scientific reports

Check for updates

OPEN

Up-regulation of MAPK14-related lncRNAs in the circulation of migraineurs

Fatemeh Ensafi Talemi[1], Bashdar Mahmud Hussen[2,3], Fatemeh Keshmir[4], Abolfazl Movafagh[1], Parisa Sarabi[4], Mohammad Rahmanian[5], Arezou Sayad[1]✉ & Soudeh Ghafouri-Fard[1]✉

Mitogen-activated protein kinase 14 (MAPK14) has a fundamental role in the development of different inflammatory and neurodegenerative disorders. However, its roles in the pathoetiology of migraine are not clear. The current case-control study focused on expression analysis of MAPK14 and related long non-coding RNAs (lncRNAs) in the circulation of migraineurs compared with healthy controls. Data showed remarkable elevation of expression levels of MAPK14, HLA Complex Group 11 (HCG11), zinc ribbon domain-containing 1-antisense 1 (ZNRD1-AS1), RAD51 antisense RNA 1 (RAD51-AS1) and long noncoding RNA-activated by DNA damage (NORAD) in both groups of migraineurs (with aura and without aura) compared with controls. The accuracy of expression levels of MAPK14, HCG11, ZNRD1-AS1, RAD51-AS1 and NORAD for differentiating migraineurs from controls was 85.71%, 81.56%, 85.11%, 77.8% and 94.33%, respectively. Thus, MAPK14 and its related lncRNAs are putative markers for migraine and might participate in the pathogenesis of this neurologic condition.

Keywords Migraine, MAPK14, LncRNA

Migraine has been regarded to be the second cause of global disability and the highest reason for disability-adjusted life years among young women in the world[1,2]. It has been estimated that there is approximately 1.1 billion cases of migraine across the world, with an increase of about 40.1% in incidence compared to 1990 [3]. Migraine is typically defined as episodes of unilateral moderate-to-severe headaches and is often associated with sound and light sensitivity and nausea[4]. While transient neurologic symptoms, known as aura, can accompany the headache; the most frequent form of migraine is the one without aura in both adults and children[5]. Migraine is a complex multifactorial disorder. According to the results of twin and family studies, the estimated heritability of this disease has been reported to be 30–60% [6,7]. Genetic predisposition, combined with various external and internal factors, underlie both heterogenicity and susceptibility to migraine disease[8,9]. In fact, the strong genetic background has been identified in the migraine that explains both monogenic and polygenic forms[9]. It has been estimated that various genes partake in the pathogenesis of the condition[10]. Due to the high burden of migraine and the important role of genetics in the disorder, investigating the genes and pathways playing a role in its pathogenesis is essential for effective targeted therapies[11–13]. A recent genome-wide analysis of migraine-associated variants has revealed enrichment of these genomic annotations in both vascular and central nervous system tissue/cell types, clearly indicating the involvement of neurovascular mechanisms in the migraine pathophysiology[14].

The Mitogen-activated protein kinase (MAPK) signaling pathways have been suggested to exert important roles in the pathogenesis of migraine[15]. Various cellular functions, including inflammation, cellular stress and neuronal signaling responses are influenced by these pathways[16]. The MAPK pathways are also implicated in central and peripheral sensitization, neuronal plasticity, pain hypersensitivity, and inflammatory responses correlated with migraine[17]. MAPK14 encodes p38α MAPK. P38α MAPK has been originally introduced as a kind of tyrosine phosphorylated protein identified in activated macrophages with an important role in inducing inflammatory cytokine, including tumor necrosis factor α (TNFα)[18]. However, beyond immune system, p38α MAPK mediated kinase activity has been correlated with several other organs[19]. More recently, MAPK and PI3K/

[1]Department of Medical Genetics, School of Medicine, Shahid Beheshti University of Medical Sciences, Tehran, Iran. [2]Department of Clinical Analysis, College of Pharmacy, Hawler Medical University, Erbil, Kurdistan Region, Iraq. [3]Department of Biomedical Sciences, College of Science, Cihan University-Erbil, Erbil, Kurdistan Region, Iraq. [4]Phytochemistry Research Centre, Shahid Beheshti University of Medical Sciences, Tehran, Iran. [5]School of Medicine, Shahid Beheshti University of Medical Sciences, Tehran, Iran. ✉email: Ar.sayad@yahoo.com; s.ghafourifard@sbmu.ac.ir

Akt signaling pathway-related gene polymorphisms have been found to be associated with migraine[20]. While, recent studies have emphasized on the role of long non-coding RNAs (lncRNAs) as epigenetic modulators and contributors to the etiology of migraine[21], the role of MAPK14-related lncRNAs in the migraine is not clear.

In this study, focusing on the MAPK pathways, we want to explore the relationship between MAPK14 and its related lncRNAs, namely HLA Complex Group 11 (HCG11), zinc ribbon domain-containing 1-antisense 1 (ZNRD1-AS1), RAD51 antisense RNA 1 (RAD51-AS1) and long noncoding RNA-activated by DNA damage (NORAD) and migraine for the first time.

Materials and methods

Migraineurs and controls

Fifty-one adult migraineurs with aura (9 males and 42 females) and 40 migraineurs without aura (6 males and 32 females) entered the study. The diagnosis was performed by a neurologist based on the International Classification of Headache Disorders[22]. In addition, 50 healthy persons were enrolled as normal control group. Cases and controls were matched in terms of sex. Controls had no history of systemic or neurodegenerative disorders or chronic headache in themselves or their family. Blood samples were obtained from all cases and controls at the same time during the day. At the time of sampling, cases were without headache for at least 24 h. The clinical data of cases and controls was obtained at the time of referral to the neurologist. Ethical committee of Shahid Beheshti University of Medical Sciences permitted the study procedure. All individuals signed informed written consent forms.

Experiments

Five milliliters of total blood were collected from enrolled individuals and stored in -70 °C. The quality and quantity of RNA were confirmed using the spectrophotometer. A260/A280 ratios of RNA samples were in the range of 1.9-2. Moreover, the 28 S and 18 S RNA bands were observed on the agarose gel. cDNA was synthesized using the obtained total RNA. Around 75 ng of RNA was used for this purpose. RNA extraction and cDNA production steps were done using SMOBIO and RiboEx kits, respectively. Expression of MAPK14, HCG11, ZNRD1-AS1, RAD51-AS1 and NORAD was measured by ABI real time PCR machine. Melting curve analyses were done to confirm the specificity of primers. *B2M* gene was used as normalizer housekeeping gene. The Ct values of this gene were not significantly different between cases and controls. Each PCR run included a negative control sample comprising all PCR reagents except for cDNA template. Primers were designed using Primer3 tool. The potential targets were checked using primer BLAST tool. All primers were designed in a way that the amplicon covers at least one exon-intron boundary to avoid amplification of possible DNA contamination. Primer sequences were the same as our previous study[23].

Statistical analysis

The statistical analysis was conducted to evaluate differences between groups concerning variables such as age, gender, age of onset, and disease duration, and to determine significant differences among these groups through appropriate statistical methods. Age differences among the three groups (healthy controls, migraineurs with aura, and migraineurs without aura) were analyzed using a one-way ANOVA. Gender differences were assessed with the Chi-square test. For the two migraine subgroups, differences in age of onset and disease duration were analyzed using independent t-tests. The normality of continuous variables was evaluated using the Shapiro-Wilk test.

Expression levels of MAPK14 and four lncRNA genes (HCG11, ZNRD1-AS1, RAD51-AS1, and NORAD) were compared between migraine patients (51 with aura and 40 without aura) and 50 controls using the comparative –delta Ct method. The normality of the –delta Ct values was evaluated using the Shapiro-Wilk test, which indicated that the data did not follow a normal distribution. Given that parametric tests, such as ANOVA, assume normality, we opted for a non-parametric approach. Accordingly, the Kruskal-Wallis test was employed to assess overall differences in gene expression among the three groups, as it is well-suited for comparing multiple independent groups when the assumption of normality is violated. To further explore pairwise differences while controlling for multiple comparisons, Dunn's post hoc test was applied with adjusted p-values. This statistical strategy ensures robust inference without relying on the assumptions required by parametric methods. Multivariate linear regression was conducted to measure the impact of disease status (with aura vs. without aura), gender, and age on gene expression levels. The influence of these variables on each gene was evaluated using the regression coefficient (β), standard error, and equivalent p-values. Receiver operating characteristic (ROC) curves were generated to determine optimal -delta Ct cut-off value for each gene, ensuring robust differentiation between migraine patients and healthy controls. The Youden index was applied to systematically identify threshold points that maximized both sensitivity and specificity. Additionally, area under the curve (AUC) and confidence intervals values were calculated to provide a more precise assessment of diagnostic performance. Sensitivity, specificity, accuracy, positive predictive value (PPV) and negative predictive value (NPV), were computed based on these refined cut-offs, enhancing the reliability and interpretability of the results. Spearman's rank correlation coefficient was used to evaluate correlations between gene expression level and different parameters, namely age, gender, age of onset, and disease duration, as the data were not normally distributed. Correlations between clinical characteristics themselves were also analyzed. The analysis was performed using SPSS software (version 20). Statistical significance was set at $p < 0.05$ for all tests. All other analyses, including ROC curve generation and additional statistical tests were conducted using GraphPad Prism version 9.0 (La Jolla, CA, USA).

Name/Gene ID	Accession number	Location	Official Full Name	Biological activity
MAPK14	NM_001315.3 , NM_139012.3, NM_139013.3, NM_139014.3	6p21.31	mitogen-activated protein kinase 14	protein coding
HCG11	NR_026790.1	6p22.2	HLA complex group 11	ncRNA
POLR1HASP (ZNRD1-AS1)	NR_026751.2, NR_145416.1 , NR_145417.1, NR_145418.1	6p22.1	POLR1H antisense, pseudogene	pseudogene
RAD51-AS1	NR_040058.1	15q15.1	RAD51 antisense RNA 1	ncRNA
NORAD	NR_027451.1	20q11.23	non-coding RNA activated by DNA damage	ncRNA

Table 1. Characteristic of MAPK14 and studied LncRNAs.

Characteristic	Controls ($N=50$)	Migraine with aura ($N=51$)	Migraine without aura ($N=40$)	P value
Age (Mean ± SD)	43.94 ± 14.01	38.89 ± 11.45	34.64 ± 11.99	**0.008**
Gender (N, %) -Male -Female	15 (30%) 35 (70%)	9 (17.64%) 42 (82.35%)	6 (12%) 32 (64%)	0.19
Age of Onset (Mean ± SD)	-	26.6 ± 9.99	25.97 ± 8.6	0.74
Disease Duration (Mean ± SD)	-	12.2 ± 9.5	9.2 ± 9.08	0.08

Table 2. General data of study participants (SD: standard deviation).

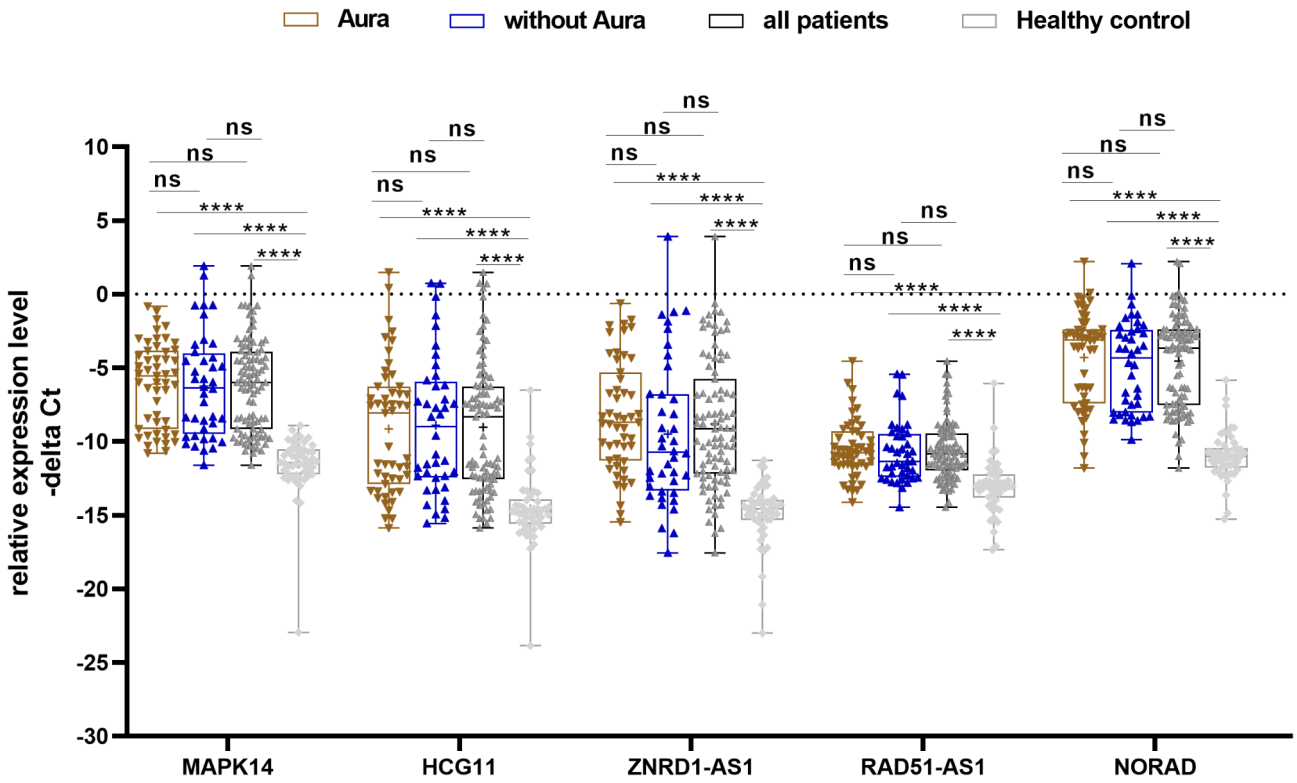

Fig. 1. Expression levels of MAPK14 gene and four lncRNA genes in total migraineurs, and two subclasses of cases and controls as defined by –delta Ct values (Ct B2M- Ct Target gene). – delta Ct values are demonstrated. Kruskal-Wallis test was used to detect differentially expression of genes between migraineurs and controls. Dunn's test was used for multiple comparisons and adjusted P values were shown (**** P value < 0.0001, ns: non-significant).

Results

Table 1 shows the characteristics of studied genes.

General data of patients and controls is shown in Table 2. All patients were Persians.

Significant differences were detected in the expressions of all studied genes between total cases and controls, as well as between both groups of patients and controls (Fig. 1). However, expression of none of genes was different between patients with and without aura.

Gene	Control Median (Interquartile Range)	Migraine with aura Median (Interquartile Range)	Migraine without aura Median (Interquartile Range)	*P*-value
MAPK14	-11.4 (-12.2) – (-10.5)	-5.5 (-9.1) – (-3.8)	-6.3 (-9.5) – (-3.9)	< 0.0001
HCG11	-14.7 (-15.5) – (-13.9)	-8.06 (-12.8) – (-6.2)	-8.9 (-12.4) – (-5.9)	< 0.0001
ZNRD1-AS1	-14.5 (-15.3) – (-13.9)	-8.7 (-11.3) – (-5.3)	-10.7 (-13.3) – (-6.8)	< 0.0001
RAD51-AS1	-13.03 (-13.8) – (-12.2)	-10.7 (-11.6) – (-9.3)	-11.3 (-12.4) – (-9.5)	< 0.0001
NORAD	-11.01 (-11.8) – (-10.4)	-3.1 (-7.4) – (-2.37)	-4.3 (-8.04) – (-2.4)	< 0.0001

Table 3. Relative expression of studied genes in healthy controls, migraineurs with Aura, and migraineurs without aura: data presented as median (Interquartile Range) based on -ΔCt values. Kruskal-Wallis test was used to detect differentially expressed genes.

Gene	Variable	Coefficient (β)	Standard Error	*P*-value
MAPK14	Age	0.003 (-0.04-0.04)	0.02	0.88
	Gender (Male/Female)	-0.48 (-1.84-0.87)	0.68	0.48
	Disease	2.88 (2.18–3.6)	0.35	< 0.0001
HCG11	Age	-0.002 (-0.06-0.05)	0.02	0.92
	Gender (Male/Female)	0.35 (-1.37-2.06)	0.86	0.69
	Disease	2.92 (2.03–3.8)	0.45	< 0.0001
ZNRD1-AS1	Age	0.02 (-0.03-0.08)	0.03	0.4
	Gender (Male/Female)	0.6 (-1.14-2.34)	0.88	0.5
	Disease	2.85 (1.94–3.76)	0.46	< 0.0001
RAD51-AS1	Age	0.004 (-0.02-0.03)	0.01	0.77
	Gender (Male/Female)	0.85 (0.001-1.7)	0.42	0.05
	Disease	1.21 (0.77–1.65)	0.22	< 0.0001
NORAD	Age	0.003 (-0.036-0.042)	0.02	0.89
	Gender (Male/Female)	0.8 (-0.43-2.03)	0.62	0.2
	Disease	3.12 (2.48–3.76)	0.32	< 0.0001

Table 4. Multivariate linear regression analysis of gene expression in migraine patients with and without Aura, and healthy controls: impact of disease status, gender, and age on gene expression levels.

Mean of –delta Ct values of genes were compared between three groups. The median values of relative expression of genes in controls were – 11.4, -14.7, -14.5, -13.03 and – 11.01 for MAPK14, HCG11, ZNRD1-AS1, RAD51-AS1 and NORAD, respectively. In both groups of patients, these values were significantly higher for all genes (P values < 0.0001 in all comparisons) (Table 3).

Then, we used multivariate linear regression analyses to measure the impact of disease status (with aura vs. without aura), gender, and age on expression levels of genes (Table 4). While disease could affect expression of all genes, age and gender did not affect expression of any of mentioned genes.

Correlation analyses revealed notable signatures in distinct groups of individuals (Table 5). For instance, correlation between MAPK14 and HCG11 was only significant among controls. However, MAPK14 expression was significantly correlated with ZNRD1-AS1 expression only among patients. Meanwhile, MAPK14 was correlated with both RAD51-AS1 and NORAD in all groups of cases and controls.

Subsequently, we assessed the diagnostic performance of mentioned genes in differentiating migraine patients from healthy controls (Table 6). The accuracy of expression levels of MAPK14, HCG11, ZNRD1-AS1, RAD51-AS1 and NORAD for this purpose was 85.71, 81.56, 85.11, 77.8 and 94.33, respectively. Moreover, the AUC values of these transcripts were 0.96, 0.88, 0.91, 0.85 and 0.96, respectively. NORAD had the highest sensitivity, PPV and NPV. Meanwhile, the highest specificity value belonged to MAPK14.

Figure 2. Combined ROC curves of five genes for distinguishing migraine patients (A: all migraine patients, B: migraine with aura, C: migraine without aura) from healthy controls. The area under the curve (AUC) for each gene is indicated on the respective curves.

We also classified patients and controls based on the –delta CT values to high and low expression groups (Table 7). In line with the former results, most of patients were grouped into the high expression subclasses for all genes, while the majority of controls were grouped into the low expression class.

Finally, we assessed correlation between expression levels of genes and clinical characteristics (Age, Gender, Age of Onset, Disease Duration). Expression of HCG11 was inversely correlated with age of onset of migraine (Correlation coefficient=-0.22). However, no other significant correlation was found between expression levels of genes and clinical characteristics.

	HCG11				ZNRD1-AS1				RAD51-AS1				NORAD			
	All patients	Migraine with aura	Migraine without aura	Controls	All patients	Migraine with aura	Migraine without aura	Controls	All patients	Migraine with aura	Migraine without aura	Controls	All patients	Migraine with aura	Migraine without aura	Controls
MAPK14	0.0009	0.02	-0.03	0.49**	0.48**	0.46**	0.49**	0.27	0.43**	0.44**	0.43**	0.33*	0.8**	0.79**	0.8**	0.59**
HCG11					0.26*	0.25	0.26	0.13	0.35**	0.32*	0.39*	0.5**	-0.02	-0.0005	-0.06	0.55**
ZNRD1-AS1									0.23*	0.24	0.21	0.27	0.5**	0.5**	0.5**	0.44**
RAD51-AS1													0.25*	0.27*	0.24	0.57**

Table 5. Spearman correlation analysis (r) of gene expression in 51 migraine patients with Aura, 40 without Aura, and 50 healthy controls, with significance levels indicated. * $p < 0.05$. ** $p < 0.001$.

Gene	ΔCt Cut-off	Sensitivity (%)	Specificity (%)	Accuracy (%)	PPV (Positive predictive value)	NPV (Negative predictive value)	AUC (Area Under Curve)
MAPK14	-9.62	81	96	85.71	96.23	73.44	0.96 (0.93–0.98)
HCG11	-13.34	81	84	81.56	89.04	70.69	0.88 (0.82–0.94)
ZNRD1-AS1	-12.84	82	90	85.11	93.75	73.77	0.91 (0.87–0.96)
RAD51-AS1	-11.8	75	86	77.8	89.47	64.62	0.85 (0.78–0.92)
NORAD	-8.86	94	94	94.33	96.7	90.38	0.96 (0.94–0.99)

Table 6. Diagnostic performance of -delta Ct cut-offs for gene expression in differentiating migraine patients from healthy controls. Sensitivity, specificity, accuracy, PPV, NPV and AUC values are shown based on ROC curve and Youden's index analysis (Ct: threshold cycle).

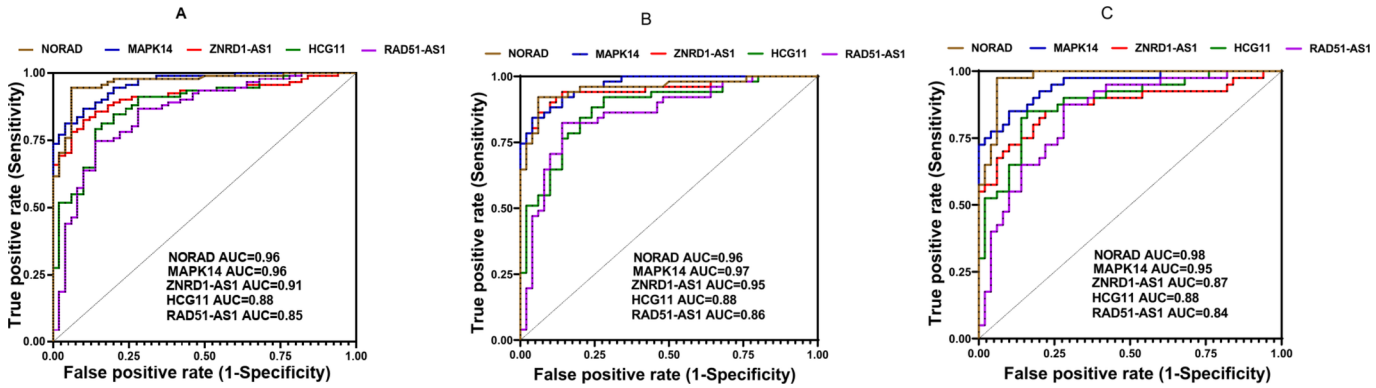

Fig. 2. depicts combined ROC curves of five genes for distinguishing migraine patients from healthy controls.

Groups	MAPK14 High	MAPK14 Low	HCG11 High	HCG11 Low	ZNRD1-AS1 High	ZNRD1-AS1 Low	RAD51-AS1 High	RAD51-AS1 Low	NORAD High	NORAD Low
Healthy individuals	3 (6%)	47 (94%)	9 (18%)	41 (82%)	5 (10%)	45 (90%)	8 (6%)	42 (84%)	3 (6%)	47 (84%)
Migraine with aura	43 (84.3%)	8 (15.7%)	40 (78.4%)	11 (21.6%)	46 (90.2%)	5 (9.8%)	42 (82.4%)	9 (17.6%)	47 (92.15%)	4 (7.84%)
Migraine without aura	31 (77.5%)	9 (22.5%)	34 (85%)	6 (15%)	29 (72.5%)	11 (27.5%)	26 (65%)	14 (35%)	39 (97.5%)	1 (2.5%)

Table 7. Classification of patients with Aura, without Aura, and controls into high and low gene expression groups according to -delta Ct cut-offs (N (%)).

Discussion

In the current study, we demonstrated remarkable over-expression of MAPK14 and its associated lncRNAs in the bloodstream of migraineurs compared with controls. MAPK14 belongs to a group of MAPKs, namely p38 kinase. These proteins are also called as cytokine-suppressive anti-inflammatory drug-binding proteins[24]. Typically, these proteins are activated by exposure to cellular stress. However, mitogens are not substantially involved in their activation. Instead, endotoxins, pro-inflammatory cytokines, osmotic shock, and heat stress strongly activate p38 kinases[24]. While the precise mechanism of migraine is not clear, studies point to contribution of endotoxins[25], pro-inflammatory cytokines[26], and heat in the induction of migraine attacks[27]. Moreover, dural afferents express osmo/mechano-sensitive channels that might contribute to the pathogenesis of migraine[28]. While evidence for reduction of plasma osmolarity prior to or during a migraine attack is not sufficient, it might be involved in the mechanical stimulation of the meninges in response to abrupt alterations in the intracranial pressure[29]. Therefore, MAPK14 might provide the mechanistical link between different cellular stresses and induction of migraine attack.

Notably, levels of a number of pro-inflammatory cytokines have been found to be increased outside migraine attacks compared to controls[26]. This finding is also in line with the fact that expression analyses in the current study were performed on blood samples of patients in headache-free periods.

Sex hormones have been found to affect induction of migraine[30]. In some cases, reduction in the estrogen levels induces a menstrual migraine attack without aura. However, elevation in the level of estrogen can induce an attack with aura in some other cases[31]. However, the gender of patients did not influence the level of MAPK14 and its related lncRNAs in our study. Thus, one can infer from these observations that the impact of MAPK14 in the pathophysiology of migraine is sex-independent.

Correlation analyses revealed notable signatures in distinct groups of patients and controls. For instance, correlation between MAPK14 and HCG11 was only significant among controls. However, MAPK14 expression

was significantly correlated with ZNRD1-AS1 expression only among patients. Meanwhile, MAPK14 was correlated with both RAD51-AS1 and NORAD in all groups of cases and controls. These findings might indicate the effect of migraine on the correlation network between MAPK14 and its associated lncRNAs.

Subsequently, we assessed the diagnostic performance of mentioned genes in differentiating migraine patients from healthy controls. Our results showed that NORAD level was a highly accurate biomarker for this purpose. The accuracy of expression levels of other genes was also beyond 80% except for RAD51-AS1. Moreover, MAPK14 and NORAD performance was excellent in terms of AUC values. Thus, these transcripts, particularly MAPK14 and NORAD might be used as biomarkers for migraine diagnosis outside the migraine attacks. However, since they were only compared with healthy controls, we cannot assert that they are specific biomarkers.

Finally, correlation analyses revealed inverse correlation between expression of HCG11 and age of onset of migraine. This miRNA has been shown to act as a competing endogenous RNA for miR-579 [32]. Notably, miR-579-3p is involved in the modulation of hypoxic ischemic encephalopathy through regulating TRAF6 expression[33]. Although the precise mechanism of contribution of HCG1 to the pathogenesis of migraine is unclear, the HCG11/miR-579 axis represents a possible candidate for further research in this field.

Taken together, MAPK14 and its related lncRNAs are putative markers for migraine and might participate in the pathogenesis of this neurologic condition. It is worth mentioning that the study did not reveal an obvious causal relationship between MAPK14-related lncRNAs and pathoetiology of migraine. From the genetic point of view, the specific mechanism of migraine is very complicated and needs to be further verified by experimental studies. Thus, we suggest conduction of further studies to elaborate the mechanistical points of participation of MAPK14 in the migraine etiology and its relation with cellular stressors that trigger migraine attack. We also state the restricted sample number and the limited population parameters as limitations of our study.

Data availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Received: 23 November 2024; Accepted: 24 February 2025
Published online: 28 February 2025

References

1. Steiner, T. J., Stovner, L. J., Jensen, R., Uluduz, D. & Katsarava, Z. Migraine remains second among the world's causes of disability, and first among young women: findings from GBD2019. *J. Headache Pain*. **21**, 137. https://doi.org/10.1186/s10194-020-01208-0 (2020).
2. Global burden of 369. diseases and injuries in 204 countries and territories, 1990–2019: a systematic analysis for the Global Burden of Disease Study 2019. *Lancet* 396, 1204–1222, (2020). https://doi.org/10.1016/s0140-6736(20)30925-9
3. Fan, L. et al. Global, regional, and National time trends in incidence for migraine, from 1990 to 2019: an age-period-cohort analysis for the GBD 2019. *J. Headache Pain*. **24**, 79. https://doi.org/10.1186/s10194-023-01619-9 (2023).
4. Amiri, P. et al. Migraine: A review on its history, global epidemiology, risk factors, and comorbidities. *Front. Neurol.* **12**, 800605. https://doi.org/10.3389/fneur.2021.800605 (2021).
5. Zalaquett, N. G. et al. The dawn and advancement of the knowledge of the genetics of migraine. *J. Clin. Med.* **13**, 2701 (2024).
6. Polderman, T. J. C. et al. Meta-analysis of the heritability of human traits based on Fifty years of twin studies. *Nat. Genet.* **47**, 702–709. https://doi.org/10.1038/ng.3285 (2015).
7. Bron, C., Sutherland, H. G. & Griffiths, L. R. Exploring the hereditary nature of migraine. *Neuropsychiatr. Dis. Treat.* **17**, 1183–1194. https://doi.org/10.2147/ndt.s282562 (2021).
8. Dodick, D. W. A Phase-by-Phase review of migraine pathophysiology. *Headache* **58** (Suppl 1), 4–16. https://doi.org/10.1111/head.13300 (2018).
9. Sutherland, H. G., Jenkins, B. & Griffiths, L. R. Genetics of migraine: complexity, implications, and potential clinical applications. *Lancet Neurol.* **23**, 429–446 (2024).
10. Grangeon, L. et al. Genetics of migraine: where are we now? *J. Headache Pain*. **24** https://doi.org/10.1186/s10194-023-01547-8 (2023).
11. Taheri, M. et al. Alteration in the expression of long non-coding RNAs in the circulation of migraineurs. *Acta Neurol. Belg.* **124**, 1295–1301. https://doi.org/10.1007/s13760-024-02513-0 (2024).
12. Ebahimzadeh, K., Gholipour, M., Samadian, M., Taheri, M. & Ghafouri-Fard, S. A comprehensive review on the role of genetic factors in the pathogenesis of migraine. *J. Mol. Neurosci.* **71**, 1987–2006. https://doi.org/10.1007/s12031-020-01788-1 (2021).
13. Ghafouri-Fard, S., Honarmand Tamizkar, K., Sayad, A., Taheri, M. & Samadian, M. Expression analysis of SOCS genes in migraine. *Front. Mol. Neurosci.* **14**, 725048. https://doi.org/10.3389/fnmol.2021.725048 (2021).
14. Hautakangas, H. et al. Genome-wide analysis of 102,084 migraine cases identifies 123 risk loci and subtype-specific risk alleles. *Nat. Genet.* **54**, 152–160. https://doi.org/10.1038/s41588-021-00990-0 (2022).
15. Haanes, K. A. & Edvinsson, L. Pathophysiological mechanisms in migraine and the identification of new therapeutic targets. *CNS Drugs*. **33**, 525–537. https://doi.org/10.1007/s40263-019-00630-6 (2019).
16. Cheng, H. et al. Caffeic acid phenethyl ester attenuates neuropathic pain by suppressing the p38/NF-κB signal pathway in microglia. *J. Pain Res.* **11**, 2709–2719. https://doi.org/10.2147/jpr.S166274 (2018).
17. Ji, R. R., Nackley, A., Huh, Y., Terrando, N. & Maixner, W. Neuroinflammation and central sensitization in chronic and widespread pain. *Anesthesiology* **129**, 343–366. https://doi.org/10.1097/aln.0000000000002130 (2018).
18. Beamer, E. & Corrêa, S. A. L. The p38(MAPK)-MK2 signaling Axis as a critical link between inflammation and synaptic transmission. *Front. Cell. Dev. Biol.* **9** (635636). https://doi.org/10.3389/fcell.2021.635636 (2021).
19. Canovas, B. & Nebreda, A. R. Diversity and versatility of p38 kinase signalling in health and disease. *Nat. Rev. Mol. Cell Biol.* **22**, 346–366. https://doi.org/10.1038/s41580-020-00322-w (2021).
20. Wang, M. et al. Association between MAPK and PI3K/Akt signaling pathway-related gene polymorphisms and migraine. *Mol. Genet. Genom. Med.* **12**, e2503. https://doi.org/10.1002/mgg3.2503 (2024).
21. dos Santos, L. C., Ratis, R. C., Piovesan, E. J. & Boldt, A. B. W. Epigenetic modulators of migraine: LncRNAs as possible new therapeutic targets. *Headache Med.*, 62–62 (2024).
22. Olesen, J. International classification of headache disorders. *Lancet Neurol.* **17**, 396–397 (2018).

23. Dashti, S. et al. In Silico identification of MAPK14-related LncRNAs and assessment of their expression in breast cancer samples. *Sci. Rep.* **10**, 8316. https://doi.org/10.1038/s41598-020-65421-2 (2020).
24. Madkour, M. M., Anbar, H. S. & El-Gamal, M. I. Current status and future prospects of p38α/MAPK14 kinase and its inhibitors. *Eur. J. Med. Chem.* **213**, 113216. https://doi.org/10.1016/j.ejmech.2021.113216 (2021).
25. Yu-Jie, D., Hai-Yan, W., Xi-Jian, W., Kaye, A. D. & Yong-Hai Potential beneficial effects of probiotics on human migraine headache: a literature review. *Pain Physician*. **20**, E251 (2017).
26. Thuraiaiyah, J., Erritzøe-Jervild, M., Al-Khazali, H. M., Schytz, H. W. & Younis, S. The role of cytokines in migraine: A systematic review. *Cephalalgia: Int. J. Headache*. **42**, 1565–1588. https://doi.org/10.1177/03331024221118924 (2022).
27. Schwedt, T. J., Krauss, M. J., Frey, K. & Gereau, R. W. t. Episodic and chronic migraineurs are hypersensitive to thermal stimuli between migraine attacks. *Cephalalgia: Int. J. Headache*. **31**, 6–12. https://doi.org/10.1177/0333102410365108 (2011).
28. Wei, X., Edelmayer, R. M., Yan, J. & Dussor, G. Activation of TRPV4 on dural afferents produces headache-related behavior in a preclinical rat model. *Cephalalgia: Int. J. Headache*. **31**, 1595–1600. https://doi.org/10.1177/0333102411427600 (2011).
29. Yan, J. & Dussor, G. Ion channels and migraine. *Headache* **54**, 619–639. https://doi.org/10.1111/head.12323 (2014).
30. Godley, F. 3 et al. How sex hormones affect migraine: an interdisciplinary preclinical research panel review. *J. Personalized Med.* **14** https://doi.org/10.3390/jpm14020184 (2024).
31. MacGregor, E. A. Migraine, menopause and hormone replacement therapy. *Post. Reproductive Health*. **24**, 11–18 (2018).
32. Wang, L., Zhou, J., Zhang, Y., Hu, T. & Sun, Y. Long non-coding RNA HCG11 aggravates osteosarcoma carcinogenesis via regulating the microRNA-579/MMP13 Axis. *Int. J. Gen. Med.* **13**, 1685–1695 (2020).
33. Wang, J. et al. Circ_0007706 downregulation ameliorates neonatal hypoxic ischemic encephalopathy via regulating the miR-579-3p/TRAF6 axis. *Brain Res. Bull.* **194**, 90–99. https://doi.org/10.1016/j.brainresbull.2023.01.013 (2023).

Acknowledgements
This study was financially supported by Shahid Beheshti University of Medical Sciences.

Author contributions
SGF and AS wrote the draft and revised it. BMH designed and supervised the study. AM and PS analyzed the data. FE, MR and FK performed the experiment and data collection. All the authors read and approved the submitted version.

Funding
Not applicable.

Declarations

Competing interests
The authors declare no competing interests.

Ethics approval and consent to participant
All procedures performed were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments. Informed consent forms were obtained from all study participants. The study protocol was approved by the ethical committee of Shahid Beheshti University of Medical Sciences.

Consent of publication
Not applicable.

Additional information
Correspondence and requests for materials should be addressed to A.S. or S.G.-F.