NIDA NATIONAL INSTITUTE ON DRUG ABUSE

National Institute on Drug Abuse

REPORT

From the NIDA Science of Genetics Council Review Work Group

May 2010

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SCHOOL OF
MEDICINE

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March 30, 2010

Nora D. Volkow, M.D., Director
National Institute on Drug Abuse
6001 Executive Boulevard
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Dear Dr. Volkow:

I am pleased to transmit the report and recommendations of the Science of Genetics Council Review Work Group that was created at your request by the National Advisory Council on Drug Abuse in 2009. The report and recommendations reflect the unanimous view of the Work Group members. We take full responsibility for the contents. We remain available to meet with you and/or members of your staff to discuss our conclusions and recommendations, although we hope the report makes our views and thinking clear on its own.

The Work Group was impressed with the dedication and knowledge of NIDA's extramural staff in helping us carry out this review. Nevertheless, as you will see, the Work Group recommends more top-down oversight of the human genetics portfolio at NIDA, with a strengthened NIDA Genetics Coordinating Committee (NGCC) more directly involved in funding decisions for any grant involving human genetics research across all NIDA Divisions. Such a role is necessary to ensure that NIDA's mission in human genetics is executed efficiently and that all human genetics research (large or small) that receives support from NIDA will adhere to the most stringent scientific standards.

The members of the Work Group and I would like to thank Denise Pintello, Ph.D., M.S.W., for her terrific and vital support throughout the process. She helped immensely by monitoring and driving the Work Group's progress and, along with Dr. Robert Katt, played a major role in editing the draft report. It was also a pleasure to work with Dr. Katt, who is an outstanding scientific writer. Thank you for this opportunity to support NIDA's mission.

Sincerely,

Eric J. Nestler

Eric J. Nestler, M.D., Ph.D.

National Institute on Drug Abuse

Science of Genetics Review

Science of Genetics
Review Work Group

May 2010

Table of Contents

EXECUTIVE SUMMARY III

INTRODUCTION: THE WORK GROUP'S REVIEW PROCESS 1

OVERVIEW OF GENE FINDING IN ADDICTION 2

Evidence for the Heritability of Addiction *2*

Genetic Tools to Identify Addiction Vulnerability Genes *4*

Deep Phenotyping in Addiction *6*

FINDINGS AND RECOMMENDATIONS 7

REFERENCES 12

Cited References *12*

Background Reading on Gene Finding in Addiction *12*

APPENDIX A: SCIENCE OF GENETICS REVIEW WORK GROUP 14

APPENDIX B: SCIENCE OF GENETICS REVIEW WORK GROUP MEETING, NOVEMBER 2009 15

APPENDIX C: SCIENCE OF GENETICS REVIEW WORK GROUP MEETING, JANUARY 2010 17

EXECUTIVE SUMMARY

The Science of Genetics Review Work Group was convened to evaluate the human genetics research portfolio of the National Institute on Drug Abuse (NIDA), to provide input to fortify NIDA's current human genetics research program through examining a range of emerging scientific opportunities, and to recommend how best to maximize the Institute's scientific investment in human genetics research. The Work Group met twice in convened meetings and interacted between and after its meetings via email and telephone conversations.

The Work Group's background review of NIDA's portfolio also serves as an overview of the state of knowledge about the genetic contributions to substance abuse and addiction. The review summarizes the evidence for the heritability of addiction and the role that current and emerging genomic methods are playing or are likely to play in identifying addiction vulnerability genes. In particular, the Work Group stresses the emerging complementarity of genome-wide association studies (GWAS) with next-generation (or deep) sequencing technologies. Deep phenotyping is discussed as an option to potentially improve the ability to identify genes associated with substance abuse and addiction disorders.

Because the available technologies are changing so rapidly, the Work Group has not specified which approaches are most appropriate to NIDA's mission or how much to invest in each. Rather, NIDA should keep abreast of new genomic technologies as they emerge. Since it is not yet known which approach(es) will be most useful for explicating the genetics of substance abuse and addiction, a balance among different approaches is needed. The Work Group's recommendations list a number of such approaches to be balanced in the portfolio.

To provide a balance of investments in research on the genetic underpinnings of substance abuse and addiction, the Work Group recommends that future investments in GWAS and other genome-wide approaches focus on other drugs of abuse in addition to nicotine, for which there is already a rich supply of early data. However, even for nicotine addiction, it is likely that additional studies will be needed as technical advances are made. To achieve a desirable balance, NIDA should develop a more coherent overall strategy for the range of substances of interest.

Given advances already made in deep sequencing methods, NIDA should consider funding deep sequencing of a select group of genes that are most proximal to the immediate effects of drugs of abuse or their metabolism. Deep sequencing may also be appropriate for genes that have been substantially implicated in the pathophysiology of substance abuse and addiction in animal models and human investigations.

Whether deep phenotyping will prove to be an important ingredient to successful gene finding efforts in substance abuse and addiction remains an open question—on which there were differences even among the Work Group members. Some members advocated that NIDA should move beyond the crude phenotypes of disease presence/absence or the level of substance consumption to include longitudinal phenotypes for the behaviors of substance abuse and addiction as well as phenotypes based on intermediate "biomarkers." Others were more skeptical and questioned whether such phenotyping was feasible for the large number of subjects needed for significant genetic analysis. Overall, the Work Group supported a balanced approach and agreed that guidelines must be established and disseminated to govern genetic studies in the context of deep phenotyping to ensure that the most rigorous genetic standards be

applied to such investigations. The Work Group strongly supported so-called genotype-driven deep phenotyping, whereby positive genetic findings are followed up aggressively to understand how a particular genetic variation alters neural and brain function to contribute to an addiction-related phenotype. The highest scientific standards required for human genetics research should, of course, apply to all such research. The Work Group's recommendation on this topic lists four study design elements for which such guidelines are needed.

NIDA should increase support for training in biostatistics, statistical genetics, and related bio-informatics disciplines—key support disciplines in which personnel shortages are already a constraint. Otherwise, as research in human genetics of substance abuse and addiction expands, shortages of expertise in these disciplines will further constrain progress and jeopardize the validity of published results. Higher priority should also be given to funding grants that support development of the statistical tools needed to optimally analyze the incoming genomic and phenotypic data.

Better top-down oversight of all NIDA grants that involve human genetics analyses—across all extramural divisions—is needed to ensure that rigorous scientific standards are used and that NIDA's strategic plan in human genetics is executed with maximal efficiency. In this regard, the effectiveness and reach of the NIDA Genetics Coordinating Committee (NGCC) should be strengthened substantially.

The Work Group recommends that a strong human genetics component be developed in the NIDA Intramural Research Program. This effort should be carefully coordinated with ongoing extramural research to avoid redundancy. It should emphasize aspects of human genetics research for which an in-house program is particularly well suited, such as longitudinal studies of highly informative populations and high-risk studies that require stable funding over a longer period than the typical research grant.

Given the high rates of comorbidity for substance abuse with other mental disorders, a great deal can be gained from greater collaborative efforts across several institutes and centers of the National Institutes of Health. These collaborations should include sharing of samples and repositories, large-scale phenotyping, and greater coordination of research initiatives, including joint funding of R01 grants examining conditions and risk factors common to these disorders.

With respect to extramural research, the Work Group makes the following across-the-board recommendations:

- Extramural grants with significant human genetic components should be reviewed by study sections that include the requisite expertise in genetics.
- Mechanisms are needed to encourage and/or require more data-sharing among NIDA (and other Institute) grantees on those project components involving human genetics data. This might include combining DNA samples and phenotyping efforts to achieve the number of subjects and depth of genotyping and phenotyping that are required to find addiction vulnerability genes.

Finally, NIDA should consider how to facilitate research on translating findings on human genetics of substance abuse and addiction: to develop improved diagnostic tests and treatments, to inform health care and public policy, and to improve clinical and community interventions for both prevention and treatment of substance abuse.

INTRODUCTION: THE WORK GROUP'S REVIEW PROCESS

In November 2009, the Director of the National Institute on Drug Abuse (NIDA), Nora D. Volkow, M.D., convened the Science of Genetics Review Work Group to evaluate NIDA's genetics research portfolio. The Work Group was asked to produce a written report with the following content:

1. A background review of the current genetics program portfolio.
2. Input into fortifying the current genetics program research mission with an emphasis on examining a range of newly developed scientific opportunities, including genome-wide association studies (GWAS), next generation (i.e., deep or high-throughput) sequencing of portions of the genome, and whole genome sequencing.
3. Recommendations as to how to best maximize the Institute's scientific investment in the genetics portfolio and help identify additional scientific approaches in which NIDA should be investing.

At its initial meeting in November 2009, the Work Group heard presentations from the Division of Basic Neuroscience and Behavioral Research (DBNBR), the Division of Clinical Neuroscience and Behavioral Research (DCNBR), the Division of Epidemiology, Services and Prevention Research (DESPR), the Division of Pharmacotherapies and Medical Consequences of Drug Abuse (DPMCDA), and NIDA's Intramural Research Program. Major topics of discussion included the collaborations across the NIDA divisions in genetics research, collaborations across the National Institutes of Health (NIH), the NIDA Genetics Consortium with its repository of samples from subjects in NIDA-funded studies, the NIDA Phenotyping Consortium, and NIDA's Genes, Environment, and Development Initiative. The Work Group discussed with the NIDA presenters and other NIDA staff such issues as the appropriate role and balance for genetics research approaches ranging from GWAS to deep sequencing of salient genes, phenotyping studies informed by well-established genotypes, and the effects of genetic variations on the molecular biology of the brain.

At the second meeting on January 25, 2010, the Work Group met briefly with the division directors of DESPR, DPMCDA, and DBNBR. The Work Group Chair reported on telephone discussions he had prior to the meeting with the director of DCNBR and the director of the NIDA Intramural Research Program (IRP). The Work Group also heard presentations on human genetics research from representatives of the National Institute of Mental Health (NIMH) and the National Human Genome Research Institute (NHGRI). The remainder of the meeting was spent in executive session to discuss the topics to be addressed in this report. The set of themes from the November meeting was reviewed and revised to develop a working draft of tentative recommendations and supporting rationale.

After the second meeting, the Work Group continued to interact via email and telephone conversations to elaborate, refine, and reach consensus on the findings and recommendations presented here.

OVERVIEW OF GENE FINDING IN ADDICTION

To respond to the request for an overview of NIDA's genetics portfolio and to set the context for the findings and recommendations in the next section, the Work Group has synthesized a high-level perspective on what is known about the genetics of substance abuse and addiction. This summary of the state of knowledge leads naturally to the formulation of directions and emphases for the next stage of research. Selections from the peer-reviewed literature that generally support this overview are listed at the end of the report as background reading.

Evidence for the Heritability of Addiction

Studies conducted over the past 15 years have provided strong evidence, initially from family studies and then from a series of large-scale clinical and population-based twin studies, that genetic factors are an important influence in the initial decision to use psychoactive substances. They play an even stronger role in the subsequent progression from substance use to abuse or addiction. While most studies find that familial-environmental factors affect the decision to use psychoactive substances initially, the impact of these factors attenuates or disappears in the processes that lead to subsequent abuse or addiction.

Several subsequent developments have further clarified the nature of these risk factors, using a widening array of more sophisticated structural equation modeling approaches applied to twin data. First, while some sharing has been seen between genetic risk factors for initiation of substance use and abuse/addiction, investigations have consistently shown that—given initiation of substance use—a unique set of genetic risk factors affects the probability of progression to abuse/addiction. Second, most of the genetic risk factors for illicit substance abuse/addiction appear to be shared across individual substances. That is, the expectation for prominent substance-specific genetic contributions (e.g., genes that influence cannabis versus cocaine versus opiate abuse/addiction) has been disproved in epidemiological samples.

Third, when substance use is expanded to include three legal psychoactive substances—caffeine, nicotine, and ethanol—the picture becomes more complicated. Genetic factors specific to legal versus illicit substances were identified but they were quite highly intercorrelated. While the illicit substances were strongly related to a common genetic factor, the pattern of the legal substances was more complex. Caffeine dependence, in particular, while heritable, shared little of its genetic risk with other disorders. A substantial proportion of the genetic risk factors for nicotine dependence were also unique to that substance. Fourth, a series of studies have shown that the genetic risks for substance use and abuse/addiction are situated within a broader context of risks for so-called externalizing disorders. Evidence from a range of twin and family investigations shows that externalizing traits, such as conduct and antisocial personality disorders, share a substantial proportion of their genetic risk factors with psychoactive substance use and misuse.

Finally, studies of the genetic risk factors for substance use, abuse, and addiction have begun to place them within their appropriate developmental context. Evidence is accumulating for gene-environment correlations in which genetic risk factors for substance abuse/addiction are partly expressed by individuals placing themselves into high risk environments. Evidence for other forms of gene-environment interactions is also growing. For example, heritability for drug use and misuse are higher in environments that have low levels of social restriction and easy access to substances of abuse. The extent to which these genetic risk factors for substance use and abuse/addiction are stable over development remains an open question.

Approaches to locating and identifying the individual genes that affect risk for psychoactive substance use and misuse have included all of the methods used with other complex biomedical and behavioral traits: linkage analysis, candidate gene association studies, and most recently GWAS and other genome-wide methods, which are reviewed below.

As with other complex traits, linkage and candidate gene studies have met with only limited success and replication has been a critical problem. One striking exception to this general pattern has been the findings in the alpha3-alpha5-beta4 cluster of nicotinic cholinergic receptor subunits on chromosome 15q, which, while found initially via a candidate gene approach, have been rapidly and robustly replicated by both further candidate-gene and GWAS approaches.

In parallel with the study of the genetic component in other complex disorders, efforts in the addiction field have increasingly focused on GWAS; an approach best suited for disorders where the etiology is unknown and where it is best to interrogate the entire genome without a priori hypotheses. It is still too early to tell how successful this method, and more advanced genome-wide approaches (whole exome, whole genome sequencing), will be for drugs of abuse. Sample size of collections has varied dramatically, being largest for nicotine dependence, mainly because of its high frequency and ready availability. In addition, many large scale surveys conducted for other reasons (e.g., cardiac disease) have included information on smoking behaviors. The quality of phenotypic information available in many GWAS studies has varied and is often modest. For example, large scale studies on smoking behavior have utilized measures like cigarettes per day. The prospects for more-refined ("deeper") phenotyping approaches are covered below. Moreover, focused sample collections of some important drugs of abuse (e.g., cannabis) remain very small in comparison to their widespread use in the population.

Initial advances have been made in developing the statistical and conceptual infrastructure needed to support the upcoming avalanche of genomics data from advances in molecular genetics, but many questions remain. One important debate is the nature of the correct control group. Are general population samples adequate, or do the controls need to have had adequate exposure to the substance to have developed problematic use if they were so predisposed? The latter type of control allows the study design to focus on the genetic component in progression to addiction, which is a very important consideration clinically. Given that we know much more about the biology of substance abuse than about many other psychiatric disorders, how can we put that information to best use in gene discovery? How can we use modern psychometric tools to extract maximal information from the currently available samples (instead of just reducing all the collected information to dichotomies of affected versus unaffected)? How can we best handle the high levels of comorbidity commonly seen in substance use disorders involving both multiple drugs of abuse and the close relationships between drugs of abuse and externalizing psychiatric disorders? As noted above, twin studies suggest that we will find some genetic variants that convey risk specific to individual substances, others that will be nonspecific with respect to a particular drug of abuse, and still others that will contribute risk to drug use and other behavioral disorders. What designs will allow us to best unravel this likely pattern of findings? How can we best combine the strengths and limitations of clinical versus epidemiological sampling in genetic studies of drug abuse? With these studies, should we focus largely on very large sample collections with minimal phenotypic information or smaller samples with rich phenotypic assessment and assessment of critical environmental risk factors? How important will gene x environment interaction and correlation be in deciphering the pathway from

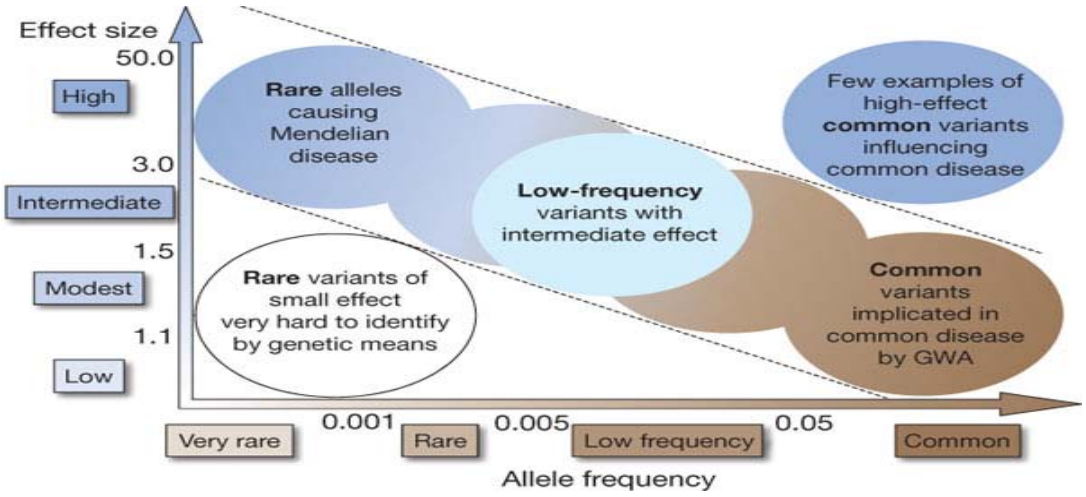
molecular variants to risk for substance use and abuse/addiction? These questions represent the range of issues that must be addressed by the field as it moves forward.

The next generation of molecular investigations of drug addiction, which involves large scale sequencing of selected candidate genes, all exons in the genome, or eventually whole genomes, has only barely begun. Much work remains to consider the optimal combination of these approaches that will maximize the likely return on research investments and identify, at long last, genes that confer risk to substance use, abuse, and addiction.

Genetic Tools to Identify Addiction Vulnerability Genes

Genomic methods to find addiction-related genes would ideally be tailored to the specific types of risk alleles expected. However, the a priori knowledge about the precise nature of those alleles remains limited. As highlighted in the figure below from Manolio et al. (2009), risk alleles of primary interest are likely to be found along the diagonal, and a variety of different methods will be effective in uncovering them. The following is a brief overview of the methods currently used to identify addiction-related genes. Much of this work has been based on growing experience in genetic studies of other complex medical syndromes.

Feasibility of identifying genetic variants (from Manolio et al. 2009)



Association of an addiction-related phenotype with a common genetic variant is now detectable by most commercially available GWAS platforms. These platforms genotype roughly 500,000 to 1 million single nucleotide polymorphisms (SNPs), based on SNPs ascertained through the International Haplotype Mapping Project. With imputation they can cover 85–90% of the common variants found in European populations (coverage will be somewhat lower for non-European populations). Given large enough patient samples, effect sizes from low to high can be detected. One caveat of current GWAS platforms is that certain putative candidate genes of interest may not be included on such platforms, which means the failure to detect such genes in GWAS is not surprising.

Association of an addiction-related phenotype with a low-frequency genetic variant will be detectable on the so-called second-generation GWAS chips. These chips are expected to genotype up to 5 million SNPs and will be based on information derived from a variety of sources including the 1000 genomes project. They will not be disease-specific. The lower boundaries of effect size will be detected by sample size, with modest to high effect sizes expected to be readily detectable.

Association of an addiction-related phenotype with a rare genetic variant is detectable by array-based methods as well as by next-generation sequencing methods. Array-based methods require advance knowledge of SNPs. In contrast, next-generation sequencing can be used as a discovery and association tool. The current balance favors array-based methods based on price and efficiency; however, this will change as deep sequencing becomes ever more affordable over the next few years. Very rare or family-specific (so-called private) mutations will be detectable only by next-generation sequencing in large sample sizes.

Next-generation sequencing technologies are evolving rapidly. Exome sequencing, for an individual gene of interest or for the entire genome (whole exome), is a viable strategy when mutations are expected in the coding regions of genes. The primary advantage of this approach is that the exome comprises only about 1% of the genome and thus is less expensive and time consuming than sequencing noncoding regions of a given gene or the entire genome. For example, a whole exome sequence can now be obtained for a few thousand dollars per individual, while a whole genome sequence is at least an order of magnitude more expensive.

However, there are at least two important limitations to the whole exome approach. First, only coding sequence is interrogated, thus missing 99% of the genome which is composed of regulatory regions, introns and inter-genic areas. There is no reason to assume a priori that disease-causing mutations are restricted to coding regions. Whole exome sequencing will also fail to detect certain copy number variations (CNVs), which have increasingly been found to be a risk factor in studies of other complex syndromes, including several mental disorders. Second, methods to identify exomic regions of the genome are currently sub-optimal. In contrast, whole genome sequencing has the advantage of automatically including all candidate genes and all intergenic regions, as well as the ability to detect CNVs and several types of structural variations (e.g., translocations). Candidate gene sequencing is a viable strategy when a strong list exists of candidates likely to harbor rare variations. This strategy would be exemplified by sequencing genes with strong, confirmed support in GWAS or CNV studies, following the idea that genes harboring one form of disease variant are more likely to harbor other forms as well.

The efficiency of detecting structural variants is related to the size of the genome region involved. Many array-based methods are effective in detecting deletions and duplications at or above 10 kb. Finer resolution is required to detect smaller insertions and deletions (indels); methods to do this, using next-generation sequencing, are evolving rapidly.

This discussion has highlighted the general principle that the experimental approach used to identify an addiction-related gene should be carefully matched to the scientific question at hand. The discussion also emphasizes the degree to which the technologies to quantify genetic variants are changing rapidly. The expectation is that next-generation sequencing will be used to an increasing extent as its cost decreases and as bioinformatics and statistical tools are developed that optimize the analysis of vast amounts of sequencing data.

Deep Phenotyping in Addiction

With some notable exceptions, there has been a high rate of nonreplication of findings from human genetic association studies of addictive disorders. Further, the few well-reproduced findings (e.g., chromosome 15q markers and nicotine dependence) have yielded small phenotypic effects—that is, small relative risks for dependence or heavy use. Given the evolving understanding of the complexity of other common medical syndromes, it is possible that no single genetic variant will account for a significant proportion of the known heritability of addictive diseases. However, another factor that may be contributing to this lack of replicable findings is the inaccuracy of the self-reported phenotypes commonly used in these studies. Consideration should therefore be given to investigating phenotypes that more closely reflect the underlying genotypes.

One approach to improved phenotyping is to move beyond simple classifications based solely on presence versus absence of addictive disorders or on the rate of drug use. More refined phenotypes might reflect different pathophysiological processes that underlie these disorders. The rationale for such a "deep phenotyping" approach is based on several features of addiction. Specifically, addiction: (a) is best reflected as a continuum, not a simple categorical classification; (b) involves developmental and longitudinal trajectories from initial exposure, to progression to abuse and addiction, to cessation and treatment response, and, unfortunately, to relapse; and (c) is a complex phenotype that can be dissected into core sub-phenotypes that may reflect its underlying pathophysiological processes. These premises support a more refined phenotyping approach that measures, for example, the consequences of genetic variation upstream from the clinical outcome, including variation in biochemical, neurophysiological, anatomical, and cognitive processes. Because these biological processes are more proximal to the genetic effects than are clinical features, this approach may provide a more sensitive means for detecting genetic association. Moreover, such variables can be measured in a more precise and objective manner, thereby reducing error and increasing statistical power for detecting associations. Among deep phenotyping approaches are measures referred to as "endophenotypes," which, in addition to being objectively assessed and heritable, exhibit a clear relationship to the clinical endpoint.

Whether such deeper phenotyping will assist in the identification of addiction-related genes will remain unknown unless and until such approaches are tested in sufficiently powered studies. NIDA should therefore consider collecting more refined-phenotype data in genetic studies of addiction. Such data would include longitudinal phenotypes that reflect dynamic processes in addictive behaviors (e.g., initiation and progression to abuse/addiction, withdrawal symptoms, cessation and relapse, and treatment response/pharmacogenetics), as well as intermediate phenotypes that reflect the underlying pathophysiology of these behaviors (e.g., brain structure and function). In addition to potentially providing more sensitive measures for genetic studies, improved phenotyping in genetic studies of addiction could enhance understanding of the mechanisms through which genetic variation affects addictive behavior. Such studies could facilitate the development of new systems for disease classification and diagnosis based on biology in contrast to the solely behavioral measures used for diagnosis today (including DSM-IVTR and DSM-V).

FINDINGS AND RECOMMENDATIONS

The study of the genetics of complex, common diseases is still in its early phases throughout the biomedical world. New technologies are rapidly advancing the ability to interrogate the entire genome without prior hypotheses, and approaches to whole genome sequencing are becoming increasingly powerful and financially feasible. In addition, for the genetics of substance abuse and addiction, there are strong biological hypotheses about underlying neurobiological mechanisms that can help drive important gene discoveries and apply them to assess risk and improve treatment outcomes.

1. The available technologies are changing so rapidly that the Work Group cannot specify which approaches are most appropriate for NIDA to support and how much of each approach is desirable. NIDA needs to keep abreast of new technologies as they emerge.

2. Since it is not yet known which approach or approaches to genetics of substance abuse and addiction in humans will be most useful, a balance among different approaches is needed. This balance will change as more dense genotyping becomes less expensive. Approaches to be balanced include but are not limited to the following:

    - Genome-wide studies versus hypothesis-driven candidate gene studies
    - GWAS versus next-generation sequencing
    - Whole exome versus whole genome sequencing
    - Cross-sectional studies of addicted individuals versus longitudinal studies of large populations continuing over many years
    - Focused studies of clinical samples versus studies of population-based cohorts
    - The several phases of substance use, abuse, and addiction—from initiation of use to progressive addiction and including spontaneous remission (i.e., unaided cessation of use), treatment response, and relapse
    - The overlaying of DNA sequencing data with epigenetic analyses (e.g., histone and DNA methylation) on peripheral tissues or postmortem brain samples

3. NIDA's GWAS efforts to date have focused heavily on nicotine addiction, although GWAS of cocaine and opioid addiction phenotypes are included in the current portfolio. The Work Group recommends that GWAS (or other genome-wide approaches) of addictions to drugs other than nicotine should continue to be supported to provide a balance of drug abuse phenotypes.

    - The Work Group believes that sufficient GWAS data for some nicotine phenotypes, such as cigarettes per day, are available for the time being. Completed studies in this area provide a rich source of data for meta-analysis and/or further deep sequencing in target regions of the genome. A caveat, however, is that the phenotypes in these prior studies are based on relatively crude measures of addiction—e.g., differences in the number of cigarettes smoked per day or retrospective self-reported cessation or use. Additional GWAS data on nicotine addiction for more richly phenotyped samples may still be needed. The same need applies to other addictions.

        a. NIDA should decide which addictions deserve the next GWAS focus, e.g., heroin, prescription drugs (e.g., stimulants, opiates), marijuana, cocaine, methamphetamine, or

others. A related question is whether caffeine use and physical dependence should also be included.

b. While discussion of such priorities for the Institute is beyond the scope of the Work Group, the Work Group recommends that NIDA develop a more coherent overall strategy (or "top-down" oversight) to genetic studies for the range of substances to study. This should include a continuing evaluation of ongoing work (both genotyping platforms and phenotyping depth) for different drugs of abuse and should establish priorities for the next series of studies to be performed. In this way, NIDA will be able to fill in the gaps, orchestrate a balanced and cost-efficient human genetics effort, and avoid redundant studies. The Work Group discussed the relative utility of investigator-initiated R01 grants, which remain very important to optimize innovation and creativity, and other more directed mechanisms to ensure that priority research is accomplished.

- The Work Group believes GWAS that are in progress should focus on using the most current array technology, ideally those that incorporate both low frequency as well as common variants. These commercial array products are expected to emerge by mid-2010.

4. NIDA should consider funding deep sequencing of a select group of genes that are most proximal to the immediate effects of drugs of abuse or their metabolism. Thus, the roughly 600 genes implicated as immediate targets for drugs of abuse (nicotinic cholinergic receptors, opioid receptors, cannabinoid receptors, dopamine transporter and receptors, as just some examples) or in the biosynthesis or catabolic breakdown of drugs or endogenous transmitters (alcohol dehydrogenase, catechol-O-methyltransferase, choline aceytltransferase, and acetylcholinesterase, among many others), might be deep-sequenced as a priority initiative.

5. NIDA should evaluate the contribution of deep phenotyping to whole-genome sequencing as well as to hypothesis-driven studies. As larger numbers of patient samples are collected for a range of substances of abuse, through the NIDA Genetic Repository or other repositories of value for research on genetics of substance abuse and addiction, there will be a need to set priorities. Greater attention to the inclusion of samples from individuals who have been more carefully phenotyped will become increasingly important. Moving beyond the crude phenotypes of disease presence and level of consumption to include longitudinal phenotypes (e.g., initiation and progression to dependence, withdrawal symptoms, cessation and relapse, and treatment response) may assist in the identification of addiction-related genes. However, such efforts must be balanced by the need to sequence large numbers of subjects to obtain statistically significant genetic findings. In contrast, there is no question that deep phenotyping, including intermediate biomarkers (e.g., drug metabolism) and effects of drug exposure on brain structure and function, could enhance understanding of the mechanisms through which genetic variation affects addictive behavior. Thus, it is essential that any positive genetic findings be followed up aggressively to understand how genetic variation alters neural and brain function to contribute to substance abuse or addiction.

6. Studies of candidate genes and pathways other than those already studied—perhaps identified from animal studies of long-term effects of drugs on the brain—should be considered for funding, but clear guidelines must be established to ensure (as for all genome-wide studies, too):

   - Study of a sufficient number of patients
   - Inclusion of all of the needed categories of patients, depending on the hypotheses (for example, drug exposed but not addicted and drug naïve controls, as well as drug-addicted)

- Use of appropriate (state of the art) genetic platform(s)
- Application of appropriate (state of the art) bioinformatics analyses

7. Guidelines must be established and disseminated to govern genetic studies in the context of deep phenotyping (brain imaging, responses to pharmacological treatments [pharmacogenomics], responses to pharmacological challenges including drugs of abuse [also pharmacogenomics], neurobehavioral and cognitive measurements, etc.). Acceptable standards for the same considerations as in recommendation 6 above must be developed to avoid investing limited funding resources in studies that are inherently underpowered.

   - The Work Group recommends that NIDA convene a technical review to establish such guidelines.
   - NIDA should then make those guidelines, once approved, readily available to individuals interested in applying for grants. Such guidelines would also be expected to help NIDA extramural staff steer potential grant applicants.

8. Support should be increased for training in biostatistics and statistical genetics in recognition of the gross shortage of such manpower available today. Otherwise, as research in human genetics of substance abuse expands, the shortage of expertise in these areas in the research workforce will constrain progress and jeopardize the scientific validity of published results. Furthermore, priority should be given to the funding of theoretical grants focused on method development in the areas of statistical genetics and bioinformatics of complex disease that would increase the efficiency and information value of the current and future large scale data collection, genotyping, and sequencing projects.

9. Better top-down oversight of all NIDA grants that involve human genetics analyses—across all extramural divisions—is needed to ensure that rigorous scientific standards are used and that NIDA's strategic plan in human genetics is executed with maximal efficiency (see recommendation 3, first bullet, item (b), page 8). The NIDA Genetics Coordinating Committee (NGCC) is a worthy start at accomplishing such oversight. However, the Work Group recommends that its effectiveness and reach be strengthened substantially. The NGCC should help NIDA leadership establish funding priorities across the full range of human genetics research and be integrally involved in funding decisions for all such grants. To achieve these aims, the Work Group recommends the following improvements:

   - Recruit additional extramural-program staff with a high level of expertise in different areas of human genetics, including areas not covered by the current membership of the NGCC (e.g., genetics of complex diseases, pharmacogenetics, genetic epidemiology, and biostatistics).
   - Scrutinize proposals under consideration, in all extramural divisions, to ensure that their genetics components are worthy of being funded and meet the high scientific standards as outlined in recommendation 6 above. Thus, representatives of the NGCC should be present at all funding meetings by analogy with the current level of participation of NIDA's Office of AIDS Research Program.
   - Maintain and update a tabulation of genetics-related grants, contracts, and IRP projects in human genetics or with human genetics components, similar to the table of extramural research projects prepared for the Work Group by DBNBR and the portfolio of HIV/AIDS–related research maintained by the NIDA Office of AIDS Research Program.

10. The Work Group recommends that a strong human genetics component be developed in the NIDA IRP, which to date has been notably lacking. This IRP effort should be carefully coordinated with ongoing extramural research to avoid redundancy. The IRP should emphasize those aspects of human genetics research for which an in-house program is particularly well suited, including but not limited to the following:
    - Longitudinal studies of highly informative populations
    - High-risk studies that require stable funding over a longer period than a typical research grant

11. Given the high rates of comorbidity for substance abuse with other mental disorders, a great deal can be gained from greater collaborative efforts across several of the NIH institutes and centers (ICs), including, NIAAA, and NIMH. These collaborations should include sharing of samples and repositories, large-scale phenotyping, and greater coordination of research initiatives, including joint funding of R01 grants examining conditions and risk factors common to these disorders.
    - The Work Group notes the diversity of psychopathological phenotypes that co-occur with substance abuse: antisocial personality and other externalizing disorders and traits typically characterized by high levels of impulsivity and/or low levels of empathy, pathological gambling and overeating, among others. In particular, there is strong evidence from genetic epidemiological studies for a substantial sharing of genetic risk between externalizing disorders such as antisocial personality disorder or conduct disorder and risk for substance use disorders. The genetic contributions of such conditions might be jointly addressed through comprehensive initiatives.

12. Extramural grants with significant human genetic components should be reviewed by study sections that include the requisite expertise in genetics. This is essential to avoid the awkward situation of a grant receiving an outstanding priority score despite fatal flaws in the quality of its genetics component (as outlined in recommendation 6). One way to achieve such a goal, if genetics expertise cannot feasibly be included in all relevant study sections, is for the NGCC to carry out a secondary review of any grant with a genetics component recommended by divisions for funding, to ensure relevance, scientific standards, consistency with NIDA genetics strategic priorities, etc.

13. Mechanisms are needed to encourage and/or require more data-sharing among NIDA grantees, as well as grantees of NIMH and other NIH ICs, on those project components involving human genetics data. This might include combining DNA samples and phenotyping efforts to achieve the number of subjects and depth of genotyping and phenotyping that are required to find addiction vulnerability genes. The Work Group favors the inclusion of "carrots" to achieve this goal, such as additional funding being made available for collaborative efforts and for making datasets available sooner rather than later, rather than a sole reliance on "sticks."

14. NIDA should consider how to facilitate research on translating the results of findings on human genetics of substance abuse: to develop improved diagnostic tests and treatments, to inform health care and public policy, and to improve clinical and community interventions for both prevention and treatment of substance abuse.
    - In considering translational studies, greater attention should be given to the effect sizes of genetic associations, since the translational utility of findings with very small relative risks, and those accounting for a very small proportion of the variance in an addiction phenotype, may be questionable.

- On the other hand, there are important precedents in the biomedical world where uncommon (even rare) genetic findings have led to dramatic improvements in treatment and in understanding disease pathogenesis. For example, introduction of the statin drugs, which have had a dramatic effect on public health, was based on rare alleles in the population. Moreover, the genetic basis of rare, familial forms of Alzheimer's and Parkinson's diseases have dramatically improved our understanding of disease pathogenesis more generally and are currently driving drug discovery efforts.
- There are many barriers to clinical translation (both Type 1 and Type 2 translation), including ethical and social issues and cost-effectiveness considerations. Research on how to apply evolving findings on the genetics of substance abuse and addiction should be carried out in parallel with the human genetics research, so that optimal methods and best practices are available as addiction vulnerability genes are identified. The challenge is to take what is learned about genetic risks and incorporate such advances into real-world settings.

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Appendices

APPENDIX A: SCIENCE OF GENETICS REVIEW WORK GROUP

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APPENDIX B: SCIENCE OF GENETICS REVIEW WORK GROUP MEETING, NOVEMBER 2009

National Institute on Drug Abuse
Science of Genetics Review Work Group
November 23–24, 2009

Hyatt Regency Bethesda
7400 Wisconsin Avenue
Bethesda, MD

Day 1—November 23, 2009

1:00– 1:10 pm	**Work Group Introductions, Opening Remarks** *Eric Nestler, M.D., Ph.D., Work Group Chair*
1:10–1:30 pm	**Work Group Charge** *Nora D. Volkow, M.D., Director, NIDA*
1:30–3:15 pm	**Overview of the Genetics and Molecular Neurobiology Research Program** *David Shurtleff, Ph.D., Director, DBNBR* *Jonathan Pollack, Ph.D., Branch Chief* *John Satterlee, Ph.D., Program Director* *Joni Rutter, Ph.D., Associate Director for Population and Applied Genetics*
3:15–3:30 pm	**Break**
3:30–5:00 pm	**Work Group Discussion** *Eric Nestler, M.D., Ph.D., Work Group Chair*
5:00 pm	**Adjourn–Day 1**

Day 2—November 24, 2009

NIDA Research Related to the Genetics Portfolio

9:00–9:30 am	**Division of Clinical Neuroscience and Behavioral Research** *Joseph Frascella, Ph.D., Director*
9:30–10:00 am	**Division of Epidemiology, Services and Prevention Research** *Wilson Compton, M.D., Director* *Kevin Conway, Ph.D., Deputy Director*
10:00–10:30 am	**Division of Pharmacotherapies & Medical Consequences of Drug Abuse** *David McCann, Ph.D., Acting Director*
10:30–10:45 am	**Break**
10:45–11:15 am	**NIDA's Intramural Research Program** *Toni Shippenberg, Ph.D., Chief, Integrative Neuroscience Section*
11:15 am–1:30 pm	**Work Group Discussion and Working Lunch**
1:30–2:45 pm	**Work Group Members' Response to the Future of NIDA Genetics of Addiction** *Work Group Members*
2:45–3:00 pm	**Break**
3:00–3:30 pm	**Follow-up Discussion with NIDA Staff** *David Shurtleff, Ph.D., Director, DBNBR* *Jonathan Pollack, Ph.D., Branch Chief* *Joni Rutter, Ph.D., Associate Director for Population and Applied Genetics*
9:00 am–5:00 pm	***EXECUTIVE SESSION***
3:30–4:30 pm	**Work Group Discussion** *Eric Nestler, M.D., Ph.D., Work Group Chair*
4:30–5:00 pm	**Work Group Recommendations and Next Steps** *Eric Nestler, M.D., Ph.D., Work Group Chair*
5:00 pm	***ADJOURN***

APPENDIX C: SCIENCE OF GENETICS REVIEW WORK GROUP MEETING, JANUARY 2010

National Institute on Drug Abuse
Science of Genetics Review Work Group
January 25, 2010

Mayflower Renaissance Hotel
Washington, D.C.

11:00–11:15 am	**Work Group Introductions; Opening Remarks** *Eric Nestler, M.D., Ph.D., Work Group Chair*
11:15 am–5:00 pm	***EXECUTIVE SESSION***
11:15–11:30 am	**Division of Epidemiology, Services and Prevention Research** *Wilson Compton, M.D., Director* *Kevin Conway, Ph.D., Deputy Director*
11:30–11:45 am	**Division of Pharmacotherapies & Medical Consequences of Drug Abuse** *David McCann, Ph.D., Acting Director* *Ivan Montoya, M.D., Acting Deputy Director* *Ahmed Elkashef, M.D., Chief, Clinical Medical Branch*
11:45 am –12:00 pm	**Division of Basic Neuroscience and Behavior Research** *David Shurtleff, Ph.D., Director, DBNBR* *Joni Rutter, Ph.D., Associate Director for Population and Applied Genetics*
12:00–12:30 pm	**Work Group Discussion**
12:30–1:00 pm	**Genetics Research at the National Institute of Mental Health** *Thomas Lehner, Ph.D., M.P.H., Chief, Genomics Research Branch, Division of Neuroscience and Basic Behavioral Research, NIMH*
1:00–2:15 pm	***Working Lunch***
2:15–2:45 pm	**New Approaches to Large Scale Sequencing** *Jim Mullikin, Ph.D., Acting Director, NIH Intramural Sequencing Center, National Human Genome Research Institute*
2:45–3:30 pm	**Work Group Discussion**
3:30–3:45 pm	***Break***
3:45–5:00 pm	**Work Group Recommendations**
5:00 pm	**Adjourn**