Unifi nootropics from the lab to the web: a story of academic (and industrial) shortcomings

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Abstract

This paper is a review of the work of my former academic group of research in the past 15 years, in the field of cognition enhancers (also called nootropics) that identified two very potent molecules: Unifiram and Sunifiram that for a variety of reasons were not protected by a patent. Some 12 years after their disclosure (2000) I casually found that on the web, there were dozens of sites offering Unifiram and Sunifiram as drugs that improve cognition in healthy individuals even if only few preclinical studies were done and their long-term toxicity was unknown.

Keywords

Cognition enhancers, online drug market, Unifiram, Sunifiram

Introduction: the back story

Three contextual aspects of this report deserve introduction before relaying the rest of the information; one is the initial laboratory approach, two is the laboratory findings, and the third is the 'market' inquiry.

Summary of lab findings: In 2000, my research group published two papers where two new compounds endowed with outstanding nootropic properties were described: Unifiram (DM232) and Sunifiram (DM235). Each presented a potency some 30,000 times higher than that of piracetam, the most representative of piracetam-like nootropics (Figure 1).

'Market' inquiry: In March 2012, I received the following e-mail. In an effort to protect the individual who contacted me, I will refer to him as John Doe.

From: John Doe
Sent: Wednesday March 21, 2012
To: fulvio.gualtieri@unifi.it
Subject: DM235 human testing

Hello, my name is John Doe
I have read many papers on DM235 and other Nootropics. Currently I am a user of Oxiracetam to combat a premature aging syndrome, which causes me severe brain fatigue, loss of memory, etc. I am 34 years old
Before Oxiracetam, I used Piracetam, but after two years it was too weak to help my mitochondria, so recently I switched to Oxiracetam which works far better! Still Oxiracetam is not good enough, so I was happy to see the invention of DM235. I have found 6 suppliers of DM235 who can provide 1000 mg sample amount for cheap-$88 USD to $200 USD. In the next few days I will place an order for 1000 mg DM235 so that I can test it on myself. I wonder, am I the first human to try it? Do you have any information about human use? If I receive the sample and ingest it, I will email you details of my experience as a human Guinea Pig.
I weight 145 lbs and plan to take 1–5 mg total dose, but my digital scale is only accurate to 100 mg so I will need to dilute a solution or guess the dose from a few crystals.
Do you have any recommendations? Thanks.

Definition and approach

Definition and evaluation of cognition enhancing activity: For those who are not familiar with the topic, cognition enhancers are drugs that should restore normal conditions in individuals that have cognitive dysfunctions due to ageing or to age-related neurodegenerative diseases like Alzheimer’s, Parkinson’s and other neurological disorders. The word nootropics is generally used for piracetam-like compounds. Both terms have also been used by street people to indicate drugs that stimulate normal mind, in this case, smart drugs, memory enhancers, and brain boosters, for their ability to produce positive effects on mental performance, are more frequently used.

The inherent complexity of neurological disorders, the uncertainty of animal models, and some unresolved questions in preclinical and clinical tests make the evaluation of nootropic activity a still difficult task, both in animals and in humans, for which satisfactory solutions have yet to be found for either.

One of the most used but much criticized assay is the mouse passive avoidance described by Jarvik and Kopp. In our research,
we used it (slightly modified) as a preliminary test to rank all our new compounds. In short, mice receive a shock when entering a dark room during a training session and remember it in the following day’s session unless their memory is impaired by the amnestic drugs. The parameter measured is the entry latency time (expressed in seconds) occurring between the time the mouse is placed in the light and the time it enters the dark room. In the first day, there is the training session, while in the second day, the mice are placed again in the light and the new latency time is measured on animals treated or untreated with the nootropic drug; comparison of the latency times of saline treated animals with those that received both the amnestic drug and the investigated one gives a measure of the cognitive activity of the compound tested. The nootropic activity is expressed as minimal effective dose (MED) usually in mg/kg.

The potency of the most active molecules that we have synthesized and evaluated is reported in Schemes 1–3. It is fair to say that the structure–activity relationships (SAR) that can be observed have to be considered with caution, as it is well known that, in vivo experiments, the pharmacokinetic properties of any molecule may affect the amount of it that reaches the target and exert its action, making questionable the building of reliable SAR. After the initial experiments, any compounds that were considered more interesting were evaluated also in the Mondadori social learning test\(^2\) and in the rat Morris water maze\(^3\), since we share the opinion that the conclusion on the

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**Figure 1.** Unifiram (DM232, 1) and Sunifiram (DM235, 2).

**Scheme 1.** Chemical modulation of Unifiram.

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**Scheme 1.** Chemical modulation of Unifiram.
nootropic activity of a drug should not rely on the results of one kind of test.

**Detailed lab findings**

**Unifi nootropics**

**Medicinal chemistry**

The first molecule identified was Unifiram (1). While studying the nicotinic activity of a series of bicyclic compounds (1,4-diazabicyclo[4.3.0]nonan-9-ones) such as A that were in fact poor nicotinic agonists, it was clear that they shared the pyrrolidin-2-one nucleus with piracetam, one of the most studied and available nootropics.

We reasoned that proper modification of the structure to allow brain tissue to penetrate could result in new nootropic compounds. Therefore, we eliminated the charge on the N4 nitrogen of piperazine, transforming it into an amide with different acid moieties (general structure B) and evaluated the compounds using the mouse passive avoidance test, obtaining very promising results. Thereafter, we optimized the lead to compound 1 that was active as nootropic at the dose of 0.001 mg/kg i.p., some thousand times lower than that of piracetam, 30 mg/kg, under the same conditions. We named the molecule Unifiram from the acronym that identifies the University of Florence on the web. The compounds of this family present also some features of another class of nootropics, the ampakines, with the consequence that our compounds are considered on one hand, ampakines the other hand, they are piracetam-like, which creates confusion.

Schemes 1–3 illustrate the most productive chemical modulations that we have performed on Unifiram: they were aimed to (i) establish SAR, (ii) find more potent molecules, and (iii) help identification of the mechanism of action.

One of the first achievements of a classical chemical modulation was the discovery that opening the pyrrolidinone ring gave a new family of compounds among which 2 completely maintained the nootropic activity of Unifiram (Scheme 1). The compound was named Sunifiram, where “S” stands for seco [6].

In Scheme 1, the main modulations made on Unifiram molecule are reported; the racemate has been resolved, finding that there is a modest enantioselectivity in favor of R- (+)-Unifiram. Then the consequences of pyrrolidinone ring expansion were evaluated and resulted in less potent nootropics; however, one of them, 3, showed an unexpected amnestic activity and, at the dose of 10 mg/kg, had the same effect of scopolamine. The same happened when the 4-F-benzenesulfonyl moiety of Unifiram was changed to isopropylsulfonyl to give 6. The use of isopropylsulfonic acid was suggested by its presence in a series of positive allosteric modulators of AMPA receptor that were active as cognition enhancers.

In the same series of expanded ring compounds, 4 and 5 were much less effective as nootropic (MED value =1 mg/kg) but presented interesting activity in neuropathic pain.

Finally, the effect of extruding the nitrogen atom of piperazine involved in the pyrrolidinone cycle was evaluated (that is, the piperazine moiety was substituted with a 4-aminopiperidine one). The rac-trans (7) and rac-cis (8) forms were prepared; the last one is the most active, showing good nootropic activity (MED value =0.1 mg/kg). Both compounds were found active in two models of chronic neuropathic pain, originated from antitumor therapy and diabetes; in the latter model, rac-7 was found to be significantly active.
three times more active than rac-8. We resolved the racemates of both 7 and 8; enantioselectivity was not observed for the nootropic activity, while the antihyperalgesic activity was mainly due to the (+) forms: (+)-8 (oxaliplatin-induced neuropathy) or (+)-7 (streptozotocin-induced neuropathy)10.

These modulations produced good (MED value = 0.1 mg/kg) or very good (MED value = 0.01 mg/kg) nootropics, however, only 2 (MED value = 0.001 mg/kg) reached the outstanding activity of 1.

Similar modulation of Sunifiram (2, DM235) is reported in Scheme 2. Many acids were used to substitute the benzoyl and propionyl moieties of Sunifiram, with mixed results on nootropic activity. Compound 9, where the propionyl and benzoyl groups were substituted by acetyl and isopropylsulfonyl moieties, respectively, although still quite potent (MED value = 0.1 mg/kg), does not reach the potency of 2. The isopropylsulfonyl group had worked well in another family of nootropics9. In this instance, it was demonstrated, for the first time, that one of the nitrogen atoms of piperazine can remain unsubstituted (and, therefore, basic) as shown by the good activity of 108. However, when we tried to exploit this finding by introducing an amino group in the chain of the propionate moiety, the result was disappointing as shown by the modest activity of 11 (MED value = 1.0 mg/kg)8. The effects of alkyl substitution on the piperazine ring were also studied: positions 2 and 3 are clearly different as a compound substituted in position 2 (14 MED value > 10 mg/kg) is practically inactive whereas that substituted in position 3 (15, MED value = 0.1 mg/kg) maintains a good activity6. The best result in this series was obtained when we substituted piperazine moiety with a 4-aminopiperidine one. In fact, compound 16, that has been named Sapunifiram, shows a very good activity (MED = 0.01 mg/kg)13.

Further chemical modulations of 2 are reported: we transformed Sunifiram (2) in a less flexible tricyclic compound carrying the same number of heavy atoms, just linking the ortho position of the benzoyl group with position 2 of piperazine nucleus and the result was an inactive compound 17 (MED value >10 mg/kg) (Scheme 3). However, if the N4 substituent of piperazine moiety was changed, as in 18, the compound showed good activity (MED value = 0.1 mg/kg) even if it did not reach the potency of parent drug12. So compound 18 would represent a less flexible model useful to study the interaction of Sunifiram with its target.

Then, we tested the consequences of extruding both nitrogen atoms of piperazine and used both isomers of 1,4-cyclohexanediamine as a scaffold for new derivatives. The cis isomer gave nootropics like 19 with fairly good activity (MED value = 0.1 mg/kg), while the trans isomer gave, again unexpectedly, potent amnestic drugs, such as 20 (MED value = 0.01 mg/kg).
Playing the same game of scaffold hopping, we shifted to a 1,4-diaminobenzene scaffold obtaining some very interesting compounds like 21 (MED value = 0.01 mg/kg).\(^{14}\) Also in the case of Sunifiram, we tried to change the size of the cycle;\(^{15}\) while reduction of the size-impaired nootropic activity (22, MED value = 1.0 mg/kg), its enlargement maintains a good activity (23, MED value = 0.1 mg/kg). Finally changing the relative position of the nitrogen atoms of piperazine was definitely detrimental for activity (24, MED value = 1.0 mg/kg).

As it can be seen from these data, at certain point of our research we dedicated ourselves to make changes on the scaffold, while keeping the propionyl/benzoyl or acetyl/4-fluorobenzenesulfonyl substituents. The reason was that some structural modification gave puzzling results; some, not exhaustive, examples are shown in Scheme 4. When decorating the diazabicyclo[4.3.0]nonan-9-one scaffold of Unifiram (MED value = 0.001 mg/kg), the benzoyl group, which conferred high potency to Sunifiram (MED value = 0.001 mg/kg), gave a large drop in activity (25, MED value = 1.0 mg/kg). On the contrary, the substitution of the benzoyl group with the 4-fluorobenzenesulfonyl moiety on the piperazine scaffold gave only a small reduction in activity (from 2, MED value = 0.001 mg/kg, to 26, MED value = 0.01 mg/kg). Changing the propionyl group on 26 with an acetyl moiety maintained activity (27, ME value = D 0.01 mg/kg); the same modification on 2 gave a large drop in potency (28, MED value = 10 mg/kg).

From the data reported in these schemes, it can be realized that SAR are inconsistent, supporting the hypothesis that pharmacokinetic properties of each molecule play a major role on the level of nootropic activity found. In this case, Unifiram and Sunifiram would represent the best compromise between the binding to the target and their pharmacokinetic properties. More reliable information for optimization would require the knowledge of the biological target to perform in vitro studies, such as binding, that at the moment are not possible.

**Pharmacology**

The most potent compounds of the series, Unifiram and Sunifiram, were selected for further studies aimed to find their pharmacological properties and possibly their mechanism of action. Summarizing the results reported in our papers, where the interested scientists are addressed for details in methods and procedures,\(^{5–8,10,12–18}\) the following were the main findings:

1. Unifiram and Sunifiram antagonized memory disruption is produced not only by the muscarinic antagonist scopolamine but also by mecamylamine (nicotinic antagonist), baclofen (GABA agonist), clonidine (alpha-2 agonist), and NBQX (AMPA antagonist)\(^ {16,18}\). The MEDs varied according to the amnesic drug and administration way.

2. Unifiram and Sunifiram at the dose of 0.001 mg/kg i.p. were able to revert the memory impairment induced by scopolamine (mouse passive avoidance test); when administered alone they were able to ameliorate unimpaired memory processes (procognitive effect) at higher doses\(^ {14}\).

3. Unifiram and Sunifiram at the dose of 0.1 mg/kg i.p. were able to prevent amnesia induced by scopolamine in the rat Morris water maze; however, when administered alone, they were inactive. (lack of procognitive effect)\(^ {16,17}\).

4. Unifiram and Sunifiram at the dose of 0.1 mg/kg i.p. were active in the rat social-learning test (procognitive effect)\(^ {6,16}\).

5. Unifiram and Sunifiram at the dose of 0.1 mg/kg i.p. significantly reduced the total sleeping time induced by pentobarbital in mice\(^ {16,17}\).

6. Unifiram slightly increased the release of ACh from parietal cortex in freely moving rats while Sunifiram was definitely more efficient. As a consequence of cholinergic potentiation, Unifiram and Sunifiram show analgesic activity in the mouse hot-plate test, where they increase the pain threshold after i.p. administration in a dose-dependent manner, but with a bell-shaped curve\(^ {6}\). The enantiomers of Unifiram showed some enantioselectivity also in this test: while both (S)-(−)-1 and (R)-(−)-1 at the dose of 0.01 mg/kg showed a similar efficacy (Figure 2), (R)-(−)-1 was able to increase ACh release also at 0.001 mg/kg\(^ {7}\).

7. Unifiram and Sunifiram elicited their nootropic effect without changing animal’s gross behaviour or modifying motor coordination as revealed by the Rotarod test (mouse, 1–10 mg/kg i.p.). The spontaneous motility and the
inspection activity were unmodified by administration of the studied compounds (mouse and rat, 1 mg/kg i.p.) as revealed by Animex and Hole Board testing in comparison with saline-treated mice.

(8) Unifiram and Sunifiram, tested within the NIMH Psychoactive Drug Screening Program by the lab of Dr Brian Roth (https://pdspdb.unc.edu/pdspWeb/), did not reveal any affinity for the most important CNS receptors, channels, and transporters (see Table 1 in Supplemental information).

(9) The results of experiments with NBQX in the kynurenate test seem to suggest that AMPA receptors are involved in the anti-amnesic activity of Unifiram and Sunifiram. Indeed Unifiram increases AMPA current in rat CA1 slices (EC50 value = 27 ± 6 nM). However, when the compounds were tested on recombinant AMPA (GluR1/GluR2) receptors, no potentiation of the AMPA-R responses was observed (unpublished results).

(10) Rat adipocytes were chosen as model to study in vitro the pharmacological activity of the compounds. Sunifiram increases NO production in rat adipocyte, with a maximal effect (2.75-fold increase over basal) at 17.5 nM, and then it decreased (bell-shaped curve). This effect was completely prevented by 100 μM l-NNAME (N’-nitro-l-arginine methyl ester, a NOS inhibitor), and it was antagonized by mecamylamine and by methyllycaconitine (10−5 M), indicating the involvement of α7 nicotinic receptor activation19. However, no agonist effect or potentiation of ACh stimulation was detected when Sunifiram was tested on rat recombinant α7 nicotinic receptor in Xenopus oocytes (unpublished results). In addition, Sunifiram was not able to revert l-NNAME-induced amnesia in the mouse passive avoidance test (unpublished results).

There is an evident pharmacological similarity between our series of compounds and piracetam-like nootropics, the main difference being the potency of Unifiram and Sunifiram: some 30,000 times higher than piracetam and more than 1000 times that of the most potent piracetam-like drugs such as oxiracetam, nefiracetam, etiracetam, and aniracetam. Moreover, our compounds seem to share the lack of toxicity of piracetam-like nootropics. However, as far as chemical structure is concerned, Unifiram has, in common with piracetam-like compounds, the 2-pyrrolidinone ring; in the equipotent Sunifiram, this feature does not seem necessary for nootropic activity.

The problem of the mechanism of action

The different families of cognition enhancers have been reviewed20–26. As far as piracetam-like nootropics are concerned, the lack of a common mechanism of action has been one of the main obstacles to their acceptance as drugs. This is a problem also for Sunifiram and Unifiram. On one hand, the high potency against scopolamine and the release of ACh6 would suggest a cholinergic mechanism of action but we were unable to evidence any interaction, orthosteric or allosteric, with muscarinic and nicotinic cholinergic receptors. On the other hand, it seems clear that Unifiram and Sunifiram can modulate the cholinergic system. However, the mechanism remains unknown even if there are strong indications that a specific target is involved. The finding that Unifiram and Sunifiram can reverse the amnesia induced by NBQX and were able to produce a NBQX sensitive reversal of the kynurenate-induced antagonism in the "kynurenate test" would support the involvement of AMPA receptors even if this is not the only mechanism of action18.

We hoped that the amnestic activity of compounds like 3, 6, and 20,14 would help to find a solution, but even for them we found no interaction with known central receptors even if the fact that their amnestic activity was contrasted by Unifiram and Sunifiram indicates a precise site of interaction. As a matter of fact when in a series of congeneric compounds agonist and antagonists are found, one can be reasonably sure that there is a specific site of interaction: receptor, enzyme, channel, etc. In conclusion, based on the results of our pharmacological research, unifi nootropics seem slightly active on a variety of biological targets with a kind of pleiotropic behavior.

Other research groups have tried to study the mechanism of action of Sunifiram and Unifiram. In 2004, Naftalin, who studied the antagonism of piracetam and like compounds on the inhibition by barbiturates of glucose transport in human erythrocytes, included Sunifiram in the experiment and found that it was a potent antagonist (Ki value = 26.0 ± 3.0 μM) of glucose transport inhibition and that the level of inhibition correlated with nootropic potency27. It was still another example of the variety of actions of Sunifiram but very likely does not represent the main mechanism of action.

Very recently, some interesting data were published in two papers by Moriguchi and coworkers. In the first paper28, using olfactory bulbectomized (OBX), mice that show in hippocampus-dependent memory impairment, it was shown that Sunifiram (0.01–1.0 mg/kg p.o.) improves cognitive deficit observed in OBX...
mice via CaM kinase II and proteine kinase C activation. Sunifiram also restored hippocampal LTP (long-term potentiation) impaired in OBX mice. From these and other experiments, the authors concluded that these data indicate that Sunifiram ameliorates OBX-induced deficits of memory-related behaviors and impaired LTP in the hippocampal CA1 region via stimulation of glycine-binding site of N-methyl-D-aspartate receptor (NMDA-R). In the second paper, the authors, to address the question of Sunifiram to gather more information. However, a few days after, I again warned him about the risks that he would face since neither the pharmacokinetic nor toxicology of such experimental compound was known.

A year later, when we had nearly forgotten about this episode, I received a message where John Doe announced that the drug was now available in the US at the online market. Moreover, Sunifiram was also available at low prices and in large quantities from a Chinese manufacturer. We were kind of shocked and a window suddenly opened on a new world when we run at the web to verify. We discovered that Sunifiram was sold by dozens of companies that provided also the documents of its purity (NMR, GC, and HPLC). At that time, Unifiram was neglected since Sunifiram is much simpler to synthesize, but was gaining credit. We realized that Unifiram and Sunifiram were quite popular in the web (in a search by the browser safari in a day of august 2014, they had 13 800 and 39 800 entries, respectively). Moreover, they were well cited in Wikipedia and Sunifiram is actually available for sale on Amazon. We also learned that our molecules are sold side-by-side online with other drugs (including other nootropics) that have passed clinical trials while ours have never been tested on humans. In addition to the usual issues on the efficacy of nootropics and the ethical concerns that arise from their use, non-approved compounds may present toxicity problems, in particular in the long term. For us, however, what was most interesting was that people consuming them were discussing their experience in several forum and blogs, which sound like clinical trial reports. Of course, John Doe did not refrain to send its own report to us; it is too long to be inserted here. The following is a tentative synopsis of his message:

(1) He was already used to take nootropics among which piracetam, oxiracetam alone, or together in grams doses.
(2) He had taken 25 mg six times a day for a total of 150 mg of Sunifiram.
(3) From the day he started assumption of Sunifiram “I have not spent one second tired” he says.
(4) The sleep duration was reduced from about 8.5 to about 5 h per night without any “sleep deficit” and no tiredness.
(5) In his mind “Sunifiram = modafinil 2.0.”
(6) Hes reports that among the members of the blogs, there has been a problem but only when the person used Sunifiram plus 5 g of caffeine.

I never heard from him again since this message. The only things I know about him is what he shared in the first email. It is possible that he used a pseudonym as his signature. I did note naïveté in his questions about chemical compound structures and pharmacology. In one of his messages, John Doe says: “your dreams have now come true”, but I disagree. In fact, our dream was to discover a medicine for sick persons and then, if possible, collect funding to support our research.

Traditionally one examines the morale of a story. My thinking is that, if our government had adequately funded our research, if our university paid more attention to the potential of in-house patents (as it is doing now), if at least one company had the foresight to put money on it, and if the pharmaceutical industry were more open to outside research, today we would have, perhaps, more of a medicine and less of a street drug. And, last but not least, we would have likely collected financial gain for our efforts.

Acknowledgements
I wish to dedicate this work to my former staff with particular acknowledgement to Serena Scapecchi who departed us too early, and to all the scientists that have contributed to its success.
Medicinal chemists

Staff: Cristina Bellucci, Silvia Dei, Dina Manetti, Maria Novella Romanelli, Serena Scapece, and Elisabetta Teodori.

PhD students and post doc: Luca Guandalini, Cecilia Martelli, Elisabetta Martini, Michele Melchiorre, and Simona Pagella.

Chemists: Gianluca Bartolucci and Carlo Bertucci.

Pharmacologists: Alessandro Bartolini, Elisabetta Baldi, Corrado Buccherelli, Nicoletta Galeotti, Carla Ghelardini, Monica Norcini, Anna Pittaluga, Annamaria Pugliese, and Alberto Salvicchi.

Many thanks to my daughter-in-law Montine Blank who revised the paper, fixing my broken English.

Declaration of interest

The author reports no conflicts of interest. The author alone is responsible for the content and writing of this article.

References

1. Jarvik ME, Kopp R. An improved one trial passive avoidance learning situation. Psychol Rep 1967;21:221–4.
2. Mondadori C, Preiswerg G, Jaekel J. Treatment with a GABA_B receptor blocker improves the cognitive performance of mice rats and rhesus monkeys. Pharmacol Commun 1992;2:93–7.
3. Morris RGM. Development of a water–maze procedure for studying spatial learning in the rat. J Neurosci Methods 1984;11:47–60.
4. Manetti D, Borea PA, Ghelardini C, et al. Reduced flexibility hybrids of the nicotinic agonist 1,1-dimethyl-4-acetylpyridazinum iodide and 2-(dimethylamino)methyl-5-methyl cyclopropenone methiodide. Med Chem 1997;7:301–12.
5. Manetti D, Ghelardini C, Bartolini A, et al. Design, synthesis and preliminary pharmacological evaluation of 1,4-diazabicyclo[4.3.0]- nonan-9-ones as a new class of highly potent nootropic drugs. J Med Chem 2000;43:1969–74.
6. Manetti D, Ghelardini C, Bartolini A, et al. Molecular simplification of 1,4-diazabicyclo[4.3.0]-nonan-9-ones gives piperazine derivatives that maintain high nootropic activity. J Med Chem 2000;43:4499–507.
7. Martini E, Ghelardini C, Bertucci C, et al. Enantioselective synthesis and preliminary pharmacological evaluation of the enantiomers of unifiram (DM232), a potent cognition-enhancing agent. Med Chem 2005;1:473–80.
8. Scapece S, Martini E, Manetti D, et al. Structure–activity relationship studies on unifiram (DM232) and sunifiram (DM235), two novel nootropic drug sunifiram improves cognitive deficits via CaM kinase II and protein kinase C activation is involved in the antiamnesic effect of DM232 (unifiram) and DM235 (sunifiram). J Med Chem 2003:368:538–45.
9. Raimondi L, Ghelardini C, Gualtieri F, et al. DM232 and DM235, two potent cognition-enhancers, increase NO levels in rat adipocytes. Proceedings of Frontiers in CNS and Oncology Medicinal Chemistry, Siena, Italy; 2007 Oct 7–9, p 47.
10. Gouliaev AH, Senning A, Piracetam and other structurally related nootropics. Brain Res Rev 1994;19:180–222.
11. Gualtieri F, Manetti D, Romanelli MN, Ghelardini C. Design and study of piracetam-like nootropics, controversial members of the problematic class of cognition-enhancing drugs. Curr Pharm Des 2002:8:125–38.
12. Gualtieri F, Guandalini L, Manetti D, et al. Cognition-enhancing drugs in mild cognitive impairment (MCI) and Alzheimer’s disease (AD): an update[1]. Med Chem Rev-Online 2005;2:471–87.
13. Romanelli MN, Galeotti N, Ghelardini C, et al. Pharmacological characterization of DM232 (unifiram) and DM235 (sunifiram) new potent cognition enhancers. CNS Drug Rev 2006:12:39–53.
14. Froestl W, Muhs A, Pfeiffer A. Cognitive enhancers (nootropics). Part 1 Drugs interacting with receptors. Update 2014. J Alzheimer’s Dis 2014;41:961–1019.
15. Froestl W, Muhs A, Pfeiffer A. Cognitive enhancers (nootropics). Part 2 Drugs interacting with enzymes. Update 2014. J Alzheimer’s Dis 2014;42:1–68.
16. Froestl W, Muhs A, Pfeiffer A. Cognitive enhancers (nootropics). Part 3 Drugs interacting with targets other than receptors and enzymes. Disease modifying drugs. J Alzheimer’s Dis 2014;42:1079–149.
17. Naftalin RJ, Cunningham P, Afzal-Ahmed I. Piracetam and TRH analogues antagonize inhibition by barbiturates, diazepam, melatonin and galanin of human erythrocyte D-glucose transport. Br J Pharmacol 2004;142:594–608.
18. Froestl W, Muhs A, Pfeiffer A. Cognitive enhancers (nootropics). Part 4 Drugs enhancing learning and cognitive performance. J Alzheimer’s Dis 2014;41:2063–72.
19. Martini E, Ghelardini C, Dei S, et al. Design, synthesis and preliminary pharmacological evaluation of new piperidines and piperazine derivatives as cognition enhancers. Bioorg Med Chem 2008;16:1431–43.
20. Manetti D, Martini E, Ghelardini C, et al. 4-Aminopiperidine derivatives as a new class of potent cognition enhancing drugs. Bioorg Med Chem Lett 2003;13:2303–6.
21. Martini E, Norcini M, Ghelardini C, et al. Design synthesis and preliminary pharmacological evaluation of new analogues of DM232 (unifiram) and DM235 (sunifiram) as cognition modulator. Bioorg Med Chem 2008;16:10034–47.
22. Martini E, Salvicchi A, Ghelardini C, et al. Design, synthesis and nootropic activity of new analogues of sunifiram and sapunifiram, two potent cognition enhancers. Bioorg Med Chem 2009;17:7606–814.
23. Ghelardini C, Galeotti N, Gualtieri F, et al. The novel nootropic compound DM232 (UNIFIRAM) ameliorates memory impairment in mice and rats. Drug Dev Res 2002;56:23–32.
24. Ghelardini C, Galeotti N, Gualtieri F; et al. DM235 (Sunifiram): a novel nootropic with potential as cognitive enhancer. Naunyn-Schmiedeberg’s Arch Pharmacol 2002;365:419–26.
25. Galeotti N, Ghelardini C, Bartolini A, et al. AMPA-receptor activation is involved in the anti-amaenic effect of DM232 (unifiram) and DM235 (sunifiram). Naunyn-Schmiedeberg’s Arch Pharmacol 2003:368:538–45.
26. Froestl W, Muhs A, Pfeiffer A. Cognitive enhancers (nootropics). Part 1 Drugs interacting with receptors. Update 2014. J Alzheimer’s Dis 2014;41:961–1019.
27. Froestl W, Muhs A, Pfeiffer A. Cognitive enhancers (nootropics). Part 2 Drugs interacting with enzymes. Update 2014. J Alzheimer’s Dis 2014;42:1–68.
28. Froestl W, Muhs A, Pfeiffer A. Cognitive enhancers (nootropics). Part 3 Drugs interacting with targets other than receptors and enzymes. Disease modifying drugs. J Alzheimer’s Dis 2014;42:1079–149.
29. Froestl W, Muhs A, Pfeiffer A. Cognitive enhancers (nootropics). Part 4 Drugs enhancing learning and cognitive performance. J Alzheimer’s Dis 2014;41:2063–72.
30. Bartolini A, Bellucci C, Galeotti N, et al. (University of Florence). Derivatives possessing nootropic activity, preparation and use. EP20000101165 20000121.
31. Lanni C, Lenzken SC, Pascale A, et al. Cognition enhancers (nootropics). Regulatory challenges. Sci Eng Ethics 2009;15:311–41.
32. Bartolini A, Bellucci C, Galeotti N, et al. (University of Florence). Derivatives possessing nootropic activity, preparation and use. EP1118612 (A1), July 25, 2001. Application number: EP20000101165 20000121.
33. Lanni C, Lenzen SC, Pascale A, et al. Cognition enhancers between treating and doping the mind. Pharmacol Res 2008;57:196–213.
34. Bostrom N, Sandberg A. Cognitive enhancement: methods, ethics, regulatory challenges. Sci Eng Ethics 2009;15:311–41.
35. Kluger J. Safety concerns raised over popular wakefulness drug. Time 2009, March 17. Available from: http://content.time.com/time/health/article/0,8599,1885825,00.html [last accessed 27 Feb 2015].

Supplementary material available online.
Supplemental Information