Psychopharmacological Neuroprotection in Neurodegenerative Disease: Assessing the Preclinical Data

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This manuscript reviews the preclinical in vitro, ex vivo, and nonhuman in vivo effects of psychopharmacological agents in clinical use on cell physiology with a view toward identifying agents with neuroprotective properties in neurodegenerative disease. These agents are routinely used in the symptomatic treatment of neurodegenerative disease. Each agent is reviewed in terms of its effects on pathogenic proteins, proteasomal function, mitochondrial viability, mitochondrial function and metabolism, mitochondrial permeability transition pore development, cellular viability, and apoptosis. Effects on the metabolism of the neurodegenerative disease pathogenic proteins alpha-synuclein, beta-amyloid, and tau, including tau phosphorylation, are particularly addressed, with application to Alzheimer's and Parkinson's diseases. Limitations of the current data are detailed and predictive criteria for translational clinical neuroprotection are proposed and discussed. Drugs that warrant further study for neuroprotection in neurodegenerative disease include pramipexole, thioridazine, risperidone, olanzapine, quetiapine, lithium, valproate, desipramine, maprotiline, fluoxetine, buspirone, clonazepam, diphenhydramine, and melatonin. Those with multiple neuroprotective mechanisms include pramipexole, thioridazine, olanzapine, quetiapine, lithium, valproate, desipramine, maprotiline, clonazepam, and melatonin. Those best viewed circumspectly in neurodegenerative disease until clinical disease course outcomes data become available, include several antipsychotics, lithium, oxcarbazepine, valproate, several tricyclic antidepressants, certain SSRIs, diazepam, and possibly diphenhydramine. A search for clinical studies of neuroprotection revealed only a single study demonstrating putatively positive results for ropinirole. An agenda for research on potentially neuroprotective agent is provided.

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This report is the first of a two-part series on psychopharmacological neuroprotection in neurodegenerative disease. Part

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Neurodegenerative diseases including Alzheimer's disease, Parkinson's disease, dementia with Lewy bodies, frontotemporal lobar degeneration (including frontotemporal dementia, semantic dementia, and primary progressive aphasia), Huntington's disease, and amyotrophic lateral sclerosis are important conditions due to their prevalence and impact upon patients, caregivers, and society. As the population ages, age-related neurodegenerative diseases such as Alzheimer's disease and Parkinson's disease will dramatically increase in prevalence. For example, Alzheimer's disease, the most common of these neurodegenerative diseases, currently affects more than 5 million people in the United States and is projected to expand to 16 million by the year 2050.[1] Neurodegenerative diseases further constitute significant sources of morbidity, diminished quality of life, caregiver burden, early nursing home placement, and cost. In Alzheimer's disease, 8.5 billion hours of unreimbursed caregiving are expended annually in the U.S. at the present time,[1] and the cost of care to the average family exceeds $90,000 per year[2] while U.S. societal costs exceed $100 billion per annum.[3,4] Thus, the impact of neurodegenerative diseases upon patients, caregivers, and society is considerable and will expand significantly over the coming years unless treatments to slow or reverse the course of these diseases can be discovered.

Psychotropics are often prescribed to control neuropsychiatric behavioral disturbances in these diseases, including apathy, agitation, aggression, disinhibition, psychosis, depression, anxiety, sleep disturbances, and other conditions (see part two of this report). Evidence of the utility of psychotropics in treating these neuropsychiatric disorders continues to increase (part two), leading clinicians to increasingly prescribe these medicines without an awareness of their impact on neurodegenerative disease cellular pathobiology. The effects of these agents on the course and progression of neurodegenerative diseases are unknown at present. Yet the possibility that these agents may have a salutary impact on the underlying disease deserves attention. It has been calculated that discovery of a treatment that could delay Alzheimer's disease onset by 1 year could lead to 12 million fewer Alzheimer's disease cases by the year 2050.[5]

We summarize recent findings regarding the effects of these psychopharmacological treatments on basic neurodegenerative disease mechanisms. In part one of this report, we confine our consideration to only the key neuroprotective mechanisms summarized below for the panoply of common first-line psychotropics used in clinical practice to treat the behavioral and neuropsychiatric manifestations of neurodegenerative disease. In contrast, in part two, we consider the wider diversity of neuroprotective mechanisms as they apply to representative candidate agents of selected psychotropic classes. Our understanding in this area has greatly advanced over the past several decades. For example, much more is known about the metabolism of beta-amyloid (Aβ) and tau proteins in Alzheimer's disease, and alpha-synuclein (αSyn) in Parkinson's disease and, to some extent, Alzheimer's disease. These proteins ultimately lead to the specific observable pathological features of neurodegenerative diseases, including amyloid plaques and intracytoplasmic inclusions, such as neurofibrillary tangles and Lewy bodies, which are associated with disease progression. These pathogenic proteins inhibit proteasomal function, an enzyme complex responsible for disposing of unwanted or damaged proteins.[6] Ubiquitin is intrinsic to proteasomal protein degradation, and ubiquitin system dysfunction is evident in certain frontotemporal lobar degenerations, *parkin*-related Parkinson's disease, and other neurodegenerative diseases. Proteasomal dysfunction and proteinopathic inclusions have been linked to mitochondrial dysfunction, the result of which can trigger apoptotic preprogrammed cell suicide pathways.[7] Protein accumulation, proteasomal and mitochondrial dysfunction, and consequent apoptosis have each been implicated as important patho-

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genetic mechanisms in neurodegenerative diseases (for a more detailed overview of these processes, see Part 2 of this report).

Apoptosis, preprogrammed cell death, is promoted by a very wide variety of mediators including p21, p38 mitogen-activated protein kinase, c-Jun-NH^2-terminal protein kinase, p53,[8] caspases 2, 3, 8, and 9,[9,10] BCL-XS, Bax, apoptosis inducing factor,[11] and Par-4,[12] whereas B-cell lymphoma/leukemia-2 protein (Bcl-2), Bcl-XL, and other mediators are antiapoptotic.[13] A mitochondrial pathway of apoptosis involves the generation of reactive oxygen species and development of the mitochondrial permeability transition pore, linked to depolarization of the mitochondrial membrane potential and release of cytochrome c from the mitochondrion to the cytoplasm.[14] Caspase 3 is then activated by cytochrome c, promoting a cascade inductive of apoptosis.[15] Nuclear DNA fragmentation is another concomitant of apoptosis.[16]

Preclinical experimental models have employed neural cell lines including neuroblastoma (N2a, dopaminergic SH-SY5Y, SK-H-SH), nigral/neuroblastoma hybrid cell lines (MES 23.5), pheochromocytoma PC12, MES 23.5 dopaminergic cells, cerebellar granule neurons, nucleus basalis cholinergic neurons, a diversity of neural tissues including hippocampus, cerebral cortex, striatum, and also cybrids. Cybrids are cytoplasmic hybrids composed of mtDNA, usually from persons with neurodegenerative disease, transplanted into host cells lacking mtDNA, allowing the study of mitochondrial gene expression that can impact apoptosis and other functions.[17] Common apoptotic mechanisms shared between neurons and other cells provoke interest in non-neuronal cells. To date, non-neuronal cell lines have included Chinese hamster ovaries, canine renal MDCK, J774.1 murine macrophages, human fibroblasts, pituitary tumors (GH3D2L, GH3D2S), and multiple adenocarcinoma lines.

Based on the known molecular pathophysiology and cellular pathobiology of neurodegenerative diseases and the available preclinical observations, we developed a tentative model of candidate agent attributes conferring clinical neuroprotection in neurodegenerative diseases. Specifically, agents likely to demonstrate translational clinical neuroprotection will possess at least one of the following properties: (a) reduce the accumulation of pathogenic proteins, including Aβ, αSyn, and hyperphosphorylated tau; (b) enhance proteasomal function; (c) improve mitochondrial function and viability including mitochondrial respiration and oxidative phosphorylation; (d) reduce free radical concentrations; (e) impede mitochondrial depolarization; (f) prevent development of the mitochondrial permeability transition pore and subsequent release of cytochrome c; (g) enhance cell viability; and (h) deter apoptosis.

We comprehensively review the literature of psychotropic drugs for their effects on the primary proteins implicated in neurodegenerative disease pathology (αSyn, Aβ, and tau), proteasome, mitochondrion, and the process of apoptosis. We review the psychiatric pharmacopoeia but do not include cholinesterase inhibitors and memantine, which have been extensively studied, with disappointing results for neuroprotection in clinical trials. Instead, this article focuses on first-line clinically effective medications used to treat apathy syndromes, personality changes, psychoses, mood disorders, anxiety conditions, and sleep disorders in the context of neurodegenerative disease. The findings from these largely preclinical data (involving in vitro, ex vivo, and nonhuman in vivo findings) are synthesized to provide an indication of whether a specific drug appears of net benefit, disadvantage, or mixed benefit and disadvantage in therapeutically modulating neurodegenerative disease pathobiology. Drugs are reviewed by their therapeutic class and also by their neuroprotective actions and we detail drugs meriting further study, those which cannot be recommended for further study due to significant limiting issues, and those with inadequate data to allow assessment. The scant literature relevant to neuroprotective clinical trials of medications is also summarized. The findings of this review can serve as a basis for the initiation of neuroprotective trials in clinical populations to determine the clinical neuroprotective effects of each medication.

METHODS

A National Library of Medicine PubMed search was conducted October 1, 2007. Specific National Library of Medicine search terms utilized were: alpha-synuclein, beta-amyloid, tau, ubiquitin, proteasome, mitochondrial viability, mitochondria, mitochondrial transition pore, cytochrome c release, leukocyte viability, and apoptosis. We examined the following drugs, which have been routinely applied as first-line symptomatic treatments for neuropsychiatric disorders arising in the con-

text of neurodegenerative disease: apathy treatments (pramipexole, ropinirole, amantadine), antipsychotics (haloperidol, fluphenazine, trifluoperazine, thiothixene, chlorpromazine, thioridazine, risperidone, olanzapine, quetiapine, ziprasidone, aripiprazole, clozapine), mood stabilizers (lithium, carbamazepine, oxcarbazepine, valproate), antidepressants (amitriptyline, imipramine, nortriptyline, desipramine, clomipramine, trimipramine, doxepin, protriptyline, maprotiline, bupropion, fluoxetine, sertraline, fluvoxamine, paroxetine, citalopram, s-citalopram, trazodone, nefazodone, venlafaxine, duloxetine, mirtazapine), anxiolytics/hypnotics (buspirone, diazepam, chlordiazepoxide, flurazepam, temazepam, chlorazepate, clonazepam, lorazepam, oxazepam, alprazolam, zaleplon, zolpidem, zopiclone, s-zopiclone), antihistamines (cyproheptadine, hydroxyzine), anticholinergics (benztropine, trihexyphenidyl), modafinil, melatonin, and ramelteon. In light of the focus of the search on common first-line psychotropics used in clinical practice to treat the behavioral and neuropsychiatric manifestations of neurodegenerative disease, certain drugs were not included in the search. For example, although certain drugs have psychotropic properties, we did not include cognitive enhancers (cholinesterase inhibitors, memantine, nootropics), mono-amine oxidase inhibitors (e.g., selegeline, rasagiline), or other anticonvulsants (e.g., lamotrigine) because these drugs do not meet the criterion of "first-line psychotropics." The search strategy joined terms of interest with specific drugs by means of an "AND" operator. All peer-reviewed articles in the database with English abstracts on or before October revealed by the search were considered; the current review (part one) was limited to articles within the database; part two of this report employed the findings of part one extended by review of bibliographies and other sources including additional National Library of Medicine searches of the literature.

RESULTS

The preclinical investigation of the impact of psychotropic drugs on molecular processes pertinent to neuroprotection varied considerably. For example, regarding dopamine agonists, we identified one paper addressing impact on αSyn, two on Aβ, one on mitochondrial function, eight on the permeability transition pore, and eight on apoptosis. No papers were identified addressing the impact of these drugs on tau, proteasomes, or cell viability. By comparison, with regard to antipsychotics, we identified six papers addressing effects on Aβ, five on tau, four on proteasomes, 28 on mitochondrion, 25 on permeability transition pore, 11 on cell viability, and 45 on apoptosis, yet no papers discussed the impact of these agents on αSyn. Only 10 total papers were identified addressing the effects of all these psychotropics on ubiquitin—one indication of the weakness of the literature in certain areas. In addition, there was considerable variability in the laboratory approaches, models, and assays utilized to examine the impact on a given molecular process. For instance, studies of mitochondrial effects used mouse, rat, or human brain cell cultures, mouse or human heart, liver or endothelial cells, and normal or neoplastic leukocytes. These studies variously assessed oxygen uptake, Complex I, II, IV, or V activity, ATP production, succinate production or succinate dehydrogenase activity, redox reaction velocity, reactive oxygen species production, and/or morphological changes on electron microscopy. Even within the papers that focused on human brain cells, different models used a variety of neuron types including those from brainstem, basal ganglia, cerebellum, and several regions of the cortex. We organized and summarized the available data making no assumptions about relative predictive translational neuroprotective merits of different models and tissues, which are not known at present (see discussion).

The most important detailed findings for each drug are briefly summarized in Table 1, Table 2, Table 3, and Table 4 (located online at http://neuro.psychiatryonline.org/cgi/content/full/22/1/8/DCI). The recently discovered TDP-43 was also considered while this project was underway, but no relevant articles were evident for this protein.

DISCUSSION

It is evident from the above that there is significant variation in degree of investigation, cell lines studied, and methodological approaches. Other limitations include the varying use of neural tissues, variance in the neuronal types studied, use of neuroblastoma lines instead of neurons, study of immature or poorly differentiated cells that may be more prone to apoptosis than more mature cells, and the infrequent characterization of effects on αSyn, tau, and Aβ. Such deficiencies in the

data significantly confound the ability to draw definitive conclusions. In particular, the deficiencies in the data raise the question as to the most valid, clinically relevant, and appropriate standards of evidence to apply in determining which preclinical findings will predictably translate into clinical neuroprotection in patients with neurodegenerative diseases.

A number of concerns impact the selection of an appropriate standard of evidence. First, there are no established general criteria for judging preclinical neuroprotective data across the diversity of neurodegenerative diseases. Second, unlike clinical evidence-based medicine (EBM) standards, there do not appear to be established uniform criteria for judging the diversity of preclinical findings. From an EBM perspective, the data considered here are even less compelling than Class II or IV[18] or Level C[19] clinical case reports since they generally do not pertain to findings in human patients. Third, there are considerable variabilities across the present preclinical findings with respect to intra- and extramodel replication, replications in neural tissue, the specific neural tissues studied, and the specific brain locus even when neurons are consistently studied. These are summarized in Table 5. Fourth, replications are still needed using the same physiological dose range, particularly because some have observed bell—shaped rather than sigmoid—shaped neuroprotective dose—response curves.[20,21] Fifth, some drugs have mixed actions, simultaneously possessing some neuroprotective actions and other neurodegenerative actions. It is not yet clear whether the various actions should receive equal weight or whether one may trump others (for example, effects on apoptotic measures may be more determinative in importance than effects on more "upstream" processes such as mitochondrial potential or proteasomal function). Sixth, there is no gold-standard preclinical model but, instead, a diversity of models that each have their own select benefits and limitations. These and other factors likely contribute to the current disconnect between preclinical findings and neuroprotective clinical trial results.

Some criteria for considering neuroprotective candidate agents have been elaborated in Parkinson's disease[22] and stroke.[23] In Parkinson's disease, scientific rationale, penetration of the blood-brain barrier, safety and tolerability, and efficacy in relevant animal models of the disease or an indication of benefit in human clinical studies constitute criteria.[22] In the case of FDA-approved psychotropics reviewed here, which essentially meet most of these criteria (with the exception of systematic, consistent application in relevant neurodegenerative disease models), the question then becomes: how good is the available preclinical evidence of neuroprotection? Ravina et al.[22] noted that the most problematic issue in Parkinson's disease was evaluating animal data given the many different models that were of uncertain value in predicting results in humans and noted further that a clinical trial would actually be needed to demonstrate the predictive validity of any preclinical model. Similarly, it is not possible to judge the quality of the present preclinical findings by the models used because the predictive validities of the models remain unclear. In stroke,[23] potentially successful drug candidates have been considered to be inferable from preclinical data by the following criteria: (a) adequately defined dose-response relations; (b) time window studies showing a benefit period; (c) adequate physiological monitoring in unbiased, replicated, randomized, blinded animal studies; (d) lesion volume and functional outcome measures determined acutely and at longer term followup; (e) demonstration in two animal species; (f) submission of findings to a peer-reviewed journal. However, even with these criteria, Gladstone et al.[24] have pointed out that translation of preclinical findings to clinical efficacy has been hampered by a lack of functional outcomes, long-term end points, permanent ischemia models, extended time windows, and selective white matter evaluation in preclinical models whereas clinical studies are plagued by insensitive outcome measures, lack of stroke subtype specificity, and inattention to the ischemic penumbra, among other concerns. Ford[25] has also pointed out that a number of compounds fulfilling these stroke neuroprotectant criteria have failed to afford translational clinical neuroprotection. Analogous concerns obtain for neurodegenerative disease preclinical models and clinical methods, particularly whether putative criteria will reliably predict translation to clinical neuroprotection. Additionally, a nearly endless array of clinical variables including gender, age, pharmacogenomics, medical history, coadministered drugs, and other factors may contribute to an inability to predict clinical neuroprotection despite preclinical success. Thus, predictive criteria remain in need of development.

Reflection upon these translational issues in regard to psychotropic neuroprotection in neurodegenerative diseases first suggests the need for replication within and between specific preclinical models in specific neu-

rons at specific loci to elucidate physiological dose-response relations that should then themselves be replicated as a first step. Additionally, other issues seem relevant to the problem of determining which candidate drugs may be most likely to effect clinical neuroprotection. We suggest preliminary neuroprotective drug selection criteria for assessing the likelihood of translational clinical neuroprotection in neurodegenerative diseases (Table 6). These criteria, including preclinical (at least two replicated neuroprotective actions at physiological doses in an established neuroprotective model, neural tissue, and disease-specific animal model in excess of the number of known neurodegenerative actions) and clinical (delayed progression on clinical markers and unexpected benign disease course not accounted for by symptomatic properties) criteria, can be evaluated over time and modified as future data indicate. Given the lack of information regarding the utility of specific preclinical paradigms in predicting clinical neuroprotective effects, it is premature to rank or weight these criteria. Rather, recent concerns[26] notwithstanding and until a better study methodology is developed, we suspect that the greater the number of criteria met by a candidate drug, the greater the likelihood of demonstrating translational clinical neuroprotective efficacy in a randomized, double-blind, placebo-controlled, delayed-start or randomized-withdrawal clinical trial.[27] Such trials are needed because agents deemed promising based upon preclinical data often fail to demonstrate neuroprotection in clinical trials for reasons identified in the above paragraph. At present, preclinical demonstration of replicable neuroprotective effects in neural tissues at clinically-relevant doses does not assure a positive result in a clinical trial, nor does the absence of such evidence necessarily exclude clinical neuroprotective benefits. Until such clinical findings obtain, it is impossible to identify preclinical determinants predictive of translational clinical success and ascertain whether patients are actually being helped or harmed in a neuroprotective sense by the use of these drugs.

Beyond the methodological concerns expressed above, a practical assessment of these preclinical findings is still possible. Given the relative infancy of this field of research, the present state of the literature, the limitations of the data described above, and our current ignorance of preclinical evidence predictive of successful clinical translation, there is the very real possibility of prematurely disregarding findings that may ultimately prove to be of clinical significance with further research (a "type II" error) by applying an overly stringent standard of evidence. It seems that, at the present time, the proper approach is to instead look at the preponderance of the available findings and attempt some generalizations that constitute general impressions to be tested in future research, similar to the process of developing and refining clinical diagnostic criteria. Accordingly, the following observations are drawn from looking at all of the studies, without any exclusions, except where there are clearly contradictory data. As noted, many of the findings have not yet been independently replicated in the same model despite apparent replication in a different model (Table 5). Until the state of the literature develops to the point where independent replications in the same model are routinely observed, appropriate assessment criteria must be very liberal, resulting in conclusions that can only be viewed as preliminary. Adopting this approach with its attending caveats, some preliminary observations can be gleaned from the data. Below, we first consider drugs with respect to their neuroprotective potentials, distinguishing drugs meriting further study from those that have limitations dissuading further investigation and those for which too little data are available to form any conclusions. (We also summarize neuroprotective effects by drug class in Appendix 1 and drugs by neuroprotective actions in Appendix 2 [located online at http://neuro.psychiatryonline.org/cgi/content/full/22/1/8/DCI]; Part 2 of this report focuses on the broader neuroprotective aspects of selected psychopharmacological classes.) Next, we assess the general properties of the various classes of psychotropics. We then consider each investigated cellular function with regard to the drugs that influence them. Finally, we detail a research agenda for drugs of interest and consider the progress made in clinical neuroprotective trials thus far, recommending a next step in their development.

Drugs of Neuroprotective Interest

Drugs meriting further study include pramipexole, thioridazine, risperidone, olanzapine, quetiapine, lithium, valproate, nortriptyline, desipramine, maprotiline, fluoxetine, paroxetine, buspirone, clonazepam, diphenhydramine, and melatonin. These are drugs with at least one significant neuroprotective action and relatively negligible countervailing neurodegeneration—promot-

ing effects, as summarized in Table 1, Table 2, and Table 3 (especially the "Comments" column summarizing the data), and particularly Table 7 (tables located online at http://neuro.psychiatryonline.org/cgi/content/full/22/1/8/DCI).

Drugs that are not recommended for further study at the present time due to more significant limiting issues (see Table 1, Table 2, and Table 3, especially "Comments" column summarizing the data). Haloperidol does not warrant further study because of tau hyperphosphorylation, reduced cell viability, and multiple proapoptotic actions, especially in hippocampus, cortex, striatum, and nigra. Fluphenazine, chlorpromazine, and clozapine, probably do not warrant further study because of multiple proapoptotic actions, and chlorpromazine inhibits tau dephosphorylation. Carbamazepine has variable neuroprotective properties. Oxcarbazepine promotes apoptosis. Clomipramine also generally promotes apoptosis. Diazepam has mixed effects on neural apoptosis, but uncouples oxidative phosphorylation, releases cytochrome c, and promotes apoptosis in a number of neuronal models, although it promoted ATP recovery and prevented cytochrome c release in a single study of ischemic hippocampal slices.

It should be emphasized that there are no convincing clinical data at present to indicate that these drugs are unsafe for clinical use due to neurodegenerative effects, only preclinical evidence to temper enthusiasm for clinical trial application as a neuroprotectant. Until such data become available, the use of these drugs continues to be guided by clinical symptomatic indications. The limiting actions described above are considered to be significant enough to likely detract from an overall neuroprotective effect, making positive findings less likely, hence our inability to recommend them at present. It must also be recognized that some of these limitations still await replication (Table 5), and that it is presently unknown precisely which neuroprotective modes of action are positively and negatively predictive of clinical neuroprotection.

Drugs for Which Limited Data Do Not Allow Recommendations There are currently insufficient data for ropinirole, amantadine, thiothixene, aripiprazole, ziprasidone, amitriptyline, imipramine, trimipramine, doxepin, protriptyline, bupropion, sertraline, fluvoxamine, citalopram, trazodone, nefazodone, venlafaxine, duloxetine, mirtazapine, chlordiazepoxide, flurazepam, temazepam, chlorazepate, lorazepam, oxazepam, alprazolam, zolpidem, cyproheptadine, hydroxyzine, modafinil, ramelteon, benztropine, trihexyphenidyl, and biperiden.

Briefly, regarding the neuroprotective effects of psychopharmacological classes, certain generalizations are apparent (see Appendix 1 for details). There is some evidence to suggest that D2 agonists, lithium, some SSRIs, and melatonin reduce *pathogenic proteins*. D2 agonists, certain atypical antipsychotics and antidepressants, and melatonin suppress *free radical formation*. Neuroleptics, lithium, certain heterocyclic antidepressants, the central benzodiazepine receptor agonist clonazepam, and melatonin inhibit *mitochondrial neurodegenerative events*. D2 agonists, atypical antipsychotics, lithium, antidepressants, the 5HT1a agonist buspirone, and melatonin inhibit *apoptosis*, whereas the peripheral benzodiazepine receptor agonist diazepam promotes apoptosis. These, however, are gross generalizations, which are better explained in Appendix 1 and Appendix 2. Moreover, it is potentially erroneous to project neuroprotective effects upon a pharmacological class because neuroprotective properties may not relate to their currently recognized pharmacodynamic effects.

Above, we have indicated which drugs merit further study, those which cannot be recommended due to significant limiting issues, and those with inadequate data to allow assessment. Among drugs meriting further study, Table 8 discloses the various agents along with evidential weights for their various neuroprotective actions. It can be seen that drugs that inhibit apoptosis and have at least one other general antiapoptotic action (each demonstrated by a net of two or more studies supporting a neuroprotective action, without consideration of their effects on specific proteins) include pramipexole, olanzapine, lithium, desipramine, and melatonin. The remaining agents have less robust findings supporting general neuroprotective actions. Considering the effects of these drugs on proteins and at least one other neuroprotective action in a disease-specific model, the most promising drugs in Alzheimer's disease would include olanzapine, lithium, and melatonin while drugs with less robust support in Alzheimer's disease include pramipexole, quetiapine, valproate, and desipramine. Applying the same criteria, drugs of promise in Parkinson's disease include pramipexole and melatonin, while drugs with less robust support in Parkinson's disease include olanzapine, lithium, valproate, desipramine and clonazepam. Similarly, in Huntington's disease, desipramine is the most promising, with less robust support for lithium, val-

proate, nortriptyline, and maprotiline. There is some support for pramipexole, olanzapine, lithium, and nortriptyline in amyotrophic lateral sclerosis. However, as we have pointed out above, it is premature to draw any clinical conclusions from these data because of the limitations we have described and because more data will be forthcoming.

Directions for Future Research

Given this inability to draw clinical conclusions, we provide the next steps that should be undertaken in developing psychotropic research to the point that results can guide the clinical application of these drugs for neuroprotection. While it is not clear what the most predictive models of clinical neuroprotection are, and what the most important neuroprotective mechanisms are, it is apparent that some drugs are further along in their preclinical research than others. It is also clear that some seemingly paradoxical neuroprotective outcomes are seen, such as modafinil's ability to increase glutamate release and yet reduce glutamate toxicity, and paroxetine's ability to reduce hippocampal Aβ production in Alzheimer's disease transgenic mice despite its anticholinergic properties that would otherwise tend to increase Aβ production. These seeming contradictions point to the need to focus on research findings rather than our current limited theoretical understanding. Thus, we outline the next research steps to be taken to elaborate findings that will move us toward establishing neuroprotective drugs that can be applied by clinicians.

Apathy Treatments It would be of interest to investigate pramipexole in normal neurons, especially dopaminergic and cholinergic neurons.

Pramipexole should be better characterized as to its effects on αSyn, Aβ, tau, and Aβ fibril and oligomer-induced reactive oxygen species formation as well as on the proteasome and on mitochondrial metabolism. It then should be investigated in clinical neuroprotection paradigms in neurodegenerative disease, particularly Parkinson's disease.

The next step for amantadine involves investigations in neurons.

Antipsychotics Risperidone needs more study to determine its neuroprotective potential. Its ability to reduce Complex I activity in regions of the brain, albeit not in the midbrain, indicates the need for further research as to its long-term safety in neurodegenerative diseases affecting the hippocampus, frontal lobe, and striatum, including Alzheimer's disease, frontotemporal lobar degeneration, and Huntington's disease. Clinical effects tend to contraindicate its use in Parkinson's disease.

Although olanzapine should be better characterized as to its multiple neuroprotective effects (especially on the proteasome and mitochondrial permeability transition pore development), antimuscarinic and parkinsonian clinical properties argue against its application in Alzheimer's disease and Parkinson's disease.

Quetiapine should be better characterized as to its effects on αSyn, Aβ, tau, the proteasome, and protection against rotenone toxicity. Further studies using Aβ and initial studies using MPP+ should be carried out, with subsequent disease-modification studies in Alzheimer's disease and Parkinson's disease if the preceding studies indicate safety, although antihistaminic and anticholinergic clinical properties can constitute a limitation to use in Alzheimer's disease.

Trifluoperazine, chlorpromazine, and thioridazine might be further studied in situations where inhibition of mitochondrial permeability transition pore development is of utility.

Aripiprazole and ziprasidone should be studied for their neuroprotective properties, given their low proclivities to induce extrapyramidal side effects in people with neurodegenerative disease.

Mood Stabilizers Lithium should be studied for neuroprotection in patients with Parkinson's disease, Huntington's disease, amyotrophic lateral sclerosis, and cerebral ischemia. A clinical trial in Alzheimer's disease is currently under way.

Investigation of valproate's ability to induce mitochondrial permeability transition pore development but not mitochondrial membrane depolarization or cytochrome c release may yield information that may help develop neuroprotective mitochondrial strategies.

Valproate might be investigated in patients with Parkinson's disease and oncological diseases for its antiapoptotic effects in the former and proapoptotic effects in microglia and the latter. Valproate's ability to increase αSyn concentrations may be either beneficial or detrimental in Parkinson's disease and other synucleinopathies, and further research is needed. Activated microglia appear to be of importance in neurodegenerative diseases, especially Alzheimer's disease. Results

of a recent clinical trial in Alzheimer's disease are not yet available.

Antidepressants Desipramine, nortriptyline, and maprotiline should be studied in other models of Huntington's disease. If effective, they might be tried in other neurodegenerative disease models and in depressed patients with Huntington's disease. Nortriptyline's effects in Huntington's disease yeast and amyotrophic lateral sclerosis mouse models deserve replication.

Fluoxetine has inhibited neural stem cell apoptosis, hippocampal apoptosis in newborn mice and rats and serotonin-induced apoptosis. Although it has some pro-apoptotic properties, fluoxetine should be studied further as a neuroprotectant in Alzheimer's disease.

Paroxetine should be studied further for neuroprotective properties, especially in regard to reductions in Aβ and hyperphosphorylated tau.

Anxiolytics and Hypnotics Buspirone has inhibited apoptosis in several neuronal models and now deserves study in regard to other related characteristics. If further studies indicate safety, studies in patients with neurodegenerative disease should then be undertaken.

Which types of GABA-A agonists protect against Aβ neurotoxicity and which do not requires clarification.

Clonazepam should be studied further for its restorative properties in Complex I deficiency, and should be better characterized in regard to apoptotic effects in neuronal models, especially on frontal lobe apoptosis in mature animals. If favorable results are forthcoming, it might then be tried in patients with neurodegenerative disease, especially Parkinson's disease, although its association with falls in the elderly is a limitation.

Diphenhydramine should be further characterized in inflammatory, malignant, hypoxic, and other models where histamine plays a role.

Melatonin might now be investigated in patients with Alzheimer's disease and in those with Parkinson's disease.

Comprehensive Strategies

Deficiencies detailed in Table 5 deserve to be addressed in future studies. Validation of Table 6 translational predictive criteria awaits investigation. The relative predictive weightings of the various criteria also await outcome studies.

Combination therapies of psychotropics with differing profiles of neuroprotective actions may yield greater clinical impact than monotherapies. These varying profiles are depicted in Table 8. For example, across neurodegenerative diseases, the combination of lithium and melatonin might provide neuroprotective synergies, as might pramipexole, olanzapine, lithium, and nortriptyline in amyotrophic lateral sclerosis, lithium, and desipramine in Huntington's disease, and pramipexole, lithium, desipramine, and melatonin in Alzheimer's disease (Table 8). In Alzheimer's disease, lithium and melatonin together might synergize efficacy at Aβ, hyperphosphorylated tau, reactive oxygen species, transition pore development, and apoptosis, with lithium perhaps improving ubiquitylation. In Parkinson's disease, this combination plus pramipexole may synergize benefits to reactive oxygen species, transition pore, and apoptosis, with lithium perhaps improving ubiquitylation and pramipexole and melatonin perhaps synergizing efficacy on αSyn. It should be remembered, however, that some combination therapies, applied in cancer chemotherapy, have sometimes resulted in a reduced efficacy of all drugs and an increase in side-effects.[28] Animal trials of proposed combinations would be a first step in evaluating their safety and efficacy.

Progress Thus Far: Clinical Trials

So far, some preliminary progress has been made in identifying the clinical neuroprotective properties of some of these agents. A search performed on October 9, 2007 using the search terms "randomized clinical trial AND (neuroprotection OR disease-modifying OR disease-modification OR disease modifying OR disease modification) for each drug revealed only one clinical neuroprotection study (ropinirole versus L-dopa), and two studies evaluating glutathione reductase and a gamma interferon, relevant to disease progression, but without evaluating actual indices of clinical neuroprotection. A 6-^{18}F-fluorodopa PET study of 186 patients with Parkinson's disease randomized to either ropinirole or L-dopa revealed a significant one third reduction in the rate of loss of dopamine terminals in subjects treated with ropinirole.[29] A study of valproate plus placebo versus valproate plus melatonin in patients with epilepsy demonstrated a significant increase in glutathione reductase in the melatonin group, but no clinical indices of actual neuroprotection were evaluated in that study.[30] A study in patients with relapsing-remitting multiple sclerosis identified a relationship between ser-

traline treatment of depression and attenuation of proinflammatory cytokine IFN-gamma, but again, actual indices of clinical neuroprotection were not assessed.[31] In addition to the findings of the search, the CALM-Parkinson's disease study involving the dopamine agonist pramipexole in Parkinson's disease found faster progression (or at least less improvement on total UPDRS score) but slower dopamine transporter signal loss than with L-dopa over 46 months,[32] although the study has been criticized for lack of a placebo, group heterogeneity, and confounding influences on dopamine transporters. In contrast, a 2-year study of ropinirole found no significant difference in fluorodopa uptake compared to L-dopa treatment (−13% versus −18%).[33]

A search of clinical trials (www.clinicaltrials.gov) on October 9, 2007 using the terms (neuroprotection OR disease-modifying OR disease-modification OR disease modifying OR disease modification) and neurodegenerative diseases revealed only a few studies in progress. These included pramipexole in amyotrophic lateral sclerosis, early versus delayed pramipexole in Parkinson's disease, and valproate in spinal muscular atrophy. Since that time, as of February 1, 2009, additional studies have been registered. In Alzheimer's disease, these include a short-term study of CSF tau epitopes with lithium, brain volume and clinical progression with valproate, and hippocampal volume, brain volume, and clinical progression with escitalopram. In frontotemporal dementia, there is a single study of CSF and brain volume with quetiapine versus D-amphetamine. In Huntington's disease, there is a study of CSF BDNF levels with lithium versus valproate. In dementia with Lewy bodies and Parkinson's disease dementia (PDD), there is a study of clinical progression with ramelteon. In Parkinson's disease, there is a study of striatal dopamine transporter by β-CIT SPECT with pramipexole versus *L*-dopa while an 8 year study of disability with pramipexole has been terminated. Only the spinal muscular atrophy and dementia with Lewy bodies/PDD studies employ clinical neuroprotective designs (delayed-start paradigm), and the validity of biomarker correlates, particularly dopamine transporter measures in Parkinson's disease, continues to be studied.

The discussion above relies on multiple investigative approaches using a number of different psychotropics in a variety of models and a diversity of cell lines. A major caveat is that preclinical results do not necessarily translate into clinical realities. For example, favorable preclinical findings for the neuroprotectant minocycline exist in Parkinson's disease, amyotrophic lateral sclerosis, Huntington's disease, stroke, spinal cord injury, and MS models, but a recent phase III trial in patients with amyotrophic lateral sclerosis was halted because of a 25% faster rate of neurological progression with the active drug than with placebo.[34] Nevertheless, some generalizations seem possible at this stage. The considerations above are offered in hopes of stimulating the identification and development of pharmaceuticals that are useful both for symptomatic improvement and for long-term neuroprotection in neurodegenerative disease. Pursuit of the directions for research suggested above may contribute to that development.

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Part II will further explore heuristic clinical applications of psychopharmacological neuroprotection in neurodegenerative disease. Look for it in the Spring 2010 issue of The Journal of Neuropsychiatry and Clinical Neurosciences, *available at http://neuro.psychiatryonline.org/.*

Full text of all data tables and appendices referenced in this article, along with the complete 548 references, accompanies the online edition of this issue at http://neuro.psychiatryonline.org/cgi/content/full/22/1/8/DCI.

References

1. Alzheimer's Association: 2007. Alzheimer's Disease Facts and Figures. Chicago, Alzheimer's Association, 2007
2. Rice DP, Fillit HM, Max W, et al: Prevalence, costs, and treatment of Alzheimer's disease and related dementia: a managed care perspective. Am J Manag Care 2001; 7:809–818
3. Meek PD, McKeithan K, Schumock GT: Economic considerations in Alzheimer's disease. Pharmacotherapy 1998; 18:68–73
4. Silverman DH, Small GW, Chang CY, et al: Positron emission tomography in evaluation of dementia: regional brain metabolism and long-term outcome. JAMA 2001; 286:2120–2127
5. Brookmeyer R, Johnson E, Ziegler-Graham K, et al: Forecasting the global burden of Alzheimer's disease. Alzheimers Dement 2007; 3:186–191

6. Layfield R, Cavey JR, Lowe J: Role of ubiquitin-mediated proteolysis in the pathogenesis of neurodegenerative disorders. Ageing Res Rev 2003; 2:343–356
7. Feany MB: New genetic insights into Parkinson's disease. N Engl J Med 2004; 351:1937–1940
8. Kharbanda S, Yuan ZM, Weichselbaum R, et al: Determination of cell fate by c-Abl activation in the response to DNA damage. Oncogene 1998; 17:3309–3318
9. Friedlander RM: Apoptosis and caspases in neurodegenerative diseases. N Engl J Med 2003; 348:1365–1375
10. Zhang JH, Zhang Y, Herman B: Caspases, apoptosis and aging. Ageing Res Rev 2003; 2:357–366
11. Le Bras M, Rouy I, Brenner C: The modulation of inter-organelle cross-talk to control apoptosis. Med Chem 2006; 2:1–12
12. El-Guendy N, Rangnekar VM: Apoptosis by Par-4 in cancer and neurodegenerative diseases. Exp Cell Res 2003; 283:51–66
13. Kim R: Unknotting the roles of Bcl-2 and Bcl-xL in cell death. Biochem Biophys Res Commun 2005; 333:336–343
14. Orrenius S, Gogvadze V, Zhivotovsky B: Mitochondrial oxidative stress: implications for cell death. Annu Rev Pharmacol Toxicol 2007; 47:143–183
15. Lemasters JJ, Qian T, Trost LC, et al: Confocal microscopy of the mitochondrial permeability transition in necrotic and apoptotic cell death. Biochem Soc Symp 1999; 66:205–222
16. Zhang JH, Xu M: DNA fragmentation in apoptosis. Cell Res 2000; 10:205–211
17. Swerdlow RH: Mitochondrial DN: A-related mitochondrial dysfunction in neurodegenerative diseases. Arch Pathol Lab Med 2002; 126:271–280
18. Shevell M, Ashwal S, Donley D, et al: Practice parameter: evaluation of the child with global developmental delay: report of the quality standards subcommittee of the American Academy of Neurology and the Practice Committee of the Child Neurology Society. Neurology 2003; 60:367–380
19. MDS Task Force: Management of Parkinson's disease: An evidence-based review: introduction. Mov Disord 2002; 17: S1–S6
20. Maurice T, Su TP, Privat A: Sigma1 (sigma 1) receptor agonists and neurosteroids attenuate B25–35-amyloid peptide-induced amnesia in mice through a common mechanism. Neuroscience 1998; 83:413–428
21. Soriano FX, Papadia S, Hofmann F, et al: Preconditioning doses of NMDA promote neuroprotection by enhancing neuronal excitability. J Neurosci 2006; 26:4509–4518
22. Ravina BM, Fagan SC, Hart RG, et al: Neuroprotective agents for clinical trials in Parkinson's disease: a systematic assessment. Neurol 2003; 60:1234–1240
23. Stroke Therapy Academic Industry Roundtable: Recommendations for standards regarding preclinical neuroprotective and restorative drug development. Stroke 1999; 30:2752–2758
24. Gladstone DJ, Black SE, Hakim AM: Heart and Stroke Foundation of Ontario Centre of Excellence in Stroke Recovery: toward wisdom from failure: lessons from neuroprotective stroke trials and new therapeutic directions Stroke 2002; 33:2123–2136
25. Ford GA: Clinical pharmacological issues in the development of acute stroke therapies. Br J Pharmacol 2008; 153(suppl 1): S112–S119
26. Clarke CE: Are delayed-start design trials to show neuroprotection in Parkinson's disease fundamentally flawed? Mov Disord 2008; 23:784–789
27. Mani RB: The evaluation of disease modifying therapies in Alzheimer's disease: a regulatory viewpoint. Stat Med 2004; 23:305–314
28. Tol J, Koopman M, Cats A, et al: Chemotherapy, bevacizumab, and cetuximab in metastatic colorectal cancer. N Engl J Med 2009; 360:563–572
29. Whone AL, Bailey DL, Remy P, et al: A technique for standardized central analysis of 6-(18)F-fluoro-L-dopa PET data from a multicenter study. J Nucl Med 2004; 45:1135–1145
30. Gupta M, Gupta YK, Agarwal S, et al: A randomized, double-blind, placebo controlled trial of melatonin add-on therapy in epileptic children on valproate monotherapy: effect on glutathione peroxidase and glutathione reductase enzymes. Br J Clin Pharmacol 2004; 58:542–547
31. Mohr DC, Goodkin DE, Islar J, et al: Treatment of depression is associated with suppression of nonspecific and antigen-specific T(H)1 responses in multiple sclerosis. Arch Neurol 2001; 58:1081–1086
32. Parkinson Study Group: Dopamine transporter brain imaging to assess the effects of pramipexole vs levodopa on Parkinson disease progression. JAMA 2002; 287:1653–1661
33. Rakshi JS, Pavese N, Uema T, et al: A comparison of the progression of early parkinson's disease in patients started on ropinirole or L-dopa: an 18F-dopa PET study. J Neural Transm 2002; 109:1433–1443
34. Gordon PH, Moore DH, Miller RG, et al: Efficacy of minocycline in patients with amyotrophic lateral sclerosis: a phase III randomised trial. Lancet Neurol 2007; 6:1045–1053