# Tacrolimus therapeutic efficacy in post-liver transplant patients with Cytochrome P450 3A5 (CYP3A5) genetic polymorphisms

## Eficácia terapêutica do Tacrolimo em pacientes pós-transplante hepático com polimorfismos genéticos do citocromo P450 3A5 (CYP3A5)

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A B S T R A C T

Genetic polymorphisms of CYP3A5 have been pointed out as factors that influenciates tacrolimus immunosuppressive efficacy in post liver transplant patients. This study aims to review the literature on the influence of cytochrome P450 3A5 (CYP3A5) genetic polymorphisms of tacrolimus in post-liver transplant patients. This study is a literature review. A combination of the descriptors "tacrolimus", "liver transplant", "cytochrome P-450 CYP3A inhibitors" and "genetic polymorphism" were used in the databases PubMed, Cochrane Library, Scopus and Scielo, being evaluated only studies between 2009 and 2019 in English, Portuguese or Spanish. A total of six studies, each from a different population were summarized. Initially, the pharmacological aspects of tacrolimus were discussed, including details on its pharmacodynamics, pharmacokinetics and toxicity After that, we analyzed the studies that correlates CYP3A5 genetic polymorphisms and tacrolimus efficacy, including the ethnical specifications and the general limittions of the studies. The CYP3A5 polymorphisms have pointed to alterations in the metabolism of tacrolimus according to the ethnic and populational genotype, specially the *1 and *3*3 alleles, reflecting in the need for dose adjustment and also in post liver transplant rejection.

Headings: Tacrolimus. Liver Transplantation. Cytochrome P-450 CYP3A Inhibitors. Polymorphism, Genetic.

## INTRODUCTION

Liver transplantation is the last treatment for several clinical conditions that affect the liver, including chronic cholestatic diseases, and hepatocellular, metabolic, vascular, and neoplastic causes[1]. Liver transplantation can be indicated both for reversing an acute or terminal condition, as well as for improving patient's quality of life[2].

Despite the increase in the number of transplants performed in Brazil in the last few years, the availability of organs remains low, with a rate of transplantation around 13.2 per million inhabitants[3].

Even so, several factors can affect prognosis after transplantation. For instance, pre-existing comorbidities such as diabetes mellitus, obesity, and advanced age are well-known risk factors for increased postoperative mortality[1].

The pharmacogenetics of the immunosuppressants, especially those associated with polymorphisms, although little considered in clinical practice, can significantly influence the post-transplant prognosis[4-6]. The polymorphisms of cytochrome P450 enzymes, for example, have been widely studied because they constitute a potential modulation factor of therapeutic efficacy, or failure, among several drugs and diseases[7,8].

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In recent studies, genetic polymorphisms of CYP3A5 have been identified as important factors affecting tacrolimus (F506) pharmacological efficacy in immunosuppression after liver[4], kidney[9] and heart[10,11] transplantation. Such polymorphisms seem to be related to epigenetic regulation, with varying prevalence in different populations and ethnicities[4,6,7]. However, it is not clear which genotype (donor and/or the recipient) contributes to the mechanism of resistance to the drug[12].

Tacrolimus is an immunosuppressive medication that started to be used in the '90s, as the primary substitute for cyclosporine to prevent rejection after solid organ transplantation[5]. Its use has been increasingly recommended, with an estimate of having been used in over 90% of liver and kidney transplants in 2012[5].

The present study aims to perform a literature review on the genetic polymorphisms of cytochrome P450 3A5 (CYP3A5) influence on tacrolimus therapeutic efficacy after liver transplantation.

## METHODS

A literature narrative review was carried out, following the PICO strategy (Population, Intervention, Comparison, and Outcome) to search for the best available medical evidence[13]. The bibliographic search covered all articles published in English, Portuguese, and Spanish from 2009 to 2019.

The following descriptors were adopted (and their Portuguese and Spanish correspondents): "tacrolimus", "liver transplant", "Cytochrome P-450 CYP3A inhibitors", "genetic polymorphism", "immunosuppression", "epigenetics" and "pharmacogenetics". The databases used included: Medline (through PubMed), Latin American and Caribbean Health Sciences Literature (LILACS), The Cochrane Library, the Scientific Electronic Library Online (Scielo) and Scopus.

In the case of duplication of articles, found in the different search strategies, suppression of the duplicates was performed. The inclusion of each manuscript occurred after an initial reading of its abstract. Those fitting the research scope were retrieved for full-text reading.

### Study population (P)

Patients, from any age, undergoing liver allografts for acute or chronic diseases, using tacrolimus as an isolated or combined immunosuppressive therapy.

### Prognostic Factor (I)

Presence of cytochrome P450 (CYP3A5) genetic polymorphism.

### Comparison (C)

Control subjects, without CYP3A5-related genetic mutation.

### Outcomes (O)

We included studies in which primary or secondary outcomes were as follow: overall mortality, organ rejection, need for retransplantation, sepsis, length of stay in the intensive care unit, length of hospital stay, other organic disorders, tacrolimus intolerance, dose ratio/serum and urinary concentration, reactivation of the underlying disease.

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# RESULTS

## Tacrolimus pharmacological properties

Tacrolimus is a macrolide immunosuppressive derived from *Streptomyces tsukubaensis* that acts as a calcineurin inhibitor[14]. Its mechanism of action, at the molecular level, occurs from its binding to the enzyme FKBP12[15]. The resulting complex is then associated with calcineurin, preventing the dephosphorylation of the nuclear factor of activated T-cells and, consequently, the interleukin-2 release, avoiding the signaling responsible for the T-lymphocytes activation[4,15].

Tacrolimus is a lipophilic drug, which has a prolonged metabolism and a highly variable and incomplete absorption rate, with bioavailability around 20 to 25%[4,6,16]. Tacrolimus metabolism is mainly mediated by CYP3A5 and CYP3A4 action[6,17,18]. Its transport is mediated by a P-glycoprotein by an efflux pump mechanism, with predominant excretion via the bile duct (95%)[18].

In addition, the binding of FKBP12 to glucocorticoid or progesterone receptors is the main mechanism related to tacrolimus safety and immunosuppressive efficacy[15,16]. This mechanism prevents the degradation or inactivation of the active molecule and allows its translocation to the nucleus. Through the DNA binding, transcription of inflammatory cytokines is inhibited, mimicking corticosteroid effects[16].

Regarding side effects, tacrolimus use is associated with nephrotoxicity and neurotoxicity[16], as detailed in Table 1. However, tacrolimus is less nephrotoxic than cyclosporin, this effect is associated with exposure for prolonged periods and at high doses[19,20]. However, the presence of confounding factors in studies evaluating the action of the drug, such as food intake close to the medication time (which alters its bioavailability), diarrhea, and different ethnicities, hinders a conclusion regarding the real risk of kidney injury[17].

Usually, the ideal tacrolimus serum concentration is targeted at 8-12ng/mL for the first three months, which progressively decreases, reaching 4-6ng/mL one year after transplantation[14,21]. Different protocols suggest maintaining a concentration of 5-10ng/mL during the first six months after the procedure[10]. However, we did not find any randomized controlled trial comparing the efficacy and safety of different protocols in post-transplant patients.

Some experts believe that serum concentrations around 2-4ng/mL are associated with fewer side effects while maintaining pharmacological efficacy[14]. However, there is no substantial evidence to establish the ideal dose in these patients.

## CYP3A5 polymorphisms and tacrolimus effectiveness

CYP3A5 is the primary enzyme responsible for tacrolimus metabolism, and it is found in the liver tissue and the small intestine[6,11,18]. Thus, its enzymatic expression at both sites influences tacrolimus pharmacokinetics[22].

**Table 1.** Main renal histology injury associated with the use of tacrolimus[17,19].

| Acute injuries | Chronic injuries |
| --- | --- |
| Acute arteriopathy | Glomerular fibrosis |
| Thrombotic microangiopathy | Interstitial fibrosis and tubular atrophy |
| Tubular vacuolization | Focal segmental glomerulosclerosis |
| | Arteriolar hyalinization |

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CYP3A5 genotype distribution is strongly related to ethnicity, as detailed in Table 2. Estimates show that about 82-95% of Europeans, 85% of Japanese, 67% of Chinese, and 14-33% of patients in Sub-Saharan Africa do not express the CYP3A5*3 gene[1]. Thus, Afro-descendants tend to be more likely intermediate or normal metabolizers when compared to Caucasians[4]. This means that the former would have a better pharmacological response and lower need for dosage adjustment than the latter. This would tend to lead to more individuals with ultra-fast (needing higher doses, possibly reaching one to two times higher than the standard dose[23]) or poor metabolism (with higher FK506 plasma levels and suffering more from the drug side effects)[4,24].

Usually, homozygous CYP3A5*1/*1 individuals tend to be normal metabolizers, unlike homozygous CYP3A5*3/*3, CYP3A5*6/*6, and CYP3A5*7/*7, which have a tendency to present a poor metabolism[6,24]. In a study with 131 Hungarians, after liver transplantation[25], the CYP3A5*3/*3 genotype was responsible for 80.92% of the sample. Among all CYP3A5 allelic variants, 39.6% were poor metabolizers compared to 14.1% ultra-fast and 46.3% normal metabolizers. In an Italian study[26], the CYP3A5*3/*3 genotype incidence was similar, being present in 84.9% of the sample. When evaluating the *3 genotype alone, the rate was as high as in 93% of the sample.

In a study carried out with 51 Chinese adults, three months after liver transplantation, the tacrolimus dose concentration observed in individuals with the CYP3A5*1 allele was lower than those with the CYP3A5*3/*3 allele[12]. Furthermore, the allelic variation of both donor and recipient also influenced tacrolimus levels. The summary of the studies is described in Table 3.

In a Japanese study[27] with 410 post-liver transplantation patients, assessing the occurrence of acute rejection between the 14th and 23rd days after surgery and its relation with the CYP3A5 enzyme assessed by intestinal biopsies, no statistical significance was found in those patients with ABO compatibility (11.5% CYP3A5*1 versus 7.4% CYP3A5*3/*3). On the other hand, when the evaluation was performed five weeks after the operation, those with CYP3A5*1 had a higher rate of acute rejection than those with the CYP3A5*3/*3 allele (14.5 versus 5.7%). Besides, the dose/concentration ratio was also higher in the latter group.

Uesugi et al.[27] also pointed out that there was an increase in the risk of rejection between 14 and 23 days after liver transplantation, especially in CYP3A5*1 (14.5% versus 5.7% CYP3A5*3/*3).

The time taken to measure tacrolimus levels after liver transplantation should also be considered. Three months after treatment, CYP3A5*1 genotype patients had higher tacrolimus doses than CYP3A5*3 patients. These results are shown in a Chinese[12], and an Italian study[26], with the latter also showing a CYP3A5*1 level increased at sixth month.

In Brazil, despite the lack of studies evaluating the relationship between CYP3A5 polymorphisms and tacrolimus use, Rodrigues-Soares et al. observed, in a systematic review[23], that the sum of alleles CYP3A5*3, CYP3A5*6 and CYP3A5*7 had a higher frequency in the black population. The exception was for the CYP3A5*3 allele, more frequent in the white population.

**Table 2.** Main CYP3A5 alleles in individuals after liver transplantation in different countries and ethnicities.

| Study | Sample size | Country | Ethnicity | Main allele |
| --- | --- | --- | --- | --- |
| Wang et al.[12], 2015 | 51 | China | Asian | *3/*3 |
| Ji et al.[18], 2018 | 58 | Korea | Asian | *1/*3 |
| Uesugi et al.[27], 2014 | 407 | Japan | Asian | *1 |
| Monostory et al.[25], 2015 | 131 | Hungary | Caucasian | *3/*3 |
| Gerárd et al.[28], 2014 | 66 | France | Caucasian | *3/*3 |
| Buendía et al.[22], 2015 | 24 | Argentina | - | *1 |
| Provenzani et al.[26], 2011 | 51 | Italy | Caucasian | *3/*3 |

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**Table 3.** Summary of studies evaluating the relationship between CYP3A5 polymorphisms and tacrolimus use after liver transplantation.

| Study | N | Country | Measure | Measurement time after transplantation | Main results |
|---|---|---|---|---|---|
| Wang et al.[12], 2015 | 51 | China | D/C ratio | Three months | The D/C ratio of tacrolimus was lower in CYP3A5*1 allele patients than in those with the CYP3A5*3/*3 allele. |
| Uesugi et al.[27], 2014 | 407 | Japan | D/C ratio | Five weeks | Tacrolimus serum level was affected by the CYP3A5*3 polymorphism in the liver after the 14th postoperative day. |
| Monostory et al.[25], 2015 | 131 | Hungary | D/C ratio, ng/mL level | - | The CYP3A5*1 allele was associated with a higher rejection rate compared to CYP3A5*3/*3 (14.5% versus 5.7%). |
| Gerárd et al.[28], 2014. | 66 | France | ng/mL level | - | Ultra-fast metabolizing receptors carrying at least one functional copy of CYP3A5*1 required an increase of approximately 100% in the initial tacrolimus dose to achieve the expected blood concentrations. |
| Buendía et al.[22], 2015 | 24 | Argentina | ng/mL level | Up to six months | Tacrolimus concentration was influenced by the following factors, in the following sequence: tacrolimus unbound plasma fraction, intrinsic clearance, donor CYP3A5 genotype, weight, and hematocrit. |
| Provenzani et al.[26], 2011 | 51 | Italy | ng/mL level | One, three and six months | Tacrolimus dose was higher in individuals expressing the CYP3A5 gene than in those who did not. |
| | | | | | Tacrolimus blood levels, after three and six months, required an increase in dose to reach the necessary concentration in those who had at least one *1 allele compared to homozygous *3*3. |

*Abreviations: D/C, Dose/Concentration; N, Sample.*

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**Limitations of the studies**

Most studies indicate the important role for CYP3A5 polymorphisms regarding mechanisms of resistance to tacrolimus therapy, in patients after liver transplantation. However, we can point out some limitations of the evaluated studies, such as the low sample sizes, the lack of dose specification as well as treatment times, and the use of convenience samples. Also, there is a mixed-use of methods to assess the postoperative tacrolimus dose concentration and a lack of description of coexisting comorbidities that can influence the drug absorption and metabolism.

## CONCLUSION

CYP3A5 genetic polymorphisms appear to be associated with changes in tacrolimus bioavailability and metabolism rate in patients after liver transplantation. There is no substantial evidence available on the influence of these polymorphisms on safety, therapeutic efficacy, and prognosis in these patients. Thus, the genetic study of patients using tacrolimus as an immunosuppressive therapy should not be performed routinely or used as an isolated parameter for adjusting therapy.

### R E S U M O

Os polimorfismos genéticos do CYP3A5 têm sido apontados enquanto fatores influenciadores na eficácia farmacológica com tacrolimo em pacientes em terapia imunossupressora no pós-transplante hepático. O presente estudo objetiva realizar uma revisão da literatura acerca da influência dos polimorfismos genéticos do citocromo P450 3A5 (CYP3A5) na eficácia terapêutica com tacrolimo em indivíduos pós-transplante hepático. Revisão da literatura. Foi utilizada a combinação dos descritores "tacrolimo", "transplante de fígado", "inibidores do citocromo P-450 CYP3A" e "polimorfismo genético", nas bases de dados: PubMed, The Cochrane Library, Scopus e Scielo, sendo avaliados apenas estudos publicados entre 2009 e 2019 em inglês, português ou espanhol. Ao todo foi feita a sumarização de seis estudos, cada um avaliando uma diferente população. Inicialmente, foram abordados os aspectos farmacológicos do tacrolimo, incluindo detalhes sobre sua farmacodinâmica, farmacocinética e toxicidade. Na seção seguinte, foi realizada a avaliação de estudos que tratam da relação entre os polimorfismos genéticos do CYP3A5 e a eficácia farmacológica com o tacrolimo, incluindo as especificações étnicas e as limitações gerais dos estudos. Os polimorfismos genéticos do CYP3A5 têm apontado para alterações no metabolismo do tacrolimo de acordo com um recorte étnico e populacional, com destaque para os alelos *1 e *3*3, refletindo na necessidade de ajuste de dose ou até mesmo nas taxas de rejeição do órgão.

**Descritores:** Tacrolimo. Transplante de Fígado. Inibidores do Citocromo P-450 CYP3A. Polimorfismo Genético.

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