DOI: 10.7860/JCDR/2024/65994.19061

Association of Platelet Indices with Diabetic Complications: A Cross-sectional Study

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ABSTRACT

Introduction: Diabetes Mellitus is a prothrombotic state characterised by enhanced platelet activity, which may lead to microvascular and macrovascular complications. Platelet indices, such as Mean Platelet Volume (MPV) and Platelet Distribution Width (PDW), are routinely available in laboratories and can serve as a prognostic markers for patients.

Aim: To evaluate platelet indices in patients with Type 2 Diabetes Mellitus (T2DM) and associated complications.

Materials and Methods: A cross-sectional study was conducted at the Department of Pathology in Krishna Vishva Vidyapeeth's Krishna Hospital and Medical Research Centre, Karad, Maharashtra, India, from July 2020 to May 2022. A total of 120 patients with type 2 diabetes, both with and without complications, were investigated. Haematological parameters (platelet count and platelet indices such as MPV and PDW) and biochemical parameters {fasting blood sugar and Haemoglobin A1c (HbA1c)} were compared between the two groups. Platelet indices were measured using an automated haematology analyser. Statistical analysis was performed using the t-test, Analysis of Variance (ANOVA), and Statistical Package for Social Sciences (SPSS) version 16.0.

Results: The study included 60 cases each of DM with complications and without. Among those with complications, 17 (28.33%) were aged 51-60 years, 14 (23.33%) were 61-70 years old, and 12 (20%) were over 70 years. The mean MPV was 12.74±3.076 fL for patients with complications and 8.65±1.58 fL for those without. The mean PDW was 15.54 15.54±3.31 fL for patients with complications and 13.94±2.66 for those without. The mean HbA1c levels were 8.33 mmol/mol for patients with complications and 6.75 mmol/mol for those without. The mean platelet count was 3.12 lakh/mm^3 for patients with complications and 2.45 lakh/mm^3 for those without. The mean fasting blood sugar levels were 219.65 mmol/L for patients with complications and 109.96 mmol/L for those without.

Conclusion: Diabetes contributes to endothelial dysfunction and platelet hyperactivity. The present study reveals that diabetic patients with uncontrolled glycaemic indices and elevated fasting blood sugar levels have higher platelet indices compared to patients without complications, where the platelet count, platelet indices, and glycaemic indices were within normal limits. These indices may also serve as useful prognostic tools.

Keywords: Diabetes mellitus, Prognostic marker, Vascular complications

INTRODUCTION

Diabetes mellitus is a group of metabolic disorders characterised by hyperglycaemia, resulting from defects in insulin secretion, insulin action, or both. It is a leading cause of end-stage renal disease, adult-onset blindness, and non traumatic lower extremity amputations. Currently, the prevalence of diabetes among Indian adults is estimated at 8.8% [1,2].

The complications associated with T2DM are primarily due to suboptimal glycometabolic control. Fasting Blood Glucose (FBG) and HbA1c are widely used to monitor glycometabolic control, with HbA1c reflecting the mean blood glucose levels over a three-month period. Factors contributing to diabetic complications include altered platelet morphology, increased platelet dysfunction, and reactivity, leading to a prothrombotic state and consequent vascular complications, which increase morbidity and mortality in diabetic patients [3,4].

Platelets in DM are hyperactive, releasing more granules, leading to increased platelet turnover and the release of large, enzymatically and metabolically active platelets with a greater tendency to form clots, resulting in both macro and microvascular complications [4,5].

Platelet indices are measured using automated haematology analysers (Nihon Kohden) as part of routine haematological procedures [6]. The normal range for MPV in the laboratory is between 7.4-10.4 fL, and for PDW, it is 9-14 fL. MPV indicates the average size and activity of platelets and is calculated by dividing the Platelet Crit (PCT) by the platelet count. PDW measures the variability in platelet size and is calculated by determining the width of the platelet histogram at the 20% height level. PDW typically elevates earlier than other indices [7,8]. The present study investigates the effects of long-standing diabetes on complications and emphasises platelet function. The objectives of the study were to analyse the distribution of platelet indices in DM patients with and without complications and to compare the levels of HbA1c and FBS in complicated and uncomplicated cases of DM.

MATERIALS AND METHODS

This cross-sectional study was conducted in the Department of Pathology, Krishna Vishwa Vidyapeeth, Karad, Maharashtra, India, with data collected over a 24-month period (July 2020 to May 2022). Blood samples were collected into K2 Ethylenediaminetetraacetic Acid (EDTA) tubes, and platelet parameters were measured within one hour after venipuncture to avoid in-vitro changes. Parameters included MPV, PDW, platelet count, FBS, and HbA1c levels, with HbA1c categorised as <7.5, 7.5 to 10, and >10 mmol/L. The study aimed to compare platelet values and HbA1c between diabetic patients with and without complications and to note the presence of microvascular (diabetic retinopathy, chronic kidney injury, and acute kidney injury) and macrovascular complications (stroke, myocardial infarction, gangrenous toe, and peripheral vasculopathy). The study protocol was approved by the Institutional Ethics Committee (Protocol Reference number KIMS/030/2020-21).

Inclusion criteria: Patients with T2DM, both with and without complications such as stroke, myocardial infarction, acute kidney injury, chronic kidney injury, gangrenous toe, diabetic nephropathy, and peripheral vasculopathy were included in the study.

Exclusion criteria: Patients with systemic diseases in addition to DM, such as hypertension, systemic lupus erythematosus, rheumatoid arthritis, those on corticosteroids or antiplatelet drugs like aspirin, patients with Cushing syndrome, and those with any diagnosed malignancy were excluded from the study.

Normal range was considered as following: MPV: 7.4-10.4 fL, PDW: 9-14 fL, FBS: <100 mg/dL, HbA1c: <6.5% [7].

Study Procedure

Methodology involved the collection of blood into K2 EDTA tubes, with platelet parameters measured within one hour postvenipuncture to minimise the risk of platelet swelling. Blood specimens were gently mixed five times before a full blood count was measured using an automated blood analyser (Nihon Kohden). Given the heterogeneity in the volume and structure of platelets, the following haematological parameters were analysed in all blood samples: platelet count, MPV, PDW, and the biochemical parameters FBS and HbA1c.

STATISTICAL ANALYSIS

Statistical analysis was performed using SPSS version 16.0. Qualitative data were described as numbers and percentages. Quantitative data were presented as ranges (maximum and minimum), means, and Standard Deviations (SD). Comparisons between different groups regarding categorical variables were performed using the Student's t-test. A p-value (probability) of <0.05 was considered statistically significant.

RESULTS

Age distribution of DM with and without complications: The present cross-sectional study was conducted over a period of two years. Among the total number of cases of DM with complications, 17 (28.33%) of 60 patients were in the age group of 51-60 years, 14 (23.33%) of 60 belonged to the age group of 61-70 years, and 12 (20%) of 60 were older than 70 years of age. In patients with DM without complications, 21 (35%) of 60 patients were between the ages of 51-60 years, 5 (8.33%) of 60 were in the age group of 61-70 years, and 8 (13.3%) of 60 were older than 70 years of age [Table/Fig-1].

Age (in years)	DM with complication (n=60)	DM without complication (n=60)
21-30	3 (5%)	2 (3.33%)
31-40	4 (6.67%)	4 (6.67%)
41-50	10 (16.67%)	20 (33.33%)
51-60	17 (28.33%)	21 (35%)
61-70	14 (23.33%)	5 (8.33%)
>70	12 (20%)	8 (13.33%)
Males	39 (65%)	21 (35%)
Females	37 (61.67%)	23 (38.33%)

[Table/Fig-1]: Age-wise distribution of DM with and without complication.

Gender-wise distribution: In DM with complications, out of 60 (100%) patients, 39 (65%) were male and 21 (35%) were female. In DM without complications, out of 60 (100%) patients, 37 (61.67%) were male and 23 (38.33%) were female. Both groups showed a nearly equal number of males and females, so the Chi-square test was used, and it was found that the p-value was not significant (p>0.05).

Comparing platelet indices with HbA1c levels in patients of DM and complications: The MPV was 11.95±2.55 for HbA1c <7.5 (n=21), 12.20±2.88 for HbA1c between 7.5 and 10 (n=29), and 15.91±3.35 for HbA1c >10 (n=10). The mean PDW was 15.10±1.56 for HbA1c <7.5, 16.08±2.11 for HbA1c between 7.5 and 10, and 17.00±1.99 for HbA1c >10. The mean platelet count was 2.93 lac/mm^3±1.11 lac/mm^3 for HbA1c <7.5, 3.54 lac/mm^3±1.12 lac/mm^3 for HbA1c between 7.5 and 10, and 3.67 lac/mm^3±1.14 lac/mm^3 for HbA1c >10 [Table/Fig-2].

	MPV		PDW		Platelet Count	
HbA1c	Mean	SD	Mean	SD	Mean	SD
<7.5 (n=21)	11.95	2.55	15.10	1.56	293762	111021
7.5 to 10 (n=29)	12.20	2.88	16.08	2.11	354095	112362
>10 (n=10)	15.91	3.35	17.00	1.99	367510	114903
ANOVA F-value	7.51		4.01		3.257	
p-value	0.0013*		0.0235*		0.0458*	

[Table/Fig-2]: Comparison of platelet indices with HbA1c levels in DM with complications.

Comparing platelet indices in DM without complications: The MPV for DM without complications was 8.63±1.51 for HbA1c <7.5 and 8.83±2.05 for HbA1c between 7.5 and 10. The mean PDW for DM without complications was 13.88±2.77 for HbA1c <7.5 and 14.23±2.09 for HbA1c between 7.5 and 10. The mean platelet count for DM without complications was 2.48 lac/mm^3±83,398 for HbA1c <7.5 and 2.25 lac/mm^3±78,757 for HbA1c between 7.5 and 10 [Table/Fig-3].

	MPV (fL)		PDW (fL)		Platelet count (U/mm^3)	
HbA1c	Mean	SD	Mean	SD	Mean	SD
<7.5 (n=51)	8.63	1.51	13.88	2.77	248843.1373	83398.4
7.5 to 10 (n=9)	8.83	2.05	14.23	2.09	225555.5556	78757.7
Unpaired t-value	0.35		0.36		7.428	
p-value	0.73		0.71		<0.0001*	

[Table/Fig-3]: Comparison of platelet indices with HbA1c in DM patients without complications.

Comparison of HbA1c and FBS in diabetic patients with and without complications: The mean HbA1c was 8.33±1.45 for diabetes with complications and 6.75±0.61 for diabetes without complications. The mean FBS was 219.65±101.32 for diabetes with complications and 109.96±26.17 for diabetes without complications [Table/Fig-4].

Diagnosis	MPV (fL) Mean±SD	PDW(fL) Mean±SD	HbA1c (mmol/moL) Mean±SD	Platelet Count (U/mm^3) Mean±SD	FBS (mmol/L) Mean±SD
DM with complication (n=60)	12.74± 3.076	15.54± 3.31	8.33± 1.45	312766.67± 107362	219.65± 101.32
DM without complication (n=60)	8.65± 1.58	13.94± 2.66	6.75± 0.61	245350± 82497	109.96± 26.17

[Table/Fig-4]: Comparison of MPV, PDW, HbA1c, platelet count and FBS in diabetic patients with and without complications.

Comparison of platelet indices and platelet count in patients of DM with complications: The mean platelet count for stroke patients was 2.97±1.022 lac/mm^3, for Myocardial Infarction (MI) it was 2.0 lac/mm^3±64500, for Acute Kidney Injury (AKI) it was 4.09±1.15 lac/mm^3, for Chronic Kidney Injury (CKI) it was 2.71 lac/mm^3±69429 for gangrene toe it was 3.70 lac/mm^3±1.13 lac/mm^3, for diabetic retinopathy it was 3.52 lac/mm^3±1.07 lac/mm^3, and for peripheral vasculopathy, it was 3.19 lac/mm^3±58174 mm^3. The mean Platelet Distribution Width (PDW) was 13.48±3.28 fL for stroke, 12.05±1.27 fL for MI, 13.78±1.38 fL for AKI, 14.05±2.29 fL for CKI, 14.8±3.15 fL for gangrenous toe, 14.1±3.31 fL for diabetic retinopathy, and 11.86±1.067 fL for peripheral vasculopathy. The mean platelet volume (MPV) was 13.98±1.37 fL for stroke, 12.82±0.12 fL for MI, 12.07±0.98 fL for AKI, 11.56±0.94 fL for CKI, 12.48±1.43 fL for gangrene toe, 13.02±1.29 fL for diabetic retinopathy, and 10.36±0.72 fL for peripheral vasculopathy [Table/Fig-5].

Complications (n=60) n (%)	Platelet count Mean±SD	PDW Mean±SD	HbA1c Mean±SD	MPV Mean±SD	FBS Mean±SD
Stroke 8 (23.33%)	297937.5±102230	13.48±3.28	14.55±2.98	13.98±1.37	213.06±131.4
Myocardial infarction 4 (6.66%)	200250±64500	12.05±1.27	16.27±1.37	12.82±0.12	182.5±23.62
Acute Kidney Injury 7 (15%)	409142.8±115132	13.78±1.38	16.17±0.95	12.07±0.98	291.42±101.23
Chronic Kidney injury 8 (13.33%)	271777.7±69429	14.05±2.29	17.58±0.59	11.56±0.94	215.5±73.67
Gangrenous toe 9 (13.33%)	370416.6±113023	14.8±3.15	18.21±0.93	12.48±1.43	240±88.93
Diabetic Retinopathy 12 (10%)	352313.1±107119	14.1±3.31	15.62±0.84	13.02±1.29	182.9±78.93
Peripheral vasculopathy 12 (10%)	31966.6±58174	11.86±1.067	15.9±1.17	10.36±0.72	130±32.24
ANOVA F-value	3.716	2.295	6.206	1.947	2.003
p-value	0.0064	0.0599	0.0002	0.004	0.0095

[Table/Fig-5]: Comparison of platelet indices, platelet count fasting blood sugar and HbA1c in patients of DM with complications.

DISCUSSION

The DM is a significant global health concern, characterised by increased prothrombotic activity, which leads to accelerated atherosclerosis, inflammation, and enhanced platelet activity [9,10]. This contributes to multi-organ involvement-including the heart, nerves, eyes, Central Nervous System (CNS), kidneys, gastrointestinal tract, and blood vessels-resulting in long-term complications associated with increased mortality and morbidity [11,12].

In DM, platelets are often immature, larger, and exhibit increased activity. Hyperglycaemia directly enhances platelet reactivity and promotes the glycation of platelet proteins. Both insulin resistance and insulin deficiency can increase platelet reactivity, as insulin normally inhibits platelet activation. Thus, a relative or absolute deficiency of insulin could lead to heightened platelet reactivity. Diabetic patients exhibit platelet hyperaggregability and activation, causing circulating platelets to release more granules, which shortens platelet lifespan and leads to the release of larger platelets from the bone marrow due to megakaryocyte activation. These larger and younger platelets have greater volume and are functionally more active because of increased surface markers. The resulting enhanced platelet aggregation and activation play a role in the development of various microvascular and macrovascular complications [13].

Age Distribution

In the present study, the mean age of the study population was 57.49±14.48 years. DM with complications was more prevalent in older age groups. A total of 17 (28.33%) out of 60 patients belonged to the age group of 51-60 years, 14 (23.33%) out of 60 were in the 61-70 years age group, and 12 (20%) out of 60 were over 70 years of age. In studies by Dwivedi T and Davangeri R, Spandana T et al., Shilpi K and Potekar RM, Bhattacharjee P et al., and Subashini S et al., the mean age of patients with DM and complications was similar to the present, as shown in [Table/Fig-6] [14-20].

Study series (author/year/place)	Males in %	Females in %	Mean age (years)
Present study	63.33	36.67	57.49±14.48
Kodiatte TA et al., [5] (2012, Kolar)	65	35	55±11.2
Dwivedi T and Davangeri R [14] (2013, Belgaum)	69.2	30.8	57.5±10.94
Bhattacharjee P et al., [17] (2016 Agartala)	51	49	57.3±14.18
Subashini S et al., [18] (2020, Puducherry)	49.6	50.4	56±11.46
Yenigun EC et al., [19] (2016, Turkey)	31.3	68.7	59.35±9.04
Taderegew M et al., [20] (2021, Ethiopia)	36.3	30.4	56.8±8.9

[Table/Fig-6]: Comparison of age-wise distribution in other and present study [5,14-20].

Platelet Indices

In the present study, the mean MPV and PDW for DM without complications were 8.65±1.58 and 13.94±2.66, respectively. The mean MPV and PDW for DM with complications were 12.74±3.076 and 15.54±3.31. These findings align with the studies by Shilpi K and Potekar RM, Bhattacharjee P et al., Hekimsoy Z et al., and Jindal S et al., as shown in [Table/Fig-7,8] [13,14,16-18,20-24]. Analysis revealed that diabetic patients had higher PDW values than MPV, as PDW is independent of platelet count, while MPV is dependent on it and is calculated using a histogram.

Study series	Mean Platelet Volume (MPV) (fL)		p-value
	Diabetics with complications	Diabetics without complications/control	
Present study	12.74±3.076	8.65±1.58	<0.0001*
Bhattacharjee P et al., [17] (2016, Agartala)	12.65±1.89	11.16±1.18	0.03
Subashini S et al., [18] 2020, Puducherry)	12.75±0.99	10.00±1.44	0.01
Taderegew M et al., [20] (2021, Ethopia)	13.6±2.2	11.8±1.9	<0.001
Hekimsoy Z et al., [21] (2004, Turkey)	10.62±1.71	9.15±0.86	0.0001

[Table/Fig-7]: Comparison of Mean Platelet Volume (MPV) in other and present study [17,18,20,21].

Study series	Platelet Distribution Width (PDW)		p-value
	Diabetes with complications	Diabetes without complications/control	
Present study	15.54	13.94	0.0040
Jabeen F et al., [13] (2013, Karachi)	15.02	14.12	0.003
Dwivedi T and Davangeri R [14] (2013, Belgaum)	17.9	14.8	<0.001
Subashini S et al., [18] (2020, Puducherry)	12.97±0.88	11.34±1.45	0.01
Taderegew M et al., [20] (2021 Ethopia)	16.6±2.5	14.9±2.4	<0.001
Dalamaga M et al., [22] (2010, Greece)	16.4	13.0	<0.001
Demirtas L et al., [23] (2015, Turkey)	16.4	15.4	<0.001

[Table/Fig-8]: Comparison of Platelet Distribution Width (PDW) in other and present study [13,14,18,20,22,23].

Platelet Count

In our study, the mean platelet count for DM with complications was 3.12±1.07 lac/mm^3, which is consistent with studies by Dwivedi T and Davangeri R, Subashini S et al., Taderegew M et al., and Jindal S et al., which reported mean platelet values of 257±75,600 lac/mm^3, 285.29±75,380 lac/mm^3, 257.9±48,500 lac/mm^3, and 229.33±70,000 lac/mm^3, respectively [14,18,20,24].

Gylcosylated Haemoglobin (HbA1c)

In the present study, the mean HbA1c in patients with DM and complications was 8.33±1.45. The mean HbA1c in patients with DM without complications was 6.75±0.61, which is similar to the findings of studies by Dwivedi T and Davangeri R, Spandana T et al., Shilpi K and Potekar RM, (2017), and Walinjkar RS et al., which reported HbA1c levels of 8.9±1.37, 7.28±0.88, 7.3±1.1, and 7.45±1.48, respectively [14-16,25]. Patients with an HbA1c level >6.5% had higher platelet indices (MPV and PDW) than patients with an HbA1c level <6.5%.

Fasting Blood Sugar (FBS)

The mean FBS was 219.65±101.32 mg/dL for patients with DM complications and 109.96±26.17 mg/dL for those without complications, which is consistent with the study by Dwivedi T and Davangeri R which reported a mean FBS of 170±83.46 mg/dL for those with complications. Shilpi K and Potekar RM, (2017) reported a mean FBS of 158.1±33.7 mg/dL, while Taderegew M et al., (2021) and Walinjkar RS et al., reported mean FBS levels of 147.9±35.2 mg/dL and 140.48±28.05 mg/dL, respectively [14,16,20,25]. Both FBS and HbA1c levels were higher in patients with DM complications than in those without complications.

Platelet Count in DM with Complications

The mean platelet count was 4.09±1.15 lac/mm^3 in cases of AKI and 2.77 lac/mm^3±69.42/mm^3 in CKI. These values are similar to the mean platelet count of 2.98 lac/mm^3±68.37/mm^3 reported by Subashini S et al., and 2.63 lac/mm^3±45.8/mm^3 by Taderegew M et al., [18,20]. The mean platelet count was 3.52±1.07 lac/mm^3 in diabetic retinopathy, which is comparable to the values reported by Subashini S et al., (2.84 lac/mm^3±68.37/mm^3) and Taderegew M et al., (2.55 lac/mm^3±55.1/mm^3) [18,20].

Platelet Indices in DM with Complications

In the present study, the mean PDW was 12.05±1.27 in cases of myocardial infarction, which was similar to the studies by Dwivedi T, Davangeri R, and Buch A et al., which reported mean PDW values of 18.7±3.43 and 14.96±3.54, respectively [14,26]. The mean PDW was 13.78±1.38 in acute kidney injury and 14.05±2.29 in chronic kidney disease, aligning with the findings by Subashini S et al., Taderegew M et al., and Buch A et al., who reported mean PDW values of 15.72±3.97, 16.6±2.8, and 15.72±3.97, respectively [18,20,26].

The mean PDW in peripheral vasculopathy was 11.86±1.067, which is comparable to the study by Buch A et al., which reported a mean PDW of 12.67±4.93 [26]. The mean PDW in diabetic retinopathy was 14.1±3.31, similar to the studies by Subashini S et al., Taderegew M et al., and Buch A et al., which reported mean PDW values of 12.97±0.97, 16.7±2.8, and 14.92±4.14, respectively [18,20,26]. The mean PDW in gangrenous toe was 14.8±3.15, which was similar to the result reported by Buch A et al., with a mean PDW of 15.82±4.51 [26].

The MPV in myocardial infarction was 12.82±0.12, which was similar to the study by Buch A et al., which had a mean MPV of 10.94±1.73 [26]. The MPV in acute kidney injury and chronic kidney disease was 12.07±0.98 and 11.56±0.94, respectively, aligning with the findings by Subashini S et al., Taderegew M et al., and Buch A et al., who reported MPVs of 12.92±0.97, 13.8±2.5, and 11.0±2.23, respectively [18,20,26]. The MPV in diabetic retinopathy was 13.02±1.29, which is similar to the values reported by Subashini S et al., Taderegew M et al., and Buch A et al., with MPVs of 12.9±0.97, 13.7±1.9, and 11.40±1.96, respectively [18,20,26]. The MPV for gangrene toe and peripheral vasculopathy was 12.48±1.43 and 10.36±0.72, which was similar to the results by Buch A et al., who reported MPVs of 12.22±1.98 in diabetic foot and 10.97±1.77 in peripheral vascular disease [26].

Therefore, platelet indices play a significant role in the early detection of microvascular and macrovascular complications in diabetes mellitus, potentially preventing associated complications.

Limitation(s)

The limitation of the present study was the lack of follow-up with a few cases, which made it impossible to compare them with current findings. Nevertheless, these cases constitute only a minimal number in this study. Additionally, patients with qualitative disorders and reactive causes for elevated platelet counts were not assessed; however, these are considered to play a minor role.

CONCLUSION(S)

Diabetes contributes to endothelial dysfunction and platelet hyperactivity, leading to microvascular and macrovascular complications. In patients with DM, platelets tend to be larger, more active, and have a higher thrombogenic potential, resulting in elevated platelet indices. The present study observed variations in the levels of platelet count and platelet indices between diabetic patients with and without complications. Diabetic patients with uncontrolled glycaemic indices and high fasting blood sugar levels had increased platelet indices, which led to complications in comparison to patients without complications, where the platelet count, platelet indices, and glycaemic indices were within normal limits.

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PLAGIARISM CHECKING METHODS: [Jain H et al.]
- Plagiarism X-checker: Jun 17, 2023
- Manual Googling: Sep 19, 2023
- iThenticate Software: Dec 07, 2023 (13%)

ETYMOLOGY: Author Origin

EMENDATIONS: 7

AUTHOR DECLARATION:
- Financial or Other Competing Interests: None
- Was Ethics Committee Approval obtained for this study? Yes
- Was informed consent obtained from the subjects involved in the study? Yes
- For any images presented appropriate consent has been obtained from the subjects. NA

Date of Submission: **Jun 16, 2023**
Date of Peer Review: **Aug 29, 2023**
Date of Acceptance: **Dec 10, 2023**
Date of Publishing: **Feb 01, 2024**