REPOSITIONING OF DRUGS FOR THE TREATMENT OF MAJOR DEPRESSIVE DISORDER BASED ON PREDICTION OF DRUG-INDUCED GENE EXPRESSION CHANGES

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Major depressive disorder (MDD) is one of the most common diseases affecting millions of people worldwide. The use of existing antidepressants in many cases does not allow achieving stable remission, probably due to insufficient understanding of pathological mechanisms. This indicates the need for the development of more effective drugs based on in-depth understanding of MDD's pathophysiology. Since the high costs and long duration of the development of new drugs, the drug repositions may be the promising alternative. In this study we have applied the recently developed DIGEP-Pred approach to identify drugs that induce changes in expression of genes associated with the etiopathogenesis of MDD, followed by identification of their potential MDD-related targets and molecular mechanisms of the antidepressive effects. The applied approach included the following steps. First, using structure-activity relationships (SARs) we predicted drug-induced gene expression changes for 3690 worldwide approved drugs. Disease enrichment analysis applied to the predicted genes allowed to identify drugs that significantly altered expression of known MDD-related genes. Second, potential drug targets, which are probable master regulators responsible for drug-induced gene expression changes, have been identified through the SAR-based prediction and network analysis. Only those drugs whose potential targets were clearly associated with MDD according to the published data, were selected for further analysis. Third, since potential new antidepressants must distribute into brain tissues, drugs with an oral route of administration were selected and their blood-brain barrier permeability was estimated using available experimental data and *in silico* predictions. As a result, we identified 19 drugs, which can be potentially repurposed for the MDD treatment. These drugs belong to various therapeutic categories, including adrenergic/dopaminergic agents, antiemetics, antihistamines, antitussives, and muscle relaxants. Many of these drugs have experimentally confirmed or predicted interactions with well-known MDD-related protein targets such as monoamine (serotonin, adrenaline, dopamine) and acetylcholine receptors and transporters as well as with less trivial targets including galanin receptor type 3 (GALR3), G-protein coupled estrogen receptor 1 (GPER1), tyrosine-protein kinase JAK3, serine/threonine-protein kinase ULK1. Importantly, that the most of 19 drugs act on two or more MDD-related targets, which may produce the stronger action on gene expression changes and achieve a potent therapeutic effect. Thus, the revealed 19 drugs may represent the promising candidates for the treatment of MDD.

Key words: drug repositioning; major depressive disorder; drug-induced gene expression; master regulators; signaling network; structure-activity relationships

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INTRODUCTION

Major depressive disorder (MDD) is responsible for a decrease in the quality of life of millions of people worldwide. According to the World Health Organization (WHO), depressive disorders are diagnosed in approximately 5% of adults [1], and with increasing age (especially in groups over 60 years old), the number of people with depression also growths. MDD is a third leading cause of disability worldwide [2] associated with 700000 suicides per year [1]. Despite the prevalence of depressive disorders, currently existing antidepressants are not effective enough and do not allow achieving complete remission in more than half of cases [3]. This indicates the need for the development of more effective drugs based on in-depth understanding of MDD's etiopathogenesis. However, the development of new drugs usually takes 10–15 years and requires nearly $2.5 billion. Drug repurposing approaches can speed up or side-step some phases of drug development, resulting in potentially faster and cheaper development programs [4–8]. Connectivity Map is one of the most widely used approaches for drug repurposing. It is used to identify drugs which induce gene expression profiles in cell lines that are reverse to those observed in disease tissues [9–12]. For example, using search for drugs that affect global gene expression in a similar manner to atypical antipsychotics Bortolasci et al. identified an ergot alkaloid metergoline to treat psychiatric disorders [9]. The one of the main limitations of the Connectivity Map approach is that

the experimental data on drug-induced gene expression profiles (DIGEPs) is absent for many existing drugs as well as for billions of organic compounds with drug-like properties. In 2013, we developed an *in silico* approach to predict DIGEPs based on structural formula of drug-like compounds using an analysis of structure-activity relationships (SARs) [13]. It was based on the published data on chemical-induced gene expression changes from the Comparative Toxicogenomics Database (CTD) [14] and SARs analysis performed by PASS software [15–17]. The approach allows predicting up- and down-regulation of particular human genes at mRNA and protein levels based on structural formula of drug-like compound. Recently, we have extended this approach with network analysis and implemented it in a freely available DIGEP-Pred 2.0 web application [18]. DIGEP-Pred 2.0 is based on significantly larger data on DIGEPs including data obtained in various cell lines. More importantly, we implemented two types of analysis that are based on the predicted genes. First, the enrichment analysis can be performed to identify pathways, biological processes, and diseases associated with the predicted expression changes. Second, in addition to prediction of gene expression changes, the structural formula of a compound is also used to predict its direct protein targets by using PASS. The network analysis is applied to reconstruct signaling and regulatory network connecting probable targets to probable genes. As a result, the user can obtain information on potential protein targets, which are responsible for drug-induced gene expression changes. All these results can be obtained using only the structural formula of a query compound.

In this study, we applied the DIGEP-Pred approach to identify drugs that induce changes in expression of genes associated with the etiopathogenesis of MDD, followed by identification of their potential targets and molecular mechanisms of antidepressive effects. The study included three main steps. First, we predicted DIGEPs for more than 3000 worldwide approved drugs. Disease enrichment analysis applied to the predicted genes allowed to identify drugs that significantly altered the expression of MDD-related genes retrieved from the literature. Second, potential drug targets, which are probable master regulators responsible for drug-induced gene expression changes, have been identified through PASS prediction and network analysis. Only those drugs whose potential targets were clearly associated with MDD in the literature, were selected for further analysis. Third, since potential new antidepressants must cause effect within brain tissues, blood-brain barrier (BBB) permeability was estimated for the selected drugs using available experimental data and *in silico* predictions. Moreover, only drugs with an oral route of administration were selected. As a result, we have identified 19 drugs which can be potentially repurposed for the treatment of MDD.

MATERIALS AND METHODS

World Wide Approved Drugs database

World Wide Approved Drugs database (WWAD) [19] was used to identify drugs that could be potentially repositioning for the treatment of MDD. The database contains information on 3776 pharmaceutical substances including structural formulas which have been approved at least in one of 71 countries and are not related to narcotic, psychotropic, and toxic compounds. To perform DIGEP prediction, we selected 3690 out of 3776 drug structures that satisfied the following conditions. First, the structure must contain at least three carbon atoms and have a molecular weight of less than 1500 Da. Second, each chemical bond in the molecule is a covalent single, double, or triple bond only. Metalloorganic and complex compounds have been excluded. Third, the total charge of the molecule is zero. Fourth, the compound has a single-component structure. These criteria are used in the PASS software and represent a standard that is widely used in creating training sets for SAR modeling and predicting the biological activity of new compounds [17].

Prediction of Drug-Induced Gene Expression Changes

The DIGEPs of 3690 drugs were predicted using SARs which were previously developed and implemented in the DIGEP-Pred 2.0 web application [18]. Briefly, SARs have been created using an information on DIGEPs at the mRNA level from the CTD database. Each compound from the CTD database was associated with a list of genes flagged with the direction of expression changes, up-regulation and down-regulation, for example, “ADGRB3 UpRegulation”. Structural formulas of compounds satisfying the above-mentioned criteria were downloaded from PubChem. The resulting training set contained 2620 structures. The corresponding SAR models for each gene and direction of its altered expression were created by the PASS software. PASS uses the Multilevel Neighborhoods of Atoms (MNA) molecular structure descriptors and a modified naive Bayes approach for simultaneous prediction of many types of biological activities, including changes in expression of particular genes. For example, compounds flagged with “ADGRB3 UpRegulation” activity were considered as “active” while other structures in the training set lacking the corresponding flag were considered as “inactive”. A binary classification SAR model was then created. The same SAR analysis was performed for all genes and direction of their altered expression presented in the training set. After the training, PASS was able to predict 18425 mRNA-related activities (up- or down-regulation of particular genes) corresponding to 13377 individual human genes with an average accuracy of 86.5% and a minimal accuracy of 75% calculated by leave-one-out

cross-validation [18]. PASS calculates two estimates of probabilities for each biological activity of a new chemical compound: Pa is a probability to be active, Pi is a probability to be inactive. If a compound has Pa > Pi, it can be considered active. The larger the Pa and Pa – Pi values are, the greater is the probability of obtaining the activity in the experiment. More information on DIGEP prediction and PASS software, could be found in our previous publications [15–18].

Disease Enrichment Analysis

Using the up- and down-regulated genes predicted with Pa values exceeding 0.5 we have performed enrichment analysis for each of 3690 drugs. The threshold of Pa > 0.5 was chosen to balance the sensitivity and specificity of prediction [17]. If we have chosen the high Pa threshold, chances to reveal experimental activity would be rather high, but the compounds thus found may be close structural analogues of compounds from the trainings set. This may result in little or no drugs identified for repositioning. In case of Pa > 0.5 as the threshold criterion, chances of detecting activity in the experiment are lower but the compounds will be less similar to the compounds in the training set [17]. This may lead to the identification of more drugs for repositioning, whereas master-regulators analysis taking into account experimentally confirmed and predicted drug targets (see below) allowed to avoid identification of many false positive results. The information on 265 genes related to MDD (see Supplementary Materials, Table S1) was obtained from a curated part of the DisGeNET database [20]. To date DisGeNET is the biggest database containing gene-disease associations. We used the DisGeNET version downloaded on January 26, 2024 and implemented in DIGEP-Pred web application. Enrichment analysis was performed for the combined lists of up- and down-regulated genes of each drug. To do this, the drug gene list was compared with 265 MDD-related genes and intersection between them was found. Next, the background list of 23335 human genes used in DIGEP-Pred was compared with 265 MDD-related genes and intersection between them was also found. Finally, the Fisher exact test was used to estimate the significance of differences between the intersection sizes for the predicted and background genes lists. This analysis allowed us to identify drugs with predicted gene lists that were "enriched" with known MDD-related genes. To perform further steps of analysis (see below), we selected 1460 drugs with enrichment *p*-values less than 0.05 and more than 50 predicted genes known to be associated with MDD.

Prediction of Direct Protein Targets and Molecular Mechanisms of Action

The data on known human protein targets of revealed 1460 drugs was obtained from DrugBank [21], DrugCentral [22], and WWAD [19] databases. These databases also provide information about molecular mechanisms of drug action, such as stimulator or inhibitor of protein function, for a part of targets. Additional human targets were predicted for 1460 drugs using SARs implemented in DIGEP-Pred 2.0 web application [18]. Briefly, to create SARs, information on ligand-protein interactions from PubChem and ChEMBL databases was used. The compounds were considered active if their K_i or IC_{50} values were less than 10 μM or if the percent of inhibition was greater than 50%. Data on molecular mechanisms of action such as agonist or antagonist was also taken into account, e.g., "Substance-P receptor antagonist", "Serine/threonine-protein kinase ULK1 activator" [18, 23]. As a result, a training set containing 656011 compounds was obtained. To perform SAR analysis, PASS software was used. After the training, PASS was able to predict 2170 mechanisms of action corresponding to 1940 individual human proteins with an average accuracy of 97.9% and a minimal accuracy of 80% calculated by leave-one-out cross validation.

Master-Regulator Analysis for the Identification of Drug Targets Responsible for Induced Gene Expression Changes

To identify protein targets of drugs which are responsible for drug-induced changes in gene expression, and, potentially, antidepressive effect, the network-based analysis was done. The analysis allowed to reconstruct the signaling regulatory network connecting known and predicted targets to predicted genes [18]. Briefly, network analysis is divided into two steps: (1) the transcription factor enrichment analysis is used to identify transcription factors potentially responsible for gene expression changes, and (2) "upstream" analysis is performed to connect revealed transcription factors with predicted protein targets. The data on transcription factor-gene interactions derived from the CollecTRI database [24] was used for enrichment analysis that was performed using Fisher exact test. Transcription factors with *p*-values less than 0.05 and associated with at least two genes were selected for "upstream" analysis in the signaling network obtained from the OmniPath database [25]. To connect the known and predicted drug targets with the enriched transcription factors, we calculated shortest paths, taking into account the direction of edges in the network. The score reflecting the probability that the action of a drug on a particular target is responsible for gene expression changes was calculated using the hyperbolic function [18]. The score represents a sum of inversed lengths of shortest paths from a target to the transcription factors. This function was chosen because the changes in cell signaling and gene expression caused by action of a drug on a target protein decrease with an increase in the distance between this protein and transcription factors [26, 27].

The significance of scores was calculated using the permutation test and protein targets with *p*-values less than 0.05 were selected. As a result, 1047 out of 1460 drugs with predicted targets — master regulators were selected for further analysis.

Estimation of Blood-Brain Barrier Permeability

The information on BBB permeability for 408 out of 1047 studied drugs was obtained from a large benchmark dataset, B3DB, complied from 50 published resources and categorized based on experimental uncertainty [28]. B3DB contains both qualitative and quantitative experimental data on BBB permeability for drugs. The drug was considered to penetrate through BBB if the log BB value was more than -1. Since many of studied compounds did not have corresponding experimental information, the BBB permeability was predicted using ADMETlab 3.0 [29] and pkCSM [30] web applications. ADMETlab 3.0 calculates probability that a compound penetrates BBB. According to recommendations of the authors, a compound with a probability greater than 0.7 is considered permeable through the BBB [29]. pkCSM calculates log BB values for a query compound. The authors recommend considering a compound as BBB-permeable, if the predicted log BB value was greater than -1 [30]. We considered a compound to penetrate the BBB if both tools predicted its permeability. We selected 325 out of 1047 drugs with positive experimental and predicted data on BBB permeability for further analysis.

Data on Routes of Administration and Drug Categories

Information on routes of administration and therapeutic categories of 325 drugs was obtained from the ATC/DDD Index 2024 and DrugBank databases. We selected only drugs with oral mode of administration for further analysis. The hormones and antineoplastic drugs were excluded because of large number of undesirable effects. We also excluded the nervous system drugs (see Supplementary Materials, Table S2) such as known antidepressants, analgesics, antiepileptics, antiparkinsonian, sedative drugs, since the most of them were studied for the antidepressive effect. Additionally, we selected the drugs whose targets — master regulators were clearly associated with MDD in the literature. As a result, we identified 19 drugs which can be potentially repurposed for the treatment of MDD.

RESULTS AND DISCUSSION

We applied the DIGEP-Pred approach to identify drugs that can be potentially repurposed for the treatment of MDD. We predicted DIGEPs for 3690 worldwide approved drugs and identified those which were significantly associated with expression changes of MDD-related genes. Network-based analysis of experimentally confirmed and predicted protein targets of drugs allowed identifying probable targets — master regulators that could be responsible for drug-induced gene expression changes. The role of many revealed drug targets in MDD pathophysiology was manually confirmed by literature analysis. We have selected only orally administrated drugs that are known or predicted to penetrate through BBB, to ensure that they may act within brain tissues. Among others we have identified 64 drugs which are used to treat nervous system disorders; these included 14 antidepressants (see Supplementary Materials, Table S2). These findings support the applicability of our approach to identify drugs for the treatment of MDD. Other nervous system drugs (analgesics, antiepileptics, antiparkinsonian, and sedative drugs) have also been previously studied in clinical and animal trials for their antidepressive effect. In the current study, we have focused on drugs whose indications are not related to nervous system disorders. As a result, we identified new 19 drugs with different indications that can be potentially repurposed for the treatment of MDD.

Table 1 contains information on 19 drugs including therapeutic categories, numbers of MDD-related genes whose expression was predicted to be changed by a drug, *p*-values from disease enrichment analysis, and data on protein targets predicted by network analysis as master regulators and potentially responsible for gene expression changes. The most of predicted drug-target interactions were supported by experimental data from DrugBank, DrugCentral, and WWAD databases. Most of interactions were also associated with information on molecular mechanism of action: stimulation or inhibition of protein targets' function, e.g., agonist or antagonist of receptor.

All targets — master regulators presented in Table 1 have known associations with MDD. Besides well-known MDD-related protein targets such as serotonin, adrenaline, dopamine, and muscarinic acetylcholine receptors, less trivial targets and related mechanisms of action have been found. These included galanin receptor type 3 (GALR3) antagonist [31, 32], G-protein coupled estrogen receptor 1 (GPER1) agonist [33–35], tyrosine-protein kinase JAK3 inhibitor [36, 37], serine/threonine-protein kinase ULK1 activator [38, 39]. Galanin is a neuropeptide involved in numerous functions such as nociception, cognition, feeding behavior, nerve regeneration, memory, neuroendocrine release, and addiction. It participates in a wide range of physiological and pathological conditions including epilepsy, chronic anxiety, depression, and pain. It is currently known that inhibition of galanin receptors 1 and 3 leads to attenuation of depressive symptoms. These receptors are promising targets for new antidepressants [31]. The G-protein coupled estrogen receptor 1 (GPER1) is a membrane receptor that plays a role in cognition, depression, homeostasis, pain, and other neurological processes. There is evidence that estrogen may accelerate the therapeutic effects of selective

Table 1. Information on 19 drugs that can be potentially repurposed for the treatment of major depressive disorder

Drug name	Drug category	Number of genes	-log10 (*p*-value)	Protein targets – master regulators
Droxidopa	Adrenergic/dopaminergic agents	97	1.69	ADRA1A-(A), ADRA1B-(A), ADRA1D-(A), ADRB3-(A)
Phenylephrine	Adrenergic/dopaminergic agents	151	3.45	ADRA1A-(A), ADRA1B-(A), ADRA1D-(A)
Methenamine	Antibacterials	173	3.66	JAK3-(I)*, TYK2-(I)*
Diphenidol	Antiemetics	170	4.01	CHRM2-(I), DRD2, DRD3, HTR2A, SIGMAR1[#]
Tropisetron	Antiemetics	159	4.46	HTR1A, HTR3-(I)[#], SLC6A4[#]
Isobromindione	Antigouts	125	3.71	GALR3-(I)*, GPER1-(A)*, HTR2B-(I)*
Cyclizine	Antihistamines	53	1.59	ADRA1D, ADRA2A, CHRM2, DRD3, HTR2A, HTR2B, HTR2C, MAOB[#], SLC6A3[#]
Cyproheptadine	Antihistamines	89	2.08	ADRA2A, ADRA2B, ADRA2C, CHRM2_I, DRD2, DRD3, HTR1A, HTR1D, HTR2A-(I), HTR2B-(I), HTR2C-(I), HTR6, HTR7-(I), SLC6A2[#], SLC6A4[#]
Diphenhydramine	Antihistamines	51	2.02	CHRM2-(I), HTR2A, HTR2B, HTR2C, HRH4, SLC6A2[#], SLC6A3[#], SLC6A4[#]
Diphenylpyraline	Antihistamines	95	1.96	SLC6A3-(I)[#]; DRD4-(I)*
Pimethixene	Antihistamines	80	2.78	CHRM2-(I), DRD2-(I), HTR1A-(I), HTR2A-(I), HTR2B-(I), HTR2C-(I), HTR6-(I), HTR7-(I)
Homarylamine	Antitussive — cough suppressants	97	2.22	GALR3-(I)*
Eprazinone	Antitussive — mucolytics	157	4.27	TACR1-(I), ULK1-(A)*
Propranolol	Beta blocking agents	95	2.16	ADRB3-(I), HTR1A-(I), HTR1B, HTR1D, HTR2A, HTR2B, HTR2C, HTR6, SIGMAR1[#], SLC6A4[#]
Lacidipine	Calcium channel blockers	81	1.86	DRD3, SLC6A2[#]
Cyclobenzaprine	Muscle relaxants	203	4.85	CHRM2, HTR2A-(I), HTR2B-(I), HTR2C-(I), HTR6-(I), HTR7-(I), SLC6A2-(I)[#], SLC6A4-(I)[#]
Pridinol	Muscle relaxants	172	4.28	CHRM2
Trospium	Urologicals — muscarinic antagonist	65	1.32	CHRM2-(I)
Dapoxetine	Urologicals — premature ejaculation treatment	81	4.79	HTR1A, HTR1B, HTR2C, SLC6A4-(I)[#]

Drug category reflects the main therapeutic indication of a drug. Number of genes represents the number of MDD-related genes, with predicted drug-included expression changes. -log10 (*p*-values) is negative decimal logarithm of *p*-values derived from a disease enrichment analysis. Protein targets – master regulators are protein targets of drugs predicted by network analysis as master regulators and potentially responsible for gene expression changes. The letters (A) and (I) mean that a drug activates or inhibits protein function, for example, a drug is an agonist or antagonist of particular receptor. Asterisk (*) shows that the target was predicted by PASS and was not confirmed in experiment. (#) shows that the target is known to be used for MDD treatment but it was not predicted by network analysis as a master regulator.

serotonin reuptake inhibitors through the GPER1 in the hypothalamic-pituitary system [33]. The JAK3 kinase is known to participate in MDD pathophysiology [36, 37]. The JAK3 gene transcription is increased in patients with depression compared to healthy controls [36]. It was shown that induction of stress in mice caused inhibition of neurogenesis and appearance of MDD symptoms. Application of JAK3 inhibitors normalized inhibition of neurogenesis and the anxious-depressive behavior [37]. The ULK1 kinase is a well-known regulator of authophagy. Chronic stress contributes to the development of depression and associates with reduced autophagy level; thus, ULK1 activators may potentially induce the antidepressive effect [38, 39].

The identified 19 drugs belong to a wide range of therapeutic categories including adrenergic/dopaminergic agents (2 drugs), antiemetics (2 drugs), antihistamines (5 drugs), antitussives (2 drugs), muscle relaxants (2 drugs). Many of these drugs have experimentally confirmed information on the interaction with monoamine (serotonin, adrenaline, dopamine) and acetylcholine receptors and transporters which were predicted as master regulators. Certain evidence exists in the literature that tropisetron [40–43], cyproheptadine [44], lacidipine [45], cyclobenzaprine [46], and dapoxetine [47], exibited the potential antidepressive effect in animal and clinical studies.

It is important that the majority of 19 drugs act on at least two MDD-related targets. We suggest that the more targets — master regulators affected by a drug, the stronger effect on gene expression should be observed. Figure 1 shows the signaling regulatory network describing mechanisms of potential therapeutic effect of Eprazinone in MDD. Eprazinone is a known antagonist of Substance P receptor that one of the perspective targets for the treatment of MDD [48, 49]. Eprazinone was also predicted by PASS as an activator of the ULK1 kinase. Both targets were predicted by network analysis as master regulators that could initiate signaling cascades leading to changes in activity of transcription factors. Figure 1 contains data on 22 most significant transcription factors revealed by enrichment analysis of predicted genes with *p*-values less than 0.05 (after Benjamini-Hochberg correction). In turn, Eprazinone predicted to change expression of 157 MDD-related genes (see Table 1) and 40 of them, which have known relationships with transcription factors in the CollecTRI database, are shown in Figure 1. The figure demonstrates that the action of Eprazinone

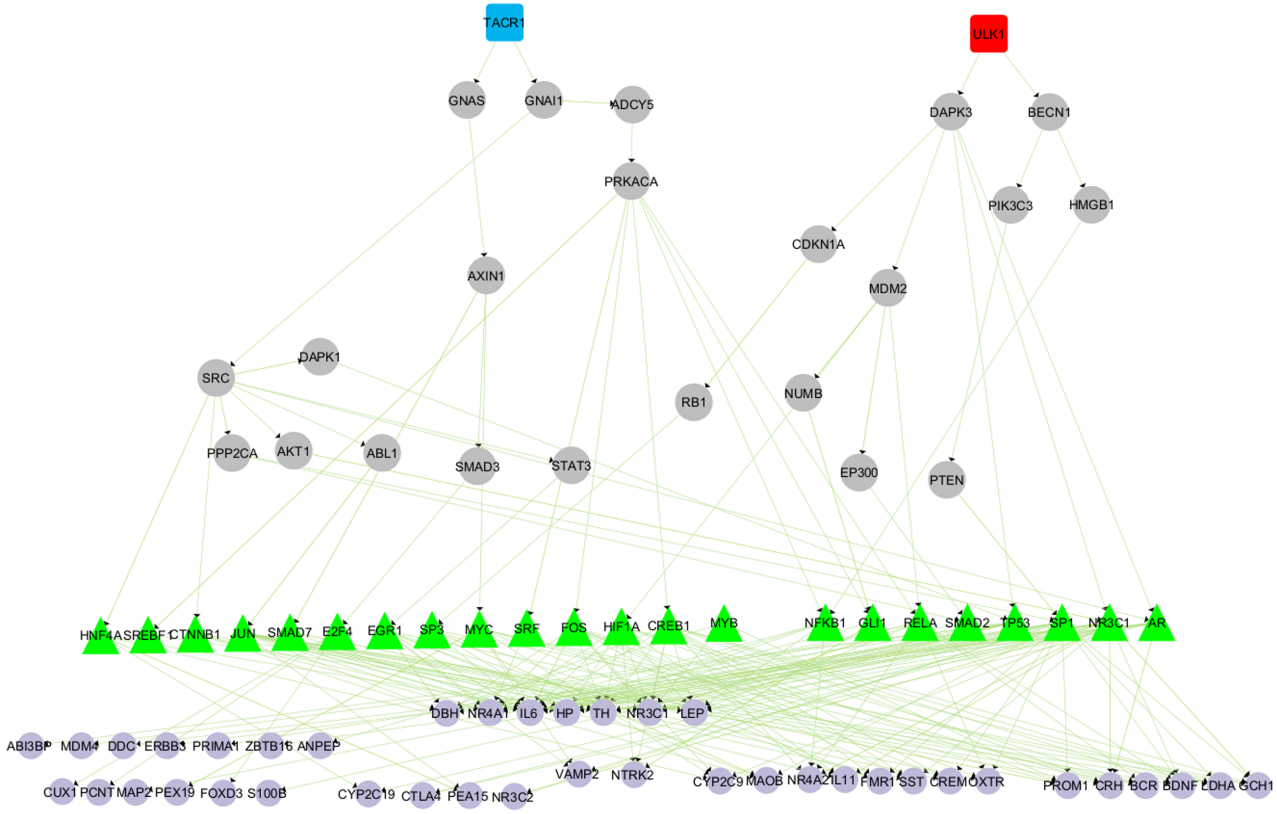

Figure 1. The signaling regulatory network describing mechanisms of potential therapeutic effect of Eprazinone on major depressive disorder. Rectangles correspond to known (TACR1) and predicted (ULK1) targets of Eprazinone. Red (blue) color of rectangle means that Eprazinone activates (inhibits) the corresponding target. Small purple circles correspond to genes whose expression was predicted to be changed by Eprazinone. Green triangles correspond to transcription factors potentially regulating expression of the genes. Grey circles correspond to intermediate proteins that link Eprazinone targets with transcription factors. The color version of the figure is available in the electronic version of the article.

on the Substance P receptor and ULK1 kinase initiates two largely separate signaling cascades and many of 22 transcription factors regulated by only one of them. The same relative specificity is true for the regulation of gene transcription by transcription factors. It potentially means that significant changes in expression of MDD-related genes may be only induced by action on several targets rather than one target. Recently, Arash Sadri proposed that the target-based drug discovery has been inefficient in the creation of new drugs for the treatment of complex diseases such as MDD. It has been shown that approved drugs mediate their therapeutic effects through numerous off-target mechanisms rather than a single target [50]. Thus, to achieve the desired therapeutic effect, multi-target drug action or the application of synergistic drug combinations are required. The effective drugs should cause permutation to a network of proteins rather than particular single targets. The proposed DIGEP-Pred approach is consistent with these principles of network pharmacology [51]. Thus, the revealed 19 drugs may represent the promising candidates for the treatment of MDD.

The proposed approach also has some limitations. Since it is based on SAR modeling, the corresponding predictions cannot be made for drugs that are significantly dissimilar to those in the training sets. The analysis also cannot be performed for drugs that do not alter gene expression and drugs that are not suitable for SAR analysis, such as inorganic, metalloorganic, and complex compounds, and macromolecules. Finally, like any other SAR-based method, the proposed approach does not allow to distinguish between drugs with similar structure but with different properties, including DIGEP.

CONCLUSIONS

We have demonstrated the usefulness of the DIGEP-Pred approach for the drug repositioning in the MDD case study. We identified 19 drugs, including adrenergic/dopaminergic agents, antiemetics, antihistamines, antitussives, and muscle relaxants, which can be potentially repurposed for the treatment of depression. Many of these drugs have experimentally confirmed or predicted interactions with monoamine (serotonin, adrenaline, dopamine) and acetylcholine receptors and transporters as well as with less trivial targets including galanin receptor type 3, G-protein coupled estrogen receptor 1, tyrosine-protein kinase JAK3, and serine/threonine-protein kinase ULK1. The most of 19 drugs act on two or more protein targets that may lead to significant changes in expression of genes participating in etiopathogenesis of major depressive disorder, and, in turn, cause pronounced therapeutic effect. Thus, the revealed 19 drugs may represent the promisimg candidates for the treatment of major depression.

COMPLIANCE WITH ETHICAL STANDARDS

This article does not contain any research involving humans or the use of animals as objects.

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CONFLICT OF INTEREST

The authors declare no conflicts of interest.

Supplementary materials are available in the electronic version at the journal site (pbmc.ibmc.msk.ru).

REFERENCES

1. WHO website. Retrieved October 9, 2024, from: https://www.who.int/ru/news-room/fact-sheets/detail/depression
2. *Malhi G.S., Mann J.J.* (2018) Depression. Lancet, **392**(10161), 2299–2312. DOI: 10.1016/S0140-6736(18)31948-2
3. *Caldiroli A., Capuzzi E., Tagliabue I., Capellazzi M., Marcatili M., Mucci F., Colmegna F., Clerici M., Buoli M., Dakanalis A.* (2021) Augmentative pharmacological strategies in treatment-resistant major depression: A comprehensive review. Int. J. Mol. Sci., **22**(23), 13070. DOI: 10.3390/ijms222313070
4. *di Masi J.A., Grabowski H.G., Hansen R.W.* (2016) Innovation in the pharmaceutical industry: New estimates of R&D costs. J. Health Econ., **47**, 20–33. DOI: 10.1016/j.jhealeco.2016.01.012
5. *Nosengo N.* (2016) Can you teach old drugs new tricks? Nature, **534**(7607), 314–316. DOI: 10.1038/534314a
6. *Mullard A.* (2012) Drug repurposing programmes get lift off. Nat. Rev. Drug Discov., **11**(7), 505–506. DOI: 10.1038/nrd3776
7. *Mullard A.* (2014) Bank tests drug development waters. Nat. Rev. Drug Discov., **13**(9), 643–644. DOI: 10.1038/nrd4412
8. *Roessler H.I., Knoers N.V.A.M., van Haelst M.M., van Haaften G.* (2021) Drug repurposing for rare diseases. Trends Pharmacol. Sci., **42**(4), 255–267. DOI: 10.1016/j.tips.2021.01.003
9. *Bortolasci C.C., Jaehne E.J., Hernández D., Spolding B., Connor T., Panizzutti B., Dean O.M., Crowley T.M., Yung A.R., Gray L., Kim J.H., van den Buuse M., Berk M., Walder K.* (2023) Metergoline shares properties with atypical antipsychotic drugs identified by gene expression signature screen. Neurotox. Res., **41**(6), 502–513. DOI: 10.1007/s12640-023-00673-0
10. *Lamb J., Crawford E.D., Peck D., Modell J.W., Blat I.C., Wrobel M.J., Lerner J., Brunet J.P., Subramanian A., Ross K.N., Reich M., Hieronymus H., Wei G., Armstrong S.A., Haggarty S.J., Clemons P.A., Wei R., Carr S.A., Lander E.S., Golub T.R.* (2006) The Connectivity Map: Using gene-expression signatures to connect small molecules, genes, and disease. Science, **313**(5795), 1929–1935. DOI: 10.1126/science.1132939

11. *Musa A., Ghoraie L.S., Zhang S.D., Glazko G., Yli-Harja O., Dehmer M., Haibe-Kains B., Emmert-Streib F.* (2018) A review of connectivity map and computational approaches in pharmacogenomics. Brief. Bioinform., **19**(3), 506–523. DOI: 10.1093/bib/bbw112
12. *Shoaib M., Giacopuzzi E., Pain O., Fabbri C., Magri C., Minelli A., Lewis C.M., Gennarelli M.* (2021) Investigating an *in silico* approach for prioritizing antidepressant drug prescription based on drug-induced expression profiles and predicted gene expression. Pharmacogenomics J., **21**(1), 85–93. DOI: 10.1038/s41397-020-00186-5
13. *Lagunin A., Ivanov S., Rudik A., Filimonov D., Poroikov V.* (2013) DIGEP-Pred: Web service for *in silico* prediction of drug-induced gene expression profiles based on structural formula. Bioinformatics, **29**(16), 2062–2063. DOI: 10.1093/bioinformatics/btt322
14. *Davis A.P., Wiegers T.C., Johnson R.J., Sciaky D., Wiegers J., Mattingly C.J.* (2023) Comparative Toxicogenomics Database (CTD): Update 2023. Nucleic Acids Res., **51**(D1), D1257–D1262. DOI: 10.1093/nar/gkac833
15. *Filimonov D.A., Lagunin A.A., Gloriozova T.A., Rudik A.V., Druzhilovskii D.S., Pogodin P.V., Poroikov V.V.* (2014) Prediction of the biological activity spectra of organic compounds using the Pass Online web resource. Chem. Heterocycl. Comp., **50**(3), 444–457. DOI: 10.1007/s10593-014-1496-1
16. *Lagunin A., Stepanchikova A., Filimonov D., Poroikov V.* (2000) PASS: Prediction of activity spectra for biologically active substances. Bioinformatics, **16**(8), 747–748. DOI: 10.1093/bioinformatics/16.8.747
17. *Poroikov V.V., Filimonov D.A., Gloriozova T.A., Lagunin A.A., Druzhilovskiy D.S., Rudik A.V., Stolbov L.A., Dmitriev A.V., Tarasova O.A., Ivanov S.M., Pogodin P.V.* (2019) Computer-aided prediction of biological activity spectra for organic compounds: the possibilities and limitations. Russ. Chem. Bull., **68**, 2143–2154. DOI: 10.1007/s11172-019-2683-0
18. *Ivanov S.M., Rudik A.V., Lagunin A.A., Filimonov D.A., Poroikov V.V.* (2024) DIGEP-Pred 2.0: A web application for predicting drug-induced cell signaling and gene expression changes. Mol. Inform., e202400032. DOI: 10.1002/minf.202400032
19. *Savosina P., Druzhilovskiy D., Filimonov D., Poroikov V.* (2024) WWAD: The most comprehensive small molecule World Wide Approved Drug database of therapeutics. Front. Pharmacol., **15**, 1473279. DOI: 10.3389/fphar.2024.1473279
20. *Piñero J., Ramírez-Anguita J.M., Saüch-Pitarch J., Ronzano F., Centeno E., Sanz F., Furlong L.I.* (2020) The DisGeNET knowledge platform for disease genomics: 2019 update. Nucleic Acids Res., **48**(D1), D845–D855. DOI: 10.1093/nar/gkz1021
21. *Knox C., Wilson M., Klinger C.M., Franklin M., Oler E., Wilson A., Pon A., Cox J., Chin N.E.L., Strawbridge S.A., Garcia-Patino M., Kruger R., Sivakumaran A., Sanford S., Doshi R., Khetarpal N., Fatokun O., Doucet D., Zubkowski A., Rayat D.Y., Jackson H., Harford K., Anjum A., Zakir M., Wang F., Tian S., Lee B., Liigand J., Peters H., Wang R.Q.R., Nguyen T., So D., Sharp M., da Silva R., Gabriel C., Scantlebury J., Jasinski M., Ackerman D., Jewison T., Sajed T., Gautam V., Wishart D.S.* (2024) DrugBank 6.0: The DrugBank knowledgebase for 2024. Nucleic Acids Res., **52**(D1), D1265–D1275. DOI: 10.1093/nar/gkad976
22. *Avram S., Wilson T.B., Curpan R., Halip L., Borota A., Bora A., Bologa C.G., Holmes J., Knockel J., Yang J.J., Oprea T.I.* (2023) DrugCentral 2023 extends human clinical data and integrates veterinary drugs. Nucleic Acids Res., **51**(D1), D1276–D1287. DOI: 10.1093/nar/gkac1085
23. *Lagunin A.A., Rudik A.V., Pogodin P.V., Savosina P.I., Tarasova O.A., Dmitriev A.V., Ivanov S.M., Biziukova N.Y., Druzhilovskiy D.S., Filimonov D.A., Poroikov V.V.* (2023) CLC-Pred 2.0: A freely available web application for *in silico* prediction of human cell line cytotoxicity and molecular mechanisms of action for druglike compounds. Int. J. Mol. Sci., **24**(2), 1689. DOI: 10.3390/ijms24021689
24. *Müller-Dott S., Tsirvouli E., Vazquez M., Ramirez Flores R.O., Badia-i-Mompel P., Fallegger R., Türei D., Lægreid A., Saez-Rodriguez J.* (2023) Expanding the coverage of regulons from high-confidence prior knowledge for accurate estimation of transcription factor activities. Nucleic Acids Res., **51**(20), 10934–10949. DOI: 10.1093/nar/gkad841
25. *Türei D., Korcsmáros T., Saez-Rodriguez J.* (2016) OmniPath: guidelines and gateway for literature-curated signaling pathway resources. Nat. Methods, **13**(12), 966–967. DOI: 10.1038/nmeth.4077
26. *Lee T., Yoon Y.* (2018) Drug repositioning using drug-disease vectors based on an integrated network. BMC Bioinformatics, **19**(1), 446. DOI: 10.1186/s12859-018-2490-x
27. *Yu H., Choo S., Park J., Jung J., Kang Y., Lee D.* (2016) Prediction of drugs having opposite effects on disease genes in a directed network. BMC Syst. Biol., **10**(Suppl 1), 2. DOI: 10.1186/s12918-015-0243-2
28. *Meng F., Xi Y., Huang J., Ayers P.W.* (2021) A curated diverse molecular database of blood-brain barrier permeability with chemical descriptors. Sci. Data, **8**(1), 289. DOI: 10.1038/s41597-021-01069-5
29. *Fu L., Shi S., Yi J., Wang N., He Y., Wu Z., Peng J., Deng Y., Wang W., Wu C., Lyu A., Zeng X., Zhao W., Hou T., Cao D.* (2024) ADMETlab 3.0: An updated comprehensive online ADMET prediction platform enhanced with broader coverage, improved performance, API functionality and decision support. Nucleic Acids Res., **52**(W1), W422–W431. DOI: 10.1093/nar/gkae236
30. *Pires D.E., Blundell T.L., Ascher D.B.* (2015) pkCSM: Predicting small-molecule pharmacokinetic and toxicity properties using graph-based signatures. J. Med. Chem., **58**(9), 4066–4072. DOI: 10.1021/acs.jmedchem.5b00104
31. *Demsie D.G., Altaye B.M., Weldekidan E., Gebremedhin H., Alema N.M., Tefera M.M., Bantie A.T.* (2020) Galanin receptors as drug target for novel antidepressants: Review. Biologics, **14**, 37–45. DOI: 10.2147/BTT.S240715
32. *Freimann K., Kurrikoff K., Langel Ü.* (2015) Galanin receptors as a potential target for neurological disease. Expert Opin. Ther. Targets, **19**(12), 1665–1676. DOI: 10.1517/14728222.2015.1072513
33. *Lu C.L., Herndon C.* (2017) New roles for neuronal estrogen receptors. Neurogastroenterol. Motil., **29**(7), e13121. DOI: 10.1111/nmo.13121
34. *Toumba M., Kythreotis A., Panayiotou K., Skordis N.* (2024) Estrogen receptor signaling and targets: Bones, breasts and brain (review). Mol. Med. Rep., **30**(2), 144. DOI: 10.3892/mmr.2024.13268
35. *Zhang Y., Tan X., Tang C.* (2024) Estrogen-immuno-neuromodulation disorders in menopausal depression. J. Neuroinflammation, **21**(1), 159. DOI: 10.1186/s12974-024-03152-1
36. *Gałecka M., Szemraj J., Su K.P., Halaris A., Maes M., Skiba A., Gałecki P., Bliźniewska-Kowalska K.* (2022) Is the JAK-STAT signaling pathway involved in the pathogenesis of depression? J. Clin. Med., **11**(7), 2056. DOI: 10.3390/jcm11072056

37. *Gulbins A., Grassme H., Hoehn R., Kohnen M., Edwards M.J., Kornhuber J., Gulbins E.* (2016) Role of janus-kinases in major depressive disorder. Neurosignals, **24**(1), 71–80. DOI: 10.1159/000442613
38. *Huang X., Wu H., Jiang R., Sun G., Shen J., Ma M., Ma C., Zhang S., Huang Z., Wu Q., Chen G., Tao W.* (2018) The antidepressant effects of α-tocopherol are related to activation of autophagy via the AMPK/mTOR pathway. Eur. J. Pharmacol., **833**, 1–7. DOI: 10.1016/j.ejphar.2018.05.020
39. *Zhang X., Bu H., Jiang Y., Sun G., Jiang R., Huang X., Duan H., Huang Z., Wu Q.* (2019) The antidepressant effects of apigenin are associated with the promotion of autophagy via the mTOR/AMPK/ULK1 pathway. Mol. Med. Rep., **20**(3), 2867–2874. DOI: 10.3892/mmr.2019.10491
40. *Garay R.P., Bourin M., de Paillette E., Samalin L., Hameg A., Llorca P.M.* (2016) Potential serotonergic agents for the treatment of schizophrenia. Expert Opin. Investig. Drugs, **25**(2), 159–170. DOI: 10.1517/13543784.2016.1121995
41. *Haus U., Varga B., Stratz T., Späth M., Müller W.* (2000) Oral treatment of fibromyalgia with tropisetron given over 28 days: Influence on functional and vegetative symptoms, psychometric parameters and pain. Scand. J. Rheumatol., **29**(sup113), 55–58. DOI: 10.1080/030097400446652
42. *Lecrubier Y., Puech A.J., Azcona A., Bailey P.E., Lataste X.* (1993) A randomized double-blind placebo-controlled study of tropisetron in the treatment of outpatients with generalized anxiety disorder. Psychopharmacology (Berlin), **112**(1), 129–133. DOI: 10.1007/BF02247373
43. *Lin J., Liu W., Guan J., Cui J., Shi R., Wang L., Chen D., Liu Y.* (2023) Latest updates on the serotonergic system in depression and anxiety. Front. Synaptic. Neurosci., **15**, 1124112. DOI: 10.3389/fnsyn.2023.1124112
44. *Greenway S.E., Pack A.T., Greenway F.L.* (1995) Treatment of depression with cyproheptadine. Pharmacotherapy, **15**(3), 357–360. DOI: 10.1002/j.1875-9114.1995.tb04374.x
45. *Khurana K., Bansal N.* (2019) Lacidipine attenuates reserpine-induced depression-like behavior and oxido-nitrosative stress in mice. Naunyn Schmiedebergs Arch. Pharmacol., **392**(10), 1265–1275. DOI: 10.1007/s00210-019-01667-6
46. *Khan I., Kahwaji C.I.* (2023) Cyclobenzaprine. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2024.
47. *Ratajczak P., Kus K., Zielińska-Przyjemska M., Skórczewska B., Zaprutko T., Kopciuch D., Paczkowska A., Nowakowska E.* (2020) Antistress and antidepressant properties of dapoxetine and vortioxetine. Acta Neurobiol. Exp. (Warsaw), **80**(3), 217–224. DOI: 10.21307/ane-2020-020
48. *Garcia-Recio S., Gascón P.* (2015) Biological and pharmacological aspects of the NK1-receptor. Biomed. Res. Int., **2015**, 495704. DOI: 10.1155/2015/495704
49. *Muñoz M., Coveñas R.* (2014) Involvement of substance P and the NK-1 receptor in human pathology. Amino Acids, **46**(7), 1727–1750. DOI: 10.1007/s00726-014-1736-9
50. *Sadri A.* (2023) Is target-based drug discovery efficient? Discovery and "off-target" mechanisms of all drugs. J. Med. Chem., **66**(18), 12651–12677. DOI: 10.1021/acs.jmedchem.2c01737
51. *Hopkins A.L.* (2008) Network pharmacology: The next paradigm in drug discovery. Nat. Chem. Biol., **4**(11), 682–690. DOI: 10.1038/nchembio.118

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РЕПОЗИЦИОНИРОВАНИЕ ЛЕКАРСТВ ДЛЯ ТЕРАПИИ БОЛЬШОГО ДЕПРЕССИВНОГО РАССТРОЙСТВА НА ОСНОВЕ ПРОГНОЗА ЛЕКАРСТВЕННО-ИНДУЦИРОВАННЫХ ИЗМЕНЕНИЙ ЭКСПРЕССИИ ГЕНОВ

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Большое депрессивное расстройство (БДР) — одно из самых распространённых заболеваний, которым страдают миллионы людей во всем мире. Использование существующих антидепрессантов во многих случаях не позволяет добиться устойчивой ремиссии, вероятно, из-за недостаточного понимания этиопатогенеза заболевания. Это указывает на необходимость разработки более эффективных лекарств на основе глубокого понимания патофизиологии БДР. Поскольку разработка новых препаратов является длительным процессом и требует больших финансовых затрат, репозиционирование лекарств является многообещающей альтернативой. В данном исследовании мы применили недавно разработанный нами подход DIGEP-Pred для поиска лекарств, вызывающих изменения в экспрессии генов, связанных с БДР, с последующей идентификацией их потенциальных мишеней, также связанных с БДР, и молекулярных механизмов антидепрессивных эффектов. Анализ включал следующие этапы. Во-первых, мы выполнили прогноз лекарственно-индуцированных изменений экспрессии генов для 3690 лекарств, зарегистрированных в разных странах мира, с использованием связей “структура-активность” (ССА). Анализ обогащения заболеваний, применённый к предсказанным генам, позволил идентифицировать лекарства, которые оказывали существенное влияние на экспрессию генов, связанных с БДР. Во-вторых, потенциальные белки-мишени лекарств, являющиеся мастер-регуляторами, которые ответственны за наблюдаемые изменения экспрессии генов, были идентифицированы с помощью прогноза на основе ССА и анализа молекулярных сетей. Для дальнейшего анализа были отобраны только те лекарства, потенциальные мишени которых, согласно опубликованным данным, были связаны с БДР. В-третьих, поскольку новые антидепрессанты должны действовать в тканях мозга, нами были выбраны лекарства с пероральным способом применения, а их проницаемость через гематоэнцефалический барьер была оценена с использованием имеющихся экспериментальных данных и прогноза *in silico*. В результате мы выявили 19 лекарств, которые потенциально могут быть репозиционированы для терапии БДР. Эти лекарства относятся к различным терапевтическим категориям, включая адренергические/дофаминергические средства, противорвотные, антигистаминные, противокашлевые средства и миорелаксанты. Для многих из найденных лекарств известны или предсказаны взаимодействия с белками-мишенями, связь которых с БДР хорошо изучена, включая моноаминовые (серотонин, адреналин, дофамин) и ацетилхолиновые рецепторы и транспортеры, а также с менее тривиальными мишенями, включая рецептор галанина типа 3 (GALR3), эстрогеновый рецептор 1, связанный с G-белком (GPER1), протеинкиназы JAK3 и ULK1. Важно, что большинство из 19 лекарств воздействуют на две или более мишени, связанные с БДР, что может приводить к более сильному воздействию на экспрессию генов и, как следствие, к более выраженному терапевтическому эффекту. Таким образом, идентифицированные 19 лекарств могут являться перспективными кандидатами для терапии БДР.

Полный текст статьи на русском языке доступен на сайте журнала (http://pbmc.ibmc.msk.ru).

Ключевые слова: репозиционирование лекарств; большое депрессивное расстройство; лекарственно-индуцированная генная экспрессия; мастер-регуляторы; сигнальная сеть; взаимосвязи структура-активность

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