When not to test JAK2; real-world JAK2 mutation testing for the investigation of erythrocytosis

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INTRODUCTION

JAK2 testing is frequently ordered for investigation of erythrocytosis.

This study aimed to perform a retrospective audit of JAK2 mutation testing at Northern Health

- Identify ways to improve and rationalise the investigation of erythrocytosis
- Compare the effectiveness of Haemoglobin (Hb) and Haematocrit (Hct) reference ranges:
  - World Health Organisation (WHO
  - Northern Pathology Victoria (NPV)

METHODS

- All episodes of JAK2 mutation testing were extracted from pathology electronic records from January 2019 to December 2023.
- Testing included JAK2 V617F, JAK2 exon 12 and MPN NGS.
- Collected information included:
  - Patient demographics
  - Indication for testing and final diagnosis
  - Other laboratory results:
    - basic haematology & biochemistry within 1 week
    - iron studies within 3 months
    - EPO & bone marrow biopsy closest to the JAK2 test

RESULTS

- Most tests were for JAK2 V617F (87.2% of tests) with 5.4% of tests being MPN NGS
- 18 diagnoses of MPN, all of which were polycythaemia vera (6.3% of patients investigated for erythrocytosis)

Figure 1: Local HB & HCT references ranges

FBE		>16 Y				65Y—150Y			
		Male		Female		Male		Female	
TEST	Unit	Low	High	Low	High	Low	High	Low	High
HB	g/L	128	175	113	160	122	170	115	155
RCC	x10^12/L	4.0	5.9	3.6	5.3	4.0	5.7	3.8	5.4
MCV	fL	80	97	80	97	80	97	80	97
HCT	L/L	0.36	0.53	0.32	0.46	0.36	0.49	0.32	0.45
MCH	pg	27	34	27	34	26	34	25	34

Table 1: Characteristics of patients investigated for erythrocytosis expressed as number of patients (% of patients) or mean (standard deviation) for continuous values.

	Polycythaemia Vera	Secondary erythrocytosis or low JAK2 allele burden
Number of patients (%)	18 (6%)	268 (94%)
Female (%)	8 (44%)	76 (28%)
Age (years)	67 (12.1)	55 (14.8)
Hb (g/L)	192 (22.1)	179 (14)
HCT (L/L)	0.59 (0.08)	0.54 (0.05)
WCC (x10^9/L)	9.9 (1.7)	8.7 (4.4)
Platelets (x10^9/L)	398 (132.3)	231 (74.4)
EPO (U/L)	4.2 (4.5)	11.5 (9.9)

Table 2: Characteristics and final diagnoses of patients according to different Hb reference range thresholds expressed as number of patients (% of patients) or mean (standard deviation) for continuous values.

CLASS 1	CLASS 2	CLASS 3
Females: less than upper limit of local reference range <160g/L for <65 <155g/L for 65+ Males: less than WHO criteria <165g/L for all males	Females: 160g/L for <65 155-160g/L for 65+ Males: 165-175g/L for <65 165-170 g/L for 65+	Females: greater than WHO criteria > 160g/L for all females Males: greater than local reference ranges > 175g/L for <65 >170g/L for 65+

	Class 1	Class 2	Class 3	p-value
Number of patients, n(%)	22 (7.7%)	45 (15.7%)	219 (76.6%)	
Females (%)	18 (81%)	5 (11%)	61 (28%)	
Age (years)	58 (18.5)	52 (14.2)	56 (14.7)	0.36
Hb (g/L)	153.5 (5.1)	165.6 (4.7)	184.9 (12.8)	**<0.01**
HCT (%)	0.46 (0.024)	0.5 (0.022)	0.56 (0.046)	**<0.01**
White cell count (x10^9/L)	8.1 (2.4)	8 (2.6)	8.9 (4.6)	0.33
Platelets (x10^9/L)	241 (89.7)	244.6 (80.3)	239 (89.3)	0.82
Diagnosis (n, %)				
No cause found	6 (27%)	12 (27%)	25 (11.5%)	**0.014**
Hypoxia (eg: smoking, chronic lung disease, OSA)	11 (50%)	23 (51%)	125 (57%)	0.64
SGLT2i	1 (4.5%)	0 (0%)	6 (2.8%)	0.16
Testosterone	1 (4.5%)	0 (0%)	7 (3.2%)	0.46
Polycythaemia vera	0 (0%)	0 (0%)	18 (8.2%)	**0.045**
Low JAK2 allele burden	0 (0%)	0 (0%)	3 (1.3%)	0.61
Other secondary cause (including multiple of above)	3 (13%)	10 (22%)	35 (16%)	0.58

Table 3: Characteristics and final diagnoses of male patients according to different Hct reference range thresholds expressed as number of patients (% of patients) or mean (standard deviation) for continuous values.

	Class 1: HCT < 0.49	Class 2: HCT 0.49-0.53 <65yrs	Class 3: HCT >0.53 <65yrs or >0.49 in 65+yrs	p-value
Number of patients, n(%)	5 (2.5%)	58 (29%)	139 (68.5%)	
Age (years)	55.4 (18.8)	47.7 (11.3)	56.6 (14.3)	**<0.01**
Hb (g/L)	156.6 (7.8)	175 (5.5)	189 (12.4)	**<0.01**
HCT (%)	0.46 (0.022)	0.52 (0.013)	0.58 (0.039)	**<0.01**
White cell count (x10^9/L)	7.5 (2.5)	8.4 (2.9)	8.8 (5.3)	0.72
Platelets (x10^9/L)	211 (27)	243 (68)	226 (81)	0.33
Diagnosis (n, %)				
No cause found	1 (20%)	14 (24%)	14 (10%)	**0.03**
Hypoxia (eg: smoking, chronic lung disease, OSA)	2 (40%)	35 (60%)	82 (59%)	0.67
SGLT2i	1 (20%)	1 (2%)	3 (2.2%)	**0.04**
Testosterone	0 (0%)	0 (0%)	7 (5%)	0.19
Polycythaemia vera	0 (0%)	0 (0%)	10 (7.2%)	0.09
Low JAK2 allele burden	0 (0%)	0 (0%)	3 (2.2%)	0.50
Other secondary causes (including multiple)	1 (20%)	8 (14%)	20 (14.4%)	0.84

Table 4: Characteristics and final diagnoses of female patients according to different Hct reference range thresholds expressed as number of patients (% of patients) or mean (standard deviation) for continuous values.

	Class 1: HCT <0.46 in <65yrs & <0.45 in 65+yrs	Class 2: HCT 0.46-0.48 in <65yrs & 0.45 - 0.48 in 65+yrs	Class 3: HCT >0.48	p-value
Number of patients (%)	6 (7%)	18 (22%)	60 (71%)	
Age (years)	68 (20.4)	55 (17.6)	60 (15)	0.22
Hb (g/L)	149.3 (7.1)	158.1 (4.5)	176 (12.7)	**<0.01**
HCT (L/L)	0.43 (0.014)	0.47 (0.08)	0.54 (0.047)	**<0.01**
White cell count (x10^9/L)	8.2 (3.2)	7.8 (2)	9.2 (3.2)	0.17
Platelets (x10^9/L)	206 (64.5)	286 (84.9)	266 (115.7)	0.29
Diagnosis (n, %)				
No cause found	2 (33.3%)	5 (28%)	8 (13%)	0.27
Hypoxia (eg: smoking, chronic lung disease, OSA)	3 (50%)	9 (50%)	31 (52%)	0.92
SGLT2i	0 (0%)	1 (5.5%)	1 (2%)	0.62
Testosterone	0 (0%)	1 (5.5%)	0 (0%)	0.18
Polycythaemia vera	0 (0%)	0 (0%)	8 (13%)	0.17
Low JAK2 allele burden	0 (0%)	0 (0%)	0 (0%)	-
Other secondary causes (including multiple)	1 (16.7%)	2 (11%)	12 (20%)	0.69

DISCUSSION

- PV was an uncommon cause of erythrocytosis (~6.3%) and no diagnoses of PV were missed using either local reference ranges or WHO criteria
- For Hb:
  - In men, the local reference range was higher for all age groups
  - In women, the WHO range was higher than local for 65+yrs, same for <65yrs
- For Hct:
  - In men, the local reference range was higher for <65yrs; same as WHO for 65+yrs
  - In women, the WHO range was higher for all age groups
- Limitations included;
  - a single centre study
  - genetic testing landscape has changed dramatically since research period, changing from predominantly single gene testing to MPN NGS panels
  - retrospective analysis
  - documentation quality varied and some patients lost to follow-up

CONCLUSIONS

- Polycythaemia vera is an uncommon cause of erythrocytosis
- In women, the WHO criteria for Hct and Hb were the most efficient at detecting PV for all ages
- In men, the local reference ranges for Hct and Hb were the most efficient at detecting PV for all ages
- Increasing the Hb and Hct thresholds for JAK2 mutation testing may better rationalise the detection of PV.

Northern Health RESEARCH WEEK 20-24 OCTOBER 2025 INSPIRED RESEARCHERS Northern Health