

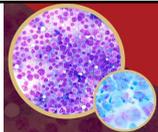
## Gânglio linfático: binómio hiperplasia/linfoma imunofentipagem de linfomas

Filipe Sampaio

Patologia Clínica

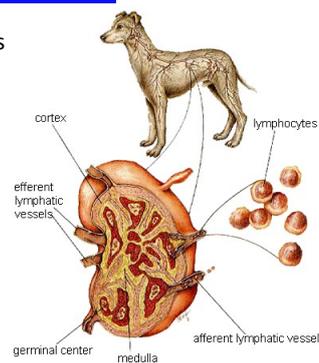
Mestrado em Medicina Interna de Animais de Companhia

1



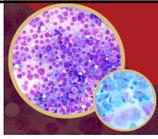
### Gânglios linfáticos = Linfonodos

- cél. apresent. de antígenos e macrófagos
- linf. B – Plasmócitos
- linf. T



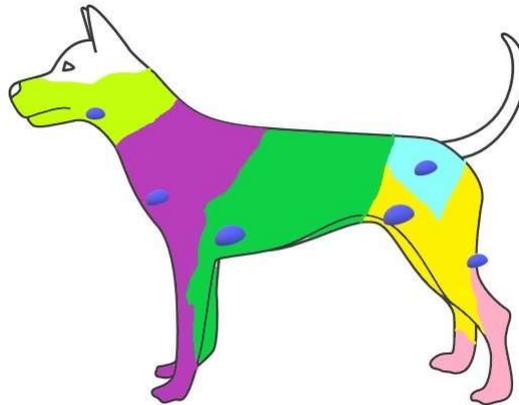
→ palpação dos linfonodos

2

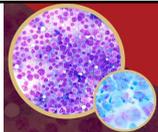


## Linfonodos e áreas de drenagem

→ relação alterações dos linfonodos c/lesões 1<sup>as</sup> na área de drenagem



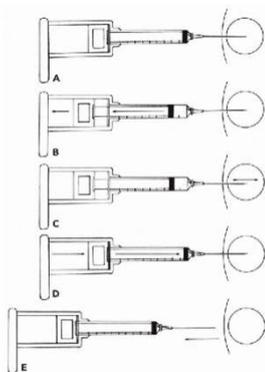
3



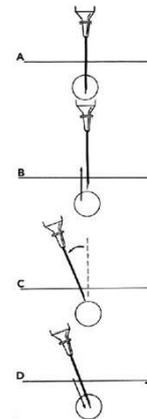
## Amostras citológicas de linfonodos

- em casos de linfadenomegália localizada, regional ou generalizada (amostras de vários LN)
- evitar linfonodo mandibular
- antes do tratamento (antibióticos, anti-inflamatórios, quimioterápicos)

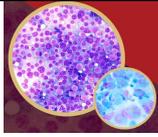
FNA technique



Fine needle sampling without aspiration/suction or Capillary method

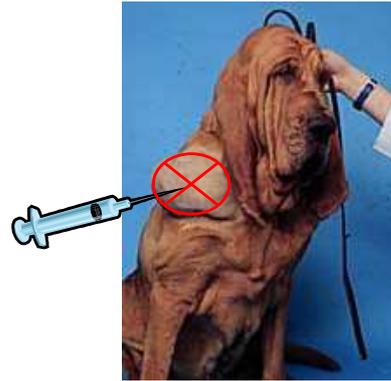
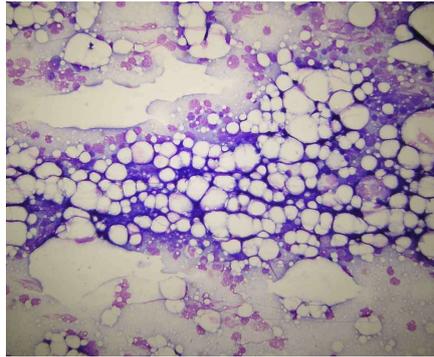


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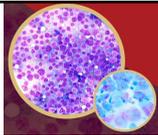


## Amostras citológicas não representativas

- amostras de LN extremamente aumentados (necrose/hemorragia)
- amostra pouco representativa (não houve redireccionamento)
- aspiração de gordura periganglionar

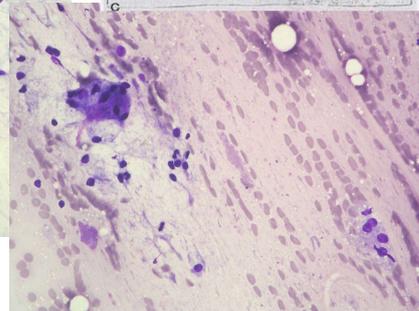
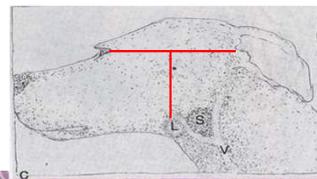
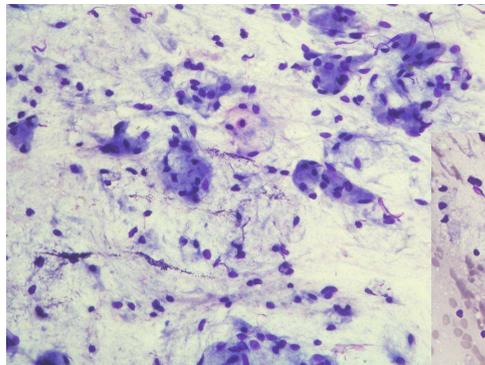


5

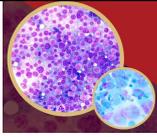


## Amostras citológicas de linfonodos

- Principal causa para amostras não representativas do LN mandibular: aspiração da glândula salivar mandibular

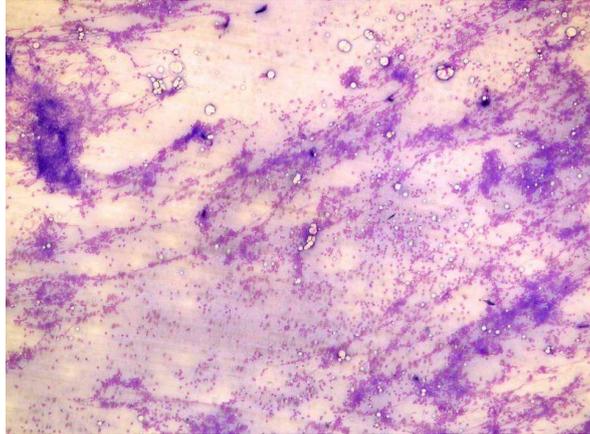


6

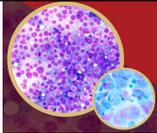


## Amostras citológicas de linfonodos

- Aplicação de força excessiva na realização do esfregaço

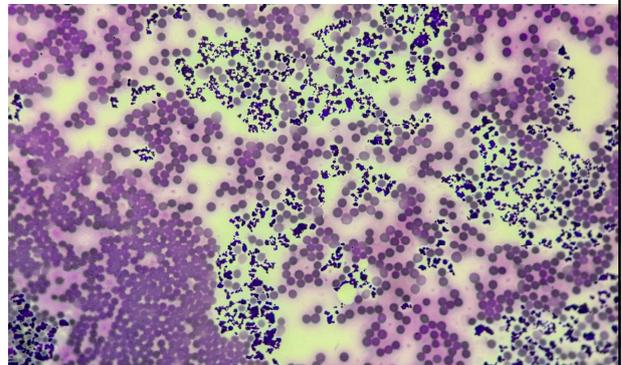
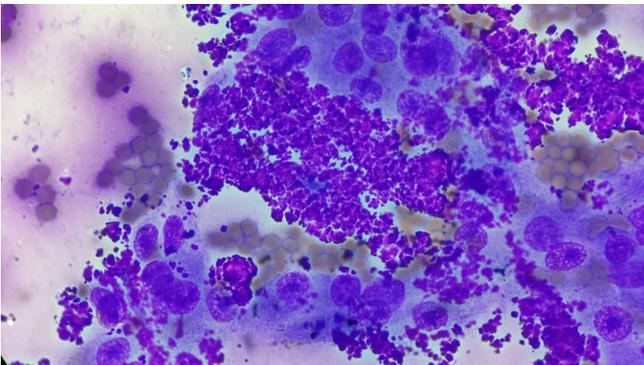


7

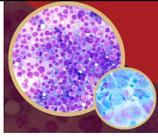


## Amostras citológicas de linfonodos

- Presença de gel ecográfico em preparações citológicas de linfonodos internos

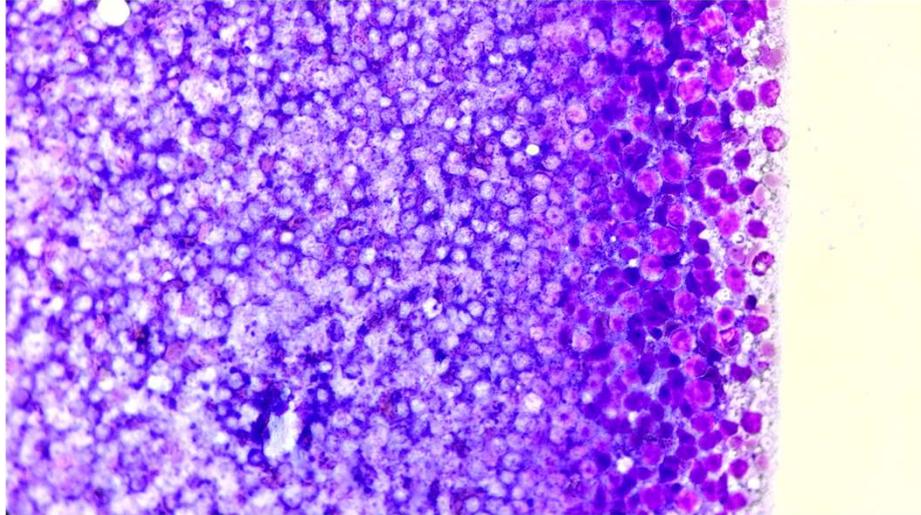


8

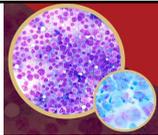


## Amostras citológicas de linfonodos

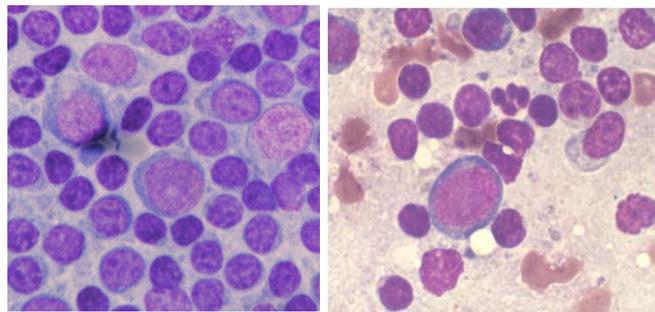
- Monocamada para avaliação citológica e eventual imunofenotipagem



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## Avaliação citológica do tipo de linfócitos



- pequenos/maduros linf. (1-1,5x)
- linf médios (2-2,5x)
- grandes linf (3x ou +)



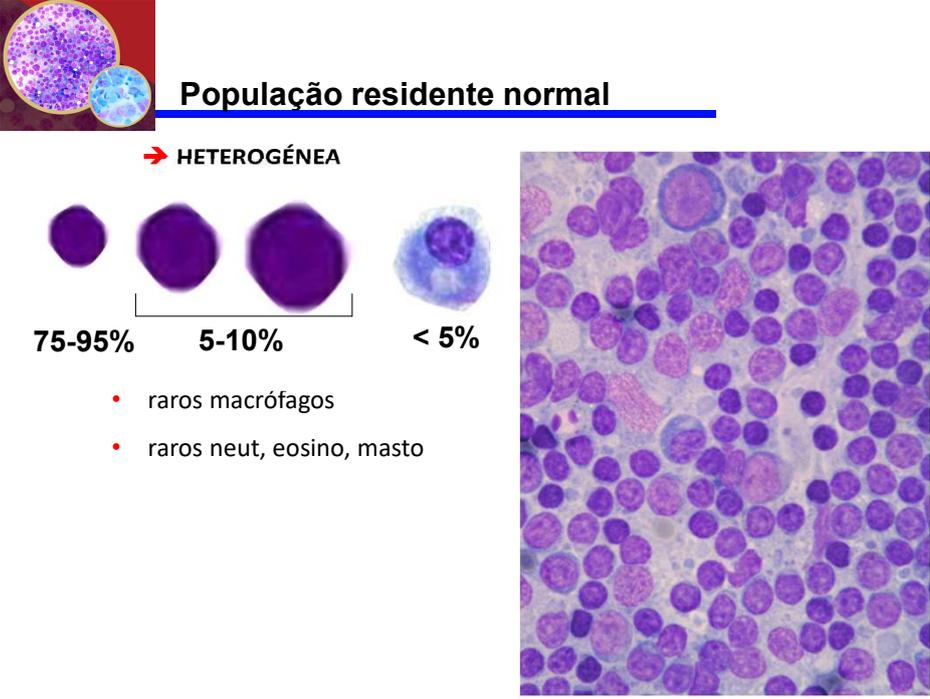
10

**População residente normal**

→ HETEROGÉNEA

75-95%      5-10%      < 5%

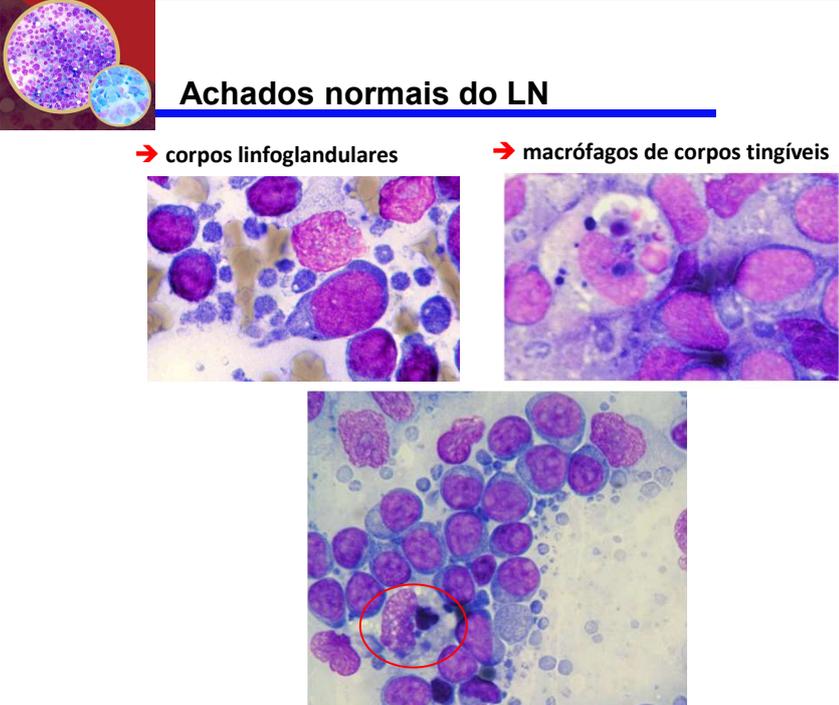
- raros macrófagos
- raros neut, eosino, masto



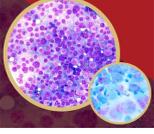
11

**Achados normais do LN**

→ corpos linfoglandulares      → macrófagos de corpos tingíveis

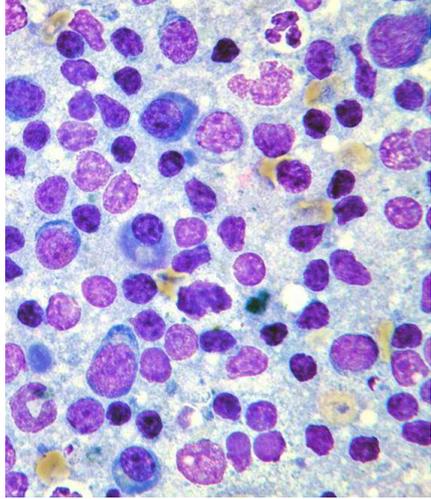


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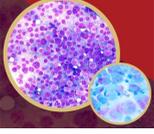
 **Hiperplasia reactiva**

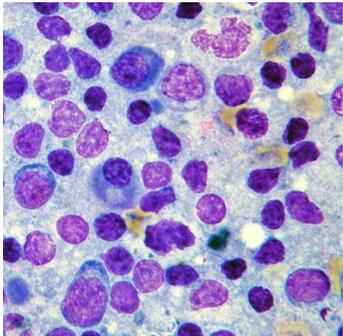
→ HETEROGÉNEA

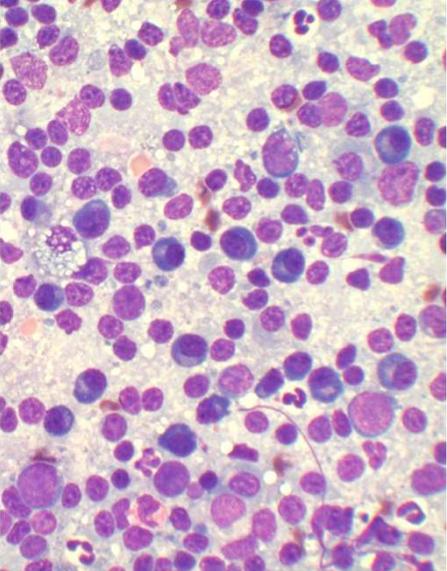
  $\geq 50\%$        $> 15\%$        $> 5-10\%$

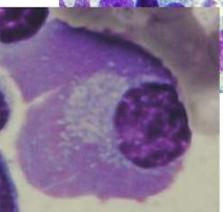


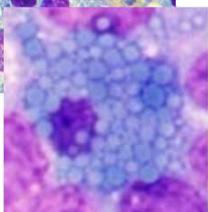
13

 **Hiperplasia reactiva**

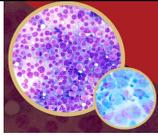




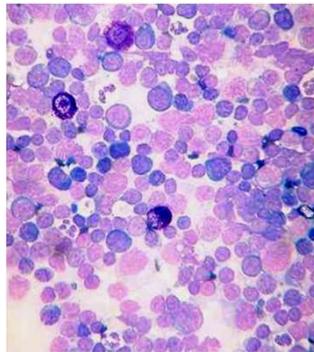




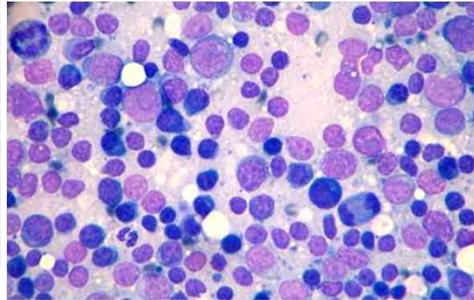
14



## Hiperplasia reativa – do gato jovem



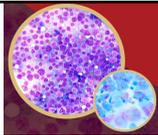
→ Gato, 2 anos, linfadenomegália generalizada



- $\geq 50\%$  pequenos linf
- $\uparrow 15\%$  linf médios/grandes
- $\uparrow$  Mastócitos

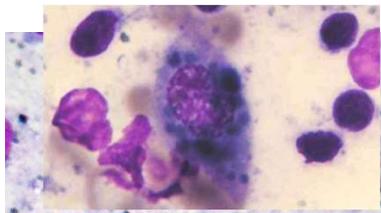
→ Etiologia desconhecida (descartar FIV, FeLV, toxoplasmose)

15

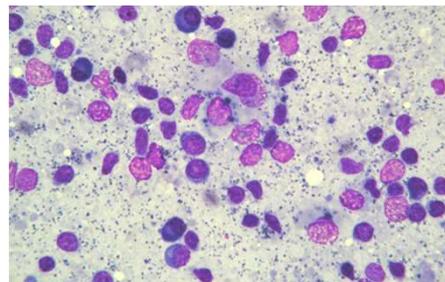
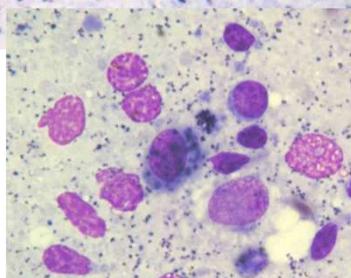


## Hiperplasia reativa/linfadenopatia dermatopática

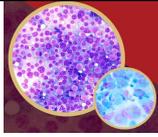
→ linfadenomegália + lesões cutâneas crônicas; camada basal atingida



- $\uparrow$  macrófagos com melanina (melanófagos)
- grânulos de melanina
- população linfóide reactiva

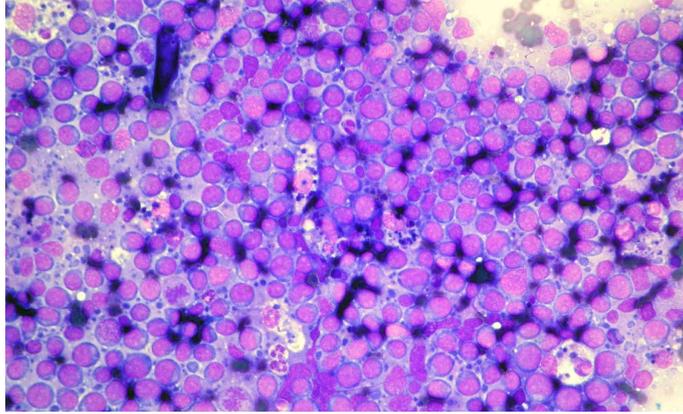


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## Linfoma

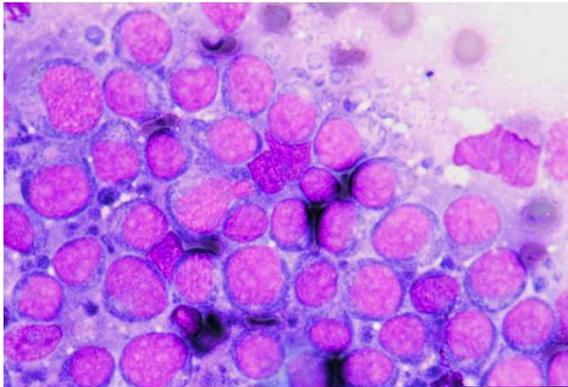
→ HOMEOGÉNEA



- ↑ macrófagos de corpos tingíveis
- ↑ corpos linfoglandulares

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→ Linfoma de células imaturas



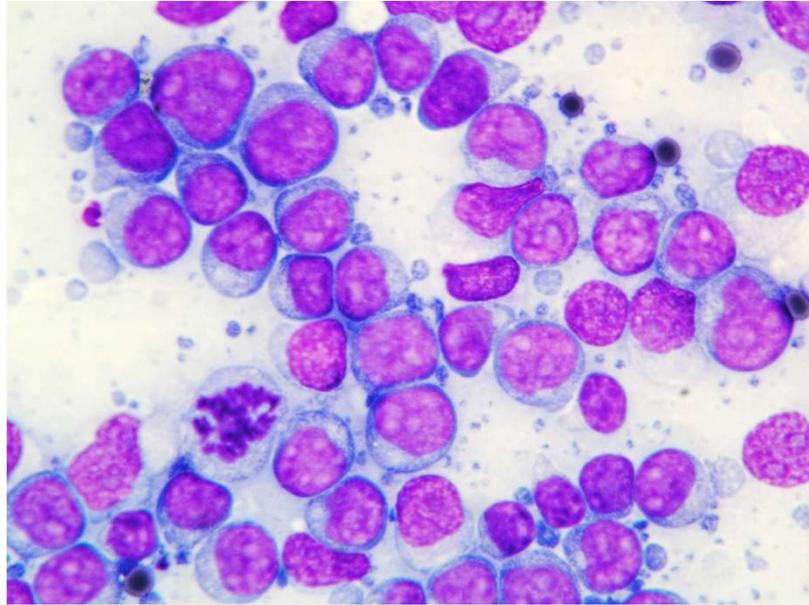
- 2,5 a 3x eritrócito
- ausência de plasmócitos



- figuras de mitose

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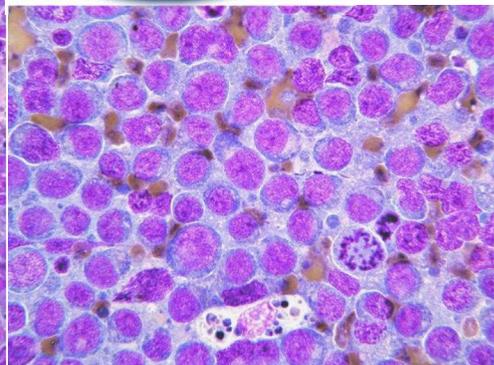
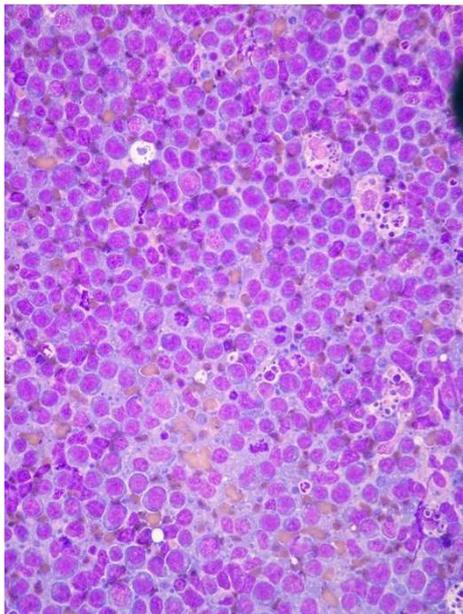
→ Linfoma de células imaturas



• Nucléolos proeminentes

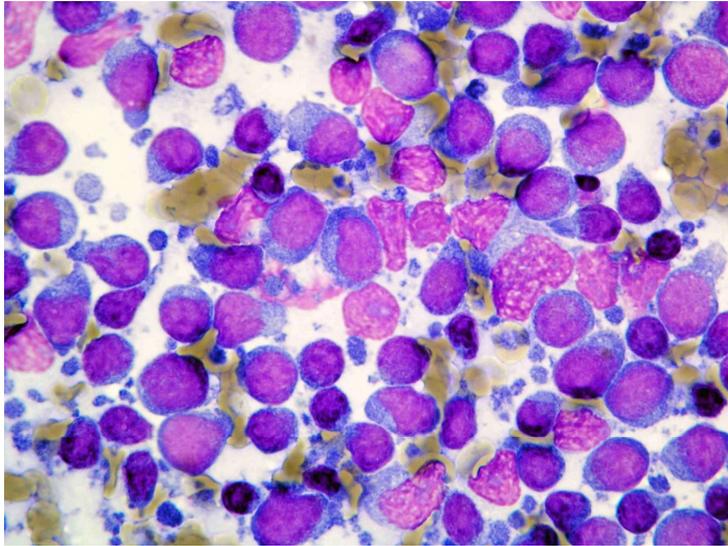
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→ Linfoma de células imaturas



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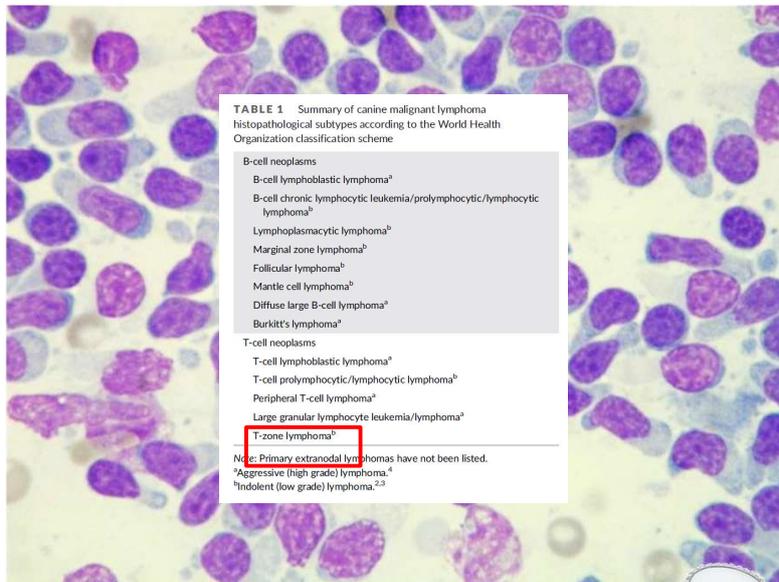
→ Linfoma T



- células “em raquete”
- núcleos irregulares
- pode haver pleomorfismo (→pedir opinião)

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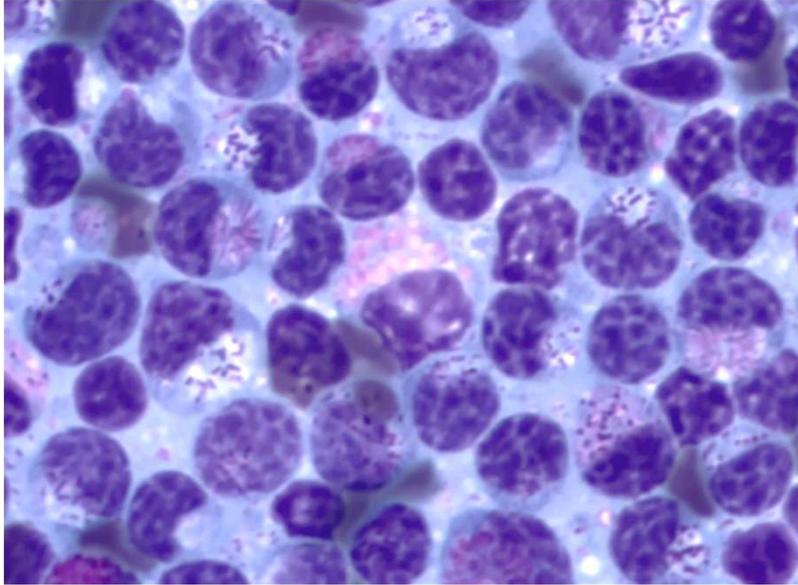
→ Linfoma T



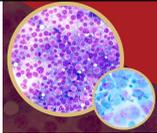
- células pequenas/medias; “em raquete”
- plasmócitos presentes

22

→ Linfoma LGL

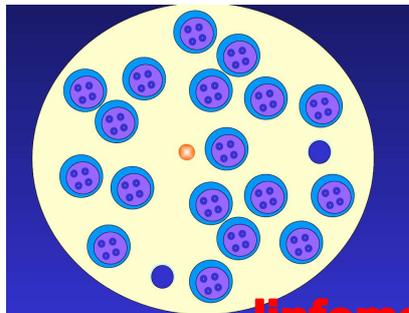


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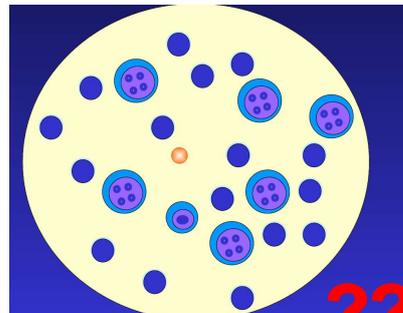


### Dificuldades no diagnóstico citológico

- de pequenas células /linfócitos maduros (raros) ↔ Histopatologia (perda da arquitetura)
- linfomas em fases iniciais (mistura de pop. residente + pop. neoplásica = heterogénea)
- envolvimento assíncrono dos gânglios
- linfomas pleomórficos

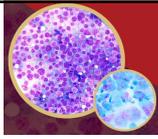


**linfoma**



**??**

24



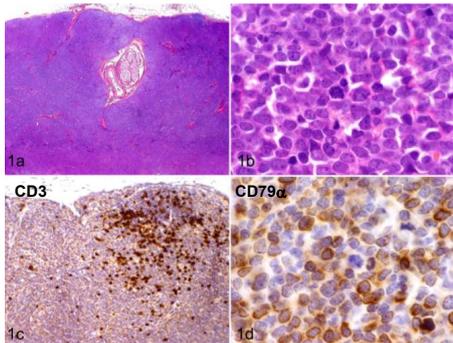
# Linfoma

- Exame *gold standard* para diagnóstico e imunofenótipo – *histopatologia*

## Classification of Canine Malignant Lymphomas According to the World Health Organization Criteria

Veterinary Pathology  
45(1) 19-211  
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Veterinary Pathologists 2011  
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sagepub.com/journalsPermissions.nav  
DOI: 10.1177/0898010110379428  
http://vcp.sagepub.com

V. E. Valli<sup>1</sup>, M. San Myint<sup>2</sup>, A. Barthel<sup>1</sup>, D. Bienzie<sup>4</sup>, J. Caswell<sup>1</sup>,  
F. Colbatzky<sup>5</sup>, A. Durham<sup>6</sup>, E. J. Ehrhart<sup>7</sup>, Y. Johnson<sup>8</sup>, C. Jones<sup>9</sup>,  
M. Kluge<sup>10</sup>, P. Labelle<sup>11</sup>, S. Lester<sup>12</sup>, M. Miller<sup>13</sup>, P. Moore<sup>14</sup>,  
S. Moroff<sup>15</sup>, P. Roccabianca<sup>16</sup>, J. Ramos-Vara<sup>17</sup>, A. Ross<sup>18</sup>,  
T. Scase<sup>19</sup>, H. Tvedten<sup>20</sup>, and W. Vernau<sup>21</sup>

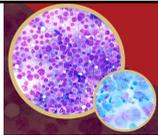


Valli et al, 2011

TABLE 1 Summary of canine malignant lymphoma histopathological subtypes according to the World Health Organization classification scheme

<b>B-cell neoplasms</b>
B-cell lymphoblastic lymphoma <sup>a</sup>
B-cell chronic lymphocytic leukemia/prolymphocytic/lymphocytic lymphoma <sup>b</sup>
Lymphoplasmacytic lymphoma <sup>b</sup>
Marginal zone lymphoma <sup>a</sup>
Follicular lymphoma <sup>b</sup>
Mantle cell lymphoma <sup>a</sup>
<b>Diffuse large B-cell lymphoma<sup>a</sup></b>
Burkitt's lymphoma <sup>a</sup>
<b>T-cell neoplasms</b>
T-cell lymphoblastic lymphoma <sup>a</sup>
T-cell prolymphocytic/lymphocytic lymphoma <sup>b</sup>
Peripheral T-cell lymphoma <sup>a</sup>
Large granular lymphocyte leukemia/lymphoma <sup>a</sup>
T-zone lymphoma <sup>a</sup>

Note: Primary extranodal lymphomas have not been listed.  
<sup>a</sup>Aggressive (high grade) lymphoma.  
<sup>b</sup>Indolent (low grade) lymphoma.<sup>2,3</sup>

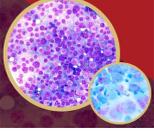


# Imunofenotipagem de Linfomas

TABLE 30.1 Comparison of Flow Cytometry, PARR, IHC, and ICC for Diagnosis of Hematopoietic Neoplasia<sup>1,2,3</sup>

	Flow Cytometry	PARR	IHC and Histopathology	ICC and Cytology
Sample	Freshly collected liquid sample with intact cells	Fluid; unstained or stained cytology slides Results from formalin-fixed tissue may not be as reliable	Formalin-fixed tissue	Usually one unstained cytology slide with intact cells for each marker
Typical size of results panel <sup>4</sup>	Largest	Smallest	Variable	Variable
Advantages in diagnosis of hematopoietic neoplasia	Typically has the shortest turnaround time Results can sometimes provide prognostic information	Can be used if other tests are equivocal (e.g., to detect a smaller number of neoplastic cells within a mixed lymphoid population)	Allows for evaluation of tissue architecture and cytoplasmic marker expression	Allows for evaluation of cytoplasmic marker expression
Limitations in diagnosis of hematopoietic neoplasia	May not be able to confirm neoplasia within a mixed cell population or if there is no aberrant marker expression	May not be as reliable as other methods for discerning B-cell neoplasia from T-cell neoplasia Some inflammatory and infectious diseases can have clonal PARR results	May have the longest turnaround time Some markers do not work well on formalin-fixed tissue	May have a long turnaround time May be difficult to interpret because of nonspecific staining or decreased marker expression

Cowell and Tyler, 2019

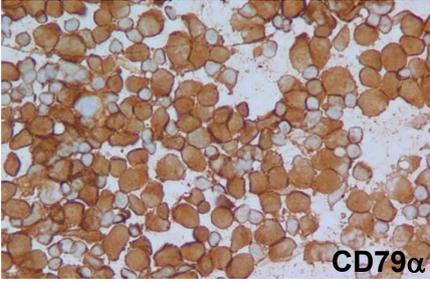
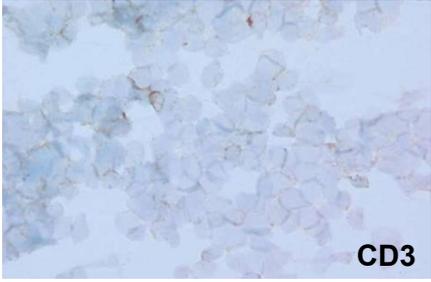


## Imunofenotipagem de Linfomas

→ Lâminas não coradas

Vet Pathol 33:204-212 (1996)

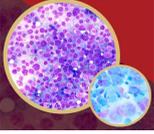
**Canine Lymphoma: Immunocytochemical Analysis of Fine-needle Aspiration Biopsy**  
M. CASIATTI, P. ROCCABIANCA, E. SCANZIANI, S. PALTRINIERI, AND P. F. MOORE

**CD79 $\alpha$**  **CD3**

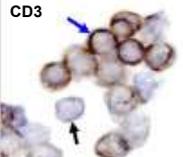
- **Necessário controles de citologia (lâminas congeladas)**
- **Variabilidade da celularidade e preservação de cada preparação**

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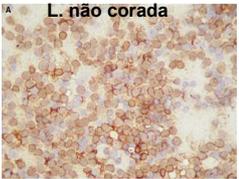
## Imunofenotipagem de Linfomas

→ Lâminas previamente coradas

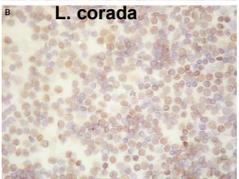



**CD3**

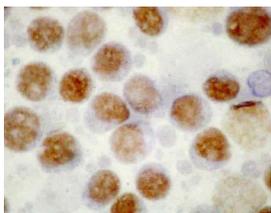
**A L. não corada**



**B L. corada**



**CD3**



**PAX5**

**TECHNICAL REPORT** [Veterinary Clinical Pathology](#) WILEY

Antigens under cover—The preservation and demasking of selected antigens for successful poststaining immunocytochemistry of effusion, brain smears, and lymph node aspirates **2019**

Stefanie Dörfelt<sup>1</sup> | Lara A. Matlasek<sup>1,2</sup> | Sandra Felten<sup>1</sup> | Laura Sangl<sup>1</sup> | Katrin Hartmann<sup>1</sup> | Kaspar Matlasek<sup>3</sup>

→ **CD3, linfoma gato, lâminas montadas com lamela**

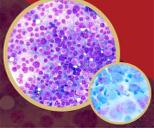
**TECHNICAL REPORT** [Veterinary Clinical Pathology](#) WILEY

Optimized immunocytochemistry using leukocyte and tissue markers on Romanowsky-stained slides from dogs and cats **2019**

Rose E. Raskin<sup>1</sup> | Julie Vickers<sup>2</sup> | Jennifer G. Ward<sup>3</sup> | Angus Toland<sup>3</sup> | Andrew G. Torrance<sup>3</sup>

→ **CD3, CD20, PAX5 linfomas cão, lâminas montadas?**

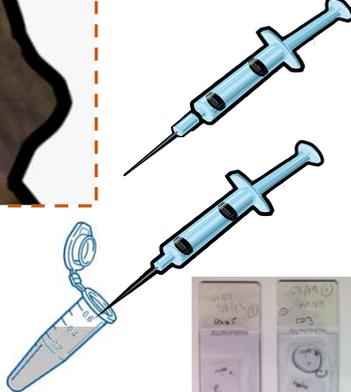
28



## Imunofenotipagem de Linfomas

➔ Em lavados da agulha e citoblocos





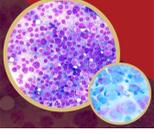





SERVIÇOS DE CITOLOGIA VETERINÁRIA  
RELATÓRIO DE ANÁLISE CITOLÓGICA

Nº registo: 58-19	Data de receção: 23-1	Cibco:
IDENTIFICAÇÃO DO ANIMAL		IDENTIFICAÇÃO DO TUTOR
Nome: Nina	Idade:	
Espécie: C	Sexo: F	IDENTIFICAÇÃO DO MÉD. VETERINÁRIO
Raça:		
<b>MATERIAL ENVIADO:</b> foram recebidos lavados de agulha para realização de citobloco e imunomarcagem para PAX-5 (marcador de linfócitos B) e CD3 (marcador de linfócitos T). <b>ESTUDO MICROSCÓPICO:</b> Nas preparações de imunohistoquímica para PAX-5 observam-se raras células marcadas (Figura 1). Por oposição, observa-se elevada quantidade de células marcadas para CD3 (linfócitos T) (Figura 2). Fig. 1: Imuno. para PAX-5 (marcador de linfócitos B). Fig. 2: Imuno para CD3 (marcador de linfócitos T).		
<b>CONCLUSÃO:</b> Linfoma T.		

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## Imunofenotipagem de Linfomas

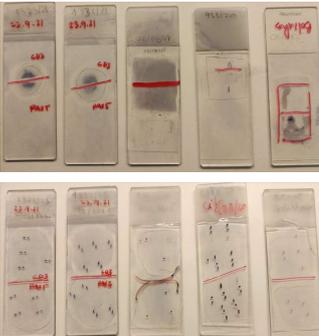
2021 ESVCP-ECVCP - Parallel Sessions
Research presentations Session

CO-05

**Immunophenotyping in stained cytology smears and matched cell blocks of canine and feline lymphoid samples**

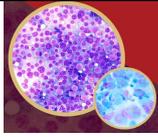
F. Sampaio<sup>1,2</sup> (samp.filipe@gmail.com), C. Marrinhas<sup>1,3</sup>, L. Fonte Oliveira<sup>1,4</sup>, F. Malhão<sup>1</sup>, C. Lopes<sup>1</sup>, H. Gregório<sup>4</sup>, C. Correia-Gomes<sup>5</sup>, R. Marcos<sup>1</sup>, M. Santos<sup>1</sup>

<sup>1</sup> Cytology Diagnostic Services, Instituto Ciências Biomédicas Abel Salazar, ICBAS – UP, University of Porto, Porto, Portugal; <sup>2</sup> Laboratório INNO, Braga, Portugal; <sup>3</sup> Hospital do Baixo Vouga, OneVet Group, Águeda, Portugal; <sup>4</sup> Anicura CHV Porto, Porto, Portugal; <sup>5</sup> Animal Health Ireland, Carrick-on-shannon, Ireland





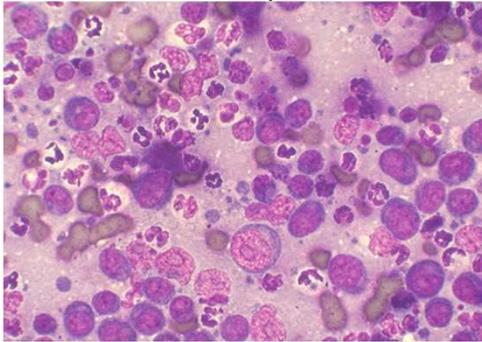
30



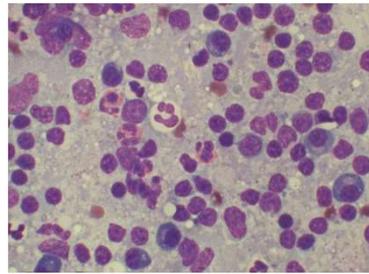
## Linfadenites

- antígenos ↔ quimiotaxia cél. inflamatórias
- envolvimento variável
- ↑ de um determinado tipo celular + população linfóide reativa
- pode haver substituição completa (ou quase) da pop. normal

### → Linfadenite purulenta

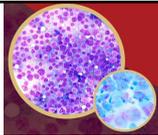


### → Linfadenite eosinofílica



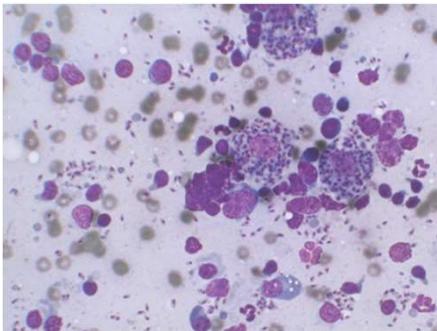
- ↳ Alergia à picada de pulga
- ↳ Síndrome eosinofílica felino
- ↳ Metástases de MCT

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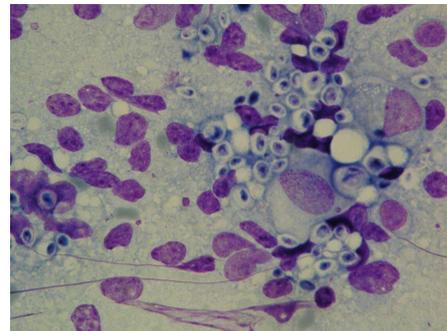


## Linfadenites

### → Linfadenite macrófaga



↳ Leishmaniose



↳ Criptococos

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# euvg

Obrigado  
Filipe Sampaio

