

ANALYSIS OF HAEMATOLOGICAL INDICATORS IN RODENTS OF THE SUPERFAMILY MUROIDEA FROM NATURAL POPULATIONS OF KYIV OBLAST, UKRAINE

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Abstract

Haematological parameters were assessed in three species of the genera *Myodes*, *Sylvaemus*, and *Apodemus*, which have long been used as bioindicators in environmental monitoring, particularly in radiobiological studies within the Chernobyl Exclusion Zone. Animals used in the study were sexually immature, aged two to three months and captured within the territory of the ‘Cherneckyi Lis’ National Park (Kyiv Oblast). The state of the haematopoietic system reflects the nature, intensity, and duration of exposure to stress factors, making these parameters the most informative biomarkers of physiological status at both individual and population levels. This study presents quantitative and qualitative characteristics of peripheral blood and bone marrow in *M. glareolus*, *S. tauricus*, and *A. agrarius*. Species-specific features of leukocyte profiles, myelograms, and bone marrow cell maturation indices were identified. Red blood parameters showed no significant interspecific differences, whereas leukogram and granulopoiesis data differed markedly among species. In leucograms of three species, a lymphocytic profile predominated (60–80%), consistent with the physiological norm for small rodents. Based on leukocyte counts in peripheral blood, the species can be ranked as follows: *M. glareolus* > *S. tauricus* > *A. agrarius*. The leukograms were characterised by a predominance of mature (segmented) neutrophils, particularly in *S. tauricus*, where their counts exceeded that of band neutrophils by 4.4-fold. In the bone marrow of *M. glareolus* and *S. tauricus*, similar distributions of granulocytic and erythroid lineages were observed, in contrast to *A. agrarius*. Notably, the myelogram of *M. glareolus* showed a relative predominance of immature neutrophils (neutrophil maturation index > 1), indicating species-specific features of granulopoiesis. The obtained data can serve as a basis for establishing reference haematological values for three murine rodent species of Kyiv Oblast. The results confirm that the formation of species- and population-specific control groups, considering age, sex, seasonality, and geographic origin, is a necessary methodological prerequisite for the correct interpretation of indicator species data in ecological monitoring programmes.

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Аналіз гематологічних показників у гризунів надродини *Muroidea* з природних популяцій Київської області

Алла Липська, Наталія Родіонова, Наталія Рябченко

Резюме. Досліджено гематологічні показники трьох видів з родів *Myodes*, *Sylvaemus* та *Apodemus*, які здавна використовуються як біоіндикаторні види в екологічному моніторингу, зокрема в радіобіологічних дослідженнях в Чорнобильській зоні відчуження. Досліди проводили на статевонезрілих тваринах дво-/тримісячного віку, виловлених на території національного парку «Чернечий ліс» (Київська обл.). Стан системи крові відображає характер, інтенсивність і тривалість дії стресових агентів, що робить такі показники інформативними біомаркерами фізіологічного стану тварин на індивідуальному та популяційному рівнях. У статті представлено кількісні та якісні характеристики периферичної крові й кістково-го мозку *M. glareolus*, *S. tauricus* та *A. agrarius*, встановлено видові особливості лейкоцитарної формули, складу мієлограми та індексів дозрівання клітин кісткового мозку. Показники червоної крові не мали достовірних міжвидових відмінностей, тоді як дані лейкограми та гранулопоезу суттєво різнилися між видами. В лейкограмі трьох видів переважав лімфоцитарний профіль (60–80%), що відповідає фізіологічній нормі для дрібних гризунів. За кількістю лейкоцитів у периферичній крові види розподіляються у ряд: *M. glareolus* > *S. tauricus* > *A. agrarius*. У лейкограмі тварин відмічали переважання зрілих (сегментоядерних) форм, особливо у особин *S. tauricus*, де їхня кількість була більшою у 4,4 рази, ніж паличкоядерних нейтрофілів. В кістковому мозку *M. glareolus* та *S. tauricus* зафіксовано схожий розподіл пулів гранулоцитарного та еритроїдного рядів, на відміну від *A. agrarius*. При цьому в мієлограмі *M. glareolus* реєстрували відносно переважання незрілих нейтрофілів (індекс дозрівання нейтрофілів > 1), що вказує на видоспецифічний гранулопоез. Отримані дані можуть бути основою для формування референтних показників системи крові у трьох видів мишоподібних гризунів Київщини. Результати підтверджують, що формування видо- та популяційно-специфічних контрольних груп з урахуванням віку, статі, сезонності та географічного походження є обов'язковою методологічною передумовою для коректної інтерпретації показників індикаторних видів у програмах екологічного моніторингу.

Ключові слова: мишоподібні гризуни, периферична кров, кістковий мозок, мієлограма, референтні значення.

Introduction

Environmental assessment based on studies of living organisms is among the most informative approaches for detecting natural and anthropogenic stressors, as it provides an integrated measure of their effects on biota. Bioindication methods enable the acquisition of data not only on the presence of environmental contaminants but also on their biological impacts. However, the reliability of such assessments depends on the availability of sensitive biomarkers and appropriately established reference groups that account for the specific characteristics of different species and populations.

Small mammals constitute a substantial component of Ukraine's mammal fauna and play a key role in the functioning of terrestrial ecosystems [Kondratenko & Zagorodniuk 2006]. Due to their high ecological plasticity, short life cycle, rapid generation turnover, and sensitivity to environmental changes, micromammals are considered effective bioindicators and convenient model organisms for studying dynamic ecological processes [Gashchak et al. 2006]. So, studies of small mammals as bioindicators in boreal forests have demonstrated that these animals play important functional roles in ecosystems, respond sensitively to environmental changes, and serve as effective tools for ecological monitoring. Different species may respond differently to the same environmental factors, limiting the use of any single species as a surrogate for another [Pearce & Venier 2005]. As inhabitants of the litter layer and upper soil horizons, small mammals are in direct contact with soil contaminants and can respond to these effects at the level of physiological systems [Talmage & Walton 1991].

Small murine rodents are of particular importance in radioecological research, as they are widely used as reference species for assessing the consequences of radionuclide contamination in accordance with the Reference Animals and Plants concept developed by the International Commission on

Radiological Protection [ICRP 2008]. Central to this framework is the requirement for baseline physiological data from reference species inhabiting uncontaminated areas. The high population density and reproductive potential of murine rodents make it possible to track radiation-induced changes under both acute and chronic exposure to ionising radiation across multiple generations, which is essential for evaluating long-term and transgenerational effects [Hancock et al. 2019]. For many years, species of the genera *Myodes*, *Sylvaemus*, and *Apodemus* have remained important model organisms in both national and international studies investigating the biological consequences of the Chernobyl accident. [Baker et al. 1996; Rodgers & Baker 2000].

For radioecological studies, the haematopoietic system is recognised as one of the most radio-sensitive and critical physiological systems, as it plays a central role in maintaining homeostasis and responds rapidly to a wide range of external stressors. Quantitative and qualitative characteristics of peripheral blood and bone marrow vary depending on the nature, intensity, and duration of environmental impacts, making these parameters sensitive and informative indicators of the functional status of animal populations.

At the same time, the use of haematological parameters from laboratory animals as reference values for wildlife populations has significant limitations. The controlled housing conditions and genetic standardisation of laboratory animals do not reflect the complex array of ecological factors affecting free-ranging populations. In natural environments, the combined influence of climatic conditions, food availability, interspecific and intraspecific competition, pathogens, and environmental contaminants create a physiological baseline that differs substantially from that observed under laboratory conditions. Therefore, direct extrapolation of laboratory-derived reference values to wild populations may lead to biased interpretations of biomonitoring and ecological assessment results [Garnier-Laplace et al. 2013; Copplestone et al. 2010].

Currently, systematic reference data on haematological values and individual variability ranges for model murid rodent species in Ukraine remain scarce in the scientific literature. Developing these reference databases is essential for effective bioindication, biodiversity conservation, and evidence-based evaluations of ecological and radioecological risks, particularly in areas exposed to different levels of anthropogenic pressure.

The present study aimed to determine reference values for key haematopoietic parameters in representatives of the genera *Apodemus*, *Sylvaemus*, and *Myodes* collected from areas of Kyiv Oblast characterised by minimal anthropogenic disturbance. These data are intended to provide a baseline reference for future radioecological research of murid rodents in the Chernobyl Exclusion Zone.

Materials and Methods

The study focused on three species of murid rodents: *Myodes glareolus* (n = 21), *Sylvaemus tauricus* (n = 19), and *Apodemus agrarius* (n = 15). Animals were captured during the decade of September in the Chornobyl National Park, located between the villages of Khodosivka and Lisnyky in the Feodosiivska Territorial Community, Kyiv Oblast, Ukraine. This park features a diverse range of environments, including pine, mixed, and locally deciduous forest communities.

In mature forest stands, the bank vole (*M. glareolus*) and the yellow-necked mouse (*S. tauricus*) predominated, whereas the striped field mouse (*A. agrarius*) was more abundant in younger forest habitats. Small mammals were captured using Sherman live traps. Pieces of rye bread fried in vegetable oil were used as bait [Zagorodniuk 2002; Zagorodniuk 2017]. Species were identified morphologically following criteria described in [Zagorodniuk 2002]. Animal age was determined using a combination of indicators, including body mass, body length, the development of reproductive organs and thymus, as well as according to the approach proposed by [Drozd & Pavlovsky 2024].

All experiments were conducted on sexually immature animals aged approximately 2–3 months.

Peripheral blood samples were collected from the carotid artery, and quantitative haematological parameters were determined using standard manual (tube-based) methods. Bone marrow for myelokaryocyte quantification was flushed from the femur using a 3% acetic acid solution to obtain a cell suspension. Bone marrow imprint smears were prepared from the sternum.

Peripheral blood cells (erythrocytes and leukocytes) and bone marrow cells were counted using a Goryaev haemocytometer. In addition, the myelokaryocyte index (MI) was calculated as the number of myelokaryocytes per milligram of body weight.

Leukograms and myelograms were analysed using light immersion microscopy of Pappenheim-stained smears at $\times 1000$ magnification. A total of 200 and 400 cells per slide were counted for leukogram and myelogram assessments, respectively [Monastyrska 2007; Zaporozhan *et al.* 2002].

The following haematological indices were calculated based on leukogram data: the adaptation stress index (lymphocytes to segmented neutrophils ratio, ASI), the immunoreactivity index (lymphocytes + eosinophils to monocytes ratio, IIR), the neutrophil maturation index (band neutrophils / segmented neutrophils, NMI), and erythrocyte maturation index (the ratio of maturing to the proliferating erythroid cells, EMI) [Raskin *et al.* 2004; Turgeon 2011].

In the myelogram, the percentage counts of blast, maturing, and mature cells of the granulocytic, erythroid, lymphoid, and megakaryocytic haematopoietic lineages were determined. Bone marrow indices characterising the kinetic parameters of cell maturation in individual haematopoietic lineages were also calculated, including: the leukocyte-to-erythroid ratio (all leukocytic lineage cells / total erythroid lineage cells); the neutrophil maturation index (myeloblasts + promyelocytes + myelocytes + metamyelocytes / band + segmented neutrophils); and the erythrocyte maturation index (haemoglobin-containing cells: polychromatophilic + orthochromic normoblasts / all erythrocyte precursors).

Experimental data were analysed using Microsoft Excel and Statistica 7.0 software. Statistical significance between experimental groups was assessed using Student's t-test. Differences were considered statistically significant at $p \leq 0.05$.

All experimental procedures complied with the requirements of the Law of Ukraine 'On Protection of Animals from Cruel Treatment' (No. 3447-IV, adopted 21.02.2006, amended on 04.08.2017).

Results

The selection of ecologically and physiologically homogeneous control groups is a key methodological consideration in ecological monitoring. This primarily includes species specificity, age and reproductive status, sex, and seasonal variability of the studied parameters, among others. In the present study, we conducted a comprehensive analysis of qualitative and quantitative parameters of peripheral blood and bone marrow in *M. glareolus*, *S. tauricus*, and *A. agrarius*—murid rodent species considered common and dominant components of forest biotopes in Ukraine. Animals subsequently used to form control groups in radiobiological studies were captured within the territory of a projected national park in Kyiv Oblast, which allows assuming relatively well-preserved habitats and limited anthropogenic impact on the studied populations.

As shown in Table 1, red blood cell parameters (erythrocytes, haemoglobin, and the colour index) did not differ significantly between species. These findings are consistent with the results reported by Kostecka-Myrcha [Kostecka-Myrcha 2002], who demonstrated that haemoglobin and erythrocyte parameters in small rodents are relatively stable within systematic groups and reflect general adaptive characteristics of blood. However, among the quantitative blood parameters, platelet counts in *M. glareolus* and *A. agrarius* were elevated compared to *S. tauricus*, by 1.8- and 2.1-fold, respectively. Leukocytes play an essential role in maintaining body homeostasis, as they are involved in the detoxification of both exogenous and endogenous toxic substances. Based on peripheral blood leukocyte counts, the animals formed the following decreasing order: *M. glareolus* > *S. tauricus* > *A. agrarius*.

According to the presented data, lymphocytes predominated among individual leukocyte fractions in all examined species. In *M. glareolus* and *S. tauricus*, lymphocytes accounted for approximately 60%, whereas in *A. agrarius* they exceeded 80%. This difference was statistically significant between *A. agrarius* and *M. glareolus*; however, no statistically significant differences were observed in absolute lymphocyte counts.

Table 1. Mean (M ± SE) quantitative and qualitative peripheral blood indices in studied species

Таблиця 1. Середні значення (M ± SE) кількісних і якісних показників периферичної крові досліджених видів

| Animal species/indicator | <i>M. glareolus</i> | <i>S. tauricus</i> | <i>A. agrarius</i> |
|-----------------------------------------|---------------------|---------------------|----------------------|
| Erythrocytes, 1012/l | 7.9 ± 0.5 | 8.4 ± 0.6 | 8.0 ± 0.8 |
| Haemoglobin, g/l | 121.8 ± 0.5 | 116.5 ± 2.4 | 118.3 ± 8.3 |
| Colour index, c.u. | 0.47 ± 0.02 | 0.43 ± 0.04 | 0.45 ± 0.02 |
| Platelets, 109/l | 205.4 ± 17.0 | 183.33 ± 17.64 | 246.7 ± 26.2 |
| Leucocytes, 109/l | 4.81 ± 0.35 | 3.85 ± 0.43 | 2.93 ± 0.16 |
| Young (myelocytes, metamyelocytes), % | 0 | 0 | 0 |
| Band neutrophils, % | 14.4 ± 2.1* | 6.69 ± 0.93 | 6.67 ± 3.43 |
| 109/l | 0.72 ± 0.12* | 0.23 ± 0.02 | 0.2 ± 0.1 |
| Segmented neutrophils (SN), % | 19.5 ± 2.5** | 29.3 ± 4.5** | 10.2 ± 3.7** |
| 109/l | 0.94 ± 0.15 | 1.09 ± 0.14 | 0.3 ± 0.1** |
| Neutrophils, total, % | 33.9 ± 3.9 | 36.0 ± 4.95 | 16.8 ± 3.93* |
| 109/l | 1.67 ± 0.24 | 1.33 ± 0.15 | 0.5 ± 0.21* |
| NMI (neutrophil maturation index), c.u. | 0.8 ± 0.1 | 0.26 ± 0.05* | 0.64 ± 0.14 |
| Eosinophils, % | 1.4 ± 0.59* | 0.06 ± 0.05 | 0.17 ± 0.12 |
| 109/l | 0.06 ± 0.02 | 0.001 ± 0.001 | 0.005 ± 0.004 |
| Basophils, % | 0.02 ± 0.01 | 0 | 0 |
| Monocytes, % | 1.41 ± 0.3 | 0.69 ± 0.16 | 0.5 ± 0.3 |
| 109/l | 0.07 ± 0.01* | 0.02 ± 0.01 | 0.02 ± 0.02 |
| Lymphocytes, % | 63.3 ± 3.94 | 63.25 ± 4.96 | 82.11 ± 3.65* |
| 109/l | 3.0 ± 0.24 | 2.5 ± 0.4 | 2.4 ± 0.15 |
| Large lymphocytes (L), % | 12.1 ± 1.9 | 8.37 ± 1.39 | 8.33 ± 2.73 |
| Large lymphocytes (S), % | 51.1 ± 4.35 | 54.9 ± 5.4 | 73.8 ± 4.2* |
| L/S ratio, c.u. | 0.27 ± 0.05 | 0.17 ± 0.04 | 0.11 ± 0.03 |
| Large granular lymphocytes, % | 2.8 ± 0.91* | 0.06 ± 0.06 | 0 |
| ASI (the adaptation stress index), c.u. | 4.48 ± 1.05 | 2.7 ± 0.5 | 14.2 ± 2.57* |
| IIR (immunoreactivity index), c.u. | 52.6 ± 8.68 | 50.5 ± 6.4 | 71.8 ± 4.9 |
| Atypical lymphocytes, % | 1.0 ± 0.63* | 0.12 ± 0.03 | 0.10 ± 0.05 |
| Aniso-poikilocytosis of erythrocytes, % | 0 | 0 | 0 |

* Statistically significant difference in the indicator of one group of animals at $p \leq 0.05$; ** statistically significant difference in the indicators between groups at $p \leq 0.05$.

Among lymphocyte subpopulations, small lymphocytes were predominant. Their relative abundance was higher than large ones in *M. glareolus*, *S. tauricus*, and *A. agrarius*, increasing by 4.2-, 6.5-, and 8.8-fold, respectively.

Regarding large lymphocytes, the highest proportion was recorded in *M. glareolus* (12.11%), while in *S. tauricus* and *A. agrarius* the values were nearly identical (8.37% and 8.33%, respectively). In the leukogram, large granular lymphocytes in *M. glareolus* constituted approximately 3%, whereas in *S. tauricus* and *A. agrarius* they were virtually absent.

The total proportion of neutrophils (the sum of segmented and band forms) among peripheral blood leukocytes in *M. glareolus* and *S. tauricus* ranged from 34% to 36%, while in *A. agrarius* it was significantly lower, at approximately 17%. In the leukogram, mature (segmented) neutrophils predominated, a pattern particularly pronounced in *S. tauricus*, where their number exceeded band neutrophils by 4.4 times. A slightly higher proportion of eosinophilic granulocytes and monocytes was recorded in *M. glareolus* compared with the other species. In all examined species, single atypical lymphocyte forms were observed, mainly represented by Rieder cells, binucleated and lobulated nuclei forms, while micronucleated cells were rare (0.01%). Overall, these results demonstrate that most peripheral blood parameters exhibited statistically significant interspecies differences.

Notably, analysis of peripheral blood and bone marrow parameters in sexually immature animals of both sexes revealed no substantial sex-related differences, similar to findings from our pre-

vious studies [Lypka *et al.* 2022]. Therefore, haematological data from both sexes were pooled into a single species-specific group. This approach provided a scientifically justified reduction in the number of animals used in the experiment, ensuring more rational and bioethical use of wild-caught specimens.

Bone marrow cytological examination provides the most comprehensive insight into the functional state of the haematopoietic system. Morphological identification of haematopoietic cells and their quantification are essential both under physiological conditions and, in particular, in pathological states. Analysis of bone marrow preparations revealed the presence of cells from all haematopoietic lineages at various stages of differentiation. The percentage composition of cellular elements in bone marrow smears is presented in Table 2.

It was established that blast cells (undifferentiated blasts, myeloblasts, erythroblasts, and lymphoblasts) collectively accounted for 3–4% of the total bone marrow cell population. A high proportion of neutrophilic granulocytes was observed, including actively proliferating, maturing, and mature cells. The highest content of band and segmented neutrophils, reflecting the status of the natural reserve pool of mature granulocytes in the bone marrow, was recorded in *A. agrarius*.

Table 2. Myelogram indices ($M \pm SE$) in representatives of studied species

Таблиця 2. Показники мієлограми ($M \pm SE$) у представників досліджених видів

| Animal species/indicator | <i>M. glareolus</i> | <i>S. tauricus</i> | <i>A. agrarius</i> |
|----------------------------------------------------------|---------------------------------------|----------------------------------------|-----------------------------------------|
| MCC counts (myelocaryocytes counts), $10^6/\text{femur}$ | 12.9 ± 1.76 | $29.9 \pm 2.01^*$ | 14.5 ± 2.26 |
| MI (myelocaryocyte index), c.u. | 0.56 ± 0.07 | $1.26 \pm 0.17^*$ | 0.75 ± 0.1 |
| Undifferentiated blasts, % | 0.44 ± 0.07 | 0.6 ± 0.12 | 0.6 ± 0.2 |
| - myeloblasts | 1.18 ± 0.234 | 1.07 ± 0.1 | 1.55 ± 0.21 |
| Neutrophilic blasts, % | | | |
| - promyelocytes | 1.16 ± 0.22 | 0.62 ± 0.13 | 1.9 ± 0.78 |
| - myelocytes | 9.06 ± 1.4 | 6.25 ± 0.55 | 9.2 ± 2.06 |
| - metamyelocytes | 5.33 ± 0.76 | 5.6 ± 0.33 | 6.05 ± 1.2 |
| - band neutrophils (BN) | 7.67 ± 1.62 | 6.5 ± 0.57 | 9.75 ± 1.57 |
| - segmented neutrophils (SN) | $5.6 \pm 0.98^{**}$ | $9.05 \pm 0.93^{**}$ | $12.65 \pm 1.02^{**}$ |
| Eosinophilic blasts, % | | | |
| - promyelocytes | 0.1 ± 0.05 | 0.08 ± 0.03 | 0.3 ± 0.06 |
| - myelocytes | 1.21 ± 0.22 | $0.65 \pm 0.15^*$ | 1.5 ± 0.13 |
| - metamyelocytes | 0.6 ± 0.13 | 0.82 ± 0.23 | $1.8 \pm 0.45^*$ |
| Eosinophils (band & segmented), % | 2.05 ± 0.24 | 2.25 ± 0.67 | 2.4 ± 0.73 |
| Basophilic (myelocytes, metamyelocytes), % | 0.83 ± 0.26 | 0.25 ± 0.12 | 0.21 ± 0.11 |
| Basophils (band & segmented), % | 0.19 ± 0.14 | 0.27 ± 0.13 | 0.35 ± 0.17 |
| Proerythroblasts, % | 0.28 ± 0.13 | 0.45 ± 0.12 | 0.72 ± 0.23 |
| Erythroblasts, % | 0.86 ± 0.12 | 1.02 ± 0.23 | 1.05 ± 0.26 |
| Normoblasts, % | | | |
| - basophilic | 3.35 ± 0.66 | 4.4 ± 0.49 | 2.95 ± 0.29 |
| - policromatophilic | 21.79 ± 2.71 | 17.0 ± 2.43 | 13.2 ± 3.23 |
| - oxyphilic | 11.8 ± 1.06 | 11.4 ± 1.94 | 9.4 ± 2.19 |
| Lymphoblasts, % | 0.38 ± 0.08 | 0.35 ± 0.14 | 0.55 ± 0.32 |
| Lymphocytes, % | 22.8 ± 4.77 | 28.8 ± 4.4 | 23.0 ± 4.85 |
| Monocytes, % | 0.32 ± 0.08 | 0.8 ± 0.22 | 0.6 ± 0.2 |
| Immunoblasts % | 2.13 ± 0.77 | 0.94 ± 0.48 | 0 |
| Plasma cells, % | $0.5 \pm 0.14^*$ | 0.15 ± 0.1 | 0 |
| Reticulocytes, % | 0.3 ± 0.06 | 0.45 ± 0.18 | $0.05 \pm 0.05^*$ |
| Megakaryoblasts, -cytes, % | 0.54 ± 0.08 | 0.2 ± 0.12 | 0.35 ± 0.18 |
| Leuko/erythroid ratio, c.o. | 1.91 ± 0.26 | 2.2 ± 0.37 | $3.2 \pm 0.5^*$ |

* Statistically significant difference in the indicator of one group of animals at $p \leq 0.05$; ** statistically significant difference in the indicators between groups at $p \leq 0.05$.

This parameter exceeded the corresponding values in *M. glareolus* and *S. tauricus* by 1.7- and 1.4-fold, respectively. Within the erythroid lineage, maturing haemoglobin-containing cells, namely polychromatic and oxyphilic normoblasts, predominated. Lymphocytes made up 20–30% of the total myelokaryocyte population, whereas thrombocytic lineage cells accounted for 0.2–0.6%. Additionally, occasional plasma cells and reticulocytes were identified in the bone marrow smears.

The overall distribution of granulocytic, erythroid, and lymphoid haematopoietic lineages is presented in Figure 1. A similar pattern of granulocytic and erythroid cell distribution was observed in *M. glareolus* and *S. tauricus*. In contrast, *A. agrarius* exhibited a significantly higher proportion of granulocytic lineage cells. In addition to quantifying individual cell populations, bone marrow examination included the calculation of indices reflecting the relationship between immature and mature cells within specific haematopoietic lineages (Table 3).

The neutrophil maturation index (NMI) demonstrated that immature neutrophil forms predominated in *M. glareolus* (NMI > 1), whereas mature cells were more abundant in *S. tauricus* and *A. agrarius*. No interspecific differences were detected for the erythroid maturation index (EMI). Mitotic activity of bone marrow cells was slightly higher in *S. tauricus* compared with *M. glareolus* and *A. agrarius*; however, these differences did not reach statistical significance.

These findings question the validity of applying laboratory-derived haematological reference values to wild populations. Genetic standardisation and controlled housing conditions of laboratory animals do not adequately reflect the complex array of environmental factors that influence a distinct physiological baseline in wild populations. In particular, our previous investigations of radiation effects on the haematopoietic system of murid rodents inhabiting natural populations of the Chernobyl Exclusion Zone demonstrated pronounced species-specific differences in haematological characteristics and in the responses of the blood system to environmental stressors [Lypyska *et al.* 2020]. At the same time, the establishment of homogeneous control groups from natural populations requires consideration of multiple factors, including age, sex, season, geographic distribution, and population-specific characteristics.

In summary, while erythrocyte parameters were relatively conserved across species, leukocyte composition and bone marrow maturation showed clear interspecific differences.

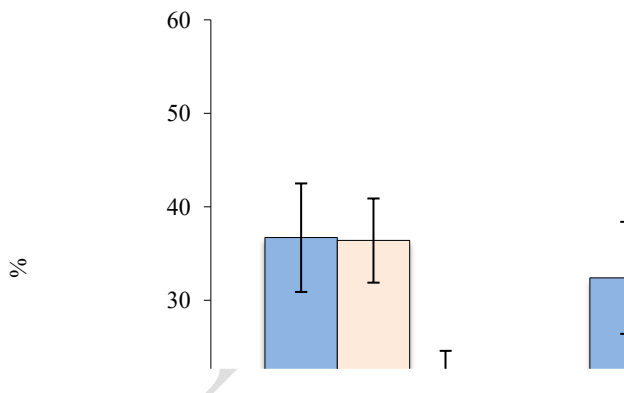


Fig. 1. Cell content (%) of individual haematopoietic pools in the bone marrow of *M. glareolus* (MGL), *S. tauricus* (STA) and *A. agrarius* (AAG).

Рис. 1. Вміст клітин (%) окремих пулів кровотворення у кістковому мозку *M. glareolus* (MGL), *S. tauricus* (STA) та *A. agrarius* (AAG).

Table 3. Bone marrow indices in studied species

Таблиця 3. Кістковомозкові індекси у досліджених видів

| Indicator | <i>M. glareolus</i> | <i>S. tauricus</i> | <i>A. agrarius</i> |
|-----------------------------------------|---------------------|--------------------|--------------------|
| NMI (neutrophil maturation index), c.u. | 1.25 ± 0.11* | 0.84 ± 0.07 | 0.82 ± 0.05 |
| EMI (erythroid maturation index), c.u. | 0.83 ± 0.06 | 0.82 ± 0.2 | 0.81 ± 0.01 |
| Mitotic index, ‰ | 13.2 ± 1.53 | 18.2 ± 2.34 | 14.0 ± 3.15 |

* Statistically significant difference at $p \leq 0.05$.

Most leukogram parameters differed significantly between species. A lymphocyte-dominated profile was characteristic of all species, with lymphocytes accounting for 60–80% of circulating leukocytes. However, *A. agrarius* exhibited the highest relative proportion of lymphocytes despite having the lowest total leukocyte count. Within the bone marrow, the principal interspecific differences were associated with the granulocytic lineage, with the largest reserve pool of mature neutrophils observed in *A. agrarius*.

The data obtained in this study may serve as a baseline reference for haematological parameters in three species of murid rodents from Kyiv Oblast, Ukraine. These reference values can be applied in future assessments of the effects of anthropogenic environmental stressors on small mammal populations, particularly in studies evaluating the impact of radiation exposure within the Chernobyl Exclusion Zone.

Author Contributions

A. Lypska: conceptualisation, implementation of the research, writing.
N. Rodionova: investigation, methodology, writing.
N. Riabchenko: investigation, writing, editing.

Declarations

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Handling of materials. All procedures involving animals were conducted in accordance with the applicable legislation of Ukraine governing the use of live animals in research.

Use of artificial intelligence. The authors confirm that no generative artificial intelligence tools were used in the preparation, writing, or analysis of this manuscript. If standard language editing tools were used, the authors retain full responsibility for the final content.

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