Drug Repurposing for Rare Diseases

Roessler, Helen; Knoers, Nine V. A. M.; van Haelst, Mieke M.; van Haaften, Gijs

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Review

Drug Repurposing for Rare Diseases

Helen I. Roessler [1], Nine V.A.M. Knoers,[1,2] Mieke M. van Haelst,[3,4,5] and Gijs van Haaften[1,5,*]

Currently, there are about 7000 identified rare diseases, together affecting 10% of the population. However, fewer than 6% of all rare diseases have an approved treatment option, highlighting their tremendous unmet needs in drug development. The process of repurposing drugs for new indications, compared with the development of novel orphan drugs, is a time-saving and cost-efficient method resulting in higher success rates, which can therefore drastically reduce the risk of drug development for rare diseases. Although drug repurposing is not novel, new strategies have been developed in recent years to do it in a systematic and rational way. Here, we review applied methodologies, recent accomplished progress, and the challenges associated in drug repurposing for rare diseases.

Highlights

The awareness and interest of the public, media and legislative bodies in the field of rare diseases has been growing consistently.

Researchers continue to successfully uncover the molecular and genomic drivers, as well as the clinical course of many rare conditions, due to advances in DNA sequencing technologies and big data analysis, resulting in high expectations for new and optimized treatments.

Due to cost effectiveness and a reduced timeline, the process of repurposing drugs for new indications represents an alternative method for finding rare disease treatments with compelling advantages over traditional drug development.

The development of systematic approaches to repurpose compounds has led to the identification of promising candidate drugs, some of which are in advanced stages of clinical trials or already approved, with the potential for use in the treatment of rare diseases.

Rare Diseases (RDs): A Unique Group of Disorders

A RD can be any heterogeneous condition affecting a small percentage of the population (Europe: 1 person per 2000[i]; USA: <200 000 individuals[ii]). RDs are often chronic, resulting in lifelong disability or early death; many RDs have a pediatric onset and about 30% of children with RD die before the age of 5 years [1]. Seventy percent of all RDs are genetic, caused by both germline and somatic gene mutations [2]. Of the RDs with a genetic origin, many have a monogenic origin [3]; they are caused by a single gene defect and follow a Mendelian inheritance pattern (dominant, recessive, X-linked). Additionally, RDs also show non-Mendelian inheritance, which includes epigenetic changes (e.g., Beckwith-Wiedemann syndrome) and mitochondrial disorders (e.g., Rett syndrome) resulting from maternal transmission of variants in mitochondrial DNA [4,5]. A minority of RDs are also caused by environmental, infectious, or immunological factors (e.g., African trypanosomiasis) [6] but will not be discussed here.

Around 7000 RDs have been identified to date. While individually rare, they globally affect 300 million people (10% of the population) worldwide, with new diseases regularly being described in medical literature [2,7,8]. Prevalence rates vary widely among RDs, where they can range from ultra-rare, with only a few cases described globally, to less rare, for which diagnosis is dependent on the experience of the individual physicians [9]. For example, Hutchinson-Gilford progeria syndrome (HGPS) , a genetic disorder resulting in premature aging, shows an incidence of merely 1 in 8 million live births [10], whereas a relatively well-known rare condition, Huntington's disease, affects an estimated 3 to 7 per 100 000 people of European ancestry [11]. Nevertheless, all RDs share similar clinical challenges, as they involve multisystem dysfunction and therefore require complex care [12]. Compared with common disorders, patients with a RD visit approximately twice as many specialists [13].

RDs: A Diagnostic Odyssey

Without an accurate (molecularly confirmed) diagnosis, it is not only difficult to identify the cause but also to design an appropriate and effective treatment strategy to suppress or reverse the condition of interest. Moreover, a molecularly confirmed clinical diagnosis often allows a clinical geneticist to give accurate genetic counseling, including providing information regarding patient prognosis, inheritance pattern, prenatal investigation options, and availability of (future) personalized treatment [14]. Diagnosis of RDs, in particular, remains a challenge for patients, doctors, and healthcare systems. Both patients and physicians have limited knowledge and experience with the disease

[1]Department of Genetics, Center for Molecular Medicine, University Medical Center Utrecht, Utrecht University, Utrecht, The Netherlands
[2]Department of Genetics, University Medical Center Groningen, Groningen, The Netherlands
[3]Department of Clinical Genetics, Amsterdam University Medical Center, Location AMC, University of Amsterdam, Amsterdam, The Netherlands
[4]Department of Clinical Genetics, Amsterdam University Medical Center, Location VUMC, VU University Amsterdam, Amsterdam, The Netherlands
[5]These authors contributed equally to this work

*Correspondence:
G.vanHaaften@umcutrecht.nl
(G. van Haaften).

they are dealing with due to insufficient characterization of the natural history of many RDs [15]. Thus, a majority of (parents of) patients undergo a downright 'diagnostic odyssey', which can be a long and frustrating journey for both patients and their families to obtain an accurate diagnosis. Typically, it can take 6 years from onset of symptoms to a correct diagnosis, where patients tend to encounter 14 diagnostic procedures and 4.5 diagnoses [16].

The identification of RD-associated genes and variants is essential for diagnosis and disease prognosis. Since 2010, the number of mutations in disease-causing genes identified per year has grown significantly due to advances in DNA sequencing technologies and big data analysis. This improvement has been transformative for diagnosing rare Mendelian diseases [17]. Currently, there are approximately 4000 RD genes known (Online Mendelian Inheritance in Man[iii]) and recent estimates suggest that over 9000 single-gene phenotypes will ultimately be discovered and molecularly defined [18]. **Next-generation sequencing (NGS)** (see Glossary), in particular **whole-exome sequencing (WES)** and **whole-genome sequencing** (**WGS**), are established diagnostic tools to unravel the genetic background of RDs. Recent advances made in these technologies make them affordable and indispensable (Box 1).

The identification of disease-causing gene mutations, however, is merely the first step in confirming a clinical diagnosis. Understanding underlying affected genetic and molecular mechanisms and pathways is essential for disease comprehension and selection of a target for therapy. This can be achieved by engineering and studying model organisms, applying multi-omics sequencing approaches from RNAseq to epigenetic information in tissue types of interest, **microarray** technology, or *in silico* techniques [19,20]. However, despite the significant increase in the speed of RD gene discovery, the gap in our understanding of the molecular and cellular mechanisms of RDs still remains [21,22].

Challenges in Drug Development for RDs

In 95% of all RDs there is no licensed treatment or cure available [23], resulting in a drastic decrease in quality of life due to stress, anxiety, chronic pain, physical impairment, or early mortality in the majority of patients [24]. Hence, the impact of therapeutic RD intervention is considerable, not only for the patients but also their family and healthcare providers. Currently, most patients merely receive symptomatic or comfort treatment, which address second-order complications instead of the underlying disease cause. Current treatment is therefore aimed at the improvement of life quality of those affected, but does not prevent the inevitable decline in function. In Fabry

Box 1. Discovering the Genetic Cause of Rare Diseases

It is fair to say that progress in identifying disease genes has been spectacular. Many commercial and customized gene-sequencing panels are available nowadays for relatively low-cost analysis of genes associated with specific diseases or groups of disorders. For a more comprehensive analysis, whole-exome sequencing (WES) has emerged as an effective tool for diagnosing patients who have already undergone comparative genomic hybridization techniques in order to exclude microdeletions or duplications as the cause of their disorder [99–101]. WES limits sequencing mainly to the protein-coding regions of the human genome, which contains 180 000 exons, merely constituting 2% of the whole genome. Nevertheless, sequencing the coding region and exon–intron boundaries reveals an estimated 85% of heritable Mendelian disease-causing mutations [102]. Within the last decade, WES has become technically feasible and more cost-effective, offering new possibilities for establishing Mendelian disease diagnosis and research [102]. Currently, the cost of sequencing an exome in a clinical setting is less than €1000. Additionally, its accuracy is remarkable, with studies reporting a sensitivity of 98.3% for detecting previously identified mutations when applying this technique [103]. However, even though the application of WES represents a promising diagnostic option for RD patients, a diagnosis in not guaranteed. In over 70% of patients with a high degree of suspicion of a monogenic RD, WES failed to provide a definitive molecular diagnosis [104]. Moreover, there are still cases of RDs that are caused by variants affecting regulatory and non-coding DNA regions, which cannot be detected by WES at all [105]. Sequencing of the whole genome (WGS) is a promising newer approach. Unlike WES, WGS produces sequence information of the coding exome as well as the other 98% of the genome, which is non-coding, but nevertheless might reveal functional relevance [106].

Glossary

Microarray: an array of hundreds of spots containing specific DNA sequences for the analysis of gene expression by hybridization.
Next-generation sequencing (NGS): the catch-all term used to describe different modern sequencing technologies resulting in sequencing of DNA and RNA much more quickly and cheaply than previous techniques.
Whole-exome sequencing (WES): a widely used sequencing method that targets only the protein-coding region of the genome (the exome).
Whole-genome sequencing (WGS): a comprehensive method that provides a base-by-base view of the entire genome.

disease, for example, previous interventions simply replaced the missing enzyme with a naturally occurring human lysosomal hydrolase enzyme, resulting in a buildup of fat in various patient tissues and therefore not providing a cure *per se*. A novel treatment, migalastat (Galafold), increases the enzyme activity by stabilizing the endogenous protein and supporting a proper folding and therefore significantly reduces associated symptoms [25]. This great unmet need makes it imperative that we find ways to accelerate the therapy development process so that we can help many RD patients who are in search of better treatments.

One way to tackle the obvious gap in clinical management of RDs is the development of novel orphan drugs. The development of such new drugs, however, is a major challenge, with often only limited knowledge available regarding disease epidemiology, manifestations, heterogeneity, natural course, and progression [26]. Taking into account that the process of developing a novel drug to treat any kind of disease is typically laborious, costly, and failure-prone [27], it is especially unappealing to do so for RDs that affect only a small number of individuals and therefore generate reduced profit. The introduction of a new compound to the market can cost as much as $2.5 billion, with further increasing numbers often including high development and manufacturing costs [28,29]. Notably, merely five out of 5000 (0.1%) experimental compounds that enter preclinical testing progress to evaluation in humans. Only one of these five compounds receives approval from the US Food and Drug Administration (FDA) for use in humans, highlighting the failure susceptibility of this process [30]. Lastly, traditional drug discovery strategies are highly time-consuming, often requiring 10 to 15 years of research and development efforts until a drug can finally enter the market (Figure 1, Key Figure). When large manufacturers invest in research and development of therapy options for RDs despite these challenges, market prices are typically extremely high, resulting in higher profit but reduced access for patients. For instance, nusinersen (Spinraza), the first approved drug for spinal muscular atrophy, is one of the most expensive drugs on the market, costing $750 000 for the first year of treatment and $375 000 every following year [31].

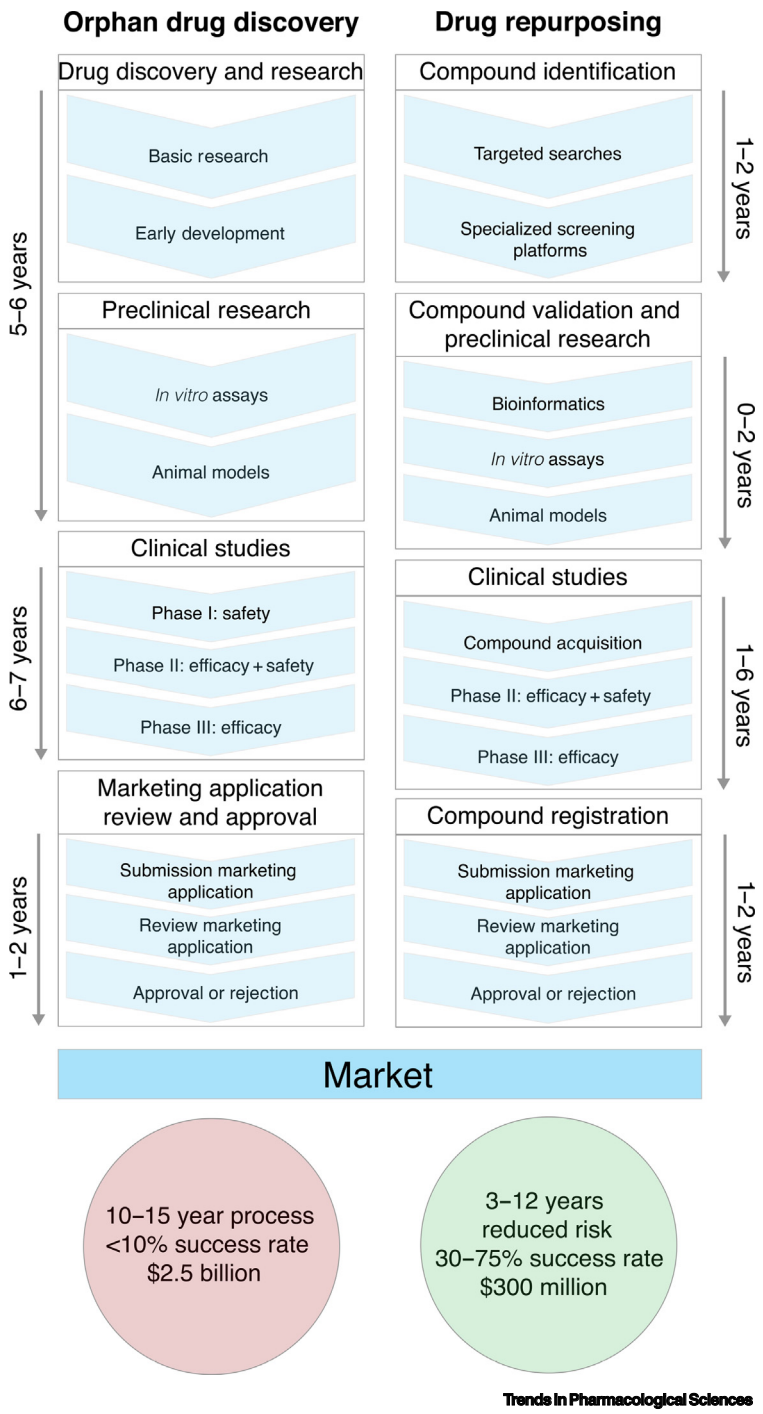
Despite the monogenic nature of the majority of RDs, many variations or disease subtypes result in different clinical manifestations and disease progressions. Depending on RD heterogeneity a compound might work for only a subset of patients. The compound choice may depend on the type of mutation the patient has, as is exemplified in cystic fibrosis where mutation-specific therapies are already in development [32,33]. In addition, designing a clinical trial can be time consuming and complicated due to absence of structured databases and patient cohorts and insufficient epidemiological and scientific data (Box 2) [34]. Despite occasional success stories, like novel therapy options for acute hepatic porphyria [35], hereditary transthyretin-mediated amyloidosis [36], cystic fibrosis [37], or spinal muscular atrophy [38], these great challenges provide little hope for obtaining a product approval for most individuals with one of the nearly 7000 RDs.

Drug Repurposing: A Cheaper and Faster Option

Drug repurposing (also known as drug repositioning or drug reprofiling) is the process of redeveloping a compound for use in a different disease [39] and is now becoming an increasingly important strategy for researchers in industry and academia [40]. Although this strategy is far from new, repurposing success stories and companies leveraging repurposing strategies are increasing in number [41,42]. They are based on the following core scientific principles: (i) single drugs often interact with multiple targets or pathways, and (ii) various drugs may act on the same target or pathway [43–45]. Growing evidence indeed suggests that any functional compound classified to be safe for human use is likely to have multiple therapeutic applications. Most repurposed drugs show little to no clear connection to their primary approved indications. For instance, compounds tend to show off-target effects triggering undesired adverse events [46]. However, these effects might be of advantage for other indications. Lastly, a target involved in a certain disease is

Key Figure

Advantages of Drug Repurposing over the Traditional Way of Orphan Drug Development



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Box 2. Challenges of Designing Clinical Trials for Rare Diseases

There are multiple requirements for designing effective clinical trials for studying therapeutics for any kind of human disorder: (i) appropriate trial design and analysis to answer the research question of interest, (ii) adequate measurements to complement the trial design, (iii) selection of correct sample size, (iv) ethical recruitment to participation, and (v) funds to support the research as well as knowledgeable study staff. Especially for RDs, these requirements can become challenging, considering the limitations of studying a small patient cohort. In the following text, we lay out the challenges arising due to studying a small patient cohort associated with RDs.

Statistical Power of Designed Clinical Trial

Typically, the randomized controlled trial that randomly assigns participants into an experimental or a control (standard of care) group is considered as gold standard for establishing efficacy in a research setting [107]. Because of high costs, time-investment, and requirement of a large disease cohort, it is usually applied in common diseases, but lesser in RDs. In the analysis of clinical trial data, RDs show two major disadvantages. Clinical studies for RDs can only enroll a small cohort (due to less patient population). In combination with high variability in phenotype and clinical course observed in a majority of RDs [108], the study's power decreases drastically. Hence, alternative trial designs and statistical techniques that maximize data from a small and heterogenous group of individuals are necessary. Possible alternative designs could be crossover studies, in which all participants receive the same two or more treatments, but the order in which they receive them depends on the group to which they are randomly assigned, or adaptive design approaches. Such an adaptive design allows adaptations to trial and/or statistical procedures of the trial after its initiation, without undermining the validity and integrity of the trial [109]. These strategies result in shortening of clinical trial duration and increased success probabilities by dropping inferior treatment arms at interim evaluations or assigning more patients to superior arms by using various randomization schemes in a play-the-winner principle. Notably, due to these measures, drugs for RDs have been approved based on studies that lacked randomization and a placebo control [110].

Natural History

In-depth knowledge of the natural history of (rare) diseases is significant for trial design as it helps to identify key milestones in disease progression, select an appropriate length of the study to monitor change in disease progression, develop adequate inclusion/exclusion criteria, and determine a clinically meaningful difference [111]. Due to small patient numbers, geographic dispersion of patients and researchers, and small number of researchers with specific expertise, collection of natural history data can be challenging. One possibility to overcome these limitations is multicenter collaboration to strengthen the knowledge of natural history and to establish a patient registry to maintain the collected data, as was done for Cantú syndrome, for instance [112].

Subject Recruitment

Another challenge for all RDs is the timely and adequate recruitment of patients meeting all inclusion criteria of the study. Often, researchers would like to focus on a specific aspect of the entire patient population (age, disease stage, etc.); however it may not be feasible to further minimize an already small cohort. In disorders with significant physical impairment, traveling to the closest research center may not be possible. Lastly, for trials investigating repurposed drugs, potential off-label applications can further threaten recruitment. In order to maximize recruited individuals, it is recommended to involve patients during trial design (patient participation in clinical research). According to interviews with representatives of RD patients, patients value involvement in trial design and thus have influence on outcomes and measurement instruments, duration of the trial, and information that is sent to potential participants [113].

also often associated with other biological processes, pathways, or phenotypes [47] and pathogenetic mechanisms of rare and common disease often influence the function of more global molecular pathways and networks rather than merely the function of single genes [48,49].

Pharmaceutical companies search for inexpensive alternatives to compensate for the high costs and disappointing success rates associated with the drug discovery pipeline [50]. Hence, they turn to already approved, discontinued, shelved, or experimental drugs and try to establish

Figure 1. Drug repurposing is shown to increase success rates, reduce development costs, shorten time to the market, and therefore reduce the overall development risk compared with traditional drug development. Repurposing candidates have already proven to be sufficiently safe in preclinical models, at least at early-stage trials in humans, thus being less likely to fail from a safety point of view in subsequent efficacy trials. Often, the only step left to accomplish is to confirm efficacy for the new indication at preclinical and clinical levels. Hence, drug repurposing is an important alternative to orphan drugs, especially in the field of rare diseases.

new medical applications. The process of drug repurposing is considered to be significantly faster and cheaper than traditional drug discovery and therefore offers hope to RD patients, for whom the conventional model is commercially unviable [51]. Specifically, development risk is reduced as repurposing candidates have already proven to be sufficiently safe in preclinical models and, at least at early-stage trials in humans, thus being less likely to fail from a safety point of view in subsequent efficacy trials. Hence, after establishing dose compatibility, a majority of preclinical testing (e.g., *in vitro* and *in vivo* screenings, chemical optimization, formulation development), safety assessment, bulk manufacturing, and even Phase I clinical trials can be bypassed. The only step left to accomplish is to confirm efficacy for the new indication at preclinical and clinical levels. However, developing a new formulation for already known drugs can also be considered an appealing strategy for drug developers, for instance, resulting in obtaining new patent protection [52]. Finasteride, for instance, was originally developed to treat benign prostatic hyperplasia, but eventually repurposed for male pattern baldness. Due to applying a new formulation, the novel method of use received a new patent, despite the fact that the old one was still active [53].

Repurposed drugs can reach the patient as a marketed treatment in 3–12 years. On average, they cost $300 million and have an estimated success rate ranging from 30% [54] to as high as a potential 75% [29]; five times higher than for developing new compounds (Figure 1). The approximately 3000 drugs that have been approved by at least one country therefore represent a vast untapped resource if they can be used against another condition, especially a RD.

An example of such a drug repurposing effort has been with HGPS, the extremely rare and fatal premature aging disease caused by variants in the Lamin A/C gene (*LMNA*), which activates a cryptic splice site and results in the production of a farnesylated mutant lamin A protein called progerin [55]. Farnesyltransferase inhibitor (FTI) drugs, which reduce the amount of permanently farnesylated progerin, are shown to hold therapeutic potential for this disorder. Based on observations from clinical trials, a new drug application has been submitted to the FDA and the process for approval of the FTI lonafarnib (Sarasar), originally used for cancer treatment, as the first ever treatment for HGPS has begun recently [56–58]. Another example where a drug was repurposed for a RD involves Muckle-Wells syndrome (MWS), an autoinflammatory disorder caused by increased interleukin-1 (IL-1). Canakinumab (Ilaris), a drug originally approved to treat rheumatoid arthritis, is a human IgG1 anti-IL-1β monoclonal antibody that provides selective and sustained blockage of IL-1β, neutralizing the effect of excess IL-1β. Various clinical studies suggested that canakinumab results in a sustained control of disease activity and a rapid remission of associated symptoms in MWS patients [59,60]. It was approved by the FDA and by the European Commission in 2009 to treat patients with MWS.

Instead of directly applying an existing drug for a new indication, as shown in the previous examples, compounds identified as hits may also be subjected to further optimization. Even though this so-called lead optimization is a more complex endeavor, as it requires full clinical trials due to the generation of new compounds, it also represents the opportunity to eliminate unwanted side-effects or off-target effects originating from the initial use. Moreover, this process can also result in the increase of potency of the chemical compound at the primary drug target protein [61]. In order to get closer to a treatment for Batten disease, a fatal nervous system RD, a library of derivates of a clinically available but, due to side-effects, restricted drugs with neuroprotective activity was synthesized. Resulting compounds were shown to reveal physicochemical features desirable for disorders involved the central nervous system [62,63].

These examples highlight that drug repurposing can be effective in finding a therapy to help RD patients in a time-efficient manner.

Approaches to Drug Repurposing for RDs

Although drug repurposing is not a novel idea, the strategy to do it in a systematic and rational way is new. Many repurposed drugs have been found serendipitously in the past in the lab or by cautiously monitoring the effect of drugs in the clinic and performing subsequentially retrospective analysis of clinical observations [64]. This has been the case for one of the most popular repurposing success stories, minoxidil, originally investigated to treat ulcers; while conducting trials in dogs, it revealed a prolonged reduction in blood pressure. Later, while undergoing clinical trials to prove its efficacy as antihypertensive medication, the drug showed an unexpected positive effect on hair loss, which subsequentially led to its application for male baldness [65]. However, as there are more than 7000 RDs in need of treatment, this approach clearly does not meet the requirements for these diseases. Thus, more systematic, organized, and data-oriented searches for candidates have been launched, profiting from technological advances like big data analytics, computer models, and high-throughput screenings [66–68]. Additionally, the availability of huge information on the genetic basis of many RDs, on gene regulation, protein structure, and drug–target interactions from the Human Genome Project, has led to great opportunities to perform drug repurposing. Profiting from the improving availability of gene annotations for both human and model organisms, newly identified causative gene defects can now be checked to see whether they share the same pathways or biological processes as common diseases. For example, genes associated with the RDs neurofibromatosis type-1, Cowden disease, and retinoblastoma share common pathways that also play a role in various types of cancer [69]. Thus, a drug discovered to be useful in treating these cancer types might have undiscovered indications for RDs.

The current process of drug repurposing typically contains the following steps: (i) identification of a target, (ii) identification of a candidate molecule, (iii) mechanistic assessment of drug effect in preclinical models, (iv) preclinical drug development, (v) evaluation of drug efficacy in Phase II clinical trials (in case sufficient safety data from Phase I studies as part of the original indication is available), and (vi) filing for marketing approval. During the process of identifying a candidate compound, various strategies that have already proven to be helpful in the field of drug repurposing can be applied. These can be broadly categorized into experimental and computational approaches (Figure 2).

Computational Pharmacology and *In Silico* Methods

Nowadays, the capacity to generate data is immense and continues to grow. The unprecedentedly large amounts of data, on both rare and common diseases, have challenged researchers trying to make sense of it [70]. Additionally, data sharing initiatives also open up access to new types of data, including patients' medical records and other data ready to be analyzed. The clear need for rational approaches to find alternative indications for existing compounds has resulted in the development of computational or *in silico* methods for drug repurposing. These methods are appealing because they nominate the most promising candidate drugs for a given indication and apply various direct and indirect evidence to generate a hypothesis, including molecular [71], literature-derived [72], and clinical data [73].

Computational techniques, including both target- and ligand-based strategies to systemically design rational repurposing processes and models, perfectly complement existing experimental techniques and are therefore widely applied in industry and academia [74,75]. In recent years, as the study of drug repurposing has become a priority in the field of drug discovery, some valuable repurposing models have been created and applied to find potential new drug indications [76]. These models primarily aim at studying the relationship between drug, target, and disease and can be generally divided into two categories: (i) structure-based models, including drug/ligand structure and target/receptor domain; and (ii) phenotypic- or network-based models [77,78]. To

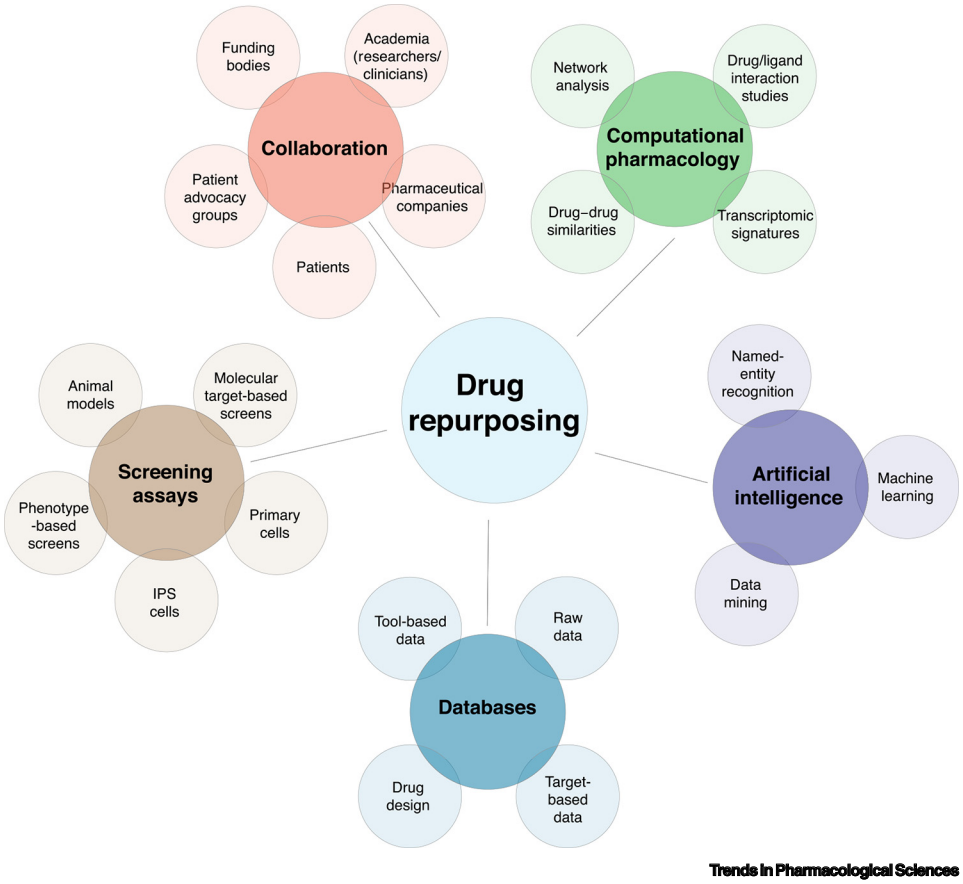

Figure 2. Approaches Applied in Drug Repurposing for Rare Diseases. Various approaches can be used individually or in combination to systematically analyze different compounds and diseases for repurposing hypotheses. These include computational and experimental approaches as well as a collaboration of various stakeholders involved in the field of rare diseases. Abbreviations: IPS, induced pluripotent stem.

identify candidate compounds, computational approaches are in need of carefully designed multistep analyses: compound prioritization coupled with well-designed validation experiments.

Artificial Intelligence (AI)

AI, holds great promise in rapidly and efficiently collecting, analyzing, and characterizing information and has already been successfully applied to basic research, diagnostics, drug discovery, and clinical trials. Due to the underrepresentation of RDs in research of treatment development, they especially can profit from AI [79].

AI, with an emphasis on deep learning or machine learning (ML), is capable of learning from data. ML algorithms are able to build mathematical models based on sample data, called training data, in order to make predictions or decisions without being explicitly programmed to perform the task. Thus, ML algorithms are able to uncover complex data [80]. If models were available for all aspects of drug discovery and development, they could be applied to predict whether a compound is likely to be used for a new indication.

Many studies have already applied ML methods in the field of drug repurposing. Lee *et al*. applied an ML unified computational framework called URSA^{HD} (unveiling RNA sample annotation for

human diseases) to apply genetic and molecular information about thousands of complex disorders, to test drug repurposing [81].

Screening Assays

The drug discovery process has been revolutionized in the last two decades, transitioning from low-throughput animal model-based techniques to high-throughput screens. The latter strategy takes advantage of the recent and rapid advances in screening technologies, which allow rescreening existing compounds against a variety of targets to identify possible therapeutic benefits or side-effects in an unbiased manner [82]. This approach can result in the discovery of novel interactions between approved drugs and previously unexplored or incompletely explored targets, like newly identified causative RD genes. The readout of such drug screens can either be based on known molecular targets or a phenotype associated with the disease. Phenotypic screens offer the opportunity to identify potential treatments for complex diseases where it might be challenging to find the primary therapeutic targets. However, screens based on a molecular target can further expedite drug discovery for RDs by addressing several diseases that share a common molecular etiology within one high-throughput screening [83].

A screening model resulting in recent advances in compound screening and evaluation of drug efficacy for neurological RDs are induced pluripotent stem cell (iPSC)-derived neurons. Whereas reliable *in vitro* models of human neurons are lacking and representation of many neurological RDs in animal models is often incomplete, patient-derived iPSCs represent a more relevant disease system in the appropriate setting [84,85]. Neuronal cells derived from iPSCs of Niemann-Pick disease type 1C, a rare lysosomal disorder, revealed cholesterol accumulation comparable with the *in vivo* situation in patients and responded to drug treatment [86]. In cystic fibrosis, mutation-specific iPSC lines were established and applied to generate organoid models for the evaluation of drug efficacies [87,88]. Lastly, Kinarivala *et al*. successfully characterized the first Batten disease patient-specific iPSC-derived model of the blood–brain barrier, resulting in the identification of small molecules modulating autophagy [89].

Collaborative Models

There is an increasing realization that repurposing drugs requires a collaborative approach, combining the strength of academia, industry, governmental bodies, and patient organizations. According to Pushpakom *et al*., there are three key components to successful drug repurposing collaborations: (i) identification of scientific experts with new ideas in RD biology, (ii) alternative funding routes, and (iii) enthusiastic engagement among all institutions involved [68]. Pharmaceutical companies not only have the necessary compound libraries of failed or shelved drug candidates but also have immensely valuable knowledge in translational research, drug development, and screening technologies that are often too expensive to acquire and maintain for many academic institutions. However, academic institutions tend to have access to patients and knowledge on disease biology and pathogenesis, which can be the basis for highly innovative drugs.

Recently, multiple repurposing initiatives have been established between pharmaceutical industry, grant funding organizations, and academic scientists to address some of the challenges in drug repurposing. One example is the Mechanisms for Human Diseases Initiative [90]. This way of working helps to reduce the risks involved and its cost is a fraction of that of creating a new therapeutic. It also has the potential to revolutionize the drug RD pipeline and empower a larger number of patient organizations to drive for treatments for their conditions.

Despite these great efforts, a clear one-way relationship is still often visible: a preclinical proof of concept for an old drug is developed by academia, which is, however, not in agreement with the

strategy of the pharmaceutical company owning the patent. As a result, no further evaluations under good industry practices follow and the drug never has a chance to reach the patient.

Another aspect of the collaborative approach is working together with patients, their families, or patient organizations. The establishment of hundreds of patient advocacy groups (PAG) and charitable foundations to support RD communities have increased public awareness, as well as interest of media and legislative bodies. PAGs give valuable insights for clinical trials such as identification of clinical end-points, assisting in patient recruitment, and can participate in meetings with regulatory agencies to discuss research and development activities.

Concluding Remarks and Future Perspectives

While individually, each RD impacts a small number of individuals, collectively, a large cohort is affected by these conditions. Once united, a well-defined patient population, a defined genetic etiology, and an appropriate model system can catalyze drug discovery. The scientific landscape for RDs has been changing rapidly and this change is expected to accelerate. RDs are now recognized as a global public health problem due to lack of treatment, number of hospitalizations over an individual's lifespan, and long-lasting diagnostic process. Researchers continue to successfully uncover the underlying molecular and genomic drivers as well as the clinical course of many conditions. Hence, people with RDs are increasingly benefiting from new therapeutics, some resulting from the break-through technologies now emerging in medicine [91,92]. Interestingly, in recent years, there has been an increase in funding for companies focusing on RDs, whereas financial support for common diseases has decreased, possibly due to a higher hurdle in developing and gaining approval for medicines that treat those conditions. Successful transition from one phase to the following in clinical studies between 2005 and 2018 was always higher for RDs compared with a group compiling all diseases or high-prevalence chronic disorders despite known challenges[iv].

Drug repurposing has gained significant traction due to its compelling advantages. According to different estimates, the number of repurposed drugs entering the regulatory-approval pipeline is rising and could account for about 20–30% of all drugs approved every year [93,94]. Recent progress and success stories have shown that drug repurposing is an efficient strategy to advance the discovery of treatments for RDs. From an economic view, the strategy of repurposing is attractive, especially when compared with the enormous costs and time investments associated with drug design and development of novel compounds. It is essential to hereby consider all possible tools and technologies available for the discovery process. NGS technologies especially have driven a dramatic shift in our understanding of RDs, increasing our knowledge of underlying causes and pathophysiological mechanisms, which can then be applied as the basis for the repurposing process. Here, computational advances and data sharing capability in particular can result in finding new indications for compounds in a more systematic manner, making drug discovery no longer dependent on serendipitous findings and observations (see Outstanding Questions).

Despite success stories, the repurposing process still encounters multiple challenges. Some pharmaceutical companies are not willing to reveal their chemical libraries of shelved or failed drugs to the public, which reduces the options for the repurposing process. Additionally, many traditional and approved drugs have poor physiochemical properties, resulting in low solubility, lack of specificity, and unrealistic dosing that cannot be achieved clinically but appears to be required for uses apart from their initial target. Hence, it can still be challenging to repurpose a certain compound despite the fact that it seems to be suitable for a new target in initial tests. Repurposed drugs still have to pass Phase II and III clinical trials for their new purpose; trials

Outstanding Questions

What measures can be taken to ensure early diagnosis of individuals with rare conditions?

How can we learn as much as possible about a rare disease from a small cohort of patients?

Can we develop more efficient and effective models for treatment development that reveal a clear readout and are sustainable?

What are the best practices to improve the collaboration between different stakeholders involved in rare disease research and drug discovery?

How can the access to industry-generated preclinical and clinical compounds for academic researchers be increased?

What are the best strategies to create further funding opportunities for drug repurposing initiatives (e.g., funding for appropriate technology)?

Will measures be developed to address patent and regulatory barriers, making drug repurposing more challenging?

resulting in the elimination of 30% and 60% of all tested drugs, respectively [95]. In some cases, repurposing drugs for new indications can even result in more severe side effects in RD patients than in the original patients. Additionally, product development also needs to be made fit-for-purpose, for example, if a drug is originally applied for adults and the new indication is a childhood disease, additional safety tests are required. Next to clinically related issues, there are multiple legal and regulatory barriers to drug repurposing. Regulatory incentives or formal guidance to encourage companies to invest in research and development of further uses for existing drugs are often lacking or, if existent, not fit for purpose [96]. Patent considerations during the drug repurposing process should not be neglected either, as the process of gaining marketing authorization for new therapeutic indications involves a great administrative burden and significant costs. For drugs that are out of basic patent and regulatory protection, the return on investment is expected to be low or absent [97]. On the contrary, pre-existing patents on the repurposing candidate might complicate commercialization. Not many pharmaceutical companies tend to consider out-licensing their discontinued programs. Interestingly, a study by Murteira *et al*. evaluating the regulatory implications of drug repurposing found that both in the EU and the US, the majority of repurposing cases were approved before patent expiry of the original product, forcing these cases to follow more complex regulatory pathways [98].

Nevertheless, by providing an overview of repurposing strategies for RDs, we hope that this article has highlighted the real potential of drug repurposing for RD patients, many of whom lack even the hope of treatment. Lastly, we believe that these strategies are not only applicable to RDs, but will most likely play an essential role in the progression of individualized and precision medicines.

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Declaration of interests

The authors have no conflict of interests.

Resources

[i]https://eur-lex.europa.eu/legalcontent/EN/TXT/?uri=CELEX%3A32000R0141

[ii]www.fda.gov/media/99546/download

[iii]https://omim.org/

[iv]www.bio.org/sites/default/files/legacy/bioorg/docs/Clinical%20Development%20Success%20Rates%202006-2015%20-%20BIO,%20Biomedtracker,%20Amplion%202016.pdf

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