Haematological Effects of NSAIDs: Microscopic Changes in Blood Cell Morphology Following Ibuprofen, Piroxicam, and Diclofenac Administration in Albino Rats

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doi: 10.51505/ijmshr.2025.9203 URL: http://dx.doi.org/10.51505/ijmshr.2025.9203

Received: Feb 07, 2025 Accepted: Feb 25, 2025 Online Published: Mar 28, 2025

Abstract

Although NSAIDs are widely used to treat pain and inflammation, little is known about how these medications may alter the shape of blood cells. Despite their widespread use, little is known about the precise effect of NSAIDs on blood cell structure. The aim of this study was to investigate how blood cell shape is affected by common pain and inflammation drugs, particularly Ibuprofen, Piroxicam, and Diclofenac. Developing suitable overdose measures and improving personal safety may be made easier with a thorough grasp of these impacts. The cross-sectional investigation was conducted at the Rivers State University animal house in Rivers State, Nigeria, using 40 albino wistar rats. 400 mg of ibuprofen, 400 mg of diclofenac, and 20 mg of piroxicam were administered in varying dosages to the rats. The eight groups of rats, each containing five Wistar albino rats weighing between 80 and 100mg were created. Rats weighing less were administered a single drug, but rats weighing more were administered multiple drugs. It was simpler to get blood samples by heart puncture when under chloroform anaesthesia. Samples of blood were placed on glass slides, treated with methanol, stained with Leishman stain, allowed to air dry, and then viewed under a microscope at different magnifications. SPSS version 27 was used for the statistical analysis, and a significance level of $p \leq 0.05$ was applied. The findings showed notable variations across the experimental groups. The red cell line was shown to be significantly impacted, as evidenced by shrinkage, target cell presence, and ovalocyte occurrence. Furthermore, a decrease in staining intensity was noted. Platelets showed a marked decrease in staining intensity, but the white cell line showed little effect. Microscopic analysis indicated substantial changes in the red cell line, including variations in size, stain retention, and the presence of ovalocytes and codocytes. White cell line changes were characterized by slight strangulation in the nucleus of some neutrophils and a hypochromic presentation in monocytes. Platelets were slightly impacted, indicating just an alteration in staining intensity. The study reveals that ibuprofen, diclofenac, and piroxicam significantly alter red and white blood cell morphology and platelet presence, potentially altering clotting

mechanisms, immunological response, and oxygen transport. Combinations can cause erythrocyte alterations, potentially causing anemia, immunological dysfunction, and bleeding disorders. Further research is needed to understand these effects.

Keywords: Impact, Non-Steroidal Anti-Inflammatory, Drugs, Blood Cell Morphology, Structure, and Wistar Albino Rats

Introduction

Due to their painkilling effect, anti-inflammatory, and antipyretic properties, non-steroidal anti-inflammatory medicines (NSAIDs) are a class of pharmaceuticals that are frequently recommended, for the frequently used to treat inflammatory diseases, musculoskeletal disorders, and arthritis (Bhala *et al*., 2013). NSAIDs can effectively relieve symptoms, but they also have a number of negative side effects, such as cardiovascular problems, renal impairment, and stomach ulcers (Cryer & Feldman, 2022). Haemostasis, immunological defence, and oxygen transport are just a few of the physiological processes that depend on blood cells, which are made up of erythrocytes (red blood cells), leukocytes (white blood cells), and thrombocytes (platelets) (Hoffbrand & Moss, 2022).

Modifications in the cell structures or function may have profound effects on a person's overall health. Transporting oxygen from the lungs to tissues and eliminating carbon dioxide depend on erythrocytes, the most occuring blood cells. Anaemia and other disorders can result from any morphological alteration, such as the conversion of normal biconcave erythrocytes into spherocytes or echinocytes, which reduces their ability to carry oxygen (Snyder & Sheehan, 2017). The loss of biconcave shape also affects their deformability. According to Delves and Roitt (2020), alterations in leukocyte shape might also impair immunological responses, increasing the body's vulnerability to infections and inflammatory conditions. Although NSAIDs are used extensively, little is known about how these medications directly affect the shape of blood cells.

Long-term NSAID use, however, may have serious effects on blood cells, according to a number of studies, especially through pathways linked to oxidative stress and membrane damage (Gonzalez-Perez *et al*., 2021). NSAID-induced cytotoxicity has been associated with oxidative stress, a disorder marked by an imbalance between the body's capacity to detoxify these hazardous byproducts via antioxidants and the generation of reactive oxygen species (ROS) (Bhattacharyya *et al*., 2014). Elevated ROS levels have the potential to cause lipid peroxidation of cell membranes, which can change the structure of platelets, leukocytes, and erythrocytes (Chatterjee *et al*., 2022). Haemolysis, immunological suppression, and an elevated risk of bleeding could result from such alterations, which could impair on the lifespan and functionality of the cells.

It has been demonstrated that NSAIDs significantly modify haematological processes in animal species, including rats. Wistar albino rats, which are frequently employed in pharmacological

ISSN: 2581-3366

and toxicological studies, are a good model to investigate these drug-induced alterations because of their genetic and physiological resemblance to humans (Turner *et al*., 2021). Researches using rat models have shown that extended exposure to NSAIDs can cause noticeable alterations in erythrocyte shape, such as heightened membrane rigidity and fragility (Singh *et al*., 2022). Long-term NSAID treatment can cause noticeable alterations in erythrocyte morphology, such as increased fragility and membrane rigidity, as studies on rat models have shown (Singh *et al*., 2022). These changes can result in microvascular problems because they lessen the erythrocytic's capacity to deform, which is essential for their passage through small capillaries. Exposure to NSAIDs has been linked to changes in leukocyte function in addition to erythrocytes. NSAIDs have been reported to change the count and shape of neutrophils and lymphocytes, which may impact the body's ability to mount effective immunological responses (Khan *et al*., 2023).

However, the effects of other NSAIDs on platelet morphology and function are less well-established, with some studies suggesting that chronic NSAID use may result in changes to the shape of platelets and impair their ability to form clots effectively, increasing the risk of bleeding disorders (Mehta *et al*., 2022). Given the potential risks associated with NSAID-induced blood cell alterations, it is imperative to thoroughly investigate these effects. Future clinical strategies regarding the safe and effective use of NSAIDs, especially in groups at risk of haematological disorders, may be informed through the findings of this study, which will also help us better understand the broader physiological implications of NSAID usage.

Therefore, this study will investigate how ibuprofen, diclofenac, and piroxicam affect blood cell structure, taking into account how they affect platelet structure, WBC morphology, Hb levels, HCT values, and RBC morphology, emphasizing how crucial it is to monitor blood cell structure in patients taking these NSAIDsand also better understand the wider physiological effects of NSAID usage and could guide future clinical approaches to the safe and efficient use of these medications, especially in populations at risk for haematological disorders.

Table 1: Morphological effects of the treatments on the Red cell lines

Groups	**Size**	**Shape**	**Staining intensity**
G1: Control	Normal (7um)	Normal	Normochromic
G2: Diclofenac	Mixed (microcytic {6um} and macrocytic cells {8um})	Normal	Hyperchromic
G3: Ibuprofen	Macrocytic cells	Codocytic cells	Hypochromic
G4: Piroxicam	Macrocytic cells	Normal	Hypochromic
G5: Diclo & Ibu	Normocytic cells	Normal	Hypochromic
G6: Diclo & Piroxicam	Microcytic cells	Ovalocyte	Hypochromic
G7: Ibu & Piroxicam	Normocytic cells	Normal	Hypochromic
G8: Diclo, Ibu & Piroxicam	Microcytic cells	Normal	Mixed

ISSN: 2581-3366

Table 2: Morphological effects of the treatments on the White cell lines

Groups	Type of WBC	Nuclear Size	Shape of nucleus	Colour of nucleus	Cytoplasmic size	Cytoplasmic appearance	Cytoplasmic colour
G1 CONTROL	**Neutrophil**	Normal	Segmented	Purple	Normal	Coarse	Pale pink
	Eosinophil	Nil	Nil	Nil	Nil	Nil	Nil
	Basophil	Nil	Nil	Nil	Nil	Nil	Nil
	Monocyte	Normal	Ellipsoidal	Pale purple	Normal	Fine	Pale pink
	Lymphocyte	Normal	Large	Purple	Normal	Fine	Light blue
G2 Diclofenac	**Neutrophil**	Band Neutrophil	Curved with slight strangulation	Hypochromic	Normal	Coarse	Pale blue
	Eosinophil	Nil	Nil	Nil	Nil	Nil	Nil
	Basophil	Nil	Nil	Nil	Nil	Nil	Nil
	Monocyte	Normal	Ellipsoidal	Purple	Normal	Presence of vacoule	Pale
	Lymphocyte	Nil	Nil	Nil	Nil	Nil	Nil
G3 Ibuprofen	**Neutrophil**	Normal	Disjointed	Dark blue	Altered	Altered	Pale
	Eosinophil	Nil	Nil	Nil	Nil	Nil	Nil
	Basophil	Nil	Nil	Nil	Nil	Nil	Nil
	Monocyte	Normal	Ellipsoidal	Blueish purple	Nil	Covered by nucleus	Nil
	Lymphocyte	Nil	Nil	Nil	Nil	Nil	Nil
G4 Piroxicam	**Neutrophil**	Nil	Nil	Nil	Nil	Nil	Nil
	Eosinophil	Nil	Nil	Nil	Nil	Nil	Nil
	Basophil	Nil	Nil	Nil	Nil	Nil	Nil
	Monocyte	Normal	Ellipsoidal	Mixed	Small	Covered by nucleus	Nil
	Lymphocyte	Nil	Nil	Nil	Nil	Nil	Nil
G5 Diclo & Ibu	**Neutrophil**	Nil	Nil	Nil	Nil	Nil	Nil
	Eosinophil	Nil	Nil	Nil	Nil	Nil	Nil
	Basophil	Nil	Nil	Nil	Nil	Nil	Nil
	Monocyte	Normal	Ellipsoidal	Pale blue	Normal	Fine	Pale blue
	Lymphocyte	Nil	Nil	Nil	Nil	Nil	Nil
G6 Diclo & Piroxicam	**Neutrophil**	Nil	Nil	Nil	Nil	Nil	Nil
	Eosinophil	Nil	Nil	Nil	Nil	Nil	Nil
	Basophil	Nil	Nil	Nil	Nil	Nil	Nil
	Monocyte	Normal	Nil	Nil	Nil	Covered by nucleus	Nil
	Lymphocyte	Nil	Nil	Nil	Nil	Nil	Nil
G7 Ibu & Piroxicam	**Neutrophil**	Small	Band nucleus	Hyperchromic	Small	Coarse	Pale blue
	Eosinophil	Nil	Nil	Nil	Nil	Nil	Nil
	Basophil	Nil	Nil	Nil	Nil	Nil	Nil
	Monocyte	Normal	Nil	Hypochromic	Small	Fine	Pale
	Lymphocyte	Nil	Nil	Nil	Nil	Nil	Nil
G8 Diclo, Ibu & Piroxicam	**Neutrophil**	Nil	Nil	Nil	Nil	Nil	Nil
	Eosinophil	Nil	Nil	Nil	Nil	Nil	Nil
	Basophil	Nil	Nil	Nil	Nil	Nil	Nil
	Monocyte	Mixed	Ellipsoidal	Bark blue	Normal	Presence of vacoule	Pale blue
	Lymphocyte	Nil	Nil	Nil	Nil	Nil	Nil

Table 3: Morphological effects of the treatments on the platelets

Groups	Morphology
G1: Control	Platelets with normal central cluster of small pink cytoplasm
G2: Diclofenac	No platelets seen in background
G3: Ibuprofen	Scanty platelets (thrombocytopenia)
G4: Piroxicam	Dense appearance of platelets over slide (thrombocytosis)
G5: Diclo & Ibu	Dense appearance of platelets
G6: Diclo & Piroxicam	Dense appearance of platelets
G7: Ibu & Piroxicam	No visible platelets seen
G8: Diclo, Ibu & Pirox.	Thrombocytosis seen

Table 4: Comparison of control group with other groups

Compared groups	Cell lines	Comparison
G1 Vs G2	Red cell line	The group 2 had a mixed appearance of red cells, a combination of hyperchromic microcytic cells and normochromic macrocytic cells as compared to the first group which was normal
	White cell line	Had presence of monocyte with presence of vacoule in the cytoplasm and a band neutrophil with a curved nucleus and slight strangulation
	Platelets	No visible platelets were seen in background
G1 Vs G3	Red cell line	Target cells or codocytes present
	White cell line	The third group had presence of neutrophil with an altered cytoplasm
	Platelets	Smaller platelets seen in scanty numbers
G1 Vs G4	Red cell line	There was presence of hypochromic macrocytic cells
	White cell line	Presence of monocyte with pale pink colouration
	Platelets	Dense appearance of platelets
G1 Vs G5	Red cell line	There was mixed appearance of hypochromic normocytic cells and few target cells seen
	White cell line	There was presence of normal monocytes
	Platelets	No platelets seen in background
G1 Vs G6	Red cell line	There was mixed appearance of hypochromic ovalocytes and hyperchromic microcytic cells seen
	White cell line	Normal monocytes with dark blue colour
	Platelets	There was appearance of platelets
G1 Vs G7	Red cell line	Hypochromic normocytic cells were present
	White cell line	Very pale monocyte was seen with hyperchromic neutrophil were present
	Platelets	No visible platelets seen
G1 Vs G8	Red cell line	Mixed presentation of hyperchromic microcytic and hypochromic microcytic cells seen
	White cell line	Presence of hyperchromic monocyte seen
	Platelets	Mild appears of platelets seen

Table 5: Intergroup comparison of morphological effects of treatment on blood cells

	Group 2 Vs Group 3	
Red cell line	-Hyperchromic microcytic cells -Normochromic macrocytic cells	-Hyperchromic codocytes or target cells scattered over the slide
White cell line	-Monocyte with vacoule in cytoplasm -Band neutrophil with slightly strangulated nucleus	-Hyperchromic Neutrophil with altered cytoplasm
Platelets	No platelets seen	Scanty platelets seen
	Group 2 Vs Group 4	
Red cell line	-Hyperchromic microcytic cells -Normochromic macrocytic cells	-Hyperchromic macrocytic cells seen
White cell line	-Monocyte with vacoule in cytoplasm -Band neutrophil with slightly strangulated nucleus	-Normal monocyte seen with pale pink coloration
Platelets	No platelets seen	Dense appearance of platelets
	Group 2 Vs Group 5	
Red cell line	-Hyperchromic microcytic cells -Normochromic macrocytic cells	-Hyperchromic normocytic cells -Target cells scattered over the slide
White cell line	-Monocyte with vacoule in cytoplasm -Band neutrophil with slightly strangulated nucleus	-Normal monocyte seen
Platelets	No platelets seen	-Dense appearance of platelets
	Group 2 Vs Group 6	
Red cell line	-Hyperchromic microcytic cells -Normochromic macrocytic cells	-Hyperchromic ovalocytes seen -Hyperchromic microcytic cells seen
White cell line	-Monocyte with vacoule in cytoplasm -Band neutrophil with slightly strangulated nucleus	Normal monocytes with dark blue colour
Platelets	No platelets seen	-Dense appearance of platelets
	Group 2 Vs Group 7	
Red cell line	-Hyperchromic microcytic cells -Normochromic macrocytic cells	-Hypochromic normocytic cells seen in clusters
White cell line	-Monocyte with vacoule in cytoplasm -Band neutrophil with slightly strangulated nucleus	-Hypochromic monocytes seen -Hyperchromic neutrophil seen
Platelets	No platelets seen	No platelets seen
	Group 2 Vs Group 8	
Red cell line	-Hyperchromic microcytic cells -Normochromic macrocytic cells	-Hyperchromic microcytic cells in slides A, B & D -Hypochromic microcytic cells in slides C
White cell line	-Monocyte with vacoule in cytoplasm -Band neutrophil with slightly	-Monocyte with presence of vacoule in slides A

	strangulated nucleus	-Hyperchromic monocyte in slides B
Platelets	No platelets seen	Mild appears of platelets seen
	Group 3 Vs Group 4	
Red cell line	-Hyperchromic codocytes or target cells scattered over the slide	-Hyperchromic macrocytic cells seen
White cell line	-Hyperchromic Neutrophil with altered cytoplasm	-Normal monocyte seen with pale pink coloration
Platelets	Scanty platelets seen	Dense appearance of platelets
	Group 3 Vs Group 5	
Red cell line	-Hyperchromic codocytes or target cells scattered over the slide	-Hyperchromic normocytic cells -Target cells scattered over the slide
White cell line	-Hyperchromic Neutrophil with altered cytoplasm	-Normal monocyte seen
Platelets	Scanty platelets seen	Dense appearance of platelets
	Group 3 Vs Group 6	
Red cell line	-Hyperchromic codocytes or target cells scattered over the slide	-Hyperchromic ovalocytes seen -Hyperchromic microcytic cells seen
White cell line	-Hyperchromic Neutrophil with altered cytoplasm	Normal monocytes with dark blue colour
Platelets	Scanty platelets seen	-Dense appearance of platelets
	Group 3 Vs Group 7	
Red cell line	-Hyperchromic codocytes or target cells scattered over the slide	-Hypochromic normocytic cells seen in clusters
White cell line	-Hyperchromic Neutrophil with altered cytoplasm	-Hypochromic monocytes seen -Hyperchromic neutrophil seen
Platelets	Scanty platelets seen	No platelets seen
	Group 3 Vs Group 8	
Red cell line	-Hyperchromic codocytes or target cells scattered over the slide	-Hyperchromic microcytic cells in slides A, B & D -Hypochromic microcytic cells in slides C
White cell line	-Hyperchromic Neutrophil with altered cytoplasm	-Monocyte with presence of vacoule in slides A -Hyperchromic monocyte in slides B
Platelets	Scanty platelets seen	Mild appears of platelets seen
	Group 4 Vs Group 5	
Red cell line	-Hyperchromic macrocytic cells seen	-Hyperchromic normocytic cells -Target cells scattered over the slide
White cell line	-Normal monocyte seen with pale pink coloration	-Normal monocyte seen
Platelets	Dense appearance of platelets	Dense appearance of platelets

	Group 4 Vs Group 6	
Red cell line	-Hyperchromic macrocytic cells seen	-Hyperchromic ovalocytes seen -Hyperchromic microcytic cells seen
White cell line	-Normal monocyte seen with pale pink coloration	Normal monocytes with dark blue colour
Platelets	Dense appearance of platelets	-Dense appearance of platelets
	Group 4 Vs Group 7	
Red cell line	-Hyperchromic macrocytic cells seen	-Hypochromic normocytic cells seen in clusters
White cell line	-Normal monocyte seen with pale pink coloration	-Hypochromic monocytes seen -Hyperchromic neutrophil seen
Platelets	Dense appearance of platelets	No platelets seen
	Group 4 Vs Group 8	
Red cell line	-Hyperchromic macrocytic cells seen	-Hyperchromic microcytic cells in slides A, B & D -Hypochromic microcytic cells in slides C
White cell line	-Normal monocyte seen with pale pink coloration	-Monocyte with presence of vacoule in slides A -Hyperchromic monocyte in slides B
Platelets	Dense appearance of platelets	Mild appears of platelets seen
	Group 5 Vs Group 6	
Red cell line	-Hyperchromic normocytic cells -Target cells scattered over the slide	-Hyperchromic ovalocytes seen -Hyperchromic microcytic cells seen
White cell line	-Normal monocyte seen	Normal monocytes with dark blue colour
Platelets	Dense appearance of platelets	-Dense appearance of platelets
	Group 5 Vs Group 7	
Red cell line	-Hyperchromic normocytic cells -Target cells scattered over the slide	-Hypochromic normocytic cells seen in clusters
White cell line	-Normal monocyte seen	-Hypochromic monocytes seen -Hyperchromic neutrophil seen
Platelets	Dense appearance of platelets	No platelets seen
	Group 5 Vs Group 8	
Red cell line	-Hyperchromic normocytic cells -Target cells scattered over the slide	-Hyperchromic microcytic cells in slides A, B & D -Hypochromic microcytic cells in slides C
White cell line	-Normal monocyte seen	-Monocyte with presence of vacoule in slides A

		-Hyperchromic monocyte in slides B
Platelets	Dense appearance of platelets	Mild appears of platelets seen
	Group 6 Vs Group 7	
Red cell line	-Hyperchromic ovalocytes seen -Hyperchromic microcytic cells seen	-Hypochromic normocytic cells seen in clusters
White cell line	Normal monocytes with dark blue colour	-Hypochromic monocytes seen -Hyperchromic neutrophil seen
Platelets	-Dense appearance of platelets	No platelets seen
	Group 6 Vs Group 8	
Red cell line	-Hyperchromic ovalocytes seen -Hyperchromic microcytic cells seen	-Hyperchromic microcytic cells in slides A, B & D -Hypochromic microcytic cells in slides C
White cell line	Normal monocytes with dark blue colour	-Monocyte with presence of vacoule in slides A -Hyperchromic monocyte in slides B
Platelets	-Dense appearance of platelets	Mild appears of platelets seen
	Group 7 Vs Group 8	
Red cell line	-Hypochromic normocytic cells seen in clusters	-Hyperchromic microcytic cells in slides A, B & D -Hypochromic microcytic cells in slides C
White cell line	-Hypochromic monocytes seen -Hyperchromic neutrophil seen	-Monocyte with presence of vacoule in slides A -Hyperchromic monocyte in slides B
Platelets	No platelets seen	Mild appears of platelets seen

Discussion

The blood films albino wistar rats for the control group (G1) seem to exhibit typical hematological traits. There are no significant anomalies, such as anisocytosis (size variation) or poikilocytosis (shape variation), as the red blood cells (RBCs) are homogeneous in both dimensions. The normocytic and normochromic appearance of RBCs on these slides is consistent with appropriate oxygen-carrying capacity and healthy erythropoiesis. The white blood cells that are visible in the smear have characteristic nuclei that are dyed purple. For the best oxygen transfer, RBCs in control animals usually keep their biconcave form. Research has demonstrated that control groups of untreated rodents do not exhibit any notable RBC abnormalities, as seen in these slides (Kushwaha, 2019). Normal RBC measures in untreated rats, such as, hemoglobin concentration, and hematocrit count, indicate the overall health of theanimals and are frequently used as benchmarks when evaluating the impact of medications or harmful chemicals (Adeyemi, 2021). The consistency in size and shape indicates that the rats have not been exposed to any stressors, such as oxidative stress or drug-induced hemolysis, that could result in RBC abnormalities.

Wright-Giemsa stain makes the WBCs in these smears appear well-stained, making the cytoplasm and nucleus stand out. It is common for healthy rats to have both granulocytes (like neutrophils) and mononuclear cells (like lymphocytes or monocytes). According to reports on control groups, normal WBC counts and morphology guarantee appropriate immune function. by Tomic *et al*. (2020).Similar to this study, prior research on animals treated with NSAIDs showed changes in WBC count and function, while the control groups maintenained their leukocyte counts with normal morphology (Bennett et al., 2018). In rats that have not had treatment, platelets usually appear as tiny, granular fragments on these slides, though not very noticeable. Normal platelet activity is essential for blood clotting in control groups; any deviations would indicate systemic issues such as drug toxicity or underlying disease (Chukwu, 2019).

It appears ibuprofen causes the development of macrocytic and codocyte-shaped cells, which may have an impact on erythropoiesis and result in the formation of larger cells with particular forms and possibly danger of anemia, as seen by the G3 exposed to Ibuprofen showing hypochromic cells due to a decrease in hemoglobin content. G4 Piroxicam also causes normal shape and bigger (macrocytic) cells. Anemia may be shown by the hypochromic staining, which indicates a decreased hemoglobin level. When diclofenac and ibuprofen are combined in G5, the result is normal-sized (normocytic) cells with a target cell shape, which may be a sign of liver dysfunction with the integrity of the cell membrane. Hypochromic staining indicates a lower count of hemoglobin. G6 was exposed to Piroxicam and Diclofenac, this combination led to smaller cells (microcytic) with an oval shape (ovalocytes), this could indicate potential iron deficiency. The hypochromic staining suggests low hemoglobin content, potentially indicating iron deficiency anemia. G8 exposed to all three drugs Diclofenac, Ibuprofen & Piroxicam, led to smaller cells (microcytic) with a normal shape, but with mixed staining intensity (both hypochromic and hyperchromic), suggesting various effects on hemoglobin content and erythropoiesis. Smith *et al*. (2023) observed similar target cell shapes in red blood cells with a diclofenac and ibuprofen combination. This shape may indicate impaired membrane integrity, consistent with this study. Jones and Lee (2020) reported macrocytic cells and codocytes with ibuprofen treatment, suggesting interference with erythropoiesis and membrane changes. This study align with their findings. Garcia *et al*. (2021) found similar macrocytic cells with piroxicam treatment, indicating a potential effect on erythropoiesis. The hypochromic staining collaborates with their observations. The microcytic and macrocytic cells observed with diclofenac suggest a possible effect on erythropoiesis, leading to variations in cell size. The hyperchromic staining with this group and the combination with ibuprofen (G5) may indicate increased hemoglobin content, possibly due to accelerated red blood cell production.

Diclofenac (G2) is well-known for its actions on neutrophils, which may indicate an inflammatory response by producing a band shape with hypochromic staining and altered cytoplasm. Ibuprofen (G3) caused neutrophils to have a fragmented nuclear structure, which may indicate cellular stress or injury. Monocytes exhibiting heterogeneous features in response to piroxicam (G4) suggest a range of responses. Each combination of G5, G6, G7, and G8 treatments has a distinct effect on monocytes; some exhibited alterations in nuclear shape and color, altered cytoplasm, and vacuole presence. Changes in neutrophil shape in Group 2 could be a sign of an impact on cell maturation. This is consistent with research by Patel et al. (2021) that showed diclofenac changed the shape of neutrophils in rats. In G3 ibuprofen exhibited fragmented neutrophil morphology, which may indicate effects on cell maturation. This is consistent with research by Khan *et al*. (2022) that showed ibuprofen changed the shape of neutrophils. The heterogeneous monocyte shape seen in G4 indicates changes in the appearance of the cells. This is in line with research by Lee (2020), which discovered that piroxicam had an impact on monocyte morphology. Neutrophils and monocytes in G5 and G7 exhibited changed shape, which may indicate that the combinations have additive effects on cell maturation. The heterogeneous monocyte shape seen in G8 suggests that the three drugs may interact. The different impacts that the combinations (G5, G7, and G8) have on cell morphology point to possible drug interactions.

The lack of visible platelets suggests acute thrombocytopenia, which may have resulted from diclofenac's suppression of COX-1 and COX-2, which lowers TXA2 production and platelet aggregation (Patrono *et al*., 2017). Additionally, immune-mediated thrombocytopenia has been connected to diclofenac (Aster & Bougie, 2017).
Scanty platelets indicated thrombocytopenia, and ibuprofen decreased platelet activation and inhibited COX-1. In contrast to aspirin's irreversible effect, its reversible action permits some platelet presence (Murray & FitzGerald, 2020).
Piroxicam, a COX-2 inhibitor, was found to cause thrombocytosis. This decreases the generation of PGI2, which raises platelet numbers and causes prothrombotic circumstances.
Ibuprofen also inhibited COX-1, reducing platelet activation but still visible. Piroxicam, a long-acting NSAID, suppressed COX-2 more than COX-1, causing elevated platelet counts and prothrombotic conditions.

The study found that groups 2, 4, 5, 6, and 8 displayed abnormalities in red blood cell morphology, including mixed appearances of hyperchromic microcytic cells, suggesting potential effects on oxygen transport and tissue perfusion. White blood cell morphology showed differences, including vacuolated monocytes and altered neutrophils, indicating potential effects on immune response and inflammation. The absence or reduced presence of platelets in some groups could indicate potential effects on hemostasis and clotting, These morphological changes could impact patients' immune response, oxygen delivery, and clotting ability.

The study analyzed the morphology of various blood samples, including those from wistar albino rat treated with Ibuprofen, Diclofenac, Piroxicam, and others. The results showed varying red cell morphology, suggesting potential effects on oxygen transport and tissue perfusion. The study also revealed alterations in white cell morphology, indicating impacts on immune response and inflammation. The presence of platelets was also observed in different groups, with some showing no visible platelets, indicating potential interactions between medications. The combined treatments in these groups showed varied effects on cell morphology. Some groups showed significant changes, such as no visible platelets, which could pose risks for patients in terms of bleeding and clotting. These findings highlight the importance of understanding the underlying mechanisms of blood clotting and hemostasis in various patient groups.

Conclusion

The study shows that either by singly or in combination, ibuprofen, diclofenac, and piroxicam have a major impact on the morphology of red and white blood cells as well as the presence of platelets, which may change clotting mechanisms, immunological response, and oxygen transport. Notably, some drug combinations caused erythrocyte alterations such macrocytosis and hypochromia, neutrophil abnormalities, and thrombocytopenia, suggesting potential risks of anemia, immunological dysfunction, and bleeding disorders. Because of the possible hematological effects of these NSAIDs, caution should be used when using these drugs, particularly when taken in combination. Patients who already have clotting challenges, anemia, or blood abnormalities should be closely monitored. To investigate the mechanisms behind these alterations and evaluate their clinical significance, more research is required.

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