Check for updates

ARTICLE OPEN

Oxidatively-induced DNA base damage and base excision repair abnormalities in siblings of individuals with bipolar disorder DNA damage and repair in bipolar disorder

Hidayet Ece Arat Çelik [1 ✉], Selda Yılmaz[2], İzel Cemre Akşahin[3,4], Burcu Kök Kendirlioğlu[1], Esma Çörekli[1], Nazlı Ecem Dal Bekar[5], Ömer Faruk Çelik [6], Neşe Yorguner[7], Bilge Targıtay Öztürk [8], Hüray İşlekel[2,9], Ayşegül Özerdem[10], Pınar Akan[11,12], Deniz Ceylan [4,13,14 ✉] and Gamze Tuna[2,12,14]

Previous evidence suggests elevated levels of oxidatively-induced DNA damage, particularly 8-hydroxy-2'-deoxyguanosine (8-OH-dG), and abnormalities in the repair of 8-OH-dG by the base excision repair (BER) in bipolar disorder (BD). However, the genetic disposition of these abnormalities remains unknown. In this study, we aimed to investigate the levels of oxidatively-induced DNA damage and BER mechanisms in individuals with BD and their siblings, as compared to healthy controls (HCs). 46 individuals with BD, 41 siblings of individuals with BD, and 51 HCs were included in the study. Liquid chromatography-tandem mass spectrometry was employed to evaluate the levels of 8-OH-dG in urine, which were then normalized based on urine creatinine levels. The real-time-polymerase chain reaction was used to measure the expression levels of *8-oxoguanine DNA glycosylase 1 (OGG1), apurinic/ apyrimidinic endonuclease 1 (APE1), poly ADP-ribose polymerase 1 (PARP1)*, and *DNA polymerase beta (POLβ)*. The levels of 8-OH-dG were found to be elevated in both individuals with BD and their siblings when compared to the HCs. The *OGG1* and *APE1* expressions were downregulated, while *POLβ* expressions were upregulated in both the patient and sibling groups compared to the HCs. Age, smoking status, and the number of depressive episodes had an impact on *APE1* expression levels in the patient group while body mass index, smoking status, and past psychiatric history had an impact on 8-OH-dG levels in siblings. Both individuals with BD and unaffected siblings presented similar abnormalities regarding oxidatively-induced DNA damage and BER, suggesting a link between abnormalities in DNA damage/BER mechanisms and familial susceptibility to BD. Our findings suggest that targeting the oxidatively-induced DNA damage and BER pathway could offer promising therapeutic strategies for reducing the risk of age-related diseases and comorbidities in individuals with a genetic predisposition to BD.

INTRODUCTION

Bipolar disorder (BD) is a chronic mood disorder that often co-occurs with various medical illnesses and is associated with premature aging, resulting in a shortened life expectancy [1]. Mounting evidence suggests that individuals with BD have elevated levels of oxidatively-induced DNA damage [2–4]. DNA is a highly susceptible molecule to oxidative insults, and it is estimated that every single cell of the human body is exposed to up to a million DNA lesions [5, 6]. Nevertheless, these damages are repaired by the cellular DNA repair machinery, which includes the base excision repair (BER). However, an overload of oxidative insults or insufficient repair can lead to persistent DNA damage, genomic instability, and ultimately, premature aging and the

development of various illnesses [6]. Therefore, oxidatively-induced DNA damage and abnormal DNA repair mechanisms have been suggested to play a crucial role in the shared pathophysiology among BD, increased cellular aging, and comorbidity [7–10].

The 8-hydroxy-2'-deoxyguanosine (8-OH-dG) is formed by the attack of the hydroxyl radical at the C8-position of guanine of dG followed by the one-electron oxidation of the OH-adduct radical of guanine [11, 12]. Since guanine is the most susceptible base to oxidation due to its low reduction potential, 8-OH-dG is the most widely used parameter to determine oxidatively-induced DNA damage [13, 14]. Alongside 8-hydroxyguanine (8-OH-Gua), various purine and pyrimidine base damages such as 8-hydroxyadenine

[1]Department of Psychiatry, School of Medicine, Maltepe University, Istanbul, Turkey. [2]Department of Molecular Medicine, Institute of Health Sciences, Dokuz Eylul University, Izmir, Turkey. [3]Graduate School of Health Sciences, Koc University, Istanbul, Turkey. [4]Research Center for Translational Medicine (KUTTAM), School of Medicine, Koc University, Istanbul, Turkey. [5]Chair of Proteomics and Bioanalytics, School of Life Sciences, Technical University of Munich, Munich, Germany. [6]Department of Medical Biochemistry, Sancaktepe Sehit Prof. Dr. Ilhan Varank Training and Research Hospital, Istanbul, Turkey. [7]Department of Psychiatry, School of Medicine, Marmara University, Istanbul, Turkey. [8]Department of Psychiatry, School of Medicine, Dokuz Eylul University, Izmir, Turkey. [9]Department of Medical Biochemistry, School of Medicine, Dokuz Eylul University, Izmir, Turkey. [10]Department of Psychiatry and Psychology, Mayo Clinic, Rochester, MN, USA. [11]Department of Neuroscience, Institute of Health Sciences, Dokuz Eylul University, Izmir, Turkey. [12]Biolzmir - Izmir Health Technologies Development and Accelerator Research and Application Center, Dokuz Eylul University, Izmir, Turkey. [13]Department of Psychiatry, School of Medicine, Koc University, Istanbul, Turkey. [14]These authors contributed equally: Deniz Ceylan, Gamze Tuna. ✉email: ecearat@yahoo.com; dozalp@ku.edu.tr

(8-OH-Ade), 4,6-diamino-5-formamidopyrimidine (FapyAde), 2,6-diamino-4-hydroxy-5-formamidopyrimidine (FapyGua), cytosine glycol (Cyt gly), thymine glycol (Thy gly), 5-hydroxymethyluracil (5-OH-MeUra), and 5,6-dihydroxycytosine exist. In contrast to the numerous well-established oxidatively-induced DNA base lesions, earlier research in psychiatric disorders exclusively concentrated on 8-OH-dG. Higher levels of 8-OH-dG were shown in individuals with BD compared to healthy controls (HCs) [2–4]. While some studies found higher levels during manic and depressive episodes, but not during euthymia [15–17], others found increased levels during all phases, including euthymia [18, 19]. A meta-analysis suggested that the increase of the 8-OH-dG is more pronounced in the depressive state of BD [4].

There are various DNA repair mechanisms, each designed to address specific types of damage or lesions, including oxidatively induced DNA lesions such as 8-OH-dG. Abnormal DNA repair processes have been observed in various medical conditions, including cancer and BD [20–23]. BER is recognized as the primary mechanism for addressing oxidatively-induced DNA damage [12], and while the number of studies on BER in BD is limited, reported abnormalities suggest its involvement in the disorder [24].

The BER pathway initiates with the recognition and removal of the damaged base by specific enzymes called DNA glycosylases, forming an apurinic/apyrimidinic (abasic) site. Among these, 8-oxoguanine DNA glycosylase 1 (OGG1) is specialized for the excision of 8-hydroxyguanine lesions. While existing research on BER gene expression in individuals with BD has predominantly focused on OGG1, showing decreased expression levels [25–27], the roles of genes involved in subsequent steps after DNA glycosylases remain less explored in BD.

In the BER pathway, endonucleases take part after DNA glycosylases and cleave the phosphodiester bonds in this abasic region. Apurinic/apyrimidinic endonuclease 1 (APE1) is a multi-functional endonuclease that creates a single-strand break on DNA, and the relationship between APE1 polymorphism and different cancer types has been identified in previous research [20, 28]. In addition, a study found that the polymorphism of APE1 is associated with the risk and onset of depression in patients with recurrent depression [29]. Subsequently, Poly-ADP-ribose poly-merase binds to the broken DNA ends, protecting them from further degradation. Finally, the gap in the site is filled by DNA polymerases, and a phosphodiester bond is formed by DNA ligases [30–33]. Downregulation of *DNA polymerase gamma (POLG)* gene expressions [26], and upregulation of *poly ADP-ribose polymerase 1 (PARP1)* gene expression [34] have been reported in mood disorders. DNA polymerase beta (POLβ) is the key polymerase enzyme of the BER mechanism responsible for the addition of nucleotides to apurinic/apyrimidinic ends. It partici-pates in various processes such as maintaining the stability of the genome [35] and telomeres [36] in cells, meiosis [37], and the ligation of non-homologous ends [38, 39]. To date, there have been no studies on the role of *POLβ* gene expression levels in psychiatric disorders.

Given that BD is a highly heritable disease, studies conducted on high-risk individuals are of great importance for the early detection of the disease and the identification of biological risk factors. The number of studies investigating oxidatively-induced DNA damage in individuals at high risk for BD is limited so far. Coello and colleagues found that the levels of 8-OH-dG were higher in newly diagnosed BD patients and their first-degree relatives compared to HCs [40]. On the other hand, another study found no difference in the levels of 8-OH-dG between twins at risk for mood disorders and HCs [41].

Currently, there is a dearth of research examining both oxidatively-induced DNA damage and BER mechanisms in individuals with BD, as well as in those at risk for the disorder, such as siblings of affected individuals. Thus, this study aims to investigate the levels of 8-OH-dG and assess the expression levels of the BER pathway genes involved in repairing 8-OH-dG lesions in individuals with BD, siblings of individuals with BD, and healthy controls.

MATERIALS AND METHODS
Study design
This is an observational study with a cross-sectional design that includes case-control groups. The study included 46 individuals with BD who were followed up at the Maltepe University, Faculty of Medicine, Department of Psychiatry between 2021 and 2022, 41 siblings of individuals with BD, and 51 HCs. The siblings of patients who are currently being followed at the hospital were invited to participate in the study via phone contact. The HCs were selected from individuals who agreed to participate in the study by accepting the distributed leaflets around the hospital vicinity. The research has been approved by the Maltepe University Faculty of Medicine Clinical Research Ethics Committee. (Date: 07/24 /2019 Number: 2019/900/48).

Participants
The individuals participating in the study were between the ages of 18–50. All participants underwent a SCID-5 interview in which the DSM-5 diagnostic criteria were examined. In addition, Health-Promoting Lifestyle Profile-II (HPLP-II) and World Health Organization Quality of Life-BREF (WHOQOL-Bref) were applied to the participants. While some studies found differences between affective episodes and euthymia in oxidatively-induced DNA damage markers [15, 17], we only included individuals with BD or siblings who were in remission for at least 4 months. The participants with HAMD-17 and YMRS scores below 7 were included in the study. Individuals who had a decompensated systemic medical condition, morbid obesity, diabetes mellitus, rheumatologic disease, active infection, or a serious neurological disease that could affect oxidative parameters, using antioxidant-containing treatments or supportive products that could affect oxidative parameters, had a serious abnormality in routine laboratory findings, had a lifelong intellectual disability or conditions affecting cognitive function (such as delirium, dementia, epilepsy, etc.), had a diagnosis of alcohol or substance abuse or addiction at any point of life, were in pregnancy, lactation period or were going through menopause were excluded from the study. Individuals who were diagnosed with schizophrenia, schizophreniform disorder, schizoaffective disorder, brief psychotic disorder, and psychotic disorder not otherwise specified after the SCID-5 interview in the sibling group were excluded from the study, because recent studies demonstrated a significant increase in oxidatively-induced DNA damage in psychotic disorders [3]. On the other hand, siblings who had a history of or currently have any psychiatric diagnosis have not been excluded from the study, as they may have a higher predisposition towards BD. In the healthy control group, individuals who had a history of or currently had any psychiatric diagnosis were excluded from the study. Urine and blood samples were collected from the participants after the diagnostic interview.

Collection and storage of the samples
Participants' first-morning urine samples and fasting blood samples (10 ml) between 08:00 and 09:00 am were collected. Daily RNA isolations were performed on the collected whole blood samples. The RNA was extracted from each whole blood sample using RNeasy Mini Kit (Qiagen Diagnostics GmbH, Germany) following the manufacturer's instructions. The amount and purity of RNA samples were measured using a Nanodrop 2000 spectro-photometer (Thermo Scientific). After isolation, RNA was stored at −80 °C until it was converted to cDNA.

8-OH-dG quantification
To evaluate oxidatively-induced DNA damage, liquid chromatography-tandem mass spectrometry (LC-MS/MS) with stable-isotope dilution using multiple reaction monitoring (MRM) acquisition mode was performed according to the previously published protocol [42]. 15.6 μL of 8-OH-dG-^{15}N$_5$ (0.002 mM) internal standard is added to 1 mL of urine samples [43], which were then centrifuged at $1000 \times g$ for 15 min. Subsequently, supernatants were filtered using nylon syringe filters. Filtered samples were loaded onto extraction cartridges and then washed with 2 mL of distilled water. 1 mL of 30% methanol was used for the elution of retained material. Eluted samples were dried in a vacuum concentrator (Thermo Scientific SpeedVac, Marietta, Ohio, USA) and then dissolved in 100 μL digestion buffer (1 mol/L sodium acetate, 10 mmol/L Tris-HCl, pH 7.5).

Table 1. Gene sequences of BER enzymes.

Gene	Gene sequences	
	F (5′-3′)	R (3′-5′)
OGG1	GGCTCAACTGTATCACCACTGG	GGCGATGTTGTTGTTGGAGGAAC
APE1	CTGCTCTTGGAATGTGGATGGG	TCCAGGCAGCTCCTGAAGTTCA
PARP1	CCAAGCCAGTTCAGGACCTCAT	GGATCTGCCTTTTGCTCAGCTTC
POLβ	TGCAGAGTCCAGTGGTGACATG	ATGAACCTTTTGTAACTGCTCCAC
GAPDH	CCCTTCATTGACCTCAACTACA	ATGACAAGCTTCCCGTTCTC
ACTB	CCCAGATCATGTTTGAGACCTT	CCAGAGGCGTACAGGGAT

Then, samples were hydrolyzed with alkaline phosphatase (22 units per sample) at 37 °C for 1 h. Samples were filtered using 3 kDa tubes by centrifugation at $5000 \times g$ for 50 min. The studies were performed by HPLC (Shimadzu, Kyoto, Japan) coupled with a mass spectrometer equipped with a triple quadrupole ion trap (4000 QTRAP Applied Biosystems, CA, USA) in the positive ionization mode. Samples were separated by an LC column with 2.1 mm × 150 mm, 3.5 μm particle size (Zorbax SB-Aq column, Agilent Technologies, California, USA) and an attached C8 guard column (2.1 mm × 12.5 mm, 5 μm particle size). Mobile phases were water containing 0.1% formic acid (mobile phase A) and acetonitrile containing 0.1% formic acid (mobile phase B). Analysis by LC-MS/MS with MRM was performed using the mass/charge (m/z) transitions m/z 284 → m/z 168, and m/z 289 → m/z 173 for 8-OH-dG and 8-OH-dG-$^{15}N_5$, respectively. Urinary creatinine concentrations were used for the normalization of the results. The results were expressed in nmol 8-OH-dG/mmol creatinine.

Base excision enzymes mRNA expression quantification using quantitative real Time-PCR

RNA samples were converted to cDNA using the RT2 First Strand Kit (Qiagen Diagnostics GmbH, Germany), and the quantitative Reverse Transcription PCR amplification was performed in triplicate by The LightCycler® 480 Instrument II (Roche) at Koc University, KUTTAM Laboratory. OGG1, APE1, PARP1, and POLβ expressions were measured by RT-qPCR according to the manufacturer's protocol (LightCycler 480 SY Green I Master Handbook Version 13, Roche Diagnostics GmbH, Mannheim, Germany). The GAPDH and β-Actin were used as housekeeping genes. The primers for OGG1, PARP1, APE1, POLβ, GAPDH, and β-Actin were obtained from the manufacturer (SentebioLab, Turkey) (Table 1). The outputs showing cycling reports and melting curves were obtained using the LightCycler® 480 Software Version 1.5.0.39. Any Ct value of more than 35 was considered negative melting curve and was analyzed to confirm the specificities of the amplicons for RT-qPCR amplification.

Each set of samples underwent normalization using two housekeeping genes, GAPDH and β-Actin. The normalization was performed using the formula $\Delta Ct = Ct_{OGG1} - [(Ct_{GAPDH} + Ct_{β-Actin})/2]$. To determine the relative changes in mRNA expression levels of the base excision enzyme genes, the term $2^{-\Delta\Delta Ct}$ was employed, where $\Delta\Delta Ct = \Delta Ct_{patient} - \Delta Ct_{the\ mean\ value\ of\ the\ healthy\ control\ group}$, and the fold changes were compared between groups. The experiments adhered to the guidelines provided by Minimum Information about Quantitative Real-Time PCR Experiments (MIQE) [44].

Statistical analyses

IBM SPSS Statistics 29.0 (Chicago IL, USA) for Windows was used for the statistical analysis. Categorical variables were analyzed with the Chi-square test. Continuous variables were checked for Gaussian distribution using quantile-quantile plots, distribution of data in histograms, skewness values (−1 to +1), kurtosis values (−2 to +2), and confirmed by the Kolmogorov–Smirnov Test. Medians and interquartile ranges were used in the figures. Both mean and SD and median and minimum–maximum values were presented in the tables when normal distribution was not achieved. The level of significance was taken as 0.05 for all tests.

Univariate Analysis of Variance (ANOVA) and Chi-square tests were applied to compare demographical and clinical variables between study groups. Levels of 8-OH-dG/creatinine and expression levels of OGG1, APE1, PARP1, POLβ were compared among study groups using the Quade Nonparametric Univariate Analyses of Covariance (ANCOVA) models which include age, sex, body mass index (BMI), smoking status, and alcohol consumption as covariates.

Spearman correlation analyses were applied in patient and sibling groups separately to evaluate correlations among continuous clinical variables (e.g., number of previous manic, hypomanic, and depressive episodes, number of episodes with mixed features, number of psychotic episodes, age of onset, duration of illness, duration of remission, number of suicide attempt, number of hospitalization, scale scores [HAMD-17, YMRS, HPLP-II total scores, WHOQOL-Bref total scores], etc.) and dependent variables (i.e., 8-OH-dG/creatinine levels, and OGG1, APE1, PARP1, POLβ gene expression levels). Point-biserial correlation analyses were applied to evaluate correlations among categorical clinical variables (e.g., current/past psychiatric history, medication type) and dependent variables (i.e., 8-OH-dG/creatinine levels, and OGG1, APE1, PARP1, POLβ gene expression levels).

Linear regression models were applied in patient and sibling groups separately, to identify the effect of sociodemographic variables (i.e., age, sex, BMI, smoking status, alcohol consumption, and HPLP-II total scores), and clinical variables that correlate with the main findings, current and past history of psychiatric illness, and medication use (i.e., mood stabilizers, antipsychotics, and antidepressants) on main findings.

RESULTS

Demographics

Comparison of the demographic and clinical characteristics between study groups is presented in Table 2. While there was no significant difference in age, sex, and years of education between groups, a significant difference was found in terms of employment status ($p < 0.001$), marital status ($p = 0.035$), BMI ($p = 0.007$) and alcohol consumption ($p = 0.033$).

Urine 8-OH-dG/creatinine levels

The median urine 8-OH-dG levels were 3.20 (1.25–6.47) nmol/mmol creatinine in individuals with BD, 3.28 (0.78–8.94) nmol/mmol creatinine in siblings, and 2.55 (0.96–7.10) nmol/mmol creatinine in HCs (Table 3, Fig. 1). The levels of 8-OH-dG of individuals with BD and siblings were found to be significantly higher than the HCs ($F = 4.520$, $t = 2.249$, $p = 0.026$; $t = 2.803$, $p = 0.006$). There was no significant difference between individuals with BD and siblings according to 8-OH-dG levels. Figure 2 illustrates representative ion-current profiles of the m/z 284 → 168 (8-OH-dG), and m/z 289 → 173 (8-OH-dG-$^{15}N_5$) mass transitions, which were recorded during the LC-MS/MS analysis of urine samples.

Base excision gene expression levels

The median OGG1 mRNA expression levels of the patient group (0.15 [0.05–3.38]) and sibling group (0.15 [0.03–1.72]) were significantly lower than the control group (0.19 [0.07–0.87]) ($F = 3.730$, $t = -2.361$, $p = 0.020$; $t = -2.278$, $p = 0.025$). There was no significant difference between individuals with BD and siblings according to OGG1 mRNA expression levels.

The median APE1 mRNA expression levels of the individuals with BD (0.34 [0.11–1.55]), and siblings (0.33 [0.07–4.08]) were significantly lower than the HCs (0.83 [0.20–10.74]) ($F = 19.928$, $t = -5.597$, $p < 0.001$; $t = -5.101$, $p < 0.001$). Individuals with BD and siblings showed no difference according to APE1 mRNA expression levels.

The median PARP1 expression levels of the individuals with BD (0.38 [0.09–1.42]), siblings (0.40 [0.09–18.32]), and healthy controls (0.40 [0.07–1.30]) did not show a significant difference ($F = 0.301$, $p = 0.741$).

The median POLβ mRNA expression levels of individuals with BD (0.19 [0.06–0.55]) and siblings (0.18 [0.06–1.03]) were significantly higher than the HCs (0.15 [0.03–0.61]) ($F = 5.642$, $t = 2.490$, $p = 0.014$; $t = 3.126$, $p = 0.002$). Individuals with BD and siblings showed no difference according to POLβ mRNA expression levels (Table 3, Fig. 1).

Effect of clinical variables on urine 8-OH-dG/creatinine and base excision gene expression levels

In the patient group, there was no correlation between demographic and clinical variables and 8-OH-dG/creatinine levels. There was a negative correlation between OGG1 mRNA expression levels and the number of suicide attempts ($r = -0.341$, $p = 0.036$),

Table 2. Comparison of the demographic and clinical characteristics between study groups.

	BD (n = 46) Median (min-max) Mean ± SD	Sibling n = 41) Median (min-max) Mean ± SD	HC (n = 51) Median (min-max) Mean ± SD	Test statistics F/χ2	p
Age	35.33 ± 7.75	36.46 ± 7.86	34.25 ± 7.22	0.971	0.384
Female (n, %)	31 (67.4)	25 (61.0)	27 (52.9)	2.353	0.346
Years of education	15 (8–12) 14.54 ± 2.47	15 (8–23) 14.12 ± 4.01	15 (7–22) 14.61 ± 2.70	0.307	0.727
Employed (n, %)	31 (67.4)	35 (85.4)	50 (98)	18.385	**<0.001**
Married (n, %)	19 (41.3)	28 (68.3)	30 (58.8)	6.131	**0.035**
BMI	27.01 ± 4.61	24.79 ± 3.45	24.83 ± 3.11	6.278	**0.007 BD > Sibling p = 0.007 BD > HC p = 0.005**
Smoker (n, %)	27 (58.7)	17 (41.5)	28 (54.9)	3.064	0.244
Using alcohol (n, %)	19 (41.3)	25 (61.0)	34 (66.7)	7.397	**0.033**
HPLP-II					
Health responsibility	21 (13–30) 21.25 ± 4.16	21 (11–27) 20.54 ± 4.13	21 (14–30) 21.12 ± 3.88	0.365	0.695
Physical activity	15 (8–27 15.25 ± 4.64	17 (9–30) 17.36 ± 5.04	18 (8–30) 17.76 ± 5.49	3.295	**0.040 BD < HC p = 0.016**
Nutrition	19 (12–29) 19.27 ± 3.83	20 (13–29) 20.66 ± 3.47	20 (13–29) 19.99 ± 3.50	1.619	0.202
Spiritual growth	26 (11–34 25.20 ± 4.65	26 (18–35) 26.22 ± 4.13	28 (18–34 27.53 ± 3.68	3.857	**0.023 BD < HC p = 0.007**
Interpersonal relations	25.08 ± 4.29	25.89 ± 3.98	27.04 ± 3.22	3.223	**0.043 BD < HC p = 0.013**
Stress management	20.02 ± 3.50	10.90 ± 3.80	19.94 ± 3.31	0.013	0.987
Total score	126.06 ± 16.22	130.57 ± 16.59	133.38 ± 16.45	2.426	0.092
WHOQOL-Bref					
Physical	26 (19–33 26.29 ± 3.49	29 (16–35) 27.86 ± 4.13	30 (22–35) 29.91 ± 3.21	12.361	**<0.001 BD < HC p < 0.001 BD < Sibling p = 0.045 Sibling < HC p = 0.007**
Psychological	22 (13–28) 21.24 ± 3.35	22 (14–28 21.90 ± 3.02	23 (15–30) 23.61 ± 2.85	7.702	**<0.001 BD < HC p < 0.001 Sibling < HC p = 0.009**
Social	10 (5–14) 10.37 ± 2.10	12 (8–14) 11.40 ± 1.80	12 (8–15) 11.82 ± 1.73	7.467	**< 0.001 BD < HC p < 0.001 BD < Sibling p = 0.012**
Environment	30 (22–40) 29.83 ± 4.08	31 (23–37) 30.72 ± 3.81	31 (23–39) 31.00 ± 3.33	1.261	0.287
Total score	94.77 ± 11.65	99.01 ± 11.63	103.91 ± 10.25	8.154	**<0.001 BD < HC p < 0.001 Sibling < HC p = 0.038**
Additional current psychiatric condition[a] (n, %)	7 (15.2)	5 (12.2)	-	-	-
Past psychiatric condition[b] (n, %)	1 (2.2)	13 (31.7)	-	-	-
Number of suicide attempts	0 (0–3) 0.28 ± 0.66	0 (0–1) 0.07 ± 0.26	-	-	-
Number of manic episodes	2 (1–10) 2.74 ± 1.96	-	-	-	-
Number of hypomanic episodes	0 (0–9) 0.80 ± 1.72	-	-	-	-
Number of depressive episodes	1 (0–10) 1.96 ± 1.91	-	-	-	-
Number of episodes with mixed features	0 (0–5) 0.43 ± 0.93	-	-	-	-
Number of psychotic episodes	2 (0–10) 2.13 ± 2.20	-	-	-	-

Table 2. continued

	BD (*n* = 46) Median (min-max) Mean ± SD	Sibling *n* = 41) Median (min-max) Mean ± SD	HC (*n* = 51) Median (min-max) Mean ± SD	Test statistics F/χ2	*p*
Total number of episodes	5 (2–20) 5.91 ± 3.60	-	-	-	-
Age of onset	22 (13–40) 22.72 ± 5.92	-	-	-	-
Duration of illness (months)	120 (24–396) 149.48 ± 94.82	-	-	-	-
Duration of remission (months)	12 (4–96) 22.22 ± 23.08	-	-	-	-
Number of hospitalizations	1 (0–8) 1.71 ± 1.73	-	-	-	-
HAMD-17	0 (0–6) 0.98 ± 1.61	0 (0–0) 0.51 ± 1.07	-	-	-
YMRS	0 (0–4) 0.59 ± 1.13	0 (0–2) 0.05 ± 0.32	-	-	-

BD bipolar disorder, *HC* Healthy control, *BMI* Body mass index, *HPLP II* Health-Promoting Lifestyle Profile-II, *WHOQOL-Bref* World Health Organization Quality of Life-BREF, *HAMD-17* Hamilton Depression Rating Scale-17, *YMRS* Young Mania Rating Scale.
[a]Additional current psychiatric conditions for BD patients: anxiety disorders (*n* = 3), obsessive-compulsive disorder (OCD, *n* = 1), attention-deficiency/hyperactivity disorder (ADHD, *n* = 2), eating disorder (*n* = 1); for siblings: anxiety disorders (*n* = 3), comorbid anxiety and ADHD (*n* = 2).
[b]Past psychiatric condition for BD patients: ADHD (*n* = 1), for siblings: major depressive disorder (MDD, *n* = 10), anxiety disorders (*n* = 2), comorbid anxiety and ADHD (*n* = 1).
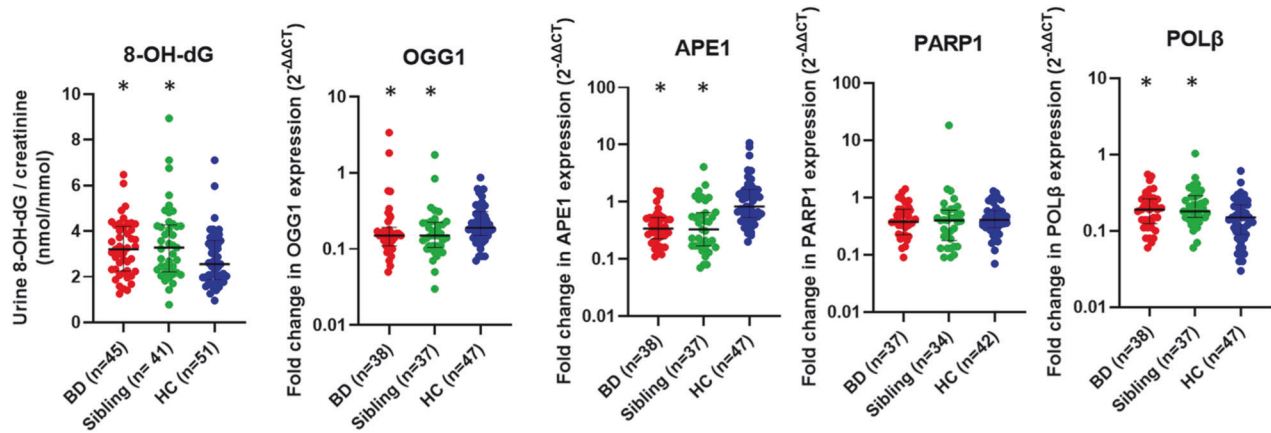All significant results have been highlighted in bold.

Table 3. Comparison of 8-OH-dG/creatinine levels and BER pathway enzymes gene expression levels among study groups.

|	BD Median (min-max) Mean ± SD	Sibling Median (min-max) Mean ± SD	HC Median (min-max) Mean ± SD	*F*	*p*
8-OH-dG/creatinine (nmol/mmol)	3.20 (1.25–6.47) 3.25 ± 1.22	3.28 (0.78–8.94) 3.51 ± 1.64	2.55 (0.96–7.10) 2.78 ± 1.19	4.520	**0.013 BD > HC *p* = 0.026 Sibling > HC *p* = 0.006**
Fold change in OGG1 mRNA expression ($2^{-\Delta\Delta Ct}$)	0.15 (0.05–3.38) 0.30 ± 0.59	0.15 (0.03–1.72) 0.22 ± 0.29	0.19 (0.07–0.87) 0.25 ± 0.17	3.730	**0.027 BD < HC *p* = 0.020 Sibling < HC *p* = 0.025**
Fold change in APE1 mRNA expression ($2^{-\Delta\Delta Ct}$)	0.34 (0.11–1.55) 0.45 ± 0.36	0.33 (0.07–4.08) 0.60 ± 0.75	0.83 (0.20–10.74) 1.60 ± 2.11	19.928	**<0.001 BD < HC *p* < 0.001 Sibling < HC *p* < 0.001**
Fold change in PARP1 mRNA expression ($2^{-\Delta\Delta Ct}$)	0.38 (0.09–1.42) 0.49 ± 0.31	0.40 (0.09–18.32) 0.96 ± 3.08	0.40 (0.07–1.30) 0.50 ± 0.30	0.301	0.741
Fold change in POLβ mRNA expression ($2^{-\Delta\Delta Ct}$)	0.19 (0.06–0.55) 0.21 ± 0.12	0.18 (0.06–1.03) 0.24 ± 0.17	0.15 (0.03–0.61) 0.16 ± 0.11	5.642	**0.005 BD > HC *p* = 0.014 Sibling > HC *p* = 0.002**

All significant results have been highlighted in bold.

HAMD-17 scores ($r = -0.410$, $p = 0.013$), and WHOQOL-Bref total scores ($r = -0.376$, $p = 0.020$). *APE1* mRNA expression levels showed positive correlations with the number of depressive episodes ($r = 0.420$, $p = 0.009$), and negative correlations with the number of hospitalizations ($r = -0.330$, $p = 0.043$) and WHOQOL-Bref total scores ($r = -0.377$, $p = 0.020$). *PARP1* mRNA expression levels showed a positive correlation with the use of antidepressants ($\rho = 0.419$, $p = 0.010$), and *POLβ* mRNA expression levels showed no correlation with the demographic and clinical variables. In the patient group, linear regression models, which included sociodemographic variables, and clinical variables that correlated with the main findings (i.e., number of depressive episodes, number of suicide attempts, number of hospitalizations, HAMD-17 scores, antidepressant use), revealed that those parameters did not have a significant impact on 8-OH-dG levels ($R^2 = 0.091$, $F = 0.635$, $p = 0.702$). Age, smoking status, and the

Fig. 1 Comparison of 8-OH-dG/creatinine levels and BER pathway enzymes gene expression levels among study groups. Levels of 8-OHdG were significantly higher in individuals with bipolar disorder and siblings of individuals with bipolar disorder compared to healthy controls. Expression levels of *OGG1* and *APE1* genes were significantly lower, and expression levels of *POLβ* gene were significantly higher in individuals with bipolar disorder and siblings of individuals with bipolar disorder compared to healthy controls.
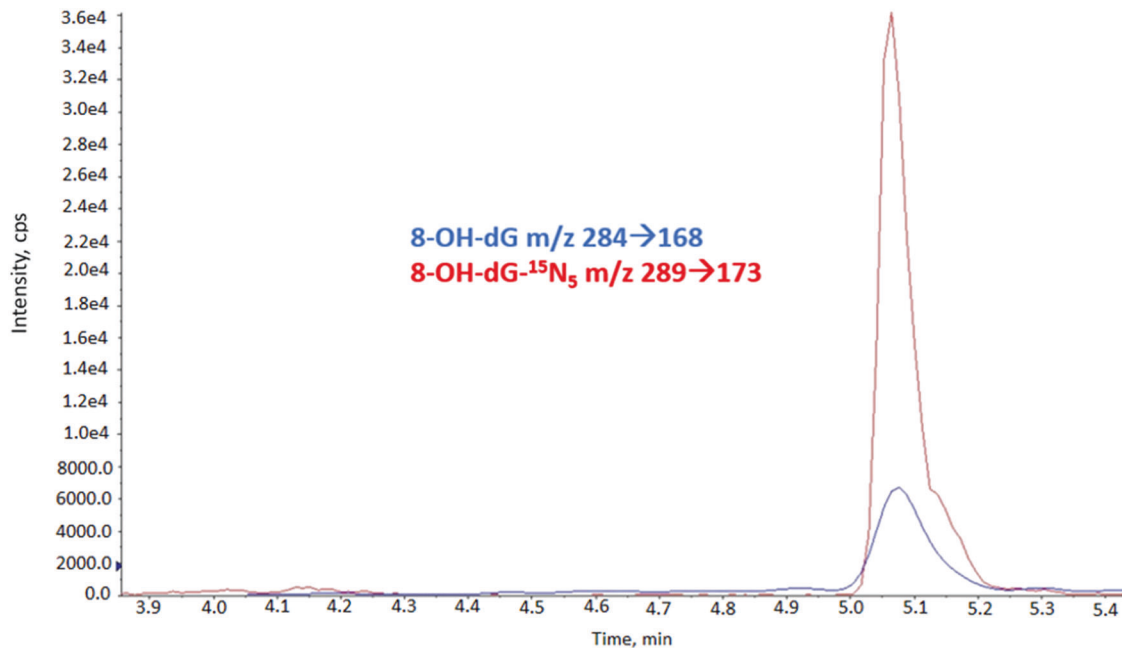
Fig. 2 Ion–current profiles of the *m/z* 284→168 (8-OH-dG), *m/z* 289→173 (8-OH-dG-$^{15}N_5$) mass transitions. Ion–current profiles for 8-OH-dG at *m/z* 284→168 (*m/z* precursor ion → product ion) and for 8-OH-dG-$^{15}N_5$ at *m/z* 289→173 (*m/z* precursor ion → product ion) were recorded during the LC-MS/MS analysis of a urine sample. The peak corresponding to 8-OH-dG in the figure originates from the analyte in the sample, and the peak for 8-OH-dG-$^{15}N_5$ comes from the stable isotope-labeled internal standard added to the sample. Quantification was performed using the ratio of the integrated peak areas of these two peaks.

number of depressive episodes had an impact on *APE1* mRNA expression levels ($R^2 = 0.610$, $F = 5.659$, $p < 0.001$; $B = -0.016$, $t = -2.815$, $p = 0.009$; $B = 0.238$, $t = 2.653$, $p = 0.013$; $B = 0.124$, $t = 5.122$, $p < 0.001$). Clinical parameters did not have an impact on *OGG1*, *PARP1* or *POLβ* mRNA expression levels ($R^2 = 0.314$, $F = 1.548$, $p = 0.187$; $R^2 = 0.256$, $F = 1.422$, $p = 0.235$; $R^2 = 0.258$, $F = 1.795$, $p = 0.133$).

In siblings, there was a positive correlation between 8-OH-dG levels and past psychiatric history ($\rho = 0.315$, $p = 0.045$), and there was no relationship between *OGG1*, *APE1*, *POLβ*, *PARP1* expression levels, and clinical variables. In sibling group, linear regression models, which included sociodemographic variables, and clinical variables that correlated with the main findings (i.e., past psychiatric history), revealed that BMI, smoking status, and past

psychiatric history had an impact on 8-OH-dG levels ($R^2 = 0.395$, $F = 3.073$, $p = 0.013$; $B = -0.174$, $t = -2.343$, $p = 0.025$; $B = 1.015$, $t = 2.129$, $p = 0.041$; $B = 1.338$, $t = 2.666$, $p = 0.012$). None of the clinical variables had an impact on *OGG1*, *APE1*, *PARP1*, *POLβ* mRNA expression levels ($R^2 = 0.176$, $F = 1.065$, $p = 0.405$; $R^2 = 0.180$, $F = 1.100$, $p = 0.385$; $R^2 = 0.091$, $F = 0.453$, $p = 0.836$; $R^2 = 0.213$, $F = 1.357$, $p = 0.264$).

DISCUSSION

This study is the first to compare both oxidatively-induced DNA damage and BER expression levels among individuals with BD, siblings of individuals with BD, and HCs. Our findings indicate higher levels of 8-OH-dG, downregulated *OGG1*, and *APE1*

expressions, and upregulated *POLβ* expressions in both the patient and sibling groups compared to HCs, with no significant differences observed between individuals with BD and their siblings.

Our findings present increased levels of 8-OH-dG in euthymic individuals with BD compared to HCs, which is supported by various studies in the literature [18, 19, 40]. Additionally, our results are consistent with a large-scale study that reported higher levels of 8-OH-dG in at-risk relatives of individuals with BD compared to HCs [40]. The elevated levels of 8-OH-dG in at-risk individuals support the role of oxidatively-induced DNA damage in BD pathogenesis. Additionally, our findings suggest that past psychiatric history has an impact on 8-OH-dG levels in siblings, most of whom had previous depressions. Increasing data suggest that there is a significant elevation in 8-OH-dG levels in depression [4, 45]. However, follow-up studies suggest that the increase in acute depression is reversible and returns to normal values after the resolution of depressive symptoms [27, 34, 46]. It should be noted that the siblings with previous episodes were euthymic at the time of inclusion in our study. Siblings with previous episodes may be more likely to share a genetic predisposition with individuals with BD, who present high levels of 8-OH-dG despite being euthymic. In addition, despite the widespread recognition of high DNA damage in obesity [47], we found that BMI has a negative impact on 8-OH-dG levels in siblings of individuals with BD. However, it is important to note that our study sample only included a small subset of siblings with obesity, and none of them had morbid obesity, which limits the generalizability of this finding.

Our finding showing a down-regulation in *OGG1* gene expression in BD is in concordance with two studies presenting consistent findings of decreased *OGG1* gene expression levels during euthymia in BD [25, 26]. In contrast, a study reported decreased *OGG1* gene expression in acute unipolar or bipolar depression compared to HCs, with a significant increase after remission [27]. However, the authors speculated that the increase in *OGG1* gene expression after remission might be temporary and may return to decreased levels over time. Our findings also present, a down-regulation of *APE1* gene expression and upregulation of *POLβ gene* expression in BD and sibling groups, while *PARP1* gene expression was comparable among study groups. As far as we know, *APE1* and *POLβ gene* expressions have not been investigated in peripheral samples of individuals with BD.

Our data revealed a significant association between *APE1* levels and the number of previous depressive episodes. Our findings also showed that *APE1* gene expression levels in patients with BD may be influenced by age, as oxidative stress has been shown to be closely related to aging and deficiencies in repair enzymes. Additionally, smoking had a positive effect on *APE1* gene expression levels in the patient group and 8-OH-dG levels in the sibling group, as smoking is associated with higher levels of 8-OH-dG. It is possible that increased expression of *APE1* levels occurs in response to increased levels of 8-OH-dG, serving as an adaptive mechanism for repair in individuals with BD. In our study, the finding of elevated *POLβ* gene expression levels in patient and sibling groups compared to HCs may indicate the significant involvement of this enzyme in the physiopathology of BD.

On the other hand, our study found comparable levels of *PARP1* gene expression in individuals with BD and their siblings to those in HCs. While previous research has not investigated *PARP1* levels in BD specifically, our negative result may be due to studying a group of individuals who were euthymic at the time of inclusion. PARP1 gene polymorphism has been found to be associated with depression [29], and increased *PARP1* gene expression levels have been observed in individuals with depression [34]. Inhibition of *PARP1* has shown antidepressant effects in both human and animal studies [48–50], suggesting that it could be a potential target for future antidepressant treatments. Further studies investigating these genes in acute phases would extend our understanding.

Lower levels of *OGG1* and *APE1* gene expression and higher levels of *POLβ* gene expression in siblings compared to HCs, indicate reduced DNA repair capacity in both patients with BD and their siblings. Many clinical, molecular, and neuroimaging studies have shown differences in first-degree relatives of individuals with BD compared to HCs [51]. Individuals who have a genetic predisposition to BD may display impairments in visual memory, verbal memory, processing speed, attention, and social cognition compared to HCs [52]. Structural and functional alterations in the prefrontal cortex [53–55] and white matter abnormalities in the body and splenium of the corpus callosum [56] have been observed in individuals with genetic predisposition to BD, as well as alterations in markers of neuroimmune dysregulation [57, 58]. Our results suggest that the BER pathway genes may serve as candidate endophenotypes for BD. In addition, dysregulated BER gene expressions may underlie the increased 8-OH-dG load in euthymic individuals with BD and their full siblings, potentially contributing to disease vulnerability and premature aging. However, further studies investigating BER genes and aging markers together are needed to confirm these associations.

The current study has several strengths. First, to the best of our knowledge, this is the first study to investigate the BER genes in the unaffected siblings of individuals with BD. Since full siblings of individuals with BD have the highest risk of developing the disorder after identical twins [59], the inclusion of full siblings of individuals with BD can provide valuable insights to demonstrate the DNA damage/repair abnormalities in individuals with a genetic predisposition of BD and can help identify potential targets for early intervention and prevention of BD. Additionally, mood-stabilizing medication is a major challenge in most studies on BD. The inclusion of full siblings who are genetically similar to individuals with BD but are not taking any medication helps to distinguish the potential impacts of medications. Second, using urine samples for 8-OH-dG measurement is another strength of the study, because it allows for the assessment of systemic oxidatively-induced DNA load in the body. Measurements of DNA damage markers in tissues can be affected by repair processes and may not fully reflect the oxidative load in the whole body. However, the products of oxidatively-induced DNA lesions, after repair, are excreted into urine without being further metabolized, making urinary 8-OH-dG levels a reliable indicator of systemic oxidatively-induced DNA damage [60, 61]. Additionally, the use of a gold standard technique for urine DNA damage measurements, liquid chromatography-tandem mass spectroscopy, for 8-OH-dG quantification is a strength due to its sensitivity and specificity compared to other techniques such as immunosorbent methods. To address a potential limitation resulting from the impact of glomerular filtration rate in measuring 8-OH-dG levels in urine samples [60], we used a normalization method by adjusting the 8-OH-dG levels to the levels of creatinine, as recommended by previous research [17]. Third, we have limited our cohort to individuals under the age of 50, as current literature defines old age as 50 in the BD population [62], in order to exclude the impact of senescence on our findings. Numerous studies have shown that age is a major confounding factor in research on oxidatively-induced DNA damage [63, 64]. Fourth, we controlled our findings for various types of confounders, including lifestyle factors, in addition to common confounders such as age, sex, BMI, smoking, and alcohol consumption. More importantly, investigating DNA damage and repair processes in combination allows us to gain a more comprehensive understanding of this dynamic process.

Limitations
The five main limitations of this study must be considered and discussed. First, the cross-sectional nature of the study and the

lack of longitudinal follow-up of patients and siblings limit the ability to draw causal conclusions and understand the direction of causality. Therefore, future follow-up of these siblings would be helpful in differentiating the roles of possible factors, such as resilience to BD, on these parameters. Second, some of the individuals recruited in the sibling group had a history of or currently presented psychiatric disorders. Although psychotic spectrum disorders and current depressive episodes were exclusion criteria due to their potential to cause a significant increase in 8-OH-dG levels, other psychiatric disorders such as past major depression, anxiety disorders, and attention deficit hyperactivity disorder were not considered as exclusion criteria in the sibling group. Although there are findings in the literature indicating increased DNA damage in these disorders [2, 46, 65], there is currently no available data on the extent to which this damage changes during the remission period. Additionally, our results showed that the current comorbid psychiatric illness in the sibling group did not affect DNA damage and BER mechanisms in the regression analysis. However, the potential influence of past or present psychiatric disorders on DNA damage and repair mechanisms should be considered in future studies with larger sample sizes. Third, another limitation of our study is that we did not exclude smokers, despite the well-established influence of smoking on these markers. However, due to the high prevalence of smoking in patients with BD and individuals with a genetic predisposition to BD, excluding smokers would introduce potential sampling bias. On the other hand, to address this limitation, we controlled for the effect of smoking in all statistical analyses. Fourth, our study focused only on the levels of 8-OH-dG lesion and four key BER genes involved in the repair of 8-OH-dG damage. Further studies investigating other types of DNA lesions, as well as RNA lesions, and the full pathway of BER genes will provide more comprehensive knowledge. Additionally, only focusing on mRNA expressions of genes is a limitation because protein levels and enzymatic activity may also affect the BER. Future studies using more comprehensive methods to investigate the BER enzymes may provide a more comprehensive understanding of the entire process. Finally, it is important to note that our study investigated DNA damage/repair status in peripheral blood cells, while brain cells have a high metabolic rate with high oxygen turnover, they may have a greater reliance on efficient and active BER than peripheral blood cells. On the other hand, studies suggest that there are consistent results among peripheral and central levels of DNA damage markers [66, 67]. Nevertheless, future studies investigating the DNA damage/repair status in brain tissue may provide a more accurate reflection of the mechanisms involved in BD.

CONCLUSION

Our findings suggest that abnormalities in DNA damage and repair mechanisms are linked to familial susceptibility to BD. Given that increased levels of 8-OH-dG have also been observed in other psychiatric and somatic diseases, elevated levels of 8-OH-dG may indicate a shared mechanism of increased oxidatively-induced DNA damage and cellular aging, which may contribute to the comorbidity risk in individuals with a genetic predisposition to BD. On the other hand, the BER pathway abnormalities might be a potential underlying accumulation of DNA damage, leading to premature aging and an increased risk of comorbidities such as metabolic disorders, cardiovascular diseases, and neurodegenerative diseases in individuals with a genetic predisposition to BD. Hence, targeting the BER pathway might offer promising therapeutic strategies for reducing the risk of age-related diseases and comorbidities in individuals with mood disorders. Further, large-scale longitudinal studies investigating both oxidatively-induced DNA damage and BER pathways in BD are required to obtain more accurate and precise results.

REFERENCES

1. Fries GR, Zamzow MJ, Andrews T, Pink O, Scaini G, Quevedo J. Accelerated aging in bipolar disorder: a comprehensive review of molecular findings and their clinical implications. Neurosci Biobehav Rev. 2020;112:107–16.
2. Jorgensen A, Baago IB, Rygner Z, Jorgensen MB, Andersen PK, Kessing LV, et al. Association of oxidative stress-induced nucleic acid damage with psychiatric disorders in adults: a systematic review and meta-analysis. JAMA Psychiatry. 2022;79:920–31.
3. Goh XX, Tang PY, Tee SF. 8-Hydroxy-2'-Deoxyguanosine and reactive oxygen species as biomarkers of oxidative stress in mental illnesses: a meta-analysis. Psychiatry Investig. 2021;18:603–18.
4. Arat Çelik HE, Tuna G, Ceylan D, Küçükgöncü S. A comparative meta-analysis of peripheral 8-hydroxy-2'-deoxyguanosine (8-OHdG) or 8-oxo-7,8-dihydro-2'-deoxyguanosine (8-oxo-dG) levels across mood episodes in bipolar disorder. Psychoneuroendocrinology. 2023;151:106078.
5. Atamna H, Cheung I, Ames BN. A method for detecting abasic sites in living cells: age-dependent changes in base excision repair. Proc Natl Acad Sci USA. 2000;97:686–91.
6. Coppedè F, Migliore L. DNA repair in premature aging disorders and neurodegeneration. Curr Aging Sci. 2010;3:3–19.
7. Berk M, Kapczinski F, Andreazza AC, Dean OM, Giorlando F, Maes M, et al. Pathways underlying neuroprogression in bipolar disorder: focus on inflammation, oxidative stress and neurotrophic factors. Neurosci Biobehav Rev. 2011;35:804–17.
8. McGorry P, Keshavan M, Goldstone S, Amminger P, Allott K, Berk M, et al. Biomarkers and clinical staging in psychiatry. World Psychiatry. 2014;13:211–23.
9. Rizzo LB, Costa LG, Mansur RB, Swardfager W, Belangero SI, Grassi-Oliveira R, et al. The theory of bipolar disorder as an illness of accelerated aging: implications for clinical care and research. Neurosci Biobehav Rev. 2014;42:157–69.
10. Özerdem A, Ceylan D. Chapter 20 - DNA damage and repair mechanisms in bipolar disorder. In: Machado-Vieira R, Soares JC, editors. Biomarkers in Bipolar Disorders. Academic Press; 2022. p. 361–83.
11. Dizdaroglu M, Jaruga P, Birincioglu M, Rodriguez H. Free radical-induced damage to DNA: mechanisms and measurement. Free Radic Biol Med. 2002;32:1102–15.
12. Dizdaroglu M. Oxidatively induced DNA damage: mechanisms, repair and disease. Cancer Lett. 2012;327:26–47.
13. Steenken S, Telo J, Novais H, Candeias L. One-electron-reduction potentials of pyrimidine bases, nucleosides, and nucleotides in aqueous solution. Consequences for DNA redox chemistry. J Am Chem Soc. 1992;114:4701–9.
14. Steenken S, Jovanovic SV. How easily oxidizable is DNA? One-electron reduction potentials of adenosine and guanosine radicals in aqueous solution. J Am Chem Soc. 1997;119:617–8.
15. Ceylan D, Scola G, Tunca Z, Isaacs-Trepanier C, Can G, Andreazza AC, et al. DNA redox modulations and global DNA methylation in bipolar disorder: effects of sex, smoking and illness state. Psychiatry Res. 2018;261:589–96.
16. Huzayyin AA, Andreazza AC, Turecki G, Cruceanu C, Rouleau GA, Alda M, et al. Decreased global methylation in patients with bipolar disorder who respond to lithium. Int J Neuropsychopharmacol. 2014;17:561–9.
17. Jacoby AS, Vinberg M, Poulsen HE, Kessing LV, Munkholm K. Increased DNA and RNA damage by oxidation in patients with bipolar I disorder. Transl Psychiatry. 2016;6:e867.
18. Munkholm K, Poulsen HE, Kessing LV, Vinberg M. Elevated levels of urinary markers of oxidatively generated DNA and RNA damage in bipolar disorder. Bipolar Disord. 2015;17:257–68.
19. Ermis B, Sagaltici E, Unal A, Alici D, Ozyurt AB. Oxidative metabolism and oxidative DNA damage in bipolar disorder: there are no difference in acute and Euthymic state. Psychiatry Behav Sci. 2021;11:249.
20. Lin C, Jin Y, Cheng S, Wang W. Association between APE1 ASP148GLU and colorectal cancer risk: a meta-analysis. Clin Investig Med. 2020;43:E24–34.
21. Wang Z, Gan L, Nie W, Geng Y. The OGG1 Ser326Cys polymorphism and the risk of esophageal cancer: a meta-analysis. Genet Test Mol Biomark. 2013;17:780–5.
22. Kumagae Y, Hirahashi M, Takizawa K, Yamamoto H, Gushima M, Esaki M, et al. Overexpression of MTH1 and OGG1 proteins in ulcerative colitis-associated carcinogenesis. Oncol Lett. 2018;16:1765–76.
23. Sampath H, Vartanian V, Rollins MR, Sakumi K, Nakabeppu Y, Lloyd RS. 8-Oxoguanine DNA glycosylase (OGG1) deficiency increases susceptibility to obesity and metabolic dysfunction. PLoS One 2012;7:e51697.

24. Kucuker MU, Ozerdem A, Ceylan D, Cabello-Arreola A, Ho AMC, Joseph B, et al. The role of base excision repair in major depressive disorder and bipolar disorder. J Affect Disord. 2022;306:288–300.

25. Ceylan D, Tuna G, Kirkali G, Tunca Z, Can G, Arat HE, et al. Oxidatively-induced DNA damage and base excision repair in euthymic patients with bipolar disorder. DNA Repair. 2018;65:64–72.

26. Munkholm K, Peijs L, Vinberg M, Kessing LV. A composite peripheral blood gene expression measure as a potential diagnostic biomarker in bipolar disorder. Transl Psychiatry. 2015;5:e614.

27. Ceylan D, Yılmaz S, Tuna G, Kant M, Er A, Ildız A, et al. Alterations in levels of 8-Oxo-2'-deoxyguanosine and 8-Oxoguanine DNA glycosylase 1 during a current episode and after remission in unipolar and bipolar depression. Psychoneuroendocrinology. 2020;114:104600.

28. Cai L, Fu Y, Zhang Y. APE1 Asp148Glu polymorphism and lung cancer susceptibility. Tumour Biol. 2014;35:5237–44.

29. Czarny P, Kwiatkowski D, Toma M, Kubiak J, Sliwinska A, Talarowska M, et al. Impact of single nucleotide polymorphisms of base excision repair genes on DNA damage and efficiency of DNA repair in recurrent depression disorder. Mol Neurobiol. 2017;54:4150–9.

30. Hoeijmakers JH. Genome maintenance mechanisms for preventing cancer. Nature 2001;411:366–74.

31. Howes TR, Tomkinson AE. DNA ligase I, the replicative DNA ligase. Subcell Biochem. 2012;62:327–41.

32. Lillenes MS, Støen M, Gómez-Muñoz M, Torp R, Günther CC, Nilsson LN, et al. Transient OGG1, APE1, PARP1 and Polβ expression in an Alzheimer's disease mouse model. Mech Ageing Dev. 2013;134:467–77.

33. Sliwinska A, Sitarek P, Toma M, Czarny P, Synowiec E, Krupa R, et al. Decreased expression level of BER genes in Alzheimer's disease patients is not derivative of their DNA methylation status. Prog Neuropsychopharmacol Biol Psychiatry. 2017;79:311–6.

34. Ahmadimanesh M, Abbaszadegan MR, Morshedi Rad D, Moallem SA, Mohammadpour AH, Ghahremani MH, et al. Effects of selective serotonin reuptake inhibitors on DNA damage in patients with depression. J Psychopharmacol. 2019;33:1364–76.

35. Almeida KH, Sobol RW. A unified view of base excision repair: lesion-dependent protein complexes regulated by post-translational modification. DNA Repair. 2007;6:695–711.

36. Fotiadou P, Henegariu O, Sweasy JB. DNA polymerase beta interacts with TRF2 and induces telomere dysfunction in a murine mammary cell line. Cancer Res. 2004;64:3830–7.

37. Kidane D, Jonason AS, Gorton TS, Mihaylov I, Pan J, Keeney S, et al. DNA polymerase beta is critical for mouse meiotic synapsis. EMBO J. 2010;29:410–23.

38. Ray S, Breuer G, DeVeaux M, Zelterman D, Bindra R, Sweasy JB. DNA polymerase beta participates in DNA End-joining. Nucleic Acids Res. 2018;46:242–55.

39. Horton JK, Srivastava DK, Zmudzka BZ, Wilson SH. Strategic down-regulation of DNA polymerase beta by antisense RNA sensitizes mammalian cells to specific DNA damaging agents. Nucleic Acids Res. 1995;23:3810–5.

40. Coello K, Bøgh HL, Stanislaus S, Kjærstad HL, Melbye SA, Ormstrup Sletved KS, et al. Higher systemic oxidatively generated DNA and RNA damage in patients with newly diagnosed bipolar disorder and their unaffected first-degree relatives. Free Radic Biol Med. 2021;168:226–33.

41. Ottesen NM, Meluken I, Frikke-Schmidt R, Plomgaard P, Scheike T, Fernandes BS, et al. Are remitted affective disorders and familial risk of affective disorders associated with metabolic syndrome, inflammation and oxidative stress? - a monozygotic twin study. Psychol Med. 2020;50:1736–45.

42. Kant M, Akış M, Çalan M, Arkan T, Bayraktar F, Dizdaroglu M, et al. Elevated urinary levels of 8-oxo-2'-deoxyguanosine, (5'R)- and (5'S)-8,5'-cyclo-2'-deoxyadenosines, and 8-iso-prostaglandin F(2α) as potential biomarkers of oxidative stress in patients with prediabetes. DNA Repair. 2016;48:1–7.

43. Tuna G, Bekar NED, İşlekel S, İşlekel GH. Urinary 8-hydroxy-2'-deoxyguanosine levels are elevated in patients with IDH1-wildtype glioblastoma and are associated with tumor recurrence in gliomas. DNA Repair. 2023;124:103463.

44. Bustin SA, Benes V, Garson JA, Hellemans J, Huggett J, Kubista M, et al. The MIQE guidelines: minimum information for publication of quantitative real-time PCR experiments. Clin Chem. 2009;55:611–22.

45. Black CN, Bot M, Scheffer PG, Cuijpers P, Penninx BW. Is depression associated with increased oxidative stress? A systematic review and meta-analysis. Psychoneuroendocrinology. 2015;51:164–75.

46. Jorgensen A, Köhler-Forsberg K, Henriksen T, Weimann A, Brandslund I, Ellervik C, et al. Systemic DNA and RNA damage from oxidation after serotonergic treatment of unipolar depression. Transl Psychiatry. 2022;12:204.

47. Włodarczyk M, Jabłonowska-Lietz B, Olejarz W, Nowicka G. Anthropometric and dietary factors as predictors of DNA damage in obese women. Nutrients. 2018;10:578.

48. Alano CC, Kauppinen TM, Valls AV, Swanson RA. Minocycline inhibits poly(ADP-ribose) polymerase-1 at nanomolar concentrations. Proc Natl Acad Sci USA. 2006;103:9685–90.

49. Rosenblat JD, McIntyre RS. Efficacy and tolerability of minocycline for depression: a systematic review and meta-analysis of clinical trials. J Affect Disord. 2018;227:219–25.

50. Ordway GA, Szebeni A, Hernandez LJ, Crawford JD, Szebeni K, Chandley MJ, et al. Antidepressant-Like Actions of Inhibitors of Poly(ADP-Ribose) polymerase in rodent models. Int J Neuropsychopharmacol. 2017;20:994–1004.

51. Steardo L Jr., Manchia M, Carpiniello B, Pisanu C, Steardo L, Squassina A. Clinical, genetic, and brain imaging predictors of risk for bipolar disorder in high-risk individuals. Expert Rev Mol Diagn. 2020;20:327–33.

52. Bora E, Özerdem A. A meta-analysis of neurocognition in youth with familial high risk for bipolar disorder. Eur Psychiatry. 2017;44:17–23.

53. Johnsen LK, Ver Loren van Themaat AH, Larsen KM, Burton BK, Baaré WFC, Madsen KS, et al. Alterations in Task-related brain activation in children, adolescents and young adults at familial high-risk for schizophrenia or bipolar disorder - a systematic review. Front Psychiatry. 2020;11:632.

54. Miskowiak KW, Kjærstad HL, Meluken I, Petersen JZ, Maciel BR, Köhler CA, et al. The search for neuroimaging and cognitive endophenotypes: a critical systematic review of studies involving unaffected first-degree relatives of individuals with bipolar disorder. Neurosci Biobehav Rev. 2017;73:1–22.

55. Mikolas P, Bröckel K, Vogelbacher C, Müller DK, Marxen M, Berndt C, et al. Individuals at increased risk for development of bipolar disorder display structural alterations similar to people with manifest disease. Transl Psychiatry. 2021;11:485.

56. Hu R, Stavish C, Leibenluft E, Linke JO. White matter microstructure in individuals with and at risk for bipolar disorder: evidence for an endophenotype from a Voxel-Based meta-analysis. Biol Psychiatry Cogn Neurosci Neuroimaging. 2020;5:1104–13.

57. Duffy A, Horrocks J, Doucette S, Keown-Stoneman C, Grof P, Andreazza A, et al. Immunological and neurotrophic markers of risk status and illness development in high-risk youth: understanding the neurobiological underpinnings of bipolar disorder. Int J Bipolar Disord. 2014;2:29.

58. Guglielmo R, Miskowiak KW, Hasler G. Evaluating endophenotypes for bipolar disorder. Int J Bipolar Disord. 2021;9:17.

59. Craddock N, Forty L. Genetics of affective (mood) disorders. Eur J Hum Genet. 2006;14:660–8.

60. Poulsen HE, Nadal LL, Broedbaek K, Nielsen PE, Weimann A. Detection and interpretation of 8-oxodG and 8-oxoGua in urine, plasma and cerebrospinal fluid. Biochim Biophys Acta. 2014;1840:801–8.

61. Wu LL, Chiou CC, Chang PY, Wu JT. Urinary 8-OHdG: a marker of oxidative stress to DNA and a risk factor for cancer, atherosclerosis and diabetics. Clin Chim Acta. 2004;339:1–9.

62. Sajatovic M, Strejilevich SA, Gildengers AG, Dols A, Al Jurdi RK, Forester BP, et al. A report on older-age bipolar disorder from the International Society for Bipolar Disorders Task Force. Bipolar Disord. 2015;17:689–704.

63. Radak Z, Zhao Z, Goto S, Koltai E. Age-associated neurodegeneration and oxidative damage to lipids, proteins and DNA. Mol Asp Med. 2011;32:305–15.

64. Liguori I, Russo G, Curcio F, Bulli G, Aran L, Della-Morte D, et al. Oxidative stress, aging, and diseases. Clin Inter Aging. 2018;13:757–72.

65. Kurhan F, Alp HH. Dynamic Thiol/Disulfide homeostasis and oxidative DNA damage in adult attention deficit hyperactivity disorder. Clin Psychopharmacol Neurosci. 2021;19:731–8.

66. Christensen MR, Poulsen HE, Henriksen T, Weimann A, Ellervik C, Lynnerup N, et al. Elevated levels of 8-oxoGuo and 8-oxodG in individuals with severe mental illness - An autopsy-based study. Free Radic Biol Med. 2018;126:372–8.

67. Knorr U, Simonsen AH, Roos P, Weimann A, Henriksen T, Christensen EM, et al. Cerebrospinal fluid oxidative stress metabolites in patients with bipolar disorder and healthy controls: a longitudinal case-control study. Transl Psychiatry. 2019;9:325.

AUTHOR CONTRIBUTIONS

Interviews with the participants were conducted by HEAÇ, BKK, EÇ, NY, and BTÖ. The laboratory analyses were conducted by HEAÇ, SY, İCA, NEDB, ÖFÇ, Hİ, PA, DC, and GT. Statistical analyses were performed by HEAÇ and DC. The conceptualization of the study was undertaken by HEAÇ, AÖ, PA, DC, and GT. The manuscript was revised by HEAÇ, SY, İCA, BKK, EÇ, NEDB, ÖFÇ, NY, BTÖ, Hİ, AÖ, PA, DC, and GT.

COMPETING INTERESTS

The authors declare no competing interests.

ADDITIONAL INFORMATION

Correspondence and requests for materials should be addressed to Hidayet Ece Arat Çelik or Deniz Ceylan.

Reprints and permission information is available at http://www.nature.com/reprints