

Ewing Sarcoma

Bone and soft-tissue pathology · morphology · fusion biology · diagnosis

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Learning objectives

- Recognize Ewing sarcoma in its epidemiologic, anatomic, and clinical context.
- Explain the EWSR1::ETS fusion-driven pathogenesis and its diagnostic relevance.
- Connect imaging, gross findings, and small round cell histology.
- Construct an integrated differential and diagnostic confirmation strategy.
- Relate systemic treatment and histologic response to clinical outcomes.

Ewing sarcoma: Defining concept

- Aggressive malignant sarcoma of bone or soft tissue.
- Classically composed of relatively uniform primitive small round cells.
- Molecularly characterized by FET::ETS fusion genes, usually EWSR1::FLI1.
- Distinct from other genetically defined undifferentiated round-cell sarcomas.

Position among primary malignant bone tumors

Ewing sarcoma

- Primitive small round cell phenotype
- Characteristic gene fusion
- Usually no malignant osteoid or cartilage

Matrix-forming bone sarcomas

- Osteosarcoma: Malignant osteoid
- Chondrosarcoma: Malignant cartilage
- Matrix helps establish lineage

Frequency and rarity

- Robbins: Approximately 10% of primary malignant bone tumors.
- Second most common bone sarcoma in children, after osteosarcoma.
- Roughly 200 new diagnoses per year in the United States.
- Denominators matter: These are not population incidence rates.

Age and sex patterns

Pediatric predominance

Robbins: Approximately 80% of patients are younger than 20 years.

Peak clinical context

Consider Ewing sarcoma in adolescents with an aggressive destructive bone lesion.

Sex distribution

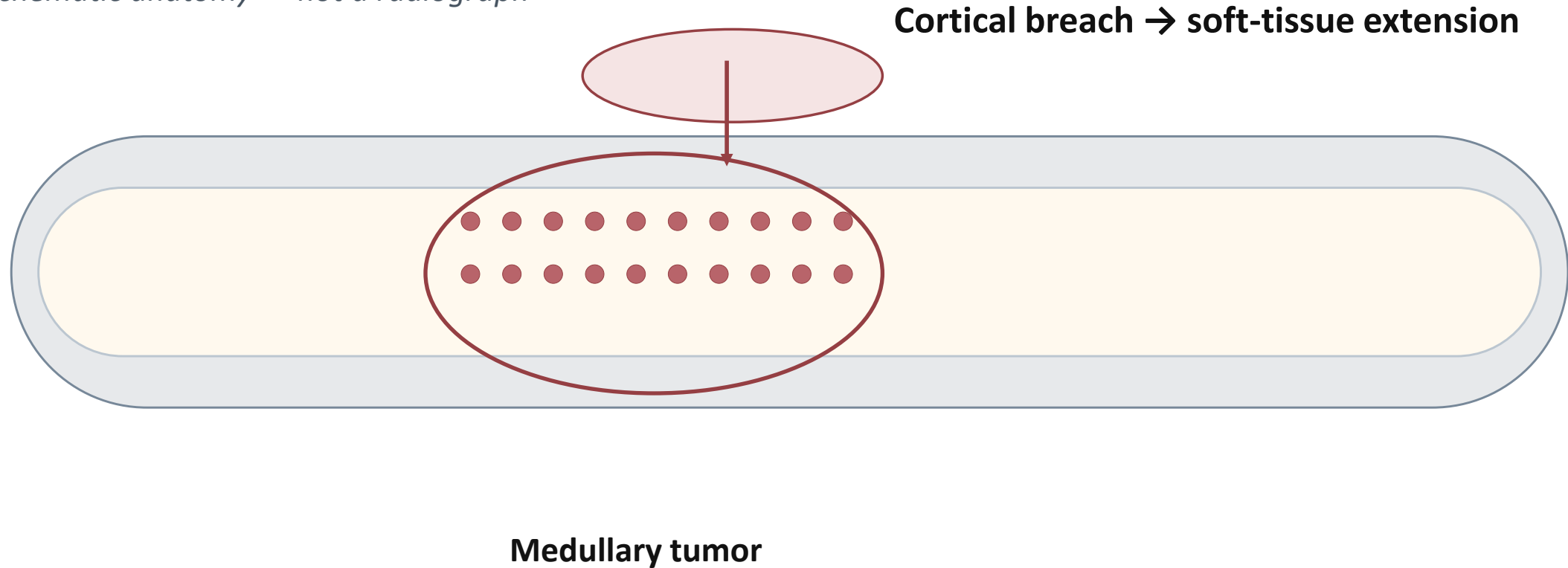
Boys are affected slightly more frequently than girls.

Common skeletal sites

- Diaphysis of long tubular bones is a characteristic location.
- Femur is a common long-bone primary site.
- Pelvis is an important flat-bone site.
- Anatomic site affects symptoms, biopsy planning, and local-control decisions.

Typical medullary origin and local extension

Schematic anatomy — not a radiograph



Extraskkeletal Ewing sarcoma

- Approximately 20% of cases arise outside bone (Robbins).
- Molecular entity remains Ewing sarcoma despite an extraosseous primary site.
- Clinical differential broadens when no osseous lesion is present.
- Tissue morphology and fusion identity become especially important.

How patients present

- Progressively painful and enlarging mass.
- Localized swelling, warmth, and tenderness are frequent.
- Symptoms reflect rapid bone destruction and surrounding tissue involvement.
- Painful symptoms may precede a clearly appreciated mass.

Ewing sarcoma can resemble infection

Shared findings

- Pain, warmth and local tenderness
- Fever may occur
- ESR elevation, anemia, or leukocytosis

Features requiring oncologic evaluation

- Destructive permeative bone lesion
- Associated enlarging soft-tissue mass
- Persistent symptoms with atypical course
- Biopsy needed for specific diagnosis

Presentation case: Pain, fever, and a bone lesion

Illustrative case

A 15-year-old has worsening thigh pain, swelling, intermittent fever, elevated ESR, and a permeative femoral lesion.

- Which clinical findings support inflammation but do not establish infection?
- Which radiographic features justify urgent specialist evaluation?
- Why must a biopsy be planned as part of definitive care?

Radiographs: An aggressive lytic pattern

- Destructive radiolucent lesion with ill-defined margins.
- Permeative or “moth-eaten” pattern indicates marrow-space infiltration.
- Tumor can breach cortex and extend beyond bone.
- The radiograph can suggest aggressiveness; It cannot specify the fusion.

Why permeative margins occur

Marrow tumor

Primitive tumor expands
within the medullary cavity



Trabecular destruction

Numerous irregular
microscopic foci coalesce

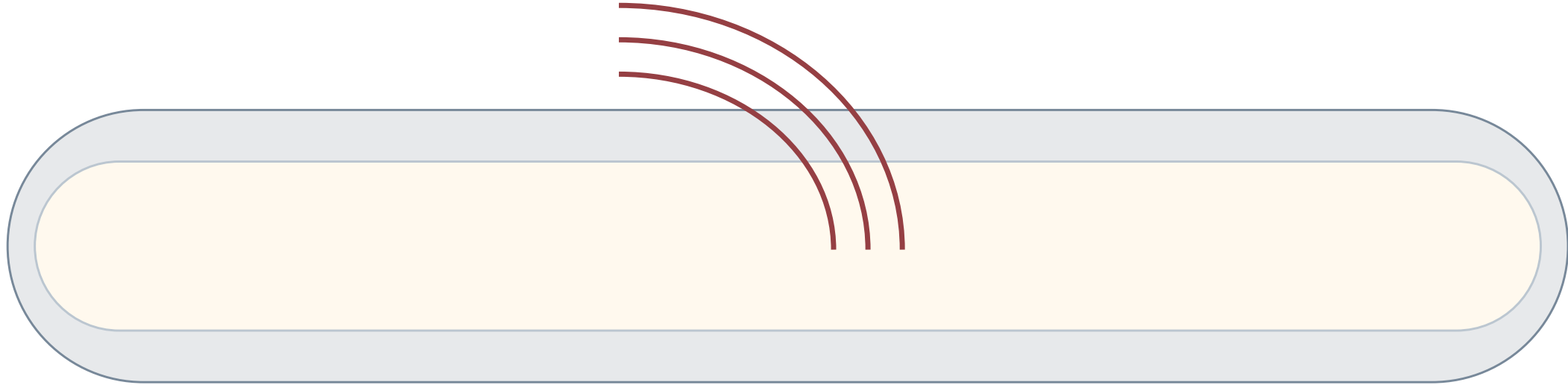


Ill-defined image

Radiographic margins appear
moth-eaten or permeative

Onion-skin periosteal reaction

Schematic anatomy — not a radiograph



Reactive periosteal lamellae

Schematic: Successive periosteal layers

Interpret the periosteal reaction

- Successive reactive layers form as tumor advances toward soft tissue.
- The laminated “onion-skin” response is a classic clue.
- A periosteal reaction is not pathognomonic for Ewing sarcoma.
- Integrate the pattern with age, tumor site, imaging, and pathology.

Imaging has three distinct purposes

Initial detection

Plain radiography characterizes the bone lesion and periosteal response.

Local extent

MRI depicts marrow, soft tissue, joint proximity, and neurovascular relations.

Distant staging

Chest CT and whole-body modalities assess metastatic dissemination.

Local imaging before tissue sampling

- MRI of the primary site is part of formal diagnostic evaluation.
- Define intramedullary extent and extraosseous components.
- Examine the full involved bone for possible discontinuous tumor deposits.
- Plan the biopsy tract with the surgical sarcoma team.

Clinical staging evaluation

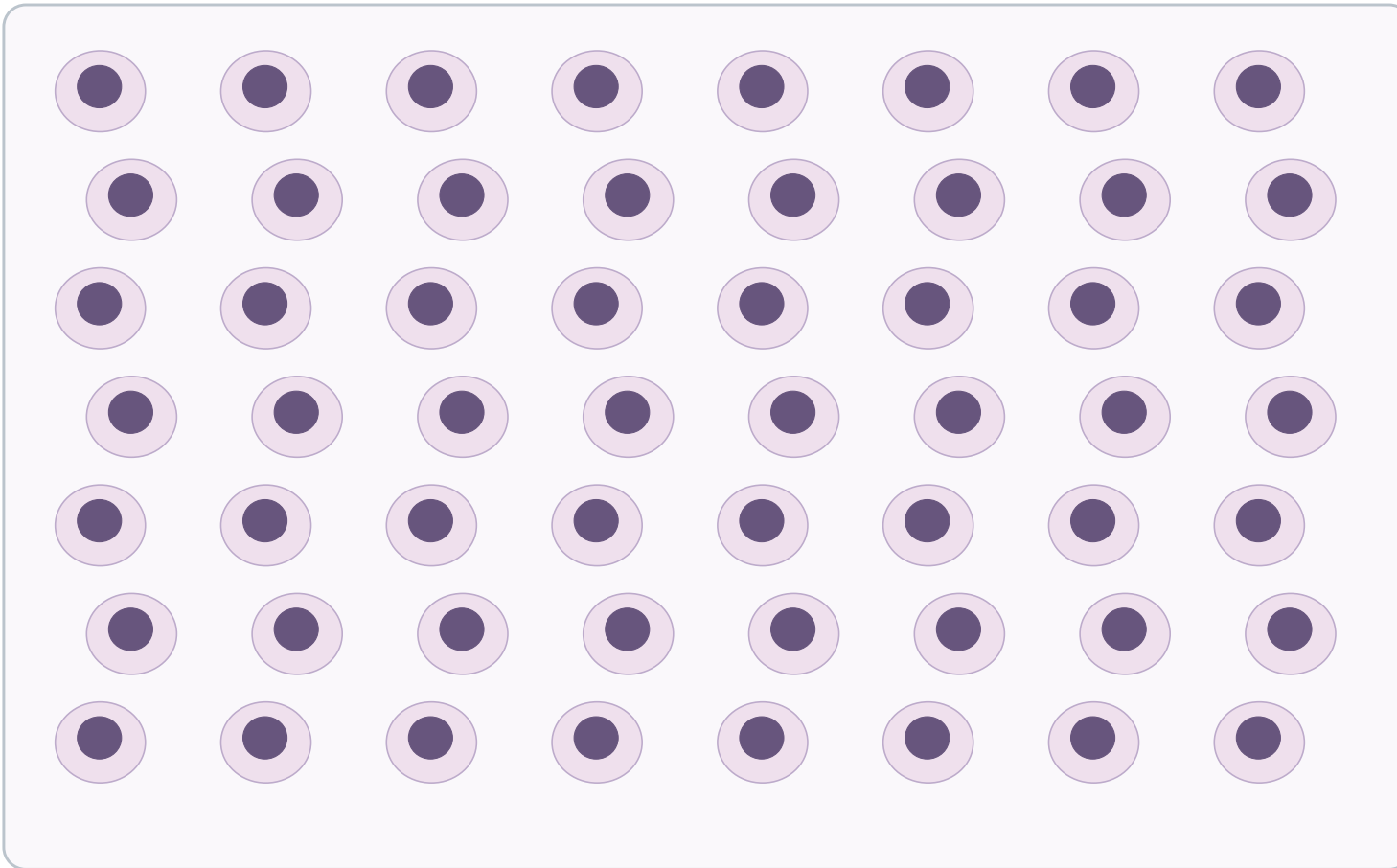
- Establish whether disease is localized or metastatic.
- Chest CT assesses pulmonary dissemination.
- PET and other body imaging are used to evaluate distant spread.
- Laboratory studies may include CBC and LDH; No blood test is diagnostic.

Gross morphology

- Typically centered in the medullary cavity.
- Soft tan-white tumor tissue replaces marrow and infiltrates cortex.
- Extension into periosteum and adjacent soft tissue is common.
- Hemorrhage and necrosis frequently contribute to gross heterogeneity.

Small round cell architecture: Conceptual map

Schematic only — not a photomicrograph



Uniform primitive round cells

- Relatively monotonous small round nuclei
- Scant, sometimes clear cytoplasm
- Tumor lacks its own bone/cartilage matrix

Microscopic pattern: Sheets of tumor cells

- Diffuse sheets of relatively uniform small round cells.
- Cells are more cohesive than typical background lymphocytes.
- Scant cytoplasm imparts a compact “small blue cell” appearance on H&E.
- Stromal fibrous septa may interrupt otherwise solid cellular sheets.

Nuclear and cytoplasmic detail

- Round to oval nuclei and limited cytoplasmic volume characterize the classic phenotype.
- Fine dispersed chromatin and inconspicuous nucleoli are often present.
- Clear cytoplasm is frequently attributable to abundant glycogen.
- Morphology supplies a strong clue—not a stand-alone molecular diagnosis.

Why glycogen matters

- Cytoplasmic clearing to glycogen accumulation.
- Periodic acid–Schiff (PAS) staining can demonstrate glycogen.
- PAS positivity is supportive but not unique to Ewing sarcoma.
- The source notes that tumor cells lack osteoid and cartilage production.

PAS and diastase: Interpret a supportive stain

H&E clue

Scant / clear cytoplasm within
round cell sheets



PAS stain

Cytoplasmic glycogen can
become magenta

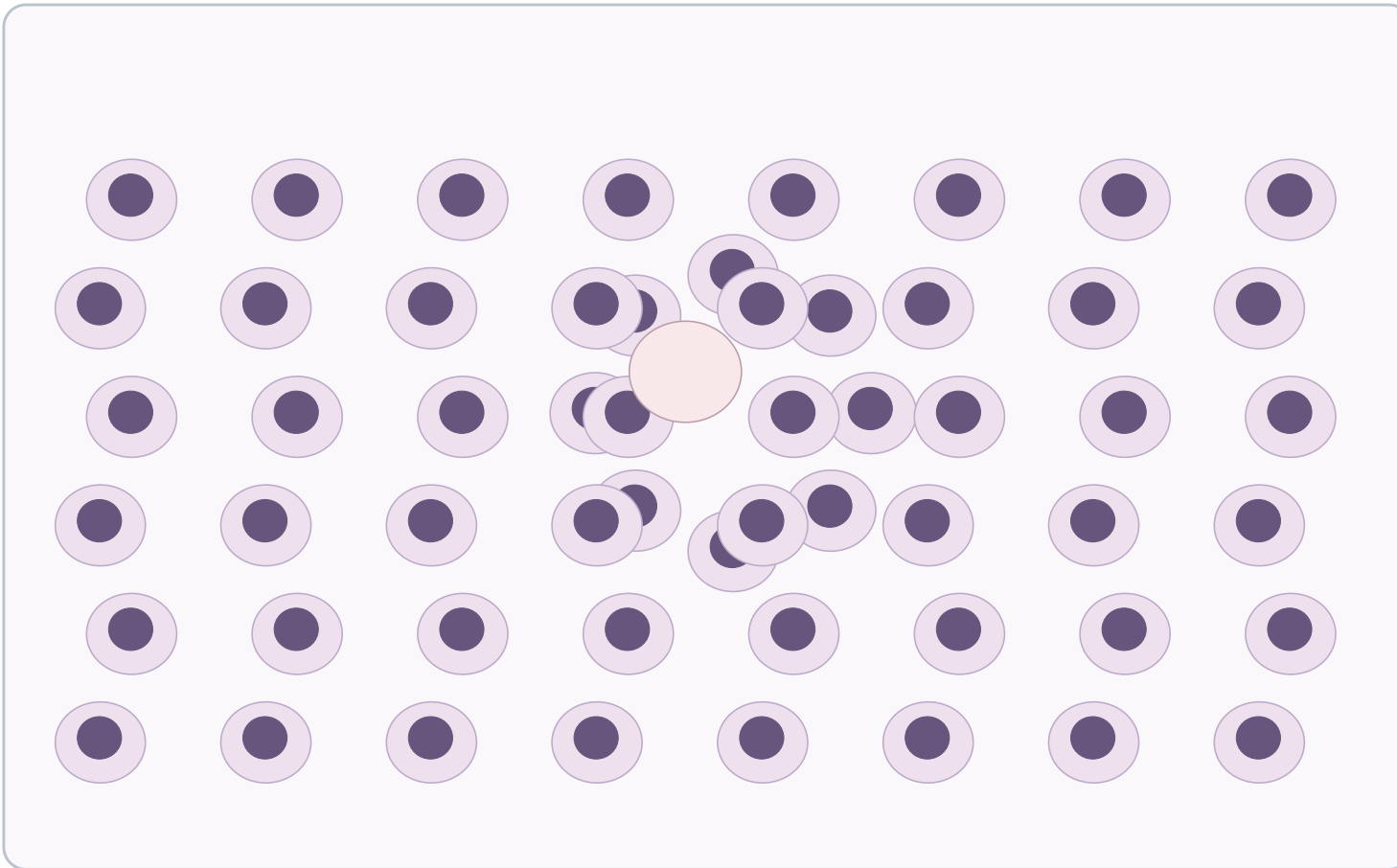


Diastase digestion

Loss of PAS reactivity supports
glycogen

Homer–Wright rosettes: Schematic

Schematic only — not a photomicrograph



Optional neuronal differentiation

- Homer–Wright rosette: Central fibrillary core
- May be present; Not obligatory
- Cannot establish the diagnosis alone

Rosettes do not redefine the tumor

- Homer–Wright rosettes may be found within Ewing sarcoma.
- They form circular cell groupings around a central fibrillary core.
- Neuronal differentiation does not imply a separate modern entity when the defining fusion is present.
- Absence of rosettes does not argue strongly against Ewing sarcoma.

Critical morphologic distinction

Ewing sarcoma

- Uniform primitive small round cell sheets
- Intracytoplasmic glycogen may be present
- No neoplastic osteoid
- No tumor cartilage

Conventional osteosarcoma

- Tumor cells deposit malignant osteoid
- Often markedly pleomorphic phenotype
- May have other matrix components
- Matrix production is diagnostic

Read a suspected Ewing biopsy systematically

Pattern

Relatively uniform small round cells in diffuse sheets

Cytoplasm

Scant or clear; Glycogen can be present

Architecture

Fibrous septa and occasional rosettes

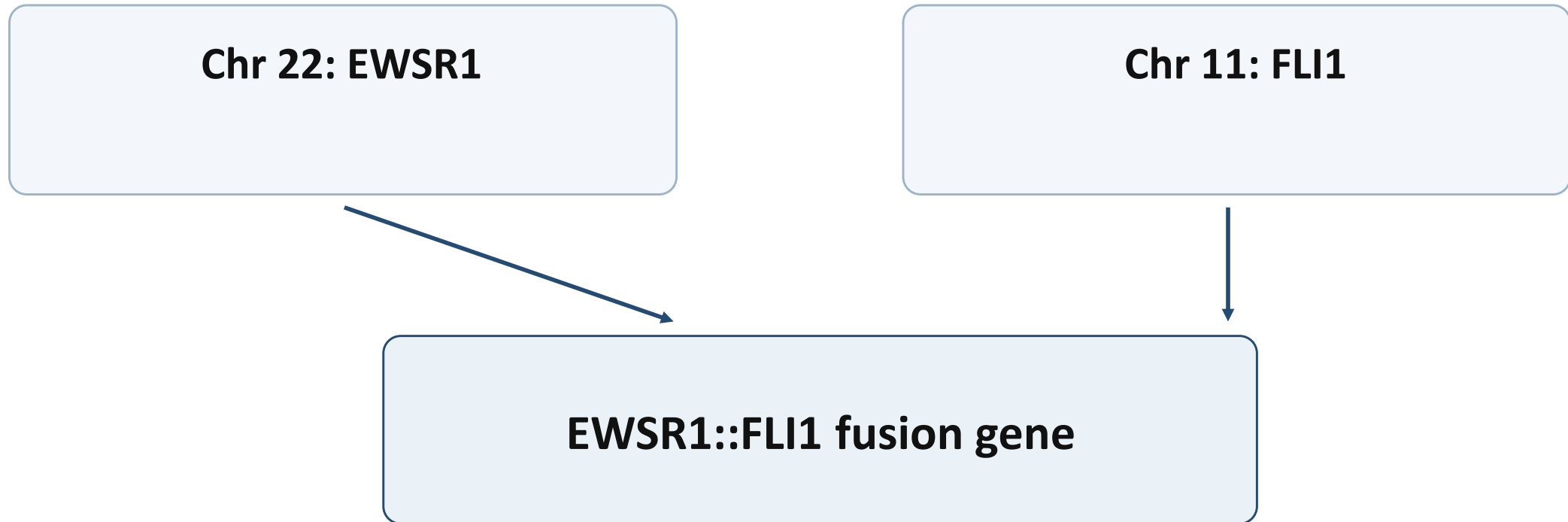
Matrix

No neoplastic bone or cartilage

Confirmation

Specific FET::ETS fusion in correct context

The canonical t(11;22) rearrangement



T(11;22)(q24;q12) • recurrent oncogenic rearrangement

Chromosomal positions and fusion identity

- EWSR1 is located at chromosome 22q12.
- FLI1 is an ETS-family transcription factor gene at chromosome 11q24.
- T(11;22)(q24;q12) brings together portions of these genes.
- The resulting EWSR1::FLI1 is a pathogenic, chimeric fusion gene.

From fusion gene to dysregulated transcription

Rearrangement

Genetic translocation creates a novel transcriptional regulator



Fusion protein

EWSR1::FLI1 associates with chromatin



Biologic effect

Aberrant transcription drives malignant cellular behavior

The partner families matter

FET component

EWSR1 is part of the FET RNA-binding protein family.

ETS component

FLI1 contributes an ETS-family DNA-binding regulatory component.

Novel function

The chimeric product changes transcriptional programs.

Fusion spectrum within Ewing sarcoma

- EWSR1::FLI1 is the predominant fusion in pediatric cases (NCI: About 85–90%).
- Other ETS partners include ERG, ETV1, ETV4, and FEV.
- Rare FUS::ETS fusions can occur.
- Fusion naming should identify both gene partners, not merely EWSR1 rearrangement.

Selected Ewing-associated gene fusions

FET partner	ETS partner	Interpretation
EWSR1	FLI1	Most common canonical fusion
EWSR1	ERG	Important less common fusion
EWSR1	ETV1 / ETV4 / FEV	Uncommon variants
FUS	ERG / FEV	Rare alternative FET partner

The cell of origin remains uncertain

- Mesenchymal stem cells a possible origin.
- Primitive neuroectodermal cells have also been proposed.
- The fusion-defined molecular identity is more secure than a single lineage-of-origin claim.
- Avoid teaching an unproven cell of origin as a settled fact.

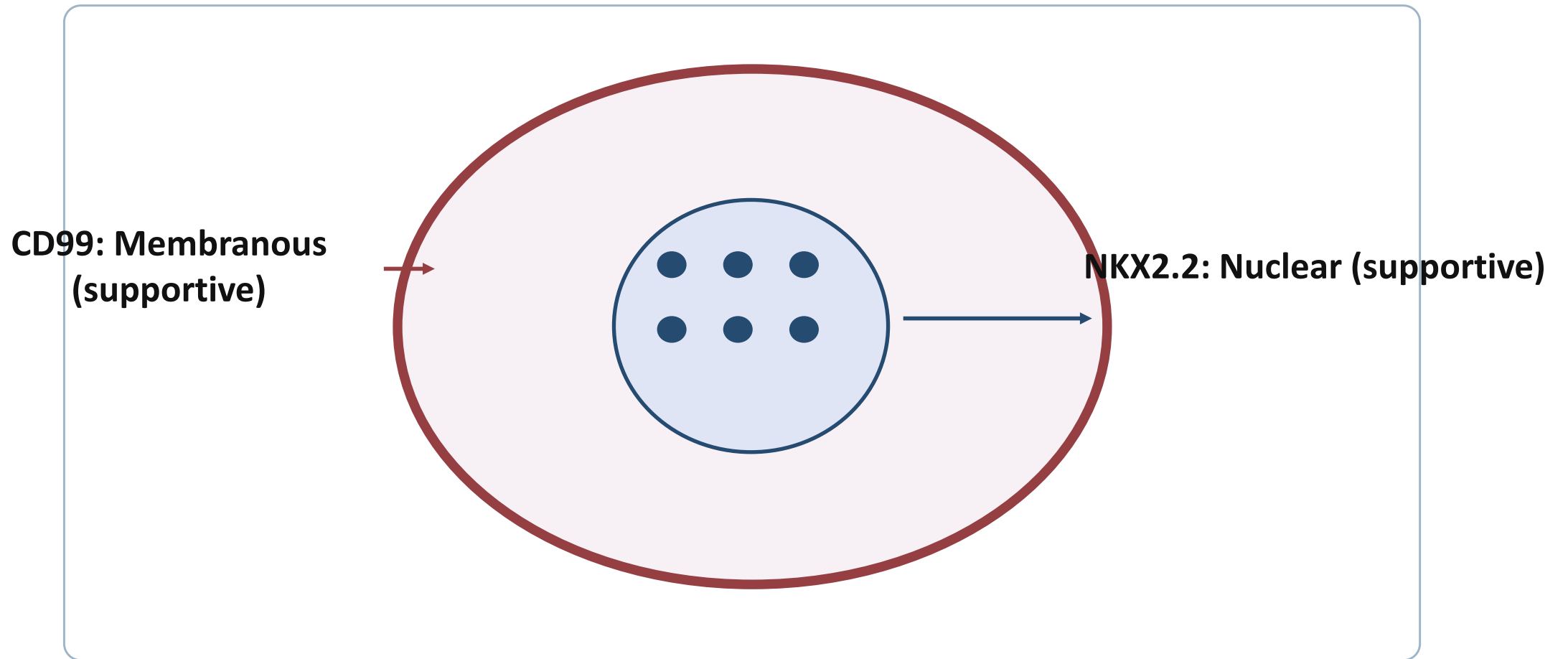
WHO-oriented round-cell sarcoma framework

Molecular class	Defining alteration
Ewing sarcoma	FET::ETS fusion
EWSR1-non-ETS round cell sarcoma	Non-ETS EWSR1-related fusion
CIC-rearranged sarcoma	CIC-related rearrangement
BCOR-altered sarcoma	BCOR genetic alteration

Older terminology in modern reporting

- “Peripheral primitive neuroectodermal tumor” historically emphasized neural features.
- “Askin tumor” referred to Ewing sarcoma of the chest wall.
- These terms are generally encompassed by Ewing sarcoma when molecular criteria are met.
- Do not group genetically distinct “Ewing-like” sarcomas under Ewing sarcoma.

Supportive IHC: Membrane versus nucleus



Neither marker is fully specific; Interpret with morphology and molecular genetics.

CD99: Diagnostic utility and pitfall

- CD99 typically shows strong membranous staining in Ewing sarcoma.
- CD99 is a useful screening marker within a small round cell lesion.
- It is not specific: Several non-Ewing tumors may express CD99.
- A positive CD99 stain does not substitute for genetic confirmation.

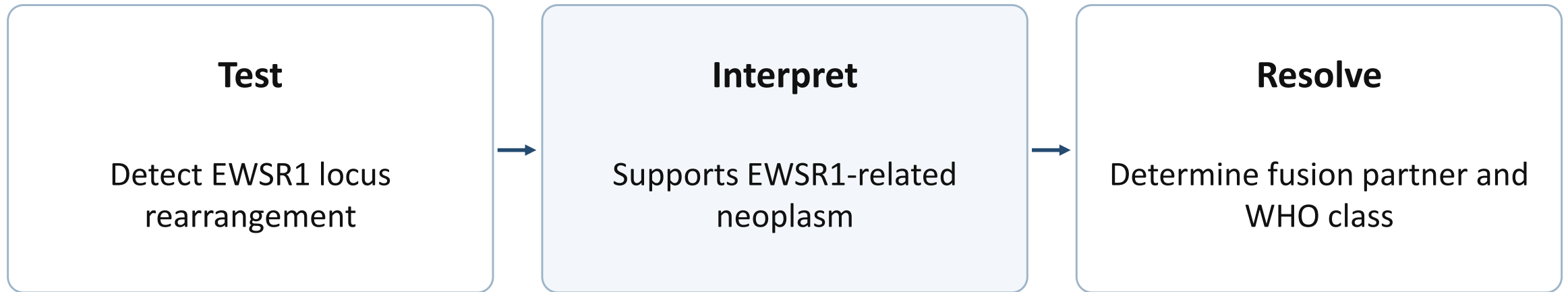
NKX2.2: Additional supporting marker

- NKX2.2 is a nuclear antigen commonly assessed in suspected Ewing sarcoma.
- Positive staining strengthens the diagnosis in the appropriate context.
- NKX2.2 is not perfectly specific.
- Its value is highest when integrated with morphology, CD99, and molecular testing.

Evidence hierarchy for diagnosis

Observation	What it contributes	What it cannot prove
Age, bone site, imaging	Raises clinical suspicion	Tumor identity
Small round cell H&E	Suggests Ewing phenotype	Unique tumor class
CD99 / NKX2.2	Supports the phenotype	Specific gene fusion
Fusion identification	Defines molecular group	Whole clinical stage

EWSR1 break-apart FISH: Correct use



FISH alone can misclassify a tumor

- EWSR1 break-apart does not reveal the fusion partner.
- Other tumor entities harbor EWSR1 rearrangements.
- FUS-driven Ewing sarcoma may be negative with an EWSR1 probe.
- A small subset of EWSR1::ERG tumors may be missed by break-apart FISH.

Targeted RNA fusion testing

- Fusion-aware assays can identify both partner genes.
- NGS fusion panels are frequently used for problematic small round cell tumors.
- A partner-aware result helps separate Ewing sarcoma from EWSR1-non-ETS neoplasms.
- Interpret negative results alongside specimen quality and assay coverage.

Integrated diagnostic algorithm

**Imaging + clinical
context**

Aggressive osseous or soft-tissue tumor



Histologic biopsy

Uniform small round cells; Exclude matrix-formers



Immunophenotype

CD99 / NKX2.2 as supportive—not definitive



Molecular test

Confirm FET::ETS fusion; Classify alternatives

Biopsy adequacy and tissue stewardship

- Plan biopsy with a multidisciplinary sarcoma service.
- Obtain viable lesional tissue for morphology and ancillary studies.
- Extensive necrosis can impair diagnostic assessment.
- Preserve suitable material for fusion testing when Ewing sarcoma is considered.

Differential: Osteosarcoma versus Ewing

Osteosarcoma

- Malignant osteoid matrix formation
- Often metaphyseal, but site alone insufficient
- Histology establishes osteoid-producing tumor

Ewing sarcoma

- Primitive small round cells without osteoid
- Often diaphyseal or pelvic primary
- Specific FET::ETS fusion supports diagnosis

Differential: Hematolymphoid malignancy

Lymphoma / leukemia

- May form small round cell sheets in bone
- CD45 / lineage studies may redirect workup
- Fusion identity differs from Ewing

Ewing sarcoma

- Primitive small round cell phenotype
- CD99 and NKX2.2 may support suspicion
- EWSR1::ETS or FUS::ETS fusion defines class

Differential: Rhabdomyosarcoma

Rhabdomyosarcoma

- May be primitive and round-cell predominant
- Myogenic differentiation assessed by specific markers
- Distinct molecular classification

Ewing sarcoma

- Often shows uniform round-cell morphology
- CD99/NKX2.2 as supportive markers
- FET::ETS molecular confirmation

Differential: Metastatic neuroblastoma

Neuroblastoma

- Pediatric small round cell tumor can infiltrate bone
- Clinical primary and neuroblastic features guide evaluation
- Neuronal marker pattern differs

Ewing sarcoma

- Can form Homer–Wright rosettes
- Rosettes therefore are not lineage-specific
- Genetic confirmation resolves ambiguity

Practical differential at first biopsy

Candidate	Discriminating question
Ewing sarcoma	Is a specific FET::ETS fusion demonstrable?
Osteosarcoma	Is malignant osteoid formed by tumor cells?
Lymphoma	Are hematolymphoid lineage markers present?
Rhabdomyosarcoma	Is true skeletal muscle differentiation demonstrated?
Metastatic neuroblastoma	Do clinical context and neuroblastic features fit?

Why EWSR1-non-ETS is not “Ewing”

- EWSR1 can participate in fusions outside the ETS transcription factor family.
- Examples include EWSR1::NFATC2 and EWSR1::PATZ1 tumors.
- Current WHO-oriented taxonomy separates these from Ewing sarcoma.
- Partner identity matters more than the shorthand “EWSR1-positive.”

Two additional WHO round-cell groups

CIC-rearranged sarcoma

A molecularly distinct undifferentiated round cell sarcoma group.

BCOR-altered sarcoma

Genetically defined group that is not conventional Ewing sarcoma.

Diagnostic consequence

Overlapping morphology requires precise fusion/alteration characterization.

Terminology: Avoid “Ewing-like” as a final diagnosis

- Historical umbrella use pooled biologically different tumors.
- WHO molecular categories provide more reproducible nomenclature.
- A tumor with nonspecific CD99 positivity should not be called Ewing sarcoma.
- Use the correct molecular category or a suitably qualified provisional diagnosis.

Molecular reporting exercise

Illustrative case

A pelvic small round cell sarcoma is CD99-positive. EWSR1 break-apart FISH is positive. RNA testing identifies EWSR1::NFATC2.

- Is the correct diagnosis necessarily Ewing sarcoma?
- Which finding is decisive for WHO-oriented classification?
- How should the initial CD99 and FISH results be described?

What makes a defensible diagnosis

Clinical setting

Compatible anatomic site and destructive growth

Morphology

Primitive uniform small round cells

Immunophenotype

Supportive but not individually specific

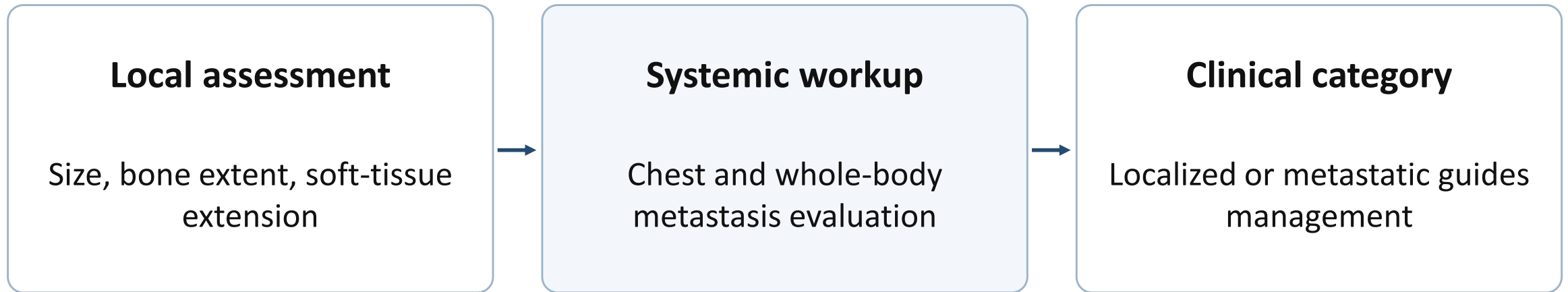
Genetics

FET::ETS fusion; Identify fusion partners

Final label

Avoid collapsing distinct WHO molecular groups

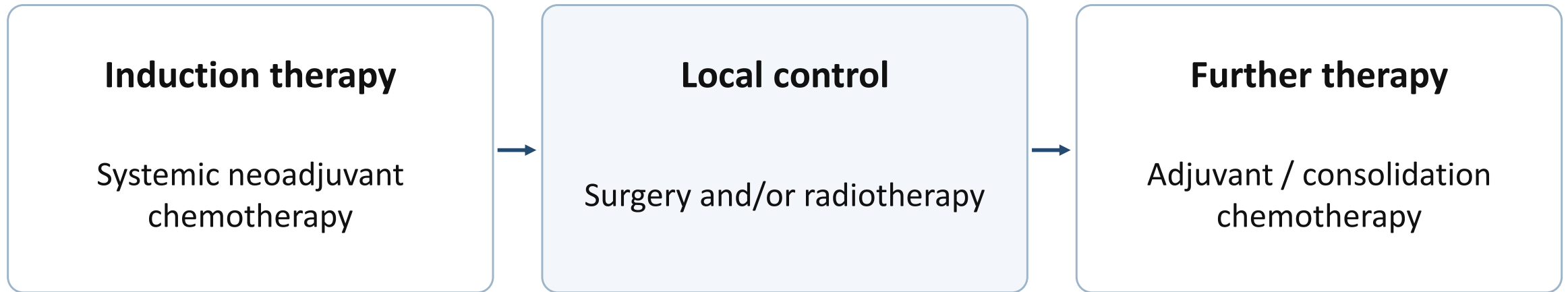
Localized versus metastatic disease



Why systemic chemotherapy is necessary

- Ewing sarcoma is clinically aggressive and may disseminate early.
- Apparently localized disease may include occult micrometastases.
- Therefore, local surgery alone is not adequate standard treatment.
- Multidrug chemotherapy is integrated with local-control strategies.

Multimodal treatment: Conceptual sequence



Surgery and radiotherapy: Complementary roles

Surgical local control

- Resection is considered when feasible with acceptable function
- Margin status influences local management
- Pathology examines viable tumor and treatment effect

Radiotherapy

- May contribute to definitive or postoperative local control
- Used in selected contexts with difficult anatomy/margins
- Treatment decisions require a specialist team

Histologic response after chemotherapy

- Chemotherapy-induced necrosis as a favorable prognostic indicator.
- The resection specimen allows estimation of residual viable tumor.
- NCI PDQ links minimal viable tumor after induction to better outcomes.
- Necrosis should be reported with method and relevant sampling limitations.

Interpreting prognosis responsibly

- Approximately 75% 5-year survival with contemporary multimodal therapy.
- It also describes approximately 50% long-term cure rates.
- These figures reflect the textbook's referenced populations and treatment era.
- Do not apply the aggregate numbers directly to an individual patient.

Major prognostic determinants

At presentation

- Localized versus metastatic disease
- Site and tumor burden
- Age and other clinical factors

After therapy begins

- Histologic response / remaining viable tumor
- Ability to achieve local control
- Relapse after initial treatment

Why survival statistics vary across sources

- Outcomes depend on localized versus metastatic case mix.
- Endpoint differs: Event-free survival, overall survival, and cure are not interchangeable.
- Age, site, disease volume, and treatment regimen influence reported results.
- NCI PDQ estimates must not be treated as directly identical to Robbins aggregates.

Integrated case: A diaphyseal tumor

Illustrative case

A 14-year-old has a painful femoral diaphyseal mass, laminated periosteal reaction, and an H&E biopsy with uniform primitive small round cells.

- List two plausible initial diagnoses and the key discriminator.
- Identify a supportive immunophenotypic panel and its limits.
- What molecular result would confirm the classic Ewing entity?

Integrated case: Molecular ambiguity

Illustrative case

An undifferentiated soft-tissue sarcoma shows diffuse membranous CD99, nuclear NKX2.2, and an EWSR1 break-apart signal, but fusion-partner testing is pending.

- Which evidence favors—but does not establish—Ewing sarcoma?
- Why is the FISH result insufficient as a final diagnosis?
- How does RNA fusion identification modify the final report?

Synthesis: Five diagnostic anchors

Who?

Predominantly patients younger than 20 years

Where?

Often long-bone diaphysis, pelvis, or soft tissue

How?

Aggressive permeative lesion ± laminated periosteal reaction

What?

Uniform small round cells, scant glycogen-rich cytoplasm

Why?

Driver FET::ETS fusion; Most often EWSR1::FLI1

Selected references

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