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ABSTRACT

Background: Spectral Data Activity Relationship (SDAR) is a computational method that uses the steric and electronic characteristics of small molecules which are measured by spectrometric data to create spectral models that reflect their biological potential. These spectral models can then be used to screen other molecules for the particular activity of the drugs used to create the model (e.g., efficacy, toxicity) or to design novel drugs having a set of desired characteristics present in the drugs used to create the model, with a high degree of accuracy. We have used the spectral modeling method to create a series of atypical antipsychotic drug candidates predicted to have targeted characteristics that would predict efficacy comparable to currently available agents but might lack some of the most troublesome side effects of these drugs. **Methods:** Training spectral models were developed from a large series of compounds (N=40-48) with activity as typical or atypical antipsychotic agents, some of which data were available for side effects such as weight gain and agranulocytosis. Spectral models were created using a random leave-out-10% process and published human experimental data. Spectral models of high accuracy were created for dopamine (DA) D₂ (q²-10% = 0.96) and 5-HT_{2A} (q²-10% = 0.99) receptor binding affinities, antipsychotic activity, agranulocytosis activity, agranulocytosis relative risk (q²-10% = 0.98), and hERG inhibition (q²-10% = 1.0). Data extracted from the spectral models were used to create 259 new atypical antipsychotic drug candidates. **Results:** When screened against the spectral models, 204 *in silico* drug candidates were predicted to have antipsychotic activity, 43 of those 204 had the desired receptor binding profiles (greater affinity for 5-HT_{2A} receptors than DA D₂ receptors), and after toxicity prediction, 12 were selected for synthesis and further development. The affinities of two of these compounds for multiple G-protein-coupled-receptors were determined by the NIMH Psychoactive Drug Screening Program and their greater affinity for 5-HT_{2A} receptors than DA D₂ receptors confirmed. These compounds were then studied using microdialysis in awake freely moving rats to determine their similarity to clozapine and other multireceptor 5-HT_{2A}/D₂ antagonists. One of the compounds, LMD-00076, produced a dose-dependent increase in DA efflux which was blocked by the 5-HT_{1A} silent antagonist, WAY 100635, as is the case with clozapine. It also produced a significant increase in cortical acetylcholine (ACh) efflux, as do direct acting 5-HT_{2A}/D₂ antagonists. A second compound, LMD-00100t, produced a significant increase in cortical DA, but at the maximum dose tested, did not increase cortical ACh efflux. This pattern resembles aripiprazole. **Discussion:** Spectral drug modeling was able to develop a large series of novel 5-HT_{2A}/D₂ antagonists in a brief period of time, and at a cost that was significantly less than associated with conventional methods of high throughput screening and directed chemical synthesis. The compounds studied to date proved to be biologically active in a rigorous model which distinguishes typical from atypical antipsychotic drugs. Further testing of these compounds in classical models of antipsychotic activity and extrapyramidal side effects in laboratory animals is in progress, along with studies of their ability to reverse the effects of phencyclidine on novel object recognition. If clinical testing were to establish the efficacy and safety of these molecules, SDAR modeling should be considered as a highly efficient and cost-effective means of developing novel drug entities for diverse clinical indications.

INTRODUCTION

In silico methods of drug design and optimization are used early in the development process to help select molecules with the most potential for further development; however, the traditional QSAR and SAR methods have limited success rates. The **Spectral Data Activity Relationship** method (SDAR, aka **Spectral Modeling™**) is a computational chemistry method used for drug design and optimization developed at FDA under a Collaborative Research and Development Agreement with LITMUS, LLC. **Spectral Modeling™** utilizes computer-based mathematical methods to analyze spectral data for identification of spectral attributes that are highly predictive of a molecule's biological, chemical, and physical activities. Further development and refinement of computational techniques has allowed LMD to extend the application of **Spectral Modeling™** to design new chemical entities (NCE) with targeted activities (e.g., improved efficacy and reduced toxicity).

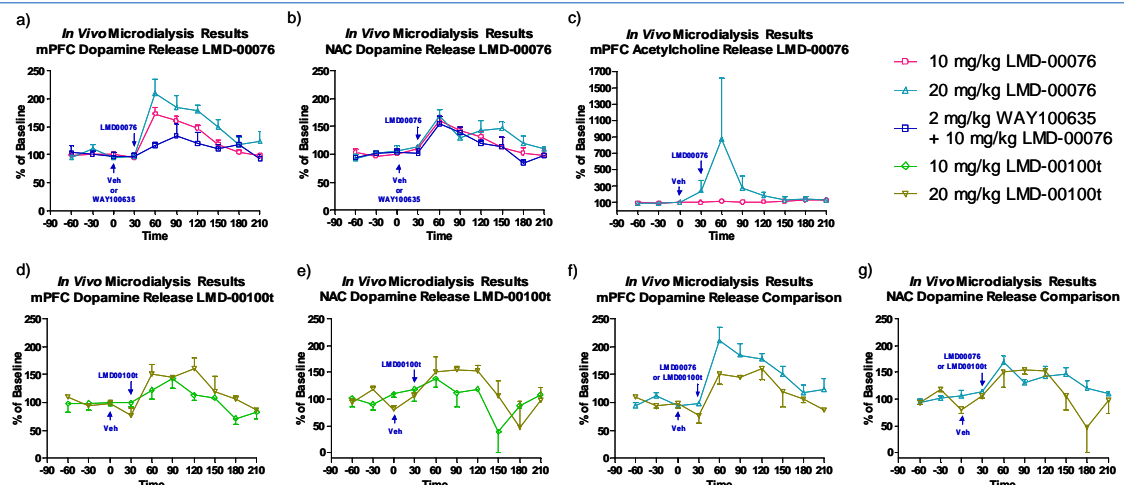
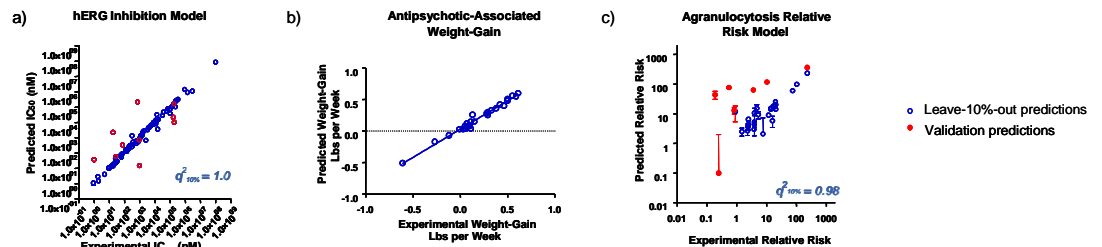
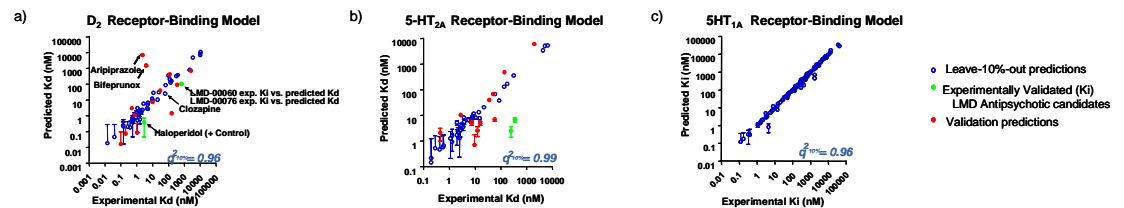
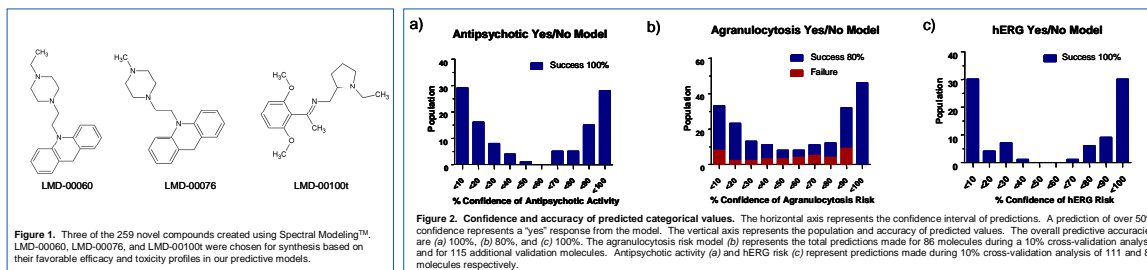
There is a tremendous unmet medical need for improved antipsychotic drugs. The goal of this project was to create new atypical antipsychotic drug candidates according to the 5-HT_{2A}/D₂ (clozapine) model, but without serious antipsychotic-associated adverse effects.

METHODS

Spectral models of key biological characteristics of atypical antipsychotic drugs were created to predict antipsychotic activity (human D₂ and 5-HT_{2A} receptor affinity) and toxicity (hERG inhibition, antipsychotic-associated weight gain, and agranulocytosis risk). Once suitable data sets had been selected, we employed *in silico* methods to determine the most significant relationships between molecular architecture and biological responses by generating formalisms that describe the activity of interest as a function of the spectral attributes of the drugs comprising the dataset. These models were subjected to leave-10%-out cross-validation and further tested by double-blind data approximately equal in number to 20% of the original data set.

By identifying key molecular regions of certain antipsychotic drugs, we were able to correlate specific molecular alterations to certain receptor affinities while simultaneously reducing antipsychotic-related toxicities. Data extracted from the models allowed assessment of molecular features predicted to be critical for atypical antipsychotic drug activity. From this data, 259 new antipsychotic drug candidates were created. New drug candidates were then screened against the battery of models. These 259 NCEs are derived from 5 new chemical genera. Functional group substitutions on these 5 new chemical genera produced all 259 NCEs. All 259 NCEs were determined patentable and patents were filed covering broad composition of matter and other claims for all 259 NCEs. Of the 259 drug candidates, 204 are predicted to have antipsychotic activity and 43 have desirable receptor-binding profiles for atypical antipsychotic activity. Of these 43 atypical antipsychotic drug candidates, 3 were selected for synthesis and *in vitro* and *in vivo* testing. The final selection was based on predicted activities and ease of synthesis. These compounds are shown in **Figure 1**.

The affinities of LMD-00060 and LMD-00076 for multiple G-protein-coupled-receptors were determined by the NIMH Psychoactive Drug Screening Program. *In vivo* microdialysis was conducted for LMD-00076 and LMD-00100t according to the method employed in Kuroki et al., 1999 [1].



RESULTS

Highly accurate categorical and empirical *in silico* models were created for predicting antipsychotic activity, D₂, 5-HT_{2A}, and 5-HT_{1A} receptor affinities, agranulocytosis relative risk, and hERG inhibition. The models represented in **Figure 2** allow assessment of the propensity of a compound to exhibit antipsychotic activity, agranulocytosis risk, and hERG inhibition risk. The graphs in **Figures 3** and **4** represent the ability to assess empirical spectral relationships among compounds that differentiate the ability to exhibit a biological response. **Figure 3** depicts empirical spectral models of binding ability at D₂, 5-HT_{2A}, and 5-HT_{1A} receptors. **Figure 4** depicts empirical spectral models of three limiting side effects of antipsychotic drugs, hERG inhibition, antipsychotic associated weight-gain, and agranulocytosis relative risk.

In Vitro and *In Vivo* Candidate Validation and Characterization

The NIMH Psychoactive Drug Screening Program confirmed greater affinity for 5-HT_{2A} receptors than D₂ receptors for LMD-00060 and LMD-00076 (**Figure 3a** and **Figure 3b**, green dots). Two of these compounds, LMD-00076 and LMD-00100t, were then studied using microdialysis in awake, freely-moving rats to determine their similarity to clozapine and other multireceptor 5-HT_{2A}/D₂ antagonists (**Figure 5**). LMD-00076 produced a modest increase in nucleus accumbens dopamine release that exhibited a ceiling effect where increasing dose did not produce additional transmitter release (**Figure 5b**). LMD-00076 also produced a significant increase in cortical acetylcholine (ACh) efflux, as do direct acting 5-HT_{2A}/D₂ antagonists (**Figure 5c**) [2]. These findings show that LMD-00076 produced neurotransmitter release patterns similar to those of clozapine in a rigorous animal model of atypical antipsychotic activity.

A second compound, LMD-00100t, produced a significant increase in cortical DA (**Figure 5d**), but at the maximum dose tested, did not increase cortical ACh efflux (data not shown). This pattern resembles aripiprazole [2]. LMD-00100t is predicted to be a very weak D₂ antagonist per the D₂ receptor-binding model (K_i value = 1200 nM) and is proportionally less potent *in vivo*. The *in vivo* microdialysis profile of LMD-00100t is like that of the partial D₂ agonist aripiprazole; however, the *in vivo* potency relative to LMD-00076 suggests that it is a weak antagonist. Determination of LMD-00100t *in vitro* receptor-binding affinities should help clarify the specific mechanism. Further *in vivo* and *in vitro* characterizations are ongoing with these initial 3 compounds. Synthesis and testing of additional candidates is currently in preparation.

DISCUSSION

The present project is the first time that **Spectral Modeling™** has been used to design new molecules with predetermined activity profiles. We used it to develop a large series of novel 5-HT_{2A}/D₂ antagonists in a brief period of time (3 months), and at a cost that was significantly less than associated with conventional methods of high throughput screening and directed chemical synthesis. In addition to demonstrating high accuracy *in silico* predictions, this work also extends the ability of **Spectral Modeling™** to predict events for which there are no direct experimental data (e.g. agranulocytosis model based on epidemiological data). This allows modeling of clinical characteristics for which predictive *in vitro* and/or *in vivo* models do not exist.

The compounds studied to date proved to be biologically active in a rigorous model which distinguishes typical from atypical antipsychotic drugs. Further testing of these compounds in classical models of antipsychotic activity and extrapyramidal side effects in laboratory animals is in progress, along with studies of their ability to reverse the effects of phencyclidine on novel object recognition. If clinical testing were to establish the efficacy and safety of these molecules, SDAR modeling should be considered as a highly efficient and cost-effective means of developing novel drug entities for diverse clinical indications.

Disclosures: Bill W. Massey, Neil C. Mitchell, Devin E. Howington, and Dwight Miller are employed by LITMUS Molecular Design.

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Zhu Li, Dan A. Buzatu, Jon Wilkes, and Richard Beger have nothing to disclose.

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