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UK NEQAS International Quality Expertise

Learning from EQA: When is debris not debris…?

Andrea Teuchert[1], Rachel Godden[1], Peter Carey[2], Barbara De la Salle[3], Bashori Rahman[1]

[1]UK NEQAS Haematology, Watford, WD18 8YX, UK; [2]Newcastle upon Tyne Hospitals NHS Foundation Trust; [3]UK NEQAS Charity

Introduction

UK NEQAS Haematology provides external quality assessment (EQA) programmes to support laboratory quality assurance in the UK and internationally. The Blood Films for Morphology (BF) EQA programme aims to help laboratories maintain and improve skills in blood film analysis, manual differential counting, and blood parasite identification.

Background Information

An interesting case of a 70-year-old female patient who was admitted to the emergency department for collapsing whilst walking in the countryside and hitting her head. She was on vacation in the UK therefore had no clinical history available. Clinical details for the patient included "generalised weakness" and a query of sepsis. The full blood count showed a low platelet count which triggered a blood film examination. Parasites were evident, and so a malaria antigen test was performed but found to be negative. Upon seeking further information about the patient, it transpired that she had recently travelled to America, spending 3 weeks walking the Appalachian trail in New England. The Parasitology Reference laboratory confirmed the patient to be infected with *Babesia microti* which is a parasite associated with the east coast of the USA.

Full Blood Count results, including automated differential count, for 2308BF2

Parameter	Result	Normal Range	Parameter	Result (x10⁹/L)	Normal Range
RBC (10^{12}/L)	3.39	3.80 – 5.80	**WBC**	3.8	4.0 – 10.0
Hb (g/L)	106	120 – 150	Neutrophils	1.9	1.8 – 7.5
Hct (L/L)	0.33	0.37 – 0.47	Lymphocytes	1.2	1.0 – 4.0
MCV (fL)	97	80.0 – 100.0	Monocytes	0.6	0.2 – 1.0
MCH (pg)	31.3	27.0 – 32.0	Eosinophils	0.0	0.0 – 0.4
MCHC (g/L)	321	280 – 350	Basophils	0.0	0.0 – 0.1
RDW (%)	12.8	12.0 – 16.0	nRBCs	0.0	0.0 – 0.0
Platelets (10^9/L)	68	150 – 400			

Further results:

Biochemistry

Parameter	Result	Normal Range	Parameter	Result	Normal Range
Sodium (mmol/L)	133	133 – 146	Albumin (g/L)	29	35 – 50
Potassium (mmol/L)	4.0	3.5 – 5.3	Total Bilirubin (µmol/L)	15	0 – 21
Urea (mmol/L)	5.1	2.5 – 7.8	Calcium (mmol/L)	1.99	2.10 – 2.55
Creatinine (µmol/L)	54	45 – 84	Adjusted Calcium (mmol/L)	2.29	2.10 – 2.55
eGFR (mL/min)	>90	>90	Phosphate (mmol/L)	1.01	0.80 – 1.50
C-reactive Protein (mg/L)	166	<5	Alkaline Phosphatase (IU/L)	78	30 – 130
Amylase (IU/L)	23	28 – 100	Aspartate Transaminase (IU/L)	64	10 – 36
Total Protein (g/L)	51	60 – 80	Alanine Transaminase (IU/L)	59	0 – 31

Parasitology
Malarial Antigen test: *Plasmodium falciparum* antigen NOT detected. *Plasmodium* species antigen NOT detected.
Babesia spp. seen. Parasitaemia 0.8%.
PCR: *Babesia microti* DNA detected.

Figure 1 – Full case details of a 70 year old female, who was brought into the Emergency Department after collapsing while walking in the English countryside

Aim

This interesting case was used in a Blood Films for Morphology EQA exercise in December 2023 (distribution 2308BF). Participants in the programme are expected to identify and report the significant morphological features seen on the film. A list of comment codes is provided from which they can make their selection. Blood films are accompanied by essential information such as fixation, stain, patient age, sex, Total Nucleated Cell count (TNC) , and Haemoglobin (Hb) level, but the clinical history of the patient is not provided to prevent bias. Participants are encouraged to indicate a possible condition ('morphological syndrome') for the cases based on the blood morphology, although this is not expected to be a final diagnosis and is not performance assessed.

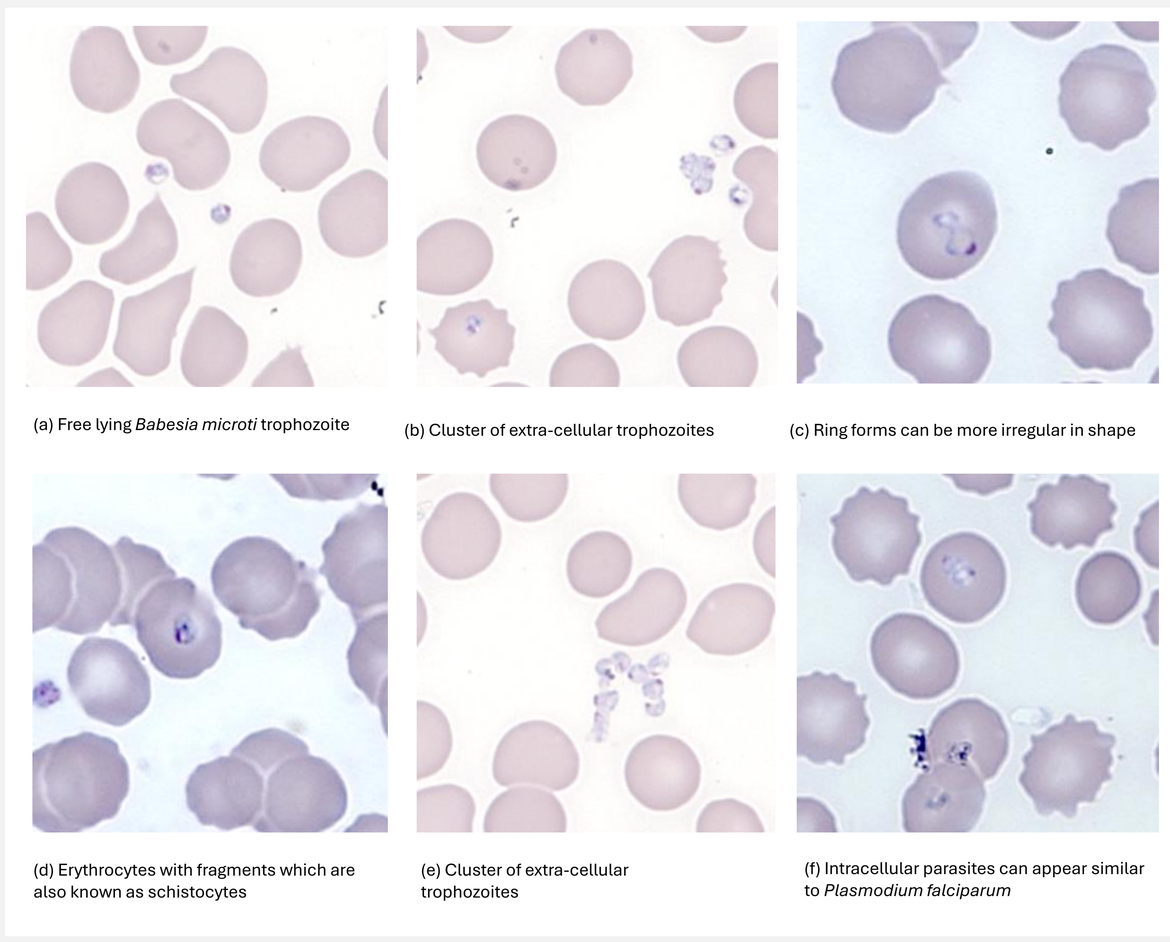
Methods

Blood films for EQA are prepared from fresh blood, generously donated from surplus laboratory material by local laboratories. The programme distributes 16 morphology cases annually across eight surveys. This case was distributed to 564 participants, and 534 participants returned results (94.6% return rate).

The clinical details provided with the film stated that this was a 70 year old female with a TNC of 3.8 x 10^9/L and a Hb concentration of 106 g/L. No further information is provided in order to test the scientists' morphology skills and report what is found on the film without bias.

Results

The most common morphological comment selected by participants was thrombocytopaenia (96.6%), echinocytes/crenated cells (70%), plasmodium falciparum (29.6%), reactive/plasmacytoid lymphocytes (28.9%) and atypical/suspect reactive lymphocytes (26.9%). 17% of the participants reported the film quality as unsatisfactory (17%), which is unusually high. The reasons provided were 'poor spreading' (48%) and 'poor staining' (34%) which are common causes for 'debris' to be seen on a film. The parasites present in this unusual case were misinterpreted as 'debris' on the film by many who took part in this exercise.



(a) Free lying *Babesia microti* trophozoite

(b) Cluster of extra-cellular trophozoites

(c) Ring forms can be more irregular in shape

(d) Erythrocytes with fragments which are also known as schistocytes

(e) Cluster of extra-cellular trophozoites

(f) Intracellular parasites can appear similar to *Plasmodium falciparum*

Figure 2 – *Babesia microti* trophozoites in both intracellular and extracellular forms

A significant number of participants suspected the presence of a parasite; 32% participants reported malaria (mainly P. falciparum), 16% reported 'other parasite' and a further 9% mentioned babesia or babesiosis.

Figure 2 shows examples of the *Babesia microti* trophozoites in both intracellular and extracellular forms seen on these blood films. A thorough morphological examination was carried out on these blood films and it was noted that the features of Babesia were difficult to find however they were present. They were possibly dismissed as "debris" by many. Furthermore, no maltese cross formations were found, which is a diagnostic feature of Babesia. Approximately 47% of participants noticed the parasites present in the red cells (figures 2c, 2d and 2f), only 9% noticed the extracellular parasites (figures 2a, 2b and 2e), which are a unique feature of babesia and not present in malarial infections. The thrombocytopaenia could trigger suspicion of a parasite present, however there are many conditions that present with thrombocytopaenia and there was limited clinical information provided. It is also important to note that the blood films were stained with May-Grunewald Giemsa stain, whereas films made for parasite identification are usually stained with Wright's Giemsa which makes visualisation of parasites easier.
This case proved to be very challenging for the participants in this EQA programme and this can be attributed to the lack of available clinical details and the problem of Babesia features being mistaken for 'debris'.

Conclusions

This exercise shows how EQA can be used to expose participants to unusual but clinically significant findings in morphology, particularly as Babesia is not endemic in the United Kingdom, although a form of babesiosis originating in England has recently been reported[1]. This is a great opportunity to remind all morphology scientists to beware of dismissing 'debris' too quickly, as demonstrated in this case '*debris' is not always debris*!

[1] Reference

Chan WY, Macdonald C, Keenan A, Xu K, Bain BJ and Chiodini PL (2021) Severe babesiosis due to *Babesia divergens* acquired in the United Kingdom. *Am J Hematol*, **96**, 889–890.