SPECTRUM OF DISEASES DIAGNOSED BY BONE MARROW EXAMINATION IN CENTRAL INDIA

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ABSTRACT: BACKGROUND: Hematological diseases are quite frequent in population and mostly present with anemia. Although diagnosed mostly by clinical examination and simple investigations, bone marrow examination is confirmatory for the diagnosis. Bone marrow aspiration is simple and safe invasive procedure. Spectrum of hematological diseases in the developing countries is different from those observed in developed countries. **OBJECTIVE:** There is a lack of information about the exact relative prevalence of hematological disorders especially in Central India that's why this study was undertaken to correlate clinico-hematological and bone marrow findings. **MATERIAL AND METHODS:** It is a retrospective study of 345 cases who underwent bone marrow examination in Department Of Pathology, Gandhi Medical College, Bhopal. **RESULTS:** In our study age range of cases was 2-80 years with male: female ratio of 1.28:1. Most common diseases observed were megaloblastic anemia (41.81%), dimorphic anemia (16.72%), hypoplastic anemia (9.09%), ITP (8.36%) and infective pathology (4.72%). In Hematological malignancies (20.28%), most common were AML (6.95% of overall & 34.28% of malignant diseases %) followed by ALL (4.92 % overall and 24.28% % of malignancies). 3 cases of Gaucher's disease were also reported. 96.81% patients were anemic while 44.92% were pancytopenic. Most common presenting symptoms were fever (44.92%) and bleeding manifestations (30.14%) which include ecchymosis, epistaxis, petechiae, melena, hematuria and hematemesis. Splenomegaly, hepatomegaly and lymphadenopathy were found in 35.65%, 21.15% and 14.78% of cases respectively. **CONCLUSION:** In our study final diagnosis could be made in majority of cases (92.76%) by bone marrow examination, although having varied clinical presentations such as fever, bleeding manifestations, ascites, hepatomegaly, splenomegaly etc and usually associated with anemia / pancytopenia/ thrombocytopenia. All hematological cases must undergo bone marrow examination to conclude the exact underlying pathology for appropriate treatment.

KEYWORDS: Bone marrow aspiration, Disease pattern, Central India.

INTRODUCTION: Hematological disorders are quite frequent in population, most of the times diagnosis can be arrived at by detailed clinical examination and few simple investigations. Most of the disorders presented with anemia. However without bone marrow examination diagnosis is usually not confirmed. Bone marrow aspiration will yield diagnostic result in majority of cases ¹. ² Although bone marrow aspiration and biopsy are an uncomfortable procedure for the patient and should be performed only when there is a clear clinical indication ³ Bone marrow aspiration examination is one of the most frequent and relatively safe invasive procedure. Though invasive it can be easily performed even in presence of thrombocytopenia with little or no risk of bleeding.

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The spectrum of hematological diseases is relatively different in the developing world than the developed countries⁴. Megaloblastic anemia is common non -malignant hematological disorder⁵, 60-80% of the world population is affected by IDA and is most common preventable nutritional anemia in the world^{6, 7, 8}.

AIM: Aim of this study was to understand the pattern of diseases encountered in bone marrow aspiration and to correlate bone marrow aspiration findings with age, gender, clinical and peripheral blood findings.

MATERIAL AND METHODS: This was a retrospective study. We reviewed all cases of bone marrow examination reported in the Department of Pathology, Gandhi Medical College, Bhopal over a period of four years from 2009-2012. Patients' clinical details, the demographic data, clinical symptoms hematological parameters were taken from medical records, were reviewed and peripheral blood and bone marrow smears stained with Leishman stain were studied. Additional stain MPO /PAS were used whenever necessary. Final diagnosis was confirmed by bone marrow examination morphologically using criteria for diagnosis as per standard norms in the text.

Inclusion criteria-All cases coming for bone marrow examination. Indications were unexplained anemia, pancytopenia, thrombocytopenia, fever, hepatosplenomegaly or bleeding manifestations.

Aspirates with inadequate material or dry tap were excluded from the study.

Ethics-Study was approved by Institutional Ethical Committee.

RESULT: Total of 345 patients who had undergone bone marrow examination were studied which consisted of 194 males and 151 females with male: female ratio-1.28:1. The age of patients ranged from 1-80 years. We found 99 (28.69 %) cases in pediatric age group (0-14 years). Maximum cases 129 (37.39%) were in age group 20-40 years, 48 (13.91%) & 20(5.79%) cases were in the age group 41-60 and 61-80 years respectively

S.No	Diagnosis	No.of cases (%) (out of 345 total cases)	Male:Female	Maximum cases in age group
1.	Megaloblastic anemia	115 (33.33%)	72/43 (1.67:1)	52 cases- 20-40 years, 24 cases- 0-14 years
2.	Dimorphic anemia	46(13.33%)	22/24 (0.91:1)	19cases- 20-40 years 12 cases- 0-14 years
3.	Hypoplastic anemia	25 (7.25%)	3/12 (1.08:1)	9 cases- 20-40 years 8 cases- 0-14 years

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4	ITP	23. (6.67%)	11/12 (0.91:1)	10 Cases- 0-14years, 5 cases- 20-40 years
5	Infective pathology	13 (3.77%)	6/7 (0.85:1)	6 cases- 20-40 years

TABLE1: DISEASEPATTERN OF MOST COMMON DISEASES WITH AGE & SEX DISTRIBUTION

Other diseases were 7, 5, 4, 3 & 2 cases of hypersplenism, megaloblastic anemia with eosinophilia, aplastic anemia, Gaucher's disease and CDA respectively. 1 case of CH syndrome, vWD, hemolytic anemia, Erythroid hyperplasia and one normal bone marrow was found. In 25(7.25%) cases diagnosis could not be made due to laboratory constraints.

S.NO	DISEASE	0-14 years	15-19 years	20-40 years	41-60 years	60-80 years	Total	M:F
1.	AML	8	3	8	4	1	24	1:2
2.	CML-accelerated phase	0	0	2	0	0	2	1:1
3.	MPD	4	1	1	1	0	7	1:6
4.	PV	0	0	1	0	0	1	1:0
5.	ET	0	0	0	1	0	1	0:1
6.	PNH	0	1	0	0	0	1	1:0
7.	MDS	0	1	1	0	0	2	2:0
8.	ALL	11	1	5	0	0	17	3.2:1
9.	NHL	1	1	0	2	0	4	3:1
10.	Hodgkin's Disease	0	0	2	0	0	2	2:0
11.	CLL	0	0	0	1	0	1	1:0
12.	MM	0	0	1	2	3	6	5:1
13.	Round cell tumor	1	0	0	0	1	2	1:1
TOTAL		25	8	21	11	5	70	

Table. 2: Distribution of Hematological Malignancies according to Age & sex.

Out of 345 cases malignancy was found in 70(20.28%) cases, out of which most common were AML 24 cases(6.95% overall; 34.28% of malignancies) followed by ALL 17 cases (4.92% overall;24.28% of malignancies), others were 7 (2.02% overall;10% of malignancies) and 6(1.73% overall;8.57% of malignancies) and 4(2.89 % overall and5.7% of malignancies) were MPD(not otherwise specified), MM and NHL respectively. 2 cases, each of Hodgkin's disease and Round cell tumor, and 1 case each of PV, ET, PNH & CLL were found.

In ALL, maximum cases 64.7%(11/17) were under 14 years and AML 33.33 %(8/24) each were found in the age group of under 14 & 20-40 years.

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In AML male: female ratio 1.4:1; maximum cases 8 were observed in both 0-14 and 20-40 years age groups followed by 3 cases each of AML M1 and unclassified AML, 2 cases of AML M6 and 1 each of AML M5 and AML M2E0. Most common were AML M3 and AML M7, maximum (4/5) each were in below 40 years age group.

Among MPD, all 6 cases were female in variable age groups and in MDS both cases were males below 40 years.

In lymphoid malignancies, male: female ratio was 3.25:1; maximum cases (11) were in 0-14 year's age group. Maximum cases in ALL were in ALL- L 2 (15 cases).

In Multiple myeloma, male: female ratio was 5:1 with maximum cases in above 40 years age group.

S.No	Symptom	No of cases out of 345(%)	Most common diseases/ %& No of cases	Other Diseases
1.	Fever	155(44.92%)	100% in CML, CLL, ET and CH syndrome 75% AA & hypersplenism Overall leukemia 48.48%(16/33) 58.82%-ALL, 50%-AML 56%-HA 47.82%-Infective pathology & ITP, 39.13%MA, 34.78% DA	Number of cases were too low to draw any conclusion
2.	Ascites	20(5.80%)	Dimorphic anemia 4/46(8.69%)	Megaloblastic anemia 5/115(4.34%)
3.	Edema	12(3.48%)	Dimorphic anemia 5/46(10.86%)	Megaloblastic anemia 6/115(5.21%)
4.	Pedal edema	29(8.41%)	Megaloblastic anemia 18/115(6.95%)	DA 3/46(6.52%)
5.	Facial edema	11(3.19%)	Megaloblastic anemia 5/115 (4.34%)	DA 2/46(4.34%)
6.	Visual disturbances	3(0.87%)	DM 1/46(2.17%)	MA 2/115(1.73%)
7.	Gall stones	6(1.74%)	1 each in DM(2.17%), ITP(4.34%), NHL (25%)	MA 2/115 (1.73%)
8.	Rash	12(3.48%)	Hypoplastic anemia 5/25(20%),	ITP 4/23(17.39%)
9.	Jaundice	14(4.06%)	Infective pathology 2/13 (15.38%)	MA 8/115(6.95%)

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10.	Icterus	10(2.90%)	Megaloblastic anemia 6/115 (5.21%)	DM2/46(4.34%)
11.	Gall stones	6 (1.74%)	NHL (25%),	1 each in DM (2.17%), ITP (4.34%), MA 2/115(1.73%)
12.	Bleeding Per Rectum	3(0.87%)	DM 1/46 (2.17. %)	MA 2/115(1.73%)
13.	Ecchymosis	6(1.74%)	ITP 4/23(17.39%)	HA2/25(8%)
14.	Epistaxis	31(8.99%)	ITP 6/23(26.08. %))	MA 9/115(7.82%)
15.	Petechiae	8(2.32%)	AA 1/4(25%),	3 Each ITP 3/23 (13.04%), HA 3/25(12%)
16.	Gum Bleed	16(4.64%)	ITP 5/23(21.73%),	MA 4/115(3.47%).
17.	Hematemesis	13(3.77%)	ITP 5/23 (21.73%)	MA 4/115(3.47%)
18.	Hematuria	9(2.61%)	L2 2/15(13.33%)	2 Each ITP2/23 (8.69%), MA4/115(3.47%)
19.	Malena	18(5.22%)	Hypersplenism 3/7(42.85%)	3 Each L2 3/15(20%), MA 6/115(5.21%)

TABLE 3: Most common symptoms with most common diseases found were as follows

In our study, most common symptoms observed were fever, bleeding manifestations followed by edema/ascites.

Most common causes found among bleeding manifestations-for ecchymosis, gum bleed, hematemesis and epistaxis were ITP followed by MA, for petechiae were Aplastic anemia followed by ITP while for malena and hematuria were hypersplenism and ALL L2 respectively.

Lymphadenopathy was found in 51cases (14.78%), (Generalized lymphadenopathy-15 cases and cervical & mesenteric-9 cases each) for which most common cause was ALL. Splenomegaly (35.65%)and hepatomegaly (21.15%) were most common in MA followed by dimorphic anemia. Osteolytic lesions(2.89%) were found in MM.

Pancytopenia was observed in 155 cases (44.92%) cases.

Most common underlying diseases (with number of cases) were Megaloblastic anemia(67), Hypoplastic anemia(20), Dimorphic anemia(13), ALL(9), AML & Infective pathology (8 each).Other causes were hypersplenism and ALL L2 (5each), Aplastic anemia(4), ITP and MDS(3 each)and one each of AML- M6, AML -M7, ALL - L1, Chediak Higashi syndrome, Congenital dyserythropoietic anemia, Pure red cell aplasia, Erythroid hyperplasia and Gaucher's disease.

Hematological parameters in our study were as follows:

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Hb (gm./dl)	Anemia			Normal	Increased
	1-5 (severe)	5-8 (moderate)	8-11(mild)	11-18	18-25
No. of cases	153(44.34%)	142 (41.15%)	39(11.30%)	10 (2.89%)	1
Most common cause	MA 56.52%(65/115) DM 47.82%(22/46)	DM 50%(23/46) MA 35.65% (41/115)	ITP 17.39% (4/23) MA 7.82% (9/115)	ITP 17.39% (4/23)	Polycythemia Vera

TABLE 4: Distribution of cases according to severity of anemia with most common causes

Diseases	Severe Hb<6 gm./dl No of cases	Moderate Hb 6-8gm/dl No of cases	Mild Hb8-11 gm./dl No of cases	Total No of cases
Megaloblastic anemia	65	41	09	115
Dimorphic anemia	22	23	01	46
Hypoplastic anemia	16	19	00	25

Table 5: Common diseases with number of cases according to severity of anemia

s.no	TLC	No of cases out of 345	Most common cause in decreasing order
1.	<4, 000 (decreased)	186(53.91%)	Hypoplastic anemia 100%(25/25), Infective pathology 69.23%(9/13), MA 64.34%(74/115), >DM 43.47%(20/46), >L240%(6/15)
2.	4, 001-11, 000 (normal)	109(31.59%)	ITP 60.86%(14/23), >DM50%(23/46), >MA 30.43%(35/115)
3.	11, 001-50, 000 (moderately increased)	38(11.01%)	MPD50% (3/6), >33.33% L2 (5/15), & MM(2/6), >ITP 26.08%(6/23), >DM 6.52% (3/46), MA5.21% (6/115)
4.	50, 001-10, 00, 000(severely increased)	12(3.47%)	100% CLL (1/1), > 50% M2 (2/40), L1 (1/2), >20% M3 & M7 (1/5)) and L2 (3/15).

Table 6: Distribution of most common diseases according to Total Leucocyte Count

Most common causes for leucopenia were hypoplastic anemia, megaloblastic anemia & infective pathology, for normal count were ITP & Dimorphic anemia while for moderate leukocytosis were MPD & MM, and for marked leukocytosis it was leukemia.

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s.no	Platelet count (per mm ³)	No of cases out of 345(%)	Causes%(no of cases out of respective disease)
1.	<50, 000	167(48.40%)	100% -AA(4/4), MDS, M6, L1 (each 2/2) M5, M2Eo(1/1), PV, PNH(1/1), 80% HA (20/25), AML M3 (4/5), 75%AML M2, NHL (3/4 each), 73.33% ALL L2(11/15), 65.21% ITP(15/23), 60% AML M7(3/5,) 57.14% hypersplenism(4/7), 53.84% Infective pathology(7/13), 30.43% DM (14/46).
2.	50, 001-1, 00, 000	86(24.92%)	100%-PNH, vWD(each1/1), 66.66%-AML(2/3), 42.85% -Hypersplenism(3/7) 30.43%- MA(35/115), 23.07% -Infective pathology (3/13), 21.73%- ITP, DM(5/23 &10/46 respectively)
3.	1, 00, 001-4, 00, 000 (normal range)	89(25.79%)	66.66% MPD(4/6), 47.82% DM(22/46), 24.34% MA(28/115)
4.	4, 00, 001-20, 00, 000	03(0.86%)	100% ET(1/1), 50%-CML in accelerated phase(1/2), 20% M7(1/5)

Table7: Distribution of most common diseases according to Platelet count

Most common causes for severe thrombocytopenia were Aplastic anemia, Leukemia & Megaloblastic anemia, for moderate thrombocytopenia were PNH, AML & hypersplenism, while for normal count were MPD & DM, and for increased count were ET & CML.

DISCUSSION: Bone marrow examination findings compared to other studies conducted were as follows-

Sl.no						
	STUDY/AUTHORS	Pudasaini S. et al (2012) ⁽⁹⁾	Fazlur Raheem et al (2005) ⁽¹⁰⁾	Anita Tahlan et.al(2008) ⁽¹¹⁾	Al-Ghazaly J et al (2006) ⁽¹²⁾	Our study (2009-2012)
	No.of cases	57	424	742	627 patients of hematologic al diseases other than lymphoma)	345

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	Age group	9 months-75 years	2-15 years	>15 years	>14years	1-80 Years
	Maximum cases in age group	31-45 years				20-40 years
	Male:female	1:1.1 (27:30)	1.96:1(281:143)		1.29:1	1.28:1
	PERIPHERAL BLOOD FINDINGS-					
	Anemia	86%				96.81%
	Pancytopenia	50%				44.92%
	Bicytopenia	36%				34.23%
	Thrombocytopenia	14%				12%
	BONE MARROW DIAGNOSIS					
	NON MALIGNANT			72%(609)		79.72%(275)
1.	Megaloblastic anemia	12.3%(7)	24.92%(103)	87%		41.81%(115)
2.	Pernicious anemia				0.74%(3)	
3	Dimorphic anemia/mixed	-	4.7%(20)		2.87%(18)	16.72%(46)
4	Microcytic anemia/IDA	7%(4)	3.53%(15)		10.36%(65)	
5	MA with malaria				9.09%(57)	
6	With excess eosinophils			2.2%		1.81%(5)
7	Infective pathology	12.3%(7)		3%		4.72%(13)
8	Erythroid hyperplasia	21%(12)				0.36%(1)
9	ITP/Megakaryocytic thrombocytopenia	10.5%(6)	9.43%(40)	2.9%	4.62%(29)	9.09%(23)
10	Hemolytic anemia		0.70%(3)			1.45%(1)
11	Hypoplastic anemia	5.3%(3)		2.6%		9.09%(25)
12	AIHA				1.43%(9)	
13	Aplastic anemia		14.15%(60)		5.74%(36)	2.54%(4)
14	Malaria falciparum		2.35%(10)			
15	Leishmaniasis	1.8%(1)	5.98%(25)		1.75%(11)	
16	Hypersplenism	-	2.59%(11)			2.03%(7)

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17	Gaucher's disease					1.09%(3)
	MALIGNANT			18% (133)		20.28% (70)
1	Acute leukemia	12.3%(7)	24.28%	41%	37.31%	58.57% (41)
2	ALL		17.92%(76)		11.96% (75)	24.28%(17)
3	AML		6.36%(27)		25.35% (159)	34.28%(24)
4	Lymphoma		1.17%(5)	14.2%		1.42%(1)
5	PV				1.11%(7)	1.42%(1)
6	ET					1.42%(1)
7	CML		0.47%(2)			2.85%(2)
8	CMPD			30%		8.57%(6)
9	MDS	3.5%(2)			2.07%(13)	2.85%(2)
10	PNH					1.42%(1)
11	Myelofibrosis				1.11%(7)	
12	HCL				0.31%(2)	
13	CLL			7.5%	5.74%(36)	1.42%(1)
14	MM	3.5%(2)		7%	1.27%(8)	8.57%(6)
15	Round cell tumor					2.85%(2)
16	Neuroblastoma		0.47%(2)			
17	Metastasis				0.31%(2)	
	Normal marrow	10.5%(6)				1.49%(1)

Table 8: Comparison of bone marrow studies with present study

Although study group was different in different studies- pediatric age group in Fazlur¹⁰, Anita¹¹ & Al -Ghazaly¹² as compared to our study where all age group was taken similar to Pudasaini S. et al⁹ all have reported male dominance.

Pudasaini S⁹ reported anemia, pancytopenia, bicytopenia & thrombocytopenia in 86%, 50%, 36% and 14% cases respectively in comparison to 96.81%, 44.92%, 34.23% & 12% respectively in anemia, pancytopenia, bicytopenia & thrombocytopenia in our series.

Our findings are similar to others as majority of cases were in nonmalignant group and most common disease in nonmalignant group was megaloblastic anemia, Anita et al¹¹ reported 87% MA unusually high, reason may be, she has studied all cases having anemia. In our series MA incidence is much higher 41.81% in comparison to 12.3% Pudasaini et al⁹ study in Nepal & 24.92% reported by Fazlur Rahman et al¹⁰, Pakistan, both are different geographically and in food behavior.

Dimorphic anemia in our study was 16.72 % much higher than 4.7% in Fazlur Raheem et al¹⁰ and 2.87% in AL-Ghazaly J et al¹² while Pudasaini S et al⁹ and Anita et al¹¹ did not report it.

Hypoplastic anemia was 9.09% in our study higher than 5.3% in Pudasaini S. et al⁹ and 2.6 % in Anita et al¹¹.

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ITP incidence in our study was 8.36 % higher than 4.62 % by AL-Ghazaly¹² and 2.9% by Anita et al¹¹ and lower than 10.5% by Pudasaini S. et al⁹ and 9.43% by Fazlur Raheem et al¹⁰.

Among infective pathology group, 4.72% was observed slightly higher than 3% reported by AL –Ghazaly et al ¹² and much lower than 12.3% reported by Pudasaini S et al⁹.

Aplastic anemia was found in 1.45% in our series as compared to 14.15% by Fuzlur Rehman et al¹⁰ and 5.74% by AL –Ghazaly et al¹¹.

Megaloblastic anemia with eosinophilia was reported 2.2 % by Anita et al ¹¹ in contrast to our 1.81%.

We found 3 cases of storage disorder –Gaucher's disease while it has not been reported by others.

Malaria was reported by AL –Ghazaly et al ¹² and Leishmaniasis was reported by Pudasaini et al ⁹, Fuzlur et al ¹⁰ and AL-Ghazaly¹² while we have not found parasitic infestation.

Pernicious anemia was found by Al Gazalye¹² alone, not by others as similar to our study.

Hematological malignancies constituted 20.28% of our cases higher than 18 % reported by Anita Tahlan et al¹¹, but lower than 27.12% by Fazlur Rahman et al¹⁰, and 47.48% by Al –Ghazaly J¹².

In our study, we observed 58.27% acute leukemia (34.28% AML, 24.28% ALL), MPD 8.57 %, MM 8.57% and NHL 1.42 % while Pudasaini S et al⁹ reported Acute leukemia 12.3%, MDS 3.5% & MM 3.5%, Fazlur Rahim et al¹⁰ found Acute leukemia 24.28% (AML 6.36 % & ALL 17.92%), Lymphoma 1.17%, and 0.47% each CML & Neuroblastoma. Anita et al¹¹ also reported 41% Acute Leukemia, CMPD 30%, Lymphoma 14.2%, CLL 7.5% & MM 7%, Al-Ghazaly J¹² found Acute leukemia in 37.31% (AML 25.35%, ALL 11.96%), CLL 5.74% & MM 1.27%.

We have not found HCL and metastasis in bone marrow while it was reported by Al-Ghazaly Jet al ¹².

This variation may be due to differences in the studied age groups & population in different areas.

Number of cases of IDA, CML & hemolytic anemia are very less in our study, as in other studies also because majority of IDA and mixed anemia, CML & hemolytic anemia are diagnosed on smear examination & needs other tests not bone marrow for confirmation of diagnosis.

CONCLUSION: Bone marrow examination is an important step to reach at confirmatory diagnosis of majority of hematological disorders. In our study, patients commonly presented with fever, bleeding manifestations and organomegaly. The most common nonmalignant diseases in decreasing order of frequencies were megaloblastic anemia, dimorphic anemia, hypoplastic anemia, ITP, Infective pathology and hematological malignancies. Among hematological malignancies, most common were AML followed by ALL.

Anemia is not a diagnosis but manifestation of an underlying disorder. Thus, even mild asymptomatic anemia should be investigated to ascertain the cause; if we wish to give appropriate treatment and bone marrow examination will clinch the diagnosis in majority of hematological diseases. Further proper documentation will definitely help in planning of health policies and National programs of our country.

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Abbreviations used:

MA-Megaloblastic Anemia
 CH-Chediak Higashi syndrome
 IDA-Iron Deficiency Anemia
 DM-Dimorphic Anemia
 CDA- Congenital Dyserythropoietic Anemia
 ITP- Immune Thrombocytopenia
 AA-Aplastic Anemia
 HA-Hypoplastic Anemia
 MM-Multiple Myeloma
 vWD-Von Willebrand Disease

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