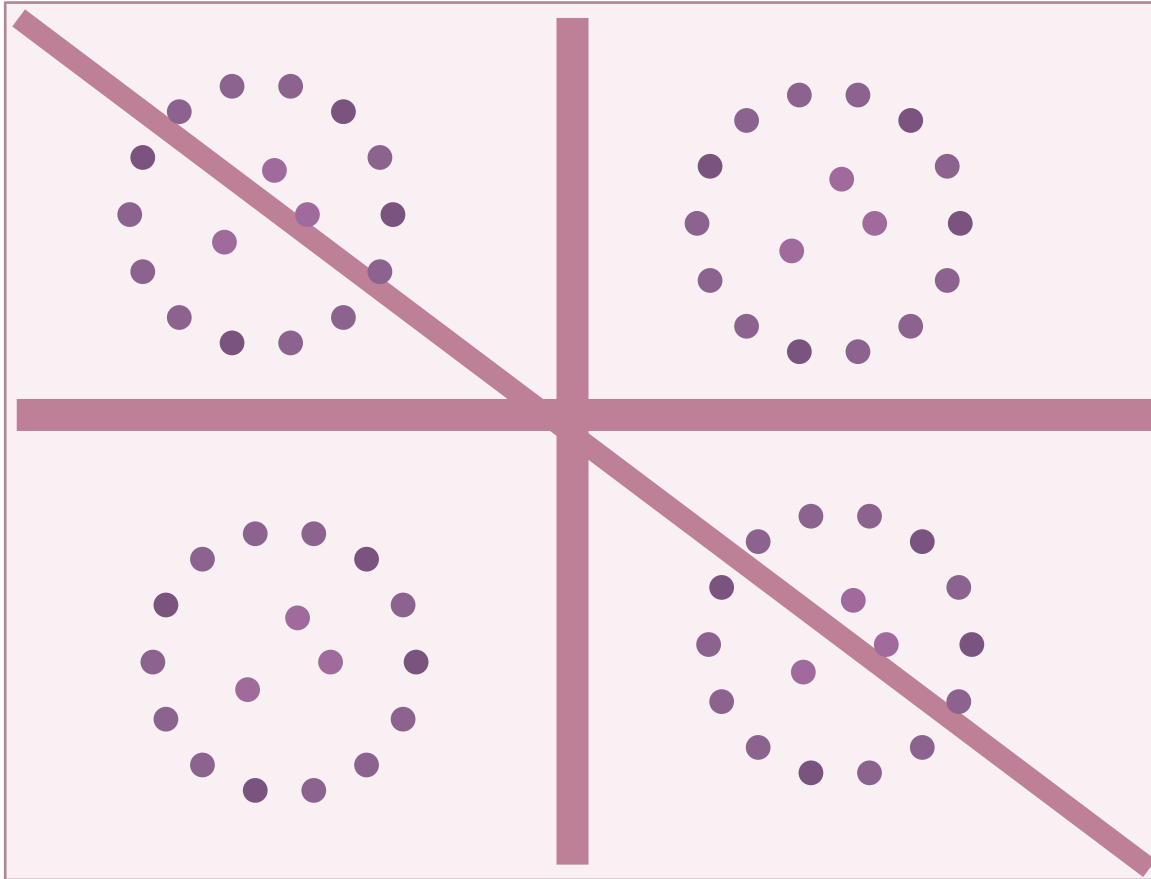
Alveolar RMS: Architecture explains its name



- Fibrous septum
- Cell aggregates
- Central loss of cohesion

"Alveolar" describes the architecture, not pulmonary origin.

Fibrous septa compartmentalize tumor cells



- Intervening septa
- Peripheral attached cells
- Central loose tumor cells

At low power, scan for septa surrounding tumor-cell nests.

Central discohesion is diagnostically useful

Central zone

- Tumor cells become minimally cohesive
- Spaces form within aggregates
- Cells may appear suspended

Peripheral zone

- Cells retain attachment to fibrous septa
- Creates the characteristic alveolar arrangement

Alveolar RMS: Cytologic profile

A relatively uniform round-cell population with scant cytoplasm.

- The architecture is as important as the cytology.
- Round, small-appearing malignant cells can resemble other neoplasms.
- Integrate myogenic immunophenotype and fusion information when available.

Embryonal versus alveolar RMS: Architecture

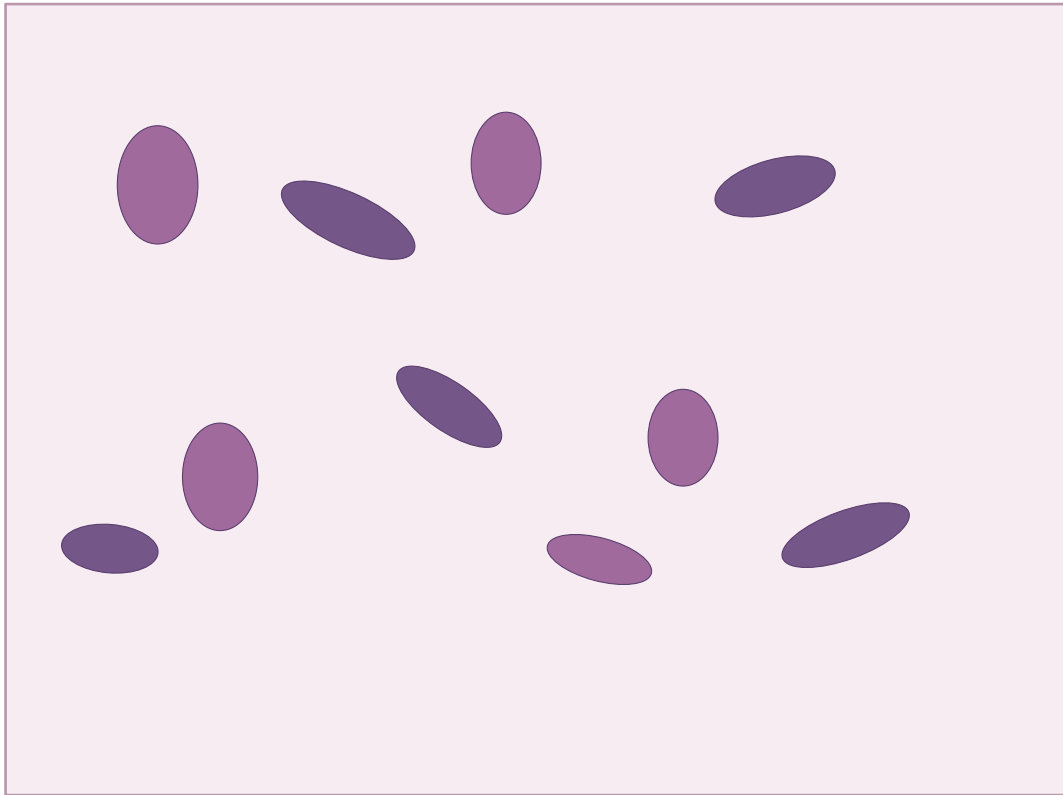
Embryonal

- Primitive round and spindle elements
- Myxoid background
- Variable stages of maturation

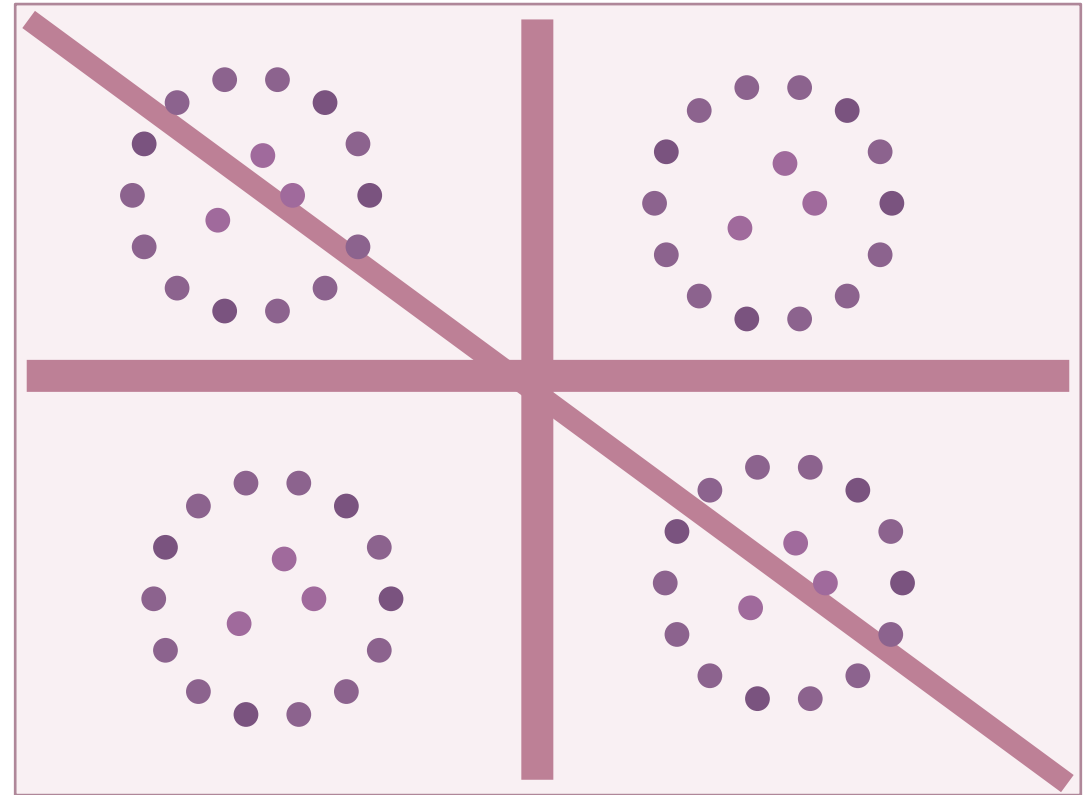
Alveolar

- Fibrous septa partition nests
- Central discohesion
- Uniform round cells with little cytoplasm

Embryonal versus alveolar RMS: Schematics



Embryonal: Mixed maturation



Alveolar: Septa and loose centers

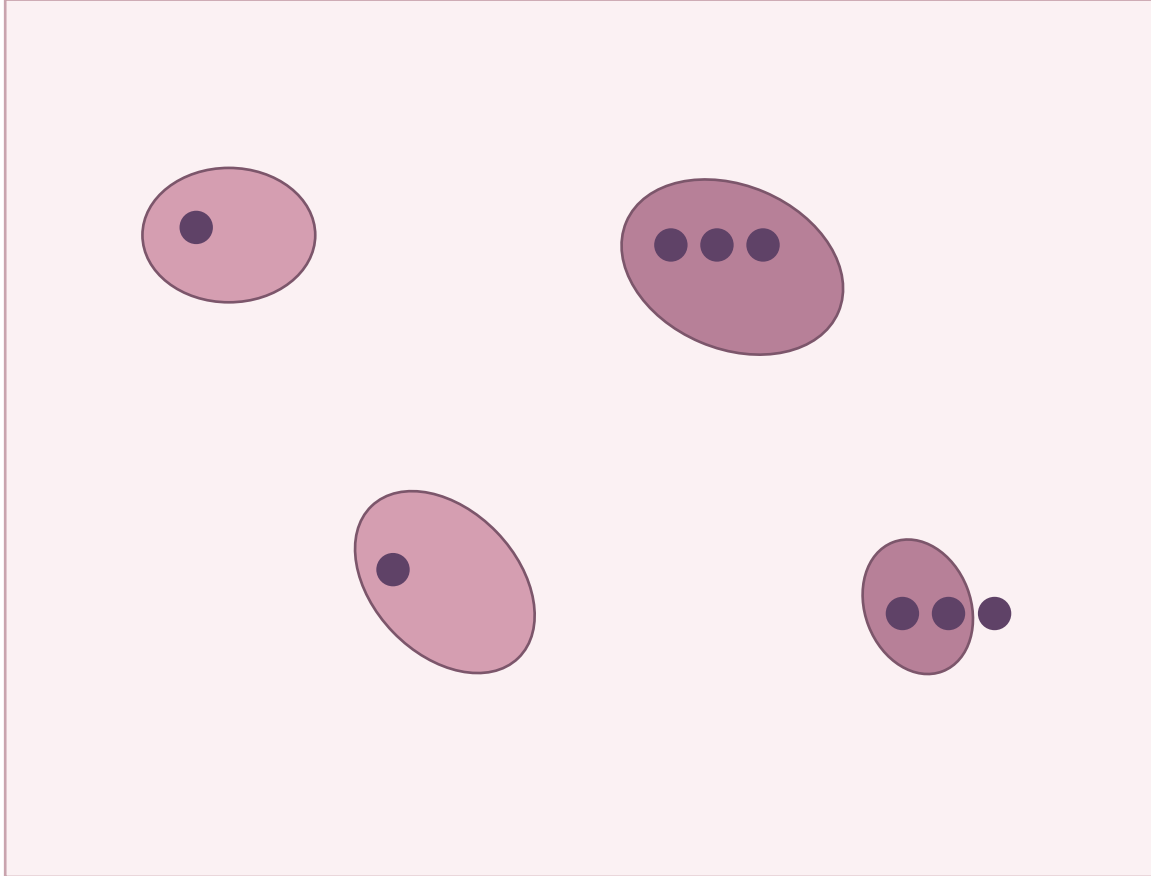
Original architectural schematics; Neither panel is a specimen.

Pleomorphic RMS: Adult tumor morphology

Large, sometimes multinucleated, bizarre eosinophilic tumor cells.

- Marked pleomorphism dominates the histology.
- A malignant spindle/pleomorphic morphology alone is not lineage-specific.
- Skeletal-muscle differentiation may require immunohistochemical confirmation.

Multinucleation and bizarre forms



- Large anaplastic tumor cell
- Multiple nuclei
- Eosinophilic cytoplasm

Stylized cytology; Do not interpret as an actual photomicrograph.

Myogenin helps confirm rhabdomyoblastic lineage

**Pleomorphic malignant
cells**

Morphologic overlap



Lineage uncertain on H&E

Differential remains broad



**Myogenin IHC supports
RMS**

Skeletal-muscle-associated
marker

Pleomorphic RMS: A differential diagnosis problem

Why difficult

- Highly atypical pleomorphic cells
- Histologic overlap with other pleomorphic sarcomas
- No single specific architectural pattern

Resolution

- Identify skeletal-muscle differentiation
- Use myogenin or appropriate myogenic immunophenotype
- Avoid diagnosis from pleomorphism alone

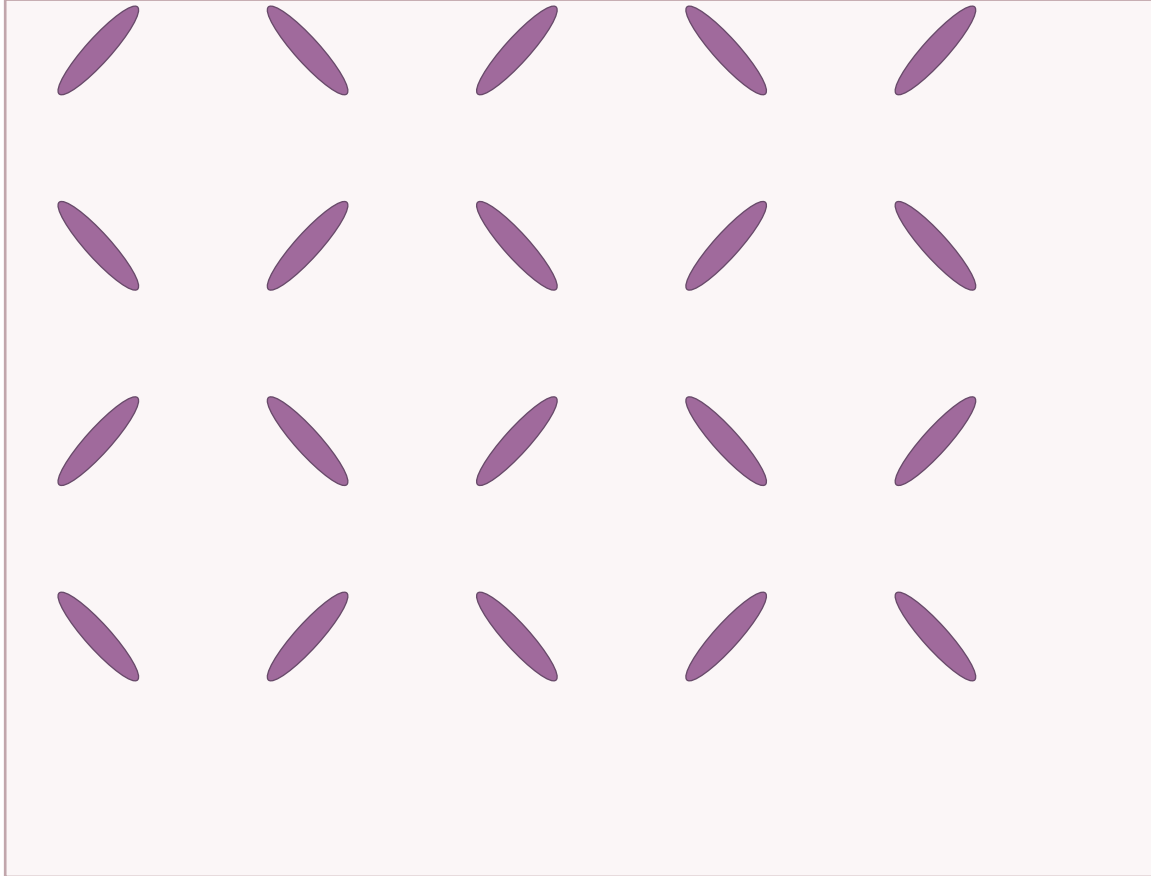
Spindle cell/sclerosing RMS: Fascicular pattern



- Fusiform tumor cells
- Long cellular fascicles
- Vesicular chromatin

Fascicular arrangement is one recognized histologic pattern.

A storiform pattern may also occur



- Intersecting cell bundles
- Storiform orientation
- Spindle-cell population

Illustrative arrangement only; Not diagnostic in isolation.

Sclerosing subtype: Collagen-rich background

Dense collagenous, sclerotic stroma may be especially prominent in adults.

- Fibrous matrix alters the appearance of the spindle-cell population.
- Rhabdomyoblasts may be encountered, but are not consistently conspicuous.
- Histology must be interpreted with the subtype-specific molecular context.

Occasional rhabdomyoblasts in spindle RMS

What to look for

- Fusiform malignant cells
- Long fascicles or storiform arrays
- Occasional cells with myogenic morphology

Interpretive limitation

- A spindle-cell pattern is nonspecific
- Absence of a visible rhabdomyoblast is not exclusionary
- Integrate myogenic lineage confirmation

Four subtypes: Morphologic discriminators

Subtype	Distinctive finding / teaching clue
Embryonal	Heterogeneous immature cells; Myxoid matrix
Alveolar	Fibrous septa; Central discohesion
Pleomorphic	Bizarre eosinophilic / multinucleated cells
Spindle/sclerosing	Fusiform fascicles / storiform; Collagen

From morphology to lineage confirmation

Recognize a malignant soft-tissue tumor



Identify dominant architectural / cytologic pattern



Seek skeletal-muscle differentiation



Assign a subtype using integrated evidence

Morphology and molecular data play different roles

Microscopy

- Identifies architecture and cell shape
- Defines degree/pattern of maturation
- Raises the subtype hypothesis

Ancillary testing

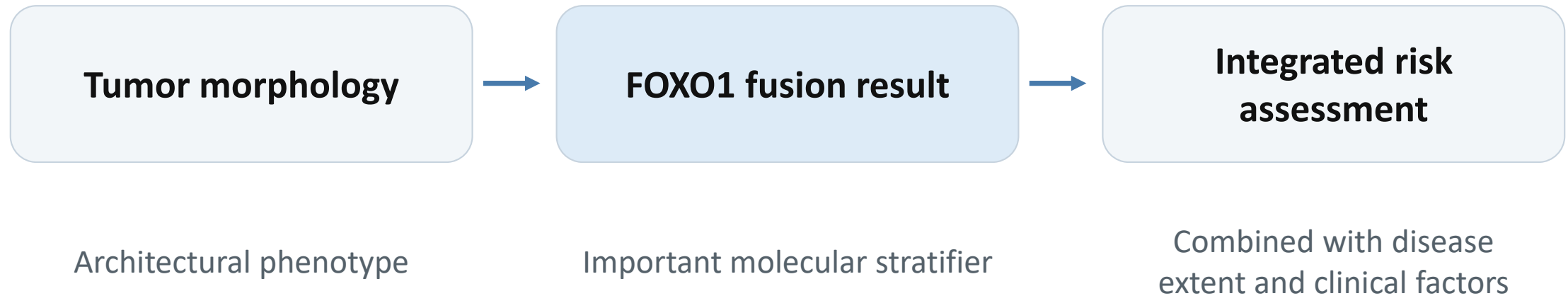
- Myogenin supports skeletal muscle lineage
- FOXO1 fusion status refines alveolar biology
- MYOD1 alteration in spindle/sclerosing subset

Fusion-negative alveolar morphology: An important caveat

Not every tumor showing alveolar histology has a FOXO1 fusion.

- NCI PDQ reports that fusion-negative alveolar-pattern tumors resemble embryonal RMS molecularly and clinically.
- Therefore, alveolar morphology and fusion-positive disease should not be treated as interchangeable.
- Avoid assigning risk from architecture alone.

FOXO1 fusion status carries prognostic information



A diagnostic biomarker is not automatically predictive

Myogenin

- Supports myogenic differentiation
- Primarily a lineage / diagnostic marker
- Does not alone quantify prognosis

FOXO1 fusion status

- Molecular association with alveolar RMS
- Contributes to risk stratification
- Not itself a treatment-response guarantee

Molecular findings require context

- FOXO1 fusion is strongly associated with alveolar rhabdomyosarcoma.
- RAS-pathway changes commonly occur in embryonal disease.
- MYOD1 alterations occur in only a subset of spindle/sclerosing lesions.
- No marker should override an internally inconsistent morphologic and clinical picture.

Clinical behavior: Aggressive malignant sarcoma

Rhabdomyosarcomas are aggressive neoplasms.

- Both local extent and disseminated disease matter clinically.
- Anatomic site influences feasibility of local treatment.
- Histologic subtype and site correlate with survival.

Risk assessment has multiple dimensions

Biology

Subtype and FOXO1

Extent

Metastatic status

Primary site

Anatomic risk

Patient factors

Age and resectability

Clinical factors beyond histologic subtype

- Age, tumor site, size, and resectability influence clinical risk.
- Nodal involvement and metastasis at presentation matter.
- FOXO1 fusion status is incorporated into contemporary pediatric risk assessment.
- A pathologic subtype is only one component of management decisions.

Primary location changes the therapeutic problem

Anatomical constraints

- Sinuses and head/neck structures
- Genitourinary organ preservation
- Potential difficulty achieving complete resection

Clinical consequence

- Local-control approach varies by site
- Multidisciplinary decisions are required
- Prognosis cannot be inferred from cell type alone

Core multimodal treatment principles

Surgical/local assessment



Systemic chemotherapy



Radiation when indicated

Resection when feasible

Addresses systemic risk

Selected for local control
needs

Favorable botryoid prognosis is relative

Textbook observation

- Botryoid variant has the most favorable prognosis
- Its typical location and recognizability aid clinical interpretation

Do not overgeneralize

- Still an invasive malignancy
- Stage and treatment remain important
- Not all embryonal lesions are botryoid

Pleomorphic RMS: An adverse adult pattern

Pleomorphic rhabdomyosarcoma often carries a grave prognosis.

- It is predominantly an adult subtype.
- Tumor cells may overlap morphologically with other high-grade pleomorphic sarcomas.
- Robbins characterizes the outcome as often fatal.

Pediatric outlook depends on fusion and extent

FOXO1 status is more informative than alveolar histology alone in modern pediatric risk assessment.

- Fusion-positive disease tends to have less favorable outcomes than fusion-negative RMS.
- Metastatic disease at diagnosis substantially worsens prognosis.
- Risk assignment also incorporates primary site, patient age, and surgical/pathologic group.

Case: Polypoid vaginal mass in a young child

A 3-year-old has a bulky, grapelike vaginal mass. The biopsy reveals small malignant cells beneath epithelium with a dense subepithelial zone and loose deeper stroma.

Diagnostic question

Which rhabdomyosarcoma variant and microscopic feature are most characteristic?

Case analysis: Botryoid embryonal RMS

Embryonal rhabdomyosarcoma, botryoid variant.

- Grapelike polypoid appearance in a hollow viscus.
- Characteristic subepithelial cambium layer.
- Must not be mistaken for a benign inflammatory polyp.

Case: Septated round-cell sarcoma

An adolescent has a soft-tissue malignancy composed of relatively uniform round cells. Fibrous septa divide tumor into aggregates with conspicuous central cellular discohesion.

Diagnostic question

What histologic subtype is suggested, and which fusion should be considered?

Case analysis: Alveolar RMS

Alveolar rhabdomyosarcoma.

- Fibrous septa and central discohesion define the classic architecture.
- Evaluate for PAX3::FOXO1 or PAX7::FOXO1 fusion.
- Fusion testing matters because not all alveolar-pattern tumors are fusion-positive.

Case: Pleomorphic adult sarcoma

An older adult has a high-grade soft-tissue tumor. The neoplasm consists of large bizarre eosinophilic cells, including multinucleated forms.

Diagnostic question

What lineage-confirming stain helps evaluate pleomorphic RMS?

Case analysis: Myogenin-supported lineage

Myogenin immunohistochemistry supports rhabdomyoblastic differentiation.

- Marked pleomorphism is not specific for RMS.
- The clinical and morphological pattern raises pleomorphic RMS.
- Lineage confirmation is important before final classification.

Case: Adult spindle-cell sarcoma

A soft-tissue tumor contains spindle-shaped malignant cells in long fascicles with areas of dense collagenous sclerosis.

Diagnostic question

Which RMS subtype should be considered, and what molecular alteration is associated with a subset?

Case analysis: Spindle cell/sclerosing RMS

Spindle cell/sclerosing rhabdomyosarcoma.

- Fascicular and sclerosing growth support the pattern.
- A subset has MYOD1 mutations.
- The spindle-cell phenotype alone is not sufficient for diagnosis.

Integrated comparison: Histology, gene, age

Subtype	Distinctive finding / teaching clue
Embryonal	Young; Primitive round/spindle; RAS activation
Alveolar	Young; Septated nests; PAX::FOXO1 often
Pleomorphic	Adults; Bizarre cells; Heterogeneous genetics
Spindle/sclerosing	All ages; Fusiform + collagen; MYOD1 subset

Five interpretive rules

- RMS is malignant and shows skeletal-muscle differentiation.
- Normal skeletal muscle is not required at the primary site.
- Subtype-specific morphology is a starting point, not the whole diagnosis.
- Myogenin supports myogenic lineage when morphology is ambiguous.
- FOXO1 fusion status adds substantial molecular and prognostic information.

Primary references and source boundaries

Robbins, Cotran & Kumar. Pathologic Basis of Disease. 11th ed. (2026). Chapter 26, pp. 1102–1103.

Robbins, Cotran & Kumar. Pathologic Basis of Disease. 11th ed. (2026). Chapter 22, p. 904 (botryoid vaginal RMS).

NCI. Childhood Rhabdomyosarcoma Treatment (PDQ®), Health Professional Version (supplementary pediatric interpretation).

NCI. Childhood Cancer Genomics (PDQ®), Health Professional Version (supplementary molecular interpretation).

Textbook subtype proportions apply to the textbook overview; pediatric-focused epidemiology may differ.