WHITE BLOOD CELLS IN POLISH ATHLETES OF VARIOUS SPORTS DISCIPLINES

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ABSTRACT: The purpose of this study was to examine the diversity of white blood cell (WBC) counts and their subsets (neutrophils, lymphocytes and monocytes) among competitive athletes of different sports disciplines. The blood samples were collected from 608 healthy, medically examined athletes (181 females and 427 males) aged 20.1 \pm 5.1 years, who represented five sport disciplines: canoeing, judo, rowing, swimming and volleyball. All blood samples were taken from the antecubital vein in the morning, after overnight fasting, in a seated position. Haematological analyses were conducted using a haematology analyser (ADVIA 120, Siemens). Neutropenia (defined as neutrophil count $< 2.0 \cdot 10^9 \cdot L^{-1}$) was found in athletes of both sexes in each discipline. There was no incident of lymphopenia (defined as a lymphocyte count $< 1.0 \cdot 10^9 \cdot L^{-1}$). Monocytopenia (defined as a monocyte count $< 0.2 \cdot 10^9 \cdot L^{-1}$) was seen only in male athletes, except judo athletes. Differences in WBC and their subset counts were related to sport disciplines: in volleyball players WBC counts were significantly higher than in athletes of canoeing and rowing (in females); neutrophil counts were the lowest in swimming athletes; lymphocyte counts were lower in athletes of canoeing than in volleyball and swimming, but only in females; monocyte counts were lower in athletes of canoeing than swimming (in females) and judo (in males). In women, counts of neutrophils were greater and counts of monocytes were smaller than in men. It seems that prevalence of neutropenia and monocytopenia and differences in WBC counts and their subsets among disciplines could be related to the adaptive response to physical exercise.

KEY WORDS: white blood cells, sports, neutropenia, monocytopenia, lymphopenia, reference values

INTRODUCTION

White blood cells (WBC) are one of the most important cells in the immune system, playing a role in defence against foreign substances. Neutrophils are first line defenders against bacterial infection, macrophages phagocyte foreign substances and enable antigen presentation, while lymphocytes activate other lymphocyte subsets and produce immunoglobulins. It was proved that physical exercise induces changes in the number of leucocytes and their subsets in the circulating blood [18]. These changes can be influenced by many factors such as catecholamines, increase in cardiac output or decreased adherence of leucocytes to endothelial tissues [9]. It is suggested that suppression of the immune system could increase susceptibility to upper respiratory tract infections (URTI) in athletes [13].

For many years researchers have examined a lot of aspects of the immune system. There are some papers concerning the influence of the selected exercise programme on immunology indicator changes [16,21]. To the author's knowledge there are only a few articles concerning WBC counts and their subsets in athletes at rest. In the

majority of these studies, WBCs are considered in only one kind of sport discipline [1,10,22].

The purpose of this study was to examine the diversity of white blood cell counts and their subsets (neutrophils, lymphocytes and monocytes) among competitive athletes of different sports disciplines.

MATERIALS AND METHODS

The blood samples were collected from 608 athletes (181 females and 427 males) of mean age 20.1 ± 5.1 years who represented five different sport disciplines: canoeing, judo, rowing, swimming and volleyball (Table 1). The majority of subjects were well-trained or elite athletes. Some samples were taken from the same athletes from two consecutive years; the data were not aggregated for those samples. The minimum break time between successive blood collecting was 6 months. Moreover, blood sample taking was done in different annual training cycles. The programme of the study was approved by the Ethical Research Committee at the Institute of Sport in Warsaw.

Accepted for publication 15.01.2012

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TABLE I. MEAN \pm STANDARD DEVIATION (SD) AGE OF SUBJECTS IN DISCIPLINES

Gender		Female		Male
Discipline	Ν	Age	Ν	Age
Judo	26	19.5 ± 3.0	43	21.5 ± 4.4
Canoeing	33	19.1 ± 4.2	62	18.5 ± 3.6
Volleyball	35	17.7 ± 4.5	156	23.3 ± 5.7
Swimming	31	16.2 ± 3.5	33	15.9 ± 2.9
Rowing	56	20.7 ± 3.3	133	21.0 ± 5.0
All	181	18.9 ± 4.0	427	21.2 ± 5.4

All blood samples were collected from the antecubital vein in the morning, after overnight fasting, in a seated position from healthy, medically examined athletes. The blood samples were taken in a preparatory period in each discipline. The break between the last training session and blood sample taking was 12 hours minimum. Subjects were informed that the last training unit should not be intensive. The influence of menstrual cycle phase and use of contraceptives on white blood cell counts has not been investigated in this study.

Haematological analyses were conducted within 2 hours after taking the blood in the Biochemistry Department's Laboratory, which has accreditation of the Polish Centre for Accreditation. A haematology analyser (ADVIA 120, Siemens) was used for analyses, which included white blood cell, lymphocyte, neutrophil and monocyte counts.

Statistical analysis

Values for total WBC, neutrophils, lymphocytes, and monocytes were log transformed. Outliers were identified as observations with a standardized residual >5.0; these data represented 0.26% of the total and were excluded. In the next step, a two-way analysis of variance (gender x sports discipline) was conducted, followed by a least significant difference (LSD) post hoc test. The level of p≤0.05 was considered significant. Reference ranges were set as values

Gender	Gender Sport n discipline n		Total WBC count, [10 ^{9.} L ⁻¹]		Neutrophil count, [10 ⁹ ·L ⁻¹]		Lymphocyte count, [10 ⁹ ·L ⁻¹]		Monocyte count, [10 ^{9.} L ⁻¹]	
		Mean	Reference range	Mean	Reference range	Mean	Reference range	Mean	Reference range	
Judo	30	6.10	4.16 – 8.95	3.14	1.90 – 5.16	2.12	1.40 – 3.21	0.42	0.25 – 0.71	
	Canoeing	44	5.73	3.52 - 9.33	2.96	1.51 – 5.78	1.98	1.19 – 3.30	0.37	0.22 - 0.65
Female	Volleyball	36	6.54	4.12 – 10.36	3.46	1.81 – 6.59	2.26	1.54 – 3.30	0.45	0.29 - 0.68
remale	Swimming	32	5.95	4.12 - 8.61	2.81	1.56 – 5.08	2.35	1.33 – 4.15	0.46	0.08 - 2.62
	Rowing	79	5.72	3.40 - 9.60	2.78	1.31 – 5.89	2.16	1.35 – 3.45	0.39	0.22 - 0.70
	All	221	5.93	3.70 - 9.52	2.97	1.51 – 5.85	2.16	1.33 – 3.49	0.41	0.18 – 0.94
	Judo	52	6.20	3.72 – 10.33	3.04	1.51 – 6.14	2.14	1.28 – 3.59	0.47	0.28 – 0.79
	Canoeing	105	5.94	3.81 – 9.26	2.91	1.53 – 5.54	2.13	1.36 – 3.34	0.43	0.22 – 0.82
Mala	Volleyball	170	5.97	3.94 - 9.04	2.97	1.71 – 5.17	2.12	1.35 – 3.33	0.44	0.26 - 0.76
Male	Swimming	34	5.70	3.50 – 9.29	2.48	1.14 – 5.37	2.26	1.36 – 3.75	0.44	0.22 – 0.90
	Rowing	181	5.96	3.91 – 9.08	2.90	1.55 – 5.42	2.16	1.38 – 3.41	0.44	0.25 – 0.78
	All	542	5.96	3.85 – 9.23	2.91	1.55 – 5.46	2.15	1.35 – 3.41	0.44	0.25 – 0.79
All Sa	amples	763	5.95	3.81 – 9.32	2.93	1.54 – 5.57	2.15	1.35 – 3.43	0.43	0.22 – 0.84
Normal refe	erence range			4.50 – 11.0		2.00 - 8.00		1.00 - 4.80		0.20 – 0.78
Ge	ender effect			ns	F=	3.97; p<0.05		ns	F=	4.44; p<0.05
Dis	cipline effect		F=2	2.65; p<0.05	F=5	5.18; p<0.001	F=	2.74; p<0.05	F=	2.76; p<0.05
I	nteraction		F=2	2.06; <i>p=0.08</i>	F=:	2.22; p=0.07		ns		ns
						Post hoc	LSD test	s		
			Volley	ball > Canoeing, p<0.05	Volley	/ball > Canoeing, p<0.05	Volley	/ball > Canoeing, p<0.05	Swimr	ning > Canoeing, p<0.05
Fer	Female		Volleyball > Rowing, p<0.01		Volleyball > Rowing, p<0.001		Swimming > Canoeing, p<0.01			
					Volley	ball > Swimming, p<0.001				
					Judo > Swimming, p<0.01				Judo > Canoeing, p<0.05	
	-1-				Canoe	eing > Swimming, p<0.01		p<0.05 p		
M	ale			_	Volley	ball > Swimming, p<0.01		_		
					Rowing > Swimming, p<0.01					

within ±2SD of arithmetic means. The logarithmic mean values and reference values of the cell counts for each sport were backtransformed. These values were also compared to previously established reference ranges [8]. The following threshold values were set to define neutropenia, lymphopenia, and monocytopenia: $<2.00 [10^9 \cdot L^{-1}], <1.00 [10^9 \cdot L^{-1}], <0.20 [10^9 \cdot L^{-1}]$, respectively. All calculations were done with SPSS version 17 (SPSS Inc., Chicago, IL).

RESULTS

Table 2 presents total WBC and their subset counts in female and male athletes from the selected disciplines. There was no gender effect on total WBC counts. In women the highest mean WBC counts were observed in volleyball players, which was significantly higher than in athletes training in canoeing (p<0.05) and rowing (p<0.05). In men, there were no significant differences related to disciplines. There was no significant interaction effect (gender x discipline) in WBC counts, but the results of statistical analysis suggest a tendency (p=0.08).

Neutrophil counts were related to gender (p<0.05) and discipline (p<0.001); there was a tendency (p=0.07) for an interaction effect (gender x discipline). Mean neutrophil count was higher (p<0.05) in female than in male athletes. In females, the highest mean neutrophil count was noted in volleyball players; it was significantly higher than among female canoeists (p<0.05), rowers (p<0.001) and swimmers (p<0.001). In men, swimmers had the lowest mean neutrophil count, significantly lower than in other sport disciplines: judo (p<0.01), canoeing (p<0.01), volleyball (p<0.01), and rowing (p<0.01). For male athletes, neutrophil counts mirrored total WBC counts, with the lowest neutrophil counts for swimming and the highest for judo. Neutropenia (defined as neutrophil count <2.0 $\cdot 10^9 \cdot L^{-1}$) was found in athletes of both sexes in each discipline (Table 3).

Lymphocyte counts did not depend on gender, but a discipline effect was noted. In female athletes, the lowest mean lymphocyte count was observed in canoeists – significantly lower than in athletes of volleyball (p<0.05) and swimming (p<0.05). In men, mean lymphocyte counts did not differ between selected disciplines. There was no sample with lymphopenia (defined as lymphocyte count <1.0 $\cdot 10^9 \cdot L^{-1}$) (Table 3).

Monocyte counts depended on gender and discipline, but no interaction effect was found. Monocyte counts were higher in male than in female athletes. Both in female and male canoeists the lowest mean monocyte counts were found, but it was only significantly higher than in swimming and judo respectively. Monocytopenia (defined as monocyte count $< 0.2 \cdot 10^9$.L⁻¹) was seen only in male athletes, except judo athletes (Table 3).

DISCUSSION

This study provides information about normal WBC counts and their subset counts for competitive, well-trained and elite athletes from different sports disciplines. In this study, the mean WBC counts for females are similar to those observed in other studies that noted lower WBC counts in individual sports rather than team sports [8,14]. In males this relationship is not that clear [14].

In accordance with contemporary studies [8,14], findings of this study also show moderately higher lymphocyte counts both in men and women in swimming, but in contrast to Horn et al. [8] lower lymphocyte counts in female canoeing and male volleyball athletes were reported.

Several studies have shown varying incidence of neutropenia in athletes from different sports disciplines: marathon runners [1], cyclists [8,10], footballers [22] and triathletes [4,8]. In this study, neutropenia was noted in every discipline, but it was observed most often in female rowers and male swimmers (13.9% and 29.4% respectively).

The differentiation of prevalence of neutropenia is widely discussed, but to the authors' knowledge the reasons for this phenomenon are not clear.

Differences in observed prevalence of neutropenia could have been related to ethnic groups [1,12]. The neutrophil counts of Africans/Afro-Caribbeans were lower than those of Caucasians. It was suggested that ethnic neutropenia is a result of diminished bone marrow reserve [1].

It was suggested that neutropenia might be due to margination of neutrophils to damaged tissues [22]. Taking into consideration the characteristics of exercise in combat sports and contact disciplines, where a larger degree of tissue damage is more frequent than in

Gender	Discipline	Neutrophils < 2.0 · 10 ⁹ ·L ⁻¹		Lymphocytes < 1.0 · 10 ⁹ ·L⁻¹	Monocytes < 0.2 ⋅ 10 ⁹ ⋅ L ⁻¹		
	Judo	2	6.7%				
Female	Canoeing	4	9.1%				
	Volleyball	1	2.8%	0	0		
	Swimming	3	9.4%				
	Rowing	11	13.9%				
Malege	Judo	7	13.5%		0		
	Canoeing	11	10.5%		2	1.9%	
	Volleyball	10	5.9%	0	1	0.6%	
	Swimming	10	29.4%		1	2.9%	
	Rowing	20	11.0%		1	0.6%	

TABLE 3. NUMBER OF ATHLETES WITH LOW WHITE BLOOD CELL SUBSETS COUNTS ENGAGED IN PARTICULAR SPORTS DISCIPLINES

swimming or rowing, probably the above theory could not explain neutropenia in this study.

One of the causes of neutropenia could be the decreased neutrophil lifespan in blood. Levada-Pires et al. [11] observed exercise-induced neutrophil death by necrosis. In contrast, Radom-Aizik et al. [17] showed that the Janus kinase/signal transducer and activator of transcription (Jak/STAT) pathway, which inhibit apoptosis, was activated after the exercise. Other genes also had higher expression after exercise. These genes are likely to accelerate apoptosis. In a study by Park et al. [15], the apoptotic index (AI) of neutrophils was greater than the AI of lymphocytes after running trials. Therefore exercise-induced neutrophil death could explain the athletes' neutropenia.

In the present study, there was no incidence of lymphopenia. This is in contrast to other studies, which noted lymphopenia in archery (5% of group) and canoeing (5% of group) [8] or cycling [10]. It is supposed that total counts of lymphocytes have not been decreased in the examined athletes, but there could be a change of counts of lymphocytes among their subsets [3]. In the present study this aspect was outside the scope of investigation.

Monocytopenia was found only among males, with the highest percentage of occurrence in swimmers (2.9% of group). Other studies also observed lower monocyte counts in athletes, for example in cycling (6% of group) [9], 5% [10] and even 53% [8] in triathlon, and 5% in gymnastics [8]. It is suggested that exercise has an influence on the common precursor for neutrophils and monocytes in the bone marrow, because often low neutrophil counts were found in the same sports that had low monocyte counts [8]. On the other hand, this correlation is not so clear, because in this study lower neutrophil counts were observed in every discipline among female athletes, but decreased monocyte counts in female samples have not been found.

In contrast to Gleeson et al. [6] and Horn et al. [8], in this study gender had a substantial effect on neutrophil and monocyte counts. Nevertheless, some authors [2, 23] also reported lower blood monocyte counts and higher neutrophil counts in women than men in the general population.

Data concerning menstrual cycle phase and use of contraceptives were not included in the analysis, which should be regarded as a study limitation. Nevertheless, in the PubMed database there are only a few articles concerning the influence of the menstrual cycle on WBC counts. A selected study where total leukocyte count, and absolute and differential counts of neutrophils and lymphocytes were analysed during menstrual, proliferative and secretory phases of the menstrual cycle among healthy women demonstrated that the variations in the various types of leukocytes during different phases of the menstrual cycle were not statistically significant [20]. The results of another study suggest that neutrophil phenotype varies minimally during the menstrual cycle and between the genders; however, the hypothesis of a potential anti-inflammatory effect of ovarian hormones on neutrophils has been supported [19]. Nevertheless, in clinical and sports medicine practice there is not a different range of reference values of white blood cells for men and women, or for women in different menstrual cycle phases. The potential influence of menstrual cycle phase and use of contraceptives on WBC count is a very interesting problem in evaluation of changes of haematological results and should be better investigated in view of sports medicine practice.

Furthermore, also some physiotherapeutic interventions, commonly used in recovery enhancement among athletes, could influence WBC counts. For example, after 10 sessions of whole body cryostimulation, greater WBC counts were observed, both in men and women [5]. Probably, also environmental factors could affects immunological parameters, e.g. negative effects of chlorinated swimming pool attendance on health of swimmers [24]. Therefore, there are at least a few factors, mentioned above, that could obscure individual and group differences in WBC counts.

CONCLUSIONS

Differences in counts of white blood cells and their subsets (neutrophils, lymphocytes and monocytes) have been related to sport disciplines. A relationship between sex and WBC subset (neutrophil and monocyte) counts was found. It seems that prevalence of neutropenia and monocytopenia and differences in WBC counts and their subsets among athletes of various disciplines could be related to the adaptive response to physical exercise.

Acknowledgments

We acknowledge with gratitude the financial support of the Ministry of Science and Higher Education of the Republic of Poland and Ministry of Sport and Tourism of the Republic of Poland.

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