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PHARMACOKINETIC VARIABILITY OF TACROLIMUS IN RENAL TRANSPLANT RECIPIENTS

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Abstract

Tacrolimus remains the cornerstone calcineurin inhibitor after kidney transplantation, but its narrow therapeutic range and large differences in exposure between and within patients make dosing difficult. This narrative review draws on published pharmacokinetic, pharmacogenetic and clinical studies to describe where this variability comes from, what it means for graft outcomes, and how dosing strategies try to contain it. CYP3A5 genotype explains the largest identifiable share of dose requirement, while diet, interacting drugs, haematocrit, diarrhoea and adherence add variability over time. Genotype-guided and model-informed dosing improve early target attainment, though clear benefit on rejection or graft survival is not established. The review reports no new data.

The consequences of missing the target run in both directions. Low exposure in the first weeks is associated with acute rejection, and persistently low levels later are linked to donor-specific antibodies and chronic injury. High exposure brings nephrotoxicity, neurotoxicity, tremor, new-onset diabetes and an increased burden of infection and

1. INTRODUCTION

Few drugs in clinical use are as awkward to dose as tacrolimus. Introduced in the 1990s, it largely replaced ciclosporin as the backbone of immunosuppression after kidney transplantation because it reduced acute rejection. It acts by binding the intracellular protein FKBP12, and the complex inhibits calcineurin, blocking activation of T cells through the NFAT pathway and cutting production of interleukin-2 [1, 2]. The benefit is real, but the margin between too little drug and too much is narrow, and the same dose given to two patients can produce blood concentrations that differ several-fold.

malignancy [2, 3]. For this reason, tacrolimus is among the drugs for which therapeutic drug monitoring (TDM) is standard practice, and consensus groups have published detailed recommendations on target ranges and sampling [3, 4].

Pharmacodynamic variability sits on top of the pharmacokinetic kind. Two patients with identical whole-blood concentrations may differ in how strongly their T cells are inhibited, because intracellular drug levels, calcineurin activity and the genes that govern the immune response also vary. Attempts to measure calcineurin activity or NFAT-regulated gene expression as direct markers of effect have so far not become routine tools. Blood concentration therefore remains a rough proxy, useful mainly because it is cheap and reproducible, and the target ranges recommended by guidelines reflect experience in populations, not individual optimal exposure [2, 3].

This article reviews the published evidence on pharmacokinetic variability of tacrolimus in adult renal transplant recipients. It first describes the pharmacokinetic features that create the problem, then discusses genetic and non-genetic sources of variation, the evidence linking within-patient variability to outcomes, and the dosing and monitoring strategies designed to respond. It is a narrative synthesis of previously published studies and reports no new data, and the literature considered is limited to that available up to 2022.

2. PHARMACOKINETIC BASIS

Several properties of tacrolimus conspire to make exposure unpredictable. Oral absorption is incomplete and highly variable, with reported bioavailability ranging from a small fraction to most of the dose and a mean around a quarter [1, 2]. Absorption depends on intestinal P-glycoprotein, which pumps the drug back into the gut lumen, and on first-pass metabolism by cytochrome P450 (CYP) 3A4 and CYP3A5 in enterocytes. Both the intestinal and hepatic enzymes are subject to large inter-individual differences in expression, so the share of drug that reaches the circulation differs from one person to the next [5, 6].

Once in blood, tacrolimus partitions heavily into red cells, where it binds FKBP12, so that whole-blood concentrations are many times higher than plasma concentrations. The small plasma fraction is largely bound to albumin and alpha-1-acid glycoprotein. Laboratories measure whole blood, and the result therefore depends on haematocrit as well as on the amount of drug present [1, 7]. After transplantation, anaemia is common and recovers over several weeks, which means that the relationship between dose and measured concentration shifts during exactly the period when dosing is most critical.

Elimination is mainly by hepatic CYP3A metabolism, with minor renal excretion of unchanged drug. The terminal half-life is long and variable, from roughly ten hours to more than twenty-four hours in different studies, and clearance differs widely between patients [1, 2]. Population pharmacokinetic analyses in adult kidney recipients have identified weight, haematocrit, time since transplantation and co-administered corticosteroid among the covariates, but a large part of the variation remains unexplained even in the best models [5, 7]. This residual variability is the target of most research.

The time course after surgery needs separate mention, since the first month is the period of greatest instability. Haematocrit is low, steroid doses are high, graft function is recovering, gut motility may be affected, and many new drugs have been started. Dose requirement per kilogram falls over the following weeks as these factors settle, and clearance estimates from early population models differ from those obtained in stable patients years later [5, 7]. For clinicians, the practical lesson is that a dose that was right in week one may not be right in week eight.

The immediate-release twice-daily capsule and the prolonged-release once-daily formulation behave differently. The prolonged-release product has a lower peak, a flatter profile and a lower total exposure for the same dose, which must be considered when switching a patient from one to the other. A randomised study showed comparable efficacy and safety of once-daily and twice-daily regimens in de novo kidney

recipients [8], and a trial using electronic monitoring found improved adherence with the once-daily regimen [9]. Switches between brands or generics, if uncontrolled, add another potential source of variability.

Analytical method adds its own noise. Immunoassays used in many laboratories show some cross-reactivity with tacrolimus metabolites and can overestimate concentrations relative to liquid chromatography with tandem mass spectrometry, particularly at low levels and in patients with hepatic impairment. Consensus reports therefore recommend that laboratories report the method used, participate in external quality assessment, and that clinicians should not switch assay without considering the effect on trend interpretation [3, 4]. A change in measured concentration may reflect a change in assay rather than in the patient.

Because trough concentration is measured so often, it is worth recalling that it is only a surrogate for total exposure. The area under the concentration-time curve (AUC) over a dosing interval is the pharmacologically preferred measure, but multiple samples are impractical. Limited sampling strategies and Bayesian estimation reduce the burden, and consensus guidance accepts the trough as a pragmatic compromise for most patients, while recommending AUC-based methods in selected situations [3, 4]. Evidence that trough alone predicts acute rejection is surprisingly modest in pooled trial data, which suggests that the question of what the best monitoring target is remains open [10].

Hepatic function influences exposure through reduced CYP3A activity. Patients with liver impairment can have markedly prolonged half-lives and need lower doses, and rises in trough concentration unexplained by interactions should prompt a review of liver tests. Albumin levels may also change the free fraction. Although these effects are well described, they account for a smaller share of the variability in renal recipients than genotype and interactions do [1, 2]. Dose reductions of one third or more are described in severe cases, with close monitoring. Cholestasis and low albumin can both shift concentrations.

Drug transporters other than P-glycoprotein have received less attention for tacrolimus, and the evidence on them is thin. Efflux in the kidney may influence intragraft concentrations independently of blood levels, and some investigators have argued that intracellular lymphocyte concentrations would be a better marker of effect. Such work remains exploratory. It does serve as a reminder that blood concentration, however carefully measured, describes the compartment that is easiest to sample, not necessarily the one where the drug acts or damages [1, 6].

3. GENETIC AND CLINICAL DETERMINANTS

The most important single genetic determinant is CYP3A5. Carriers of at least one functional CYP3A5*1 allele express the enzyme and clear tacrolimus faster, while people homozygous for the common loss-of-function CYP3A5*3 allele express little or none. The frequency of expressers varies by ancestry, being a minority among people of European descent and a majority among people of African descent [6, 11]. Early studies showed that expressers needed higher doses to reach the same concentration, and the finding has been replicated in many cohorts and in a meta-analysis of observational work [11, 12, 13].

Other genetic variants contribute less. A functional polymorphism in CYP3A4 intron 6 (CYP3A4*22) reduces enzyme expression and is associated with lower dose requirement, though its effect is smaller and mainly relevant in CYP3A5 non-expressers [14]. Polymorphisms in the ABCB1 gene that encodes P-glycoprotein were examined in early reports, but results have been inconsistent and the clinical relevance is doubtful. Dosing equations that combine genotype with clinical covariates explain only part of the variation in dose, which reflects both the contribution of non-genetic factors and the limits of current models [15, 16].

The CPIC guideline translates the genotype findings into advice. For adult recipients who are intermediate or extensive metabolisers, meaning they carry one or two functional CYP3A5*1 alleles, a starting dose of 1.5 to 2

times the standard, not exceeding 0.3 mg/kg/day, is suggested, with subsequent adjustment guided by trough concentration [17]. Poor metabolisers, who carry two non-functional alleles, are given the standard starting dose. The guideline says explicitly that monitoring remains necessary, since genotype explains only part of the variability and acts as a starting point rather than a replacement for measurement [6, 17].

Food and drugs are well-recognised non-genetic influences. Food reduces both the rate and extent of tacrolimus absorption, with a high-fat meal having more effect than a low-fat one, so consistent timing relative to meals matters [18]. Inhibitors of CYP3A4 and P-glycoprotein, including azole antifungals, macrolide antibiotics such as clarithromycin, and non-dihydropyridine calcium channel blockers, raise concentrations and sometimes by large amounts. Inducers such as rifampicin, phenytoin, carbamazepine and St John's wort cut them sharply. Corticosteroids have a modest inducing effect, which is why falling prednisolone doses after transplantation lower the dose requirement [19, 20].

Gastrointestinal events are an underappreciated cause of fluctuation. Persistent diarrhoea, which is common after transplantation because of infection or mycophenolate, has been associated with increased tacrolimus concentrations, probably through altered gut enzyme and transporter activity and changes in absorption [21]. A patient with diarrhoea may therefore need a dose reduction, despite concerns about under-absorption, and a sudden jump in trough concentration without a change in dose should prompt enquiry into bowel habit as well as drug interactions and adherence.

Adherence is arguably the largest uncontrolled clinical factor. Immunosuppressive regimens require strict timing, and transplant recipients, particularly adolescents and young adults, may miss or delay doses in ways that are rarely admitted. Electronic monitoring studies show that missed doses and irregular dose timing are common, and that simplifying to a once-daily formulation improves taking adherence in at least some populations [9, 22]. Because non-adherence mimics pharmacokinetic variability on routine monitoring, interpretation of unexplained fluctuations should include a candid, non-judgemental conversation with the patient.

Clinical features such as age, sex, body mass index and time since transplantation have also been examined. Dose requirement per kilogram is generally higher in the first days and decreases over the following months as haematocrit rises and steroid doses fall. Evidence for effects of age and sex is mixed, with some analyses reporting that older patients need lower doses, but the contribution is minor compared with genotype [16]. Studies using theory-based pharmacokinetic models suggest that body size scaling, haematocrit and CYP3A5 together predict early clearance better than any single factor [7].

4. VARIABILITY AND GRAFT OUTCOMES

Variability within the same patient over time has attracted the most attention in recent years. It is typically summarised as the coefficient of variation (CV) of trough concentrations (or of dose-corrected concentrations) measured at stable doses over months. Reviews describe a long list of causes, including non-adherence, changes in diet, intercurrent illness, interacting drugs, formulation switches and measurement error, with adherence thought to account for much of the high end [22, 23]. The measure is attractive because it is simple to calculate from routine monitoring data.

Retrospective cohort studies have linked high within-patient variability to poor outcomes. In a Dutch cohort, high variability in clearance was a risk factor for chronic rejection [24]. A Spanish analysis found that greater variability predicted graft loss and the development of donor-specific antibodies [25], and a Canadian study reported that time-dependent variability in trough levels was a risk factor for late graft failure [26]. These findings are consistent across countries and across analytic methods, which strengthens confidence that the association is real [23].

What the association means is debated. Variability may cause harm directly, because

periods of under-exposure allow alloimmune responses to escape suppression. Alternatively, it may be a marker of other risks, particularly poor adherence, which would still cause rejection if exposure were stabilised with another formulation. Observational data cannot easily separate the two possibilities. High clearance, with consistently low dose-adjusted concentrations, has also been linked to early acute rejection, so mean exposure matters in addition to the spread around it [27].

Choosing a target concentration adds a further layer. The ELITE-Symphony trial, in which a low-dose tacrolimus regimen with induction therapy gave good renal function and low rejection rates, showed that lower exposure than previously used can be effective when combined with other immunosuppressants [28]. Guidelines, including those of KDIGO, therefore specify target ranges that fall with time after transplantation and vary by the combined regimen and immunological risk, instead of a single number [3, 29]. Interpreting variability in this context means asking whether fluctuations bring the patient below the range for that period, not whether the numbers vary at all.

Nephrotoxicity links exposure to outcome from the opposite side. Calcineurin inhibitors cause afferent arteriolar vasoconstriction, and prolonged high exposure leads to arteriolar hyalinosis and interstitial fibrosis in biopsies. Distinguishing toxicity from rejection on the basis of the trough alone is unreliable, since both occur across overlapping concentration ranges, and biopsy is often needed. Reduction of exposure to limit toxicity is the logic behind the low-dose regimens in modern practice, and it makes accurate dosing even more important, because the margin for error shrinks as targets are lowered [2, 28].

5. DOSING AND MONITORING STRATEGIES

Standard practice is a weight-based starting dose followed by adjustment according to trough concentration. The approach is simple and works well enough for many patients but leaves a large proportion outside the target at the first measurement, so that several adjustments are needed before the range is reached. During that period, patients are exposed to either rejection risk or toxicity, which has motivated alternatives [3, 5].

Timing of sampling has practical consequences. Steady state is reached about two to three days after a dose change in most patients, and trough samples are intended to be taken immediately before the next dose, around twelve hours after the evening dose for the twice-daily capsule and twenty-four hours for the once-daily product. Samples drawn late give falsely low troughs and prompt unnecessary dose increases. Written instructions for patients, and clinic procedures that record the time of the last dose, are cheap measures that reduce this avoidable error [3, 4, 29].

Genotype-guided dosing is the best studied alternative. A randomised study in France showed that adjusting the initial dose according to CYP3A5 and ABCB1 genotype increased the proportion of patients with a first trough in the target range [30]. A larger Dutch randomised trial in living-donor transplant recipients found that CYP3A5 genotype-based dosing improved target attainment at the first measurement, but did not reduce the incidence of the composite endpoint of acute rejection, graft loss or death [31]. The Clinical Pharmacogenetics Implementation Consortium recommends higher starting doses for expressers [17].

Model-informed precision dosing goes further by combining genotype and clinical covariates with measured concentrations to give individual dose predictions. A prospective randomised study of computerised dosing, using a population model including genotype, haematocrit and steroid dose, found that more patients reached the target concentration early than with standard care [32]. Such tools require software, validated models and staff trained in their use, and their cost-effectiveness has not been demonstrated outside a few centres. Nevertheless, they address the main weakness of simple weight-based dosing, which ignores the factors known to matter [7, 32].

Conversion between formulations needs particular care. Patients moved from the twice-daily capsule to the prolonged-release product are usually given the same total daily

dose, then monitored closely, since the lower exposure may require an increase of a few tens of per cent in some patients. Conversion in the other direction can raise troughs. Pivotal trials supported equivalence of efficacy at the population level, but individual responses vary, and the first weeks after conversion are a high-risk period for out-of-range troughs [3, 8]. Pharmacy records should flag any unintended brand substitution.

One more practical point concerns communication. Concentration results are only useful if dose changes reach the patient reliably, and errors in transcribing instructions are a recognised source of harm after discharge. Many units now give patients a written dose card, and some use mobile applications to record doses and troughs. Clear instructions on what to do when a dose is vomited or missed, when diarrhoea begins, or when a new antibiotic is prescribed by another clinician all help to limit avoidable swings in exposure [3, 22].

6. LIMITATIONS AND FUTURE DIRECTIONS

The principal findings reviewed here are brought together in table 1, and the spread of entries by theme is displayed in fig. 1. The table makes clear that sources of variability are better characterised than the effect of reducing them on hard clinical outcomes. Studies of genotype-guided and model-based dosing have mostly used target attainment as the primary endpoint, which is a surrogate, and the trials were not powered to detect differences in rejection or graft survival.

Several caveats apply across the evidence. Most cohorts are single-centre and retrospective, with different assays, target ranges and co-immunosuppression, so comparisons between them are imprecise. Ancestry is poorly represented in many genetic studies. Measures of variability are calculated in a variety of ways, and there is no agreed threshold at which variability becomes clinically important. Prospective work will need to define such a threshold and test whether interventions that reduce variability, such as adherence support or a switch of formulation, improve graft survival [9, 22, 23, 24].

7. CONCLUSION

Variability in tacrolimus exposure has identifiable origins. Absorption and first-pass metabolism by CYP3A4, CYP3A5 and P-glycoprotein set the baseline; CYP3A5 genotype is the largest known genetic factor; and food, interacting drugs, haematocrit, diarrhoea and adherence modify exposure over time. Associations between within-patient variability and chronic rejection, donor-specific antibodies and graft failure are consistent in retrospective cohorts, though whether variability is a cause or only a marker is not settled [6, 23, 24, 25, 26].

Practical strategies have improved how quickly patients reach the target range, especially genotype-guided starting doses and model-informed dosing, yet trials have not shown a clear benefit on rejection or graft survival [30, 31, 32]. Until that evidence exists, a sensible approach is to combine genotype or model-based initial dosing where available, consistent timing with food, careful review of interacting drugs, attention to adherence, and use of consensus-based monitoring with trough levels and, when needed, exposure estimates [3, 4, 15, 16, 17, 29].

8. ABBREVIATIONS

AUC-Area under the concentration-time curve, CV-Coefficient of variation, CYP-Cytochrome P450, KDIGO-Kidney Disease: Improving Global Outcomes, NFAT-Nuclear factor of activated T cells, TDM-Therapeutic drug monitoring, CPIC-Clinical Pharmacogenetics Implementation Consortium, ELITE-Efficacy Limiting Toxicity Elimination

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10. AUTHORS CONTRIBUTIONS

All the authors have contributed equally.

11. CONFLICT OF INTERESTS

Declared none

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