Chapter 9

Summary, general discussion and directions for future research
Summary

Many acute and chronic lung diseases can cause pulmonary fibrosis and are commonly referred to as interstitial lung disease (ILD) or diffuse lung disease. In most cases the origin of ILD is not known. Genetic predisposition and environmental antigens are indicated as risk factors in the majority of these ILDs. Several studies have explored the association of genetic polymorphisms or the presence of certain variant alleles, with the occurrence and/or progression of ILD of unknown etiology.\textsuperscript{1-7} Moreover, the search for more specific ‘markers’ still continues.

The aim of the studies presented in this thesis was to evaluate the clinical and prognostic importance of genetic testing in a group of patients with drug-induced ILD (DI-ILD), a group of patients with oral anticoagulants induced diffuse alveolar hemorrhage (DAH), and a group of sarcoidosis patients. All these patients were referred to the ILD care center of the Maastricht University Medical Centre, the Netherlands.

The analyses performed were executed on whole blood samples of drug-induced ILD, idiopathic pulmonary fibrosis (IPF), and sarcoidosis patients. Of patients suffering from diffuse alveolar hemorrhage (DAH) analyses were done on whole blood samples and cells present in bronchoalveolar lavage fluid (BALF). For the analyses performed on healthy volunteer (HV) groups, whole blood and buccal swab (BS) samples were used. In afore mentioned populations the value of genotyping cytochrome P450 (CYP) enzymes (i.e. CYP2D6, CYP2C9 and CYP2C19) and/or vitamin K epoxide reductase (VKORC1) was examined. In sarcoidosis patients, next to tumor necrosis factor alpha (\textit{TNF-\alpha} G-308A) and butyrophilin-like 2 (\textit{BTNL2} G16071A), human leukocyte antigen (HLA) DRB1 and DQB1 were typed.

\textit{Chapter 1}, the general introduction, presents a summary of various ILDs, with an emphasis on drug-induced interstitial lung disease (DI-ILD), diffuse alveolar hemorrhage (DAH), and sarcoidosis. Furthermore, the term polymorphism and the possible ways in how to detect them are pointed out. In addition, the fluorescence resonance energy transfer (FRET) detection method used in this thesis is explained more in detail.

In \textit{chapter 2} the possible role of cytochrome P450 (CYP) enzymes in ILD, especially in drug-induced ILD (DI-ILD) was reviewed. The CYP enzyme family plays an important role in the metabolization of a variety of ingested, injected or inhaled xenobiotic substances. Polymorphisms in the CYP genes can
influence the metabolic activity of the subsequent enzymes, which in turn may lead to localized (toxic) reactions and tissue damage. Drug toxicity can be due to either absence or to very poor enzyme activity. In case of reduced enzyme activity, dose reduction or prescribing an alternative drug metabolized by a different, unaffected CYP enzyme is recommended to prevent toxic side effects. This is particularly important in multi drug-use. Knowing a patient’s CYP profile before drug prescription could prevent drug-induced ILD and other drug-induced toxicity. Moreover, it might be helpful in explaining serious adverse effects from inhaled, injected or ingested xenobiotic substances.

Chapter 3 describes an easy DNA isolation method to be used prior to genetic testing. Several commercial DNA isolation kits are available for extracting genomic DNA from whole blood samples, but these procedures are time consuming and expensive. An alternative technique could be dried blood spot (DBS) sampling. With the subsequent simple isolation method DNA isolation is faster, cheaper, and logistics are easier. This DBS DNA isolation method, an alternative to the commercially available DNA isolation kits, is practical and it discriminates between genotypes. It can also be used for buccal swabs resulting in good DNA yields and giving completely concurrent results with samples isolated using commercial DNA isolation kits. This expands the possibilities of this quick and easy DNA isolation procedure, especially in combination with the noninvasive patient friendly buccal swab sampling method. In addition, buccal swab sampling appeared to be a good alternative to invasive sampling methods.

Chapter 4 describes drug-induced pulmonary toxicity, a serious and expanding problem. Many drugs are metabolized by cytochrome P450 (CYP) enzymes. To establish whether allelic variation in CYP polymorphic genes contributes to variability in drug response and unexpected toxicity, a case-control study was conducted. The cases consisted of patients with drug-induced interstitial lung disease. Furthermore, two control groups were used: one group of healthy volunteers and one group of patients with idiopathic pulmonary fibrosis (IPF). Of the patients with drug-induced ILD 91.5% had at least one of the studied variant genes compared with 70.5% of the healthy volunteers and 69.1% of the IPF patients (both p’s<0.001). The percentage of individuals with one or more variant CYP genes was higher in the drug-induced ILD group. A significant association between the development of drug-induced ILD and the presence of one or more variant CYP genes was found. Drug-induced ILD was associated with the presence of at least one variant CYP allele. This study supports the potential usefulness of ‘personalized’ medicine by genotyping, aiming to improve efficacy, tolerability and drug safety.
The case presented in chapter 5 illustrates that understanding the mechanisms of drug metabolism and interactions may help to prevent side effects. The patient was a poor metabolizer for CYP2D6 corroborating that polymorphisms in this CYP gene influenced the metabolic activity of the corresponding enzymes. In addition, this affected the subsequent serum drug levels of venlafaxine and to a much lesser degree metoprolol and their metabolites. This achieved that toxic serum levels of venlafaxine were present and no active metabolite (O-desmethylenlafaxine) could be detected. Besides therapeutic drug monitoring, genotyping some important cytochrome P450 (CYP) enzymes was of additional value in explaining why the patient developed severe adverse effects. It also helped us to understand why the patient did not experience any therapeutical effect of the prescribed venlafaxine. This case highlights the potential benefit of clinical and genetic risk stratification (pharmacogenetics) before starting treatment.

In chapter 6 diffuse alveolar hemorrhage (DAH) is highlighted. DAH is a serious bleeding complication that can occur as a result of, among others, oral anticoagulation therapy. This study explored the hypothesis that in patients treated with coumarins DAH may be associated with vitamin K epoxide reductase complex1 (VKORC1) and cytochrome P450 (CYP) variant alleles in CYP2C9, and in case of acenocoumarol use also with CYP2C19 variants. Clinical information of patients using coumarins with at least one episode of DAH was gathered retrospectively during a seven year period. Out of 173 confirmed DAH cases, 75 had received oral anticoagulants and 63 of these 75 (84%) patients were included because DNA was available. Of these samples CYP and VKORC1 single nucleotide polymorphisms (SNPs) were genotyped. In 62 out of 63 DAH cases VKORC1, CYP2C9 variant genes, or both were found. This indicates that genotyping appears to be useful in predicting a high risk of serious side effects related to oral anticoagulant use, including DAH. Consequently, in concurrence with the American Food and Drug Administration (FDA), we recommend this genotyping, in order to provide a safer and more individualized anticoagulant therapy.

Chapter 7 describes a study designed to evaluate the relationship between the presence of tumor necrosis factor alpha (TNF-α) polymorphisms and the prognosis of sarcoidosis. In a retrospective case-control study TNF-α G-308A, TNF-α G-238A and lymphotoxin-α (LTA) were genotyped in 625 sarcoidosis patients. These patients were classified into 327 patients with non-persistent (regressing to or stable at chest X-ray stage 0 or I) disease and 298 patients with persistent disease using chest X-ray appearances and lung function test results after a period of at least two years of follow-up. The TNF-α -308A variant allele was observed in 25.5% of patients with persistent disease
compared with 44.0% of patients with non-persistent disease. Consequently, the presence of a $\text{TNF-}\alpha$ -308A variant allele is associated with a favorable prognosis. Because of the strong linkage between $\text{TNF-}\alpha$ G-308A and HLA-DRB1*03, genotyping of one relatively simple TNF-\(\alpha\) SNP is useful in predicting the prognosis of pulmonary sarcoidosis.

In chapter 8 the association between butyrophiline-like 2 ($\text{BTNL2}$) G16071A with the course of pulmonary sarcoidosis was assessed and the association with disease predisposition was verified in 632 sarcoidosis patients. In addition, the linkage between $\text{BTNL2}$ G16071A and certain HLA-DRB1 and HLA-DQB1 types was investigated. It appeared that the $\text{BTNL2}$ 16071A variant allele was significantly more often present in patients with persistent disease (92.4%) compared with patients having non-persistent disease (86.6%). The presence of a $\text{BTNL2}$ 16071A variant allele was found to be associated with an almost twofold increased risk of progressing to more severe and persistent pulmonary sarcoidosis. Furthermore, the predisposition to develop sarcoidosis was confirmed, as well as the strong linkage between the $\text{BTNL2}$ 16071A variant allele and DRB1*15 positivity. It also became apparent that typing for DRB1 is sufficient because of the lack of additional information obtained by typing the DQB1*06, to establish the 15Q6 haplotype. Whether or not to determine the DRB1 type or test the $\text{BTNL2}$ G16071A SNP therefore, depends on the ability and/or availability to perform either test. Additional research will be necessary to explore the role of these findings in the clinical management of sarcoidosis patients.
General discussion

Both genetic and non-genetic information is important in the susceptibility, development, cause, and treatment response of diseases. The more we know about a patient’s genes and context, the better disease management decisions can be made.\(^8\) Nevertheless, often the cause of a disease appears to be not only associated with a single nucleotide polymorphism (SNP), but is much more complicated. Some patients will continue to react unpredictably to therapy even though, according to obtained tests results, problems were not expected. Extending mapping of transcription factor binding SNPs and structural variants associated with transcription factor binding might gather useful information about the role of genetics in the phenotypic diversity in humans and provide insight into genetic causes of human disease.\(^9,10\)

The success of modern medicine is partly the result of effective medical treatment. Although the overall advantage of many drugs outweighs the side effects, substantial costs are still made as a result of complications of drug therapy. The variability in drug response among patients is multi-factorial. These factors include extrinsic features like environmental aspects or co-medications, but also genetic and intrinsic factors that affect the disposition (absorption, distribution, metabolism and excretion) of individual drugs. The existence of large population differences with small intra-patient variability is consistent with inheritance as determinant of drug response. It is estimated that genetics can account for 20-95% of the variability in drug disposition and effects.\(^11\) Together clinical and genetic risk stratification (pharmacogenetics) may lead to more accurate prevention of drug-induced damage in the future.

In a recent editorial the results of almost ten years after the revealing of a draft sequence of the human genome were assessed. Although detailed maps of genetic markers of human variation, have facilitated many remarkable genomewide efforts to associate known SNPs with disease predisposition, more than one decade of genomics will be required to understand the inborn risks of most common disorders.\(^8\) Nevertheless, reviews that highlight successful applications of gene-based medicine might hasten adoption of the beneficial changes in medicine that will eventually, if gradually, come from gene-based sciences.\(^8\) All the same, our increased understanding of the interactions between the entire genome and non-genomic factors that result in health and disease is paving the way for an era of ‘genomic medicine’.\(^12\)

There are an increasing number of examples where pharmacogenetic studies have indicated that a genetic test prior to treatment may be useful either for setting the individual dose or choosing a certain drug.\(^13-18\) The ability to identify
individuals who are susceptible to adverse drug reactions has the potential to reduce the personal and population costs of drug-related morbidity and the potential to attribute to the patients’ safety. Especially persons with more than one cytochrome P450 (CYP) polymorphism and/or other relevant polymorphisms may be susceptible to develop adverse effects, such as drug-induced interstitial lung disease (ILD), when (multiple) drugs are prescribed.\textsuperscript{19} Moreover, other pharmacogenetic factors might be involved, such as those involved in methotrexate induced pneumonitis.\textsuperscript{20-22}

One of the major conclusions of this thesis is that genetic screening prior to drug prescription may potentially prevent serious adverse effects such as diffuse alveolar hemorrhage (DAH) or drug-induced ILD. Another important finding is that the results obtained by genetic testing appeared to be useful in disease management, because of the prognostic value of the absence or presence of specific polymorphisms.

Polymorphisms in drug-induced lung diseases

Not only drug interactions, environmental factors, disease processes, and aging are factors in the inter-individual metabolic capacity variance, genetic factors also play an important role in developing adverse effects in general. Considering the fact that bio-(in)activation by CYP enzymes play an important role in human drug toxicity, polymorphisms in the CYP enzyme system may result in large inter-individual variations in the metabolism and toxicity of xenobiotics. The presence of CYP variant alleles adds a substantial susceptibility risk factor to the development of drug-induced pulmonary adverse events.\textsuperscript{19,23,24} In the study presented in chapter 6, genetic allelic variants appeared to be one of the determinants of variability in sensitivity to coumarins. Furthermore, others found that even in patients with stable international normalized ratios (INRs) without adverse (bleeding) effects these polymorphisms had a profound influence on dose requirements and time to reach therapeutic INRs.\textsuperscript{15,25} Despite numerous attempts to standardize the management of oral anticoagulant therapy, a high proportion of patients are still inadequately anticoagulated and the optimal means to initiate the therapy is still a matter of debate. It has been suggested that dosing algorithms incorporating genetics might outperform usual care. However, a major obstacle to the widespread adoption of genetic based coumarin dose modelling is that access to timely genetic testing is currently not widely available.\textsuperscript{26} Nevertheless, even despite its limitations, prospective genotyping for CYP2C9 and VKORC1 of patients taking oral anticoagulants has the clear potential to significantly optimize the safety of drug therapy and set a promising example of ‘personalized’ medicine.\textsuperscript{10}
Drugs, oxidative stress and pulmonary damage

Beside anticoagulants, D-penicillamine, nitrofurantoin, amiodarone, propylthiouracil, cocaine, or sirolimus, and/or exposure to toxic agents such as trimellitic anhydride, insecticides, and pesticides may also cause bleeding complications including DAH events. It has been suggested that oxidative damage plays a role in the pathophysiology of various diseases, including DAH and pulmonary fibrosis. DAH is characterized by pulmonary alveolar cell death, inflammation, and hyaline membranes composed of fibrin and cellular debris. Idiopathic pulmonary fibrosis (IPF) is a chronic, progressive and frequently fatal form of ILD, characterized by impaired fibrinolysis and a complex pathogenesis. As a result of diffuse alveolar bleeding events iron is released in the lung. In turn this free toxic iron causes oxidative stress, inflammation, and finally irreversible damage or fibrosis. Moreover, people with VKORC1 and/or CYP2C9 variant alleles might have a predisposition for a greater risk of bleeding events in case they use oral anticoagulants, which might make them more susceptible to iron-catalysed toxicity caused by oxidants. This is important as anticoagulant therapy is considered to be an additional new strategy to treat IPF patients. In contrast, Thomassetti et al. did not find a beneficial effect of anticoagulants. It is tempting to speculate that an association with VKORC1 and/or CYP2C9 variant alleles might even be a risk factor for the development or exacerbation of IPF. Furthermore, it was accentuated that in DAH cases early recognition of the presence of one of the studied polymorphisms is important, because of a potential lethal outcome and the fact that simple vitamin K supplementation can be life-saving. Increased availability of vitamin K through daily supplementation, is expected to diminish the inhibitory activity and the relative day-to-day variability of coumarins and can significantly improve anticoagulation control in unstable patients.

Importance of various genetic variants

It was emphasized that early recognition of the presence of certain polymorphisms is important. Not only in case of anticoagulant use, also in psychiatry, pharmacogenetic testing has become more common practice in the United States (US) with FDA approval of the AmpliChip CYP450 and CYP2D6 and CYP2C19 testing helping patients with a history of excessive difficulties with antidepressants. This was also illustrated by the case described in chapter 5. Besides CYP and VKORC1 polymorphisms other genetic variants are also important. Two main immunosuppressive drugs that are often used in the treatment of severe ILD are methotrexate (MTX) and azathioprine. MTX is frequently used in the treatment of autoimmune diseases such as rheumatoid arthritis, psoriasis, Crohn’s disease, as well as in sarcoidosis. MTX toxicity and efficacy is associated with a number of polymorphic enzymes and testing for
variants was found to be predictive of response to and safety of MTX treatment.17,21,22,41 Moreover, when a patient develops an adverse drug reaction after starting MTX treatment, testing the C677T and A1298C variants in the methylenetetrahydrofolate reductase \( (MTHFR) \) gene involved in the MTX metabolism, should be considered.42 In case of an azathioprine indication, also used as treatment for certain ILDs including IPF, testing thiopurine methyltransferase \( (TPMT) \) variants involved in the azathioprine metabolism is advised before starting treatment.43-45 In the US, drug labels for azathioprine now include information on TPMT polymorphisms and recommend determining patients’ phenotype or genotype prior to drug treatment.46

Sarcoidosis

Sarcoidosis is a chronic granulomatous disorder of unknown cause with a highly variable course, characterized by activation of T-lymphocytes and macrophages. In an effort to determine the etiology of sarcoidosis the multicenter study A Case Control Etiologic Study of Sarcoidosis (ACCESS) was designed.47 One of the lessons learned from ACCESS was that genetic associations with sarcoidosis were observed and appeared to be stronger predictors of the course of sarcoidosis than environmental factors.48 The assumption that genes contribute to the etiology of sarcoidosis comes from the observation that prevalence and incidence rates of sarcoidosis are different between ethnic groups and that the disease tends to cluster in families. Interactions of exposures with genetic predispositions would have important implications for the understanding of immune responses as well as the pathogenesis of sarcoidosis.47 Prognostic factors of sarcoidosis are very important because certain obstacles confound the accurate prediction of the prognosis of sarcoidosis, such as the lack of reliable activity markers, the intensity level of granulomatous response, and the inability to differentiate the response from the dys-regulated repair process leading to fibrosis.47

The role of genes in the etiology of sarcoidosis

Association of sarcoidosis and class I and II HLA antigens is well known and in several studies the risk of progression of sarcoidosis, and the presence or absence of a polymorphism has been established.1,3,24,49,50 In addition to HLA types, co-stimulatory molecules of the immunoglobulin superfamily are also necessary to activate T-cells. The butyrophilin-like 2 \( (BTNL2) \) gene is located close to and in linkage with HLA-DRB1, which in turn is implicated in the etiology of sarcoidosis.51-54 Moreover, the presence of the \( BTNL2 \) 16071A variant allele was recently associated with an increased risk of developing sarcoidosis.55-58 In T-helper cell 1 (Th1) dominated granulomatous diseases similar to sarcoidosis, an association between the presence of \( BTNL2 \) G16071A
and disease predisposition was found. This association was attributed to the strong linkage with HLA-DRB1/DQB1 haplotypes. Nevertheless, because of this strong linkage between BTNL2 and HLA-DRB1/DQB1, BTNL2 G16071A should be considered relevant to any immune-related disease associated with HLA-DRB1/DQB1. In the study presented in this thesis, the strong linkage between the BTNL2 16071A variant allele and HLA-DRB1*15 was confirmed. Another conclusion was that the presence of the BTNL2 A-allele was also associated with an increased risk of progressing to a persistent form of pulmonary sarcoidosis. Another SNP that was confirmed to influence sarcoidosis susceptibility is cyclooxygenase-2 (COX2) T8473C, however, no association with progression was found. Therefore, determining the BTNL2 G16071A genotype even prior to the development of complaints, can be beneficial, for example when there is a family history of sarcoidosis. Not only to establish whether or not someone is more prone to develop sarcoidosis, but also whether there is an additional risk to progress to a more persistent form. Recently, strong evidence was found for the postulation that several SNPs in the vascular endothelial growth factor (VEGF) and its receptor genes VEGFR-1 and VEGFR-2 also possessed the ability to predict predisposition and progression of sarcoidosis, but further studies are needed to evaluate the clinical relevance.

The influence of genes on the course of sarcoidosis

Recently, it was found that serum amyloid A plays an important role in granuloma formation in sarcoidosis as well as tumor necrosis factor (TNF), interleukin-10 (IL-10), interferon-gamma (INF-γ), and Toll-like receptor 2 (TLR2). In turn, these cytokines and enzymes not only influence the serum amyloid A production, but their expression can also be influenced by possible polymorphisms in their coding or non-coding sequence, and thus influence the cause or course of sarcoidosis. The potent pro-inflammatory cytokine TNF-alpha (TNF-α) for instance plays a pivotal role in inflammatory and immune responses, and regulates and sustains granuloma formation in sarcoidosis. Several SNPs are identified in the TNF-α gene and especially the variant A-allele of the TNF-α G-308A gene is associated with higher TNF-α serum levels and an acute course of sarcoidosis. The findings presented in chapter 7 of this thesis demonstrate that it is sometimes beneficial to possess a variant allele (i.e. TNF-α -308A), opposed to not having a polymorphism, in the course of sarcoidosis. The risk of progressing to a more severe pulmonary involvement or persistent form of sarcoidosis was found to be higher in the absence of a TNF-α G-308A allelic variant, corroborating findings of other studies conducted on smaller populations. In line with this, bearing the HLA-DRB1*03 type also predicted a more favorable outcome, as was previously established in sarcoidosis patients with Löfgren's syndrome, the
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acute form of sarcoidosis with predominantly spontaneous remission.\textsuperscript{1,2,66,68} What is more, both \textit{TNF-\alpha} G-308A and HLA-DRB1*03 typing appear to be interchangeable as far as the outcome or prediction of the course of sarcoidosis is concerned, consequently confirming the strong linkage between both genes.\textsuperscript{68,69}

The role of genes in therapeutic management

Because of the strong linkage disequilibrium found between several genes and HLA-types, the choice of whether or not to test for a simple SNP (for example \textit{BTNL2} or \textit{TNF-\alpha} gene SNPs) or perform HLA typing appears to depend more on the availability of the technique or test. Conclusions drawn from studies presented in this thesis can be useful in furthering the clinical validation of applying genotyping in predicting the clinical course of sarcoidosis in individual patients and fine-tune disease management. Since available sarcoidosis therapies are not without risk and sometimes even toxic, patient care would benefit from the ability to predict the progression of the disease.\textsuperscript{70} Therefore, identifying those cases that might have a chance to develop a more severe manifestation of sarcoidosis is important and treatment aimed at avoiding irreversible damage can be started early.\textsuperscript{71} Furthermore, if the course of the disease has already progressed to severe or refractory sarcoidosis it would be of great clinical relevance to be able to select those cases who might benefit from a certain drug and who will not. For instance, when corticosteroids, most often the first drug of choice in sarcoidosis, are not effective, MTX might be considered. It has been beneficial in certain cases, however, it has a variety of clinical efficacies and toxicities, which are difficult to predict.\textsuperscript{22,72} Several reports have suggested that the use of pharmacogenetics might help to improve the understanding of drug efficacy and toxicity. However, studies in rheumatoid arthritis or psoriasis patients showed conflicting data whether or not these side effects were caused by the altered expression of genes by MTX.\textsuperscript{22,73,74}

Recently, anti-TNF-\alpha therapy has proven to be useful in the treatment of refractory sarcoidosis. However, response to this therapy is not always as promising as expected.\textsuperscript{70,75} Moreover, it takes some time before the effect of these so-called biologicals is apparent, substantial side-effects are reported and additionally, this therapy is expensive. In refractory sarcoidosis, bearing a \textit{TNF-\alpha} G-308A variant allele appears to be a disadvantage. In previous studies conducted in rheumatoid arthritis patients not possessing a variant allele seem to respond to the therapy in contrast to variant allele carriers.\textsuperscript{76,77} Insight into the patient’s probable response might provide important information, especially for those patients, who very likely will not benefit from this anti-TNF-\alpha therapy, and therefore should be treated with alternative medication without delay.
Directions for future research

There is still a need for well designed prospective clinical trials that measure patient-oriented outcomes of selected genomic applications, and studies that evaluate the role of genomic variation in disease susceptibility, predicting prognosis, treatment response and in tailoring drug treatment for individual patients. These investigations are aimed to help to bridge the gap between ‘personalized’ and ‘evidence-based’ medicine\textsuperscript{12}

In order to ascertain anticoagulation levels quicker and safer, without the risk of serious side effects and identify coumarin sensitive cases, genotyping for \textit{VKORC1}, \textit{CYP2C9}, and \textit{CYP2C19} polymorphisms would be a preferable cause of action. This genotyping should be performed ideally prior to oral anticoagulant therapy, but certainly in case of high and unstable or even overshoot target INRs. A cost-effectiveness study, evaluating whether or not screening for \textit{VKORC1} and \textit{CYP2C9} polymorphisms prior to administering oral anticoagulant therapy is of clinical relevance, is therefore highly recommended.

Furthermore, a prospective study to determine if, when variant alleles are present, simultaneous oral vitamin K supplementation can prevent diffuse alveolar hemorrhage (DAH) events is needed. Whether or not an association exists between \textit{CYP} and \textit{VKORC1} polymorphisms and the development and/or progression of pulmonary fibrosis is another important issue for future studies. Hypothesizing that fibrosis could be associated with repeated episodes of diffuse alveolar damage to a certain extent is intriguing. Prospective clinical trials are needed to clarify the role of anticoagulant therapy and other alveolar bleeding initiating agents in the development of pulmonary fibrosis. Further investigations are also needed to assess if anticoagulant therapy is a friend or foe in the therapeutic strategies of pulmonary fibrosis. Therefore, it would be interesting to evaluate whether screening of patients before initiating anticoagulant therapy might be of clinical relevance.

MTX, frequently used in sarcoidosis in combination with or without anti-TNF-\textalpha therapy, is also influenced by the presence of variant alleles. For that reason, a study into the influence of these polymorphisms on the efficiency and efficacy of this drug in sarcoidosis therapy should certainly be considered. Moreover, these studies might even give an answer to the question about which patients would benefit from MTX and for whom no advantage can be achieved or for whom the result will even be detrimental. It might be fascinating to develop a screening system based on both clinical and genetic information.
Next to the information about the progression of sarcoidosis, a patient’s TNF-α status or genetic make-up can also play a significant role in therapeutic disease management. Assessing a possible association in individual cases between the absence of a \( TNF-\alpha \) -308A variant allele and being a responder or non-responder to anti-TNF-α therapy would be of great clinical relevance. The results could provide important information about whether or not a patient would possibly benefit from anti-TNF-α therapy. Therefore, research should continue to depict the role of \( TNF \) genes in the immunogenetics and clinical management of sarcoidosis.

The ability to identify individuals who are susceptible to adverse drug reactions, with the inclusion of both clinical and genetic risk stratification, may lead to a more accurate prevention of drug-induced damage and has the potential to reduce the personal and population costs of drug-related morbidity.\textsuperscript{78,79} The introduction of a genetic medical passport for each patient aimed to achieve a more appropriate individual pharmacotherapy seems promising in therapeutic drug monitoring. However, whether this might reduce the risk of side effects and related medical consumption needs to be evaluated. Collaboration between medical specialists, clinical pharmacists, pharmacologists and laboratory specialists will be necessary to accomplish individualize pharmacotherapy based on the pharmacogenetic profile.\textsuperscript{80}
References


