

# BMJ Open Effect of FTY720 (fingolimod) on graft survival in renal transplant recipients: a systematic review protocol

Reza Gholamnezhadjafari,<sup>1</sup> Reza Falak,<sup>2,3</sup> Nader Tajik,<sup>2,3</sup> Reza Aflatoonian,<sup>4</sup> Abbas Ali Keshkar,<sup>5</sup> Abbas Rezaei<sup>1</sup>

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## ABSTRACT

**Introduction:** Studies have shown that FTY720 has inconsistent effects in kidney transplant recipients. Several review articles on FTY720 have been published, but most have focused on the mechanism of action of FTY720. Therefore, this review aims to evaluate and determine the beneficial and harmful effects of FTY720 therapy in kidney transplant recipients.

**Methods and analysis:** We electronically searched the following databases: PubMed, Scopus, the Web of Sciences, EMBASE, Cochrane databases and the Cochrane Central Registry of Controlled Trials. Any clinical, randomised controlled trials relating to FTY720 for treating kidney transplant recipients were included without publication status or language restriction. Study selection, data extraction and assessment of study quality were performed independently by two researchers. Data were synthesised by either the fixed effects or the random effects model according to a heterogeneity test. If the extracted data were suitable for meta-analysis, STATA software was used to combine the relative risks for dichotomous outcomes, and the mean differences for continuous outcomes with 95% CIs were measured. Death, loss of function and incidence of acute kidney rejection were assessed as the primary outcomes. Renal graft function, malignancy, delayed graft function and infection were evaluated as secondary outcomes.

**Ethics/dissemination:** This review does not require formal ethics approval because the data are not individualised. The resulting review article will be submitted for publication in a peer-reviewed journal.

**Trial registration number:** CRD42015024648.

## INTRODUCTION

Kidney transplantation is a cost-effective treatment for patients with end-stage renal disease. However, the immune response of the host to the grafted tissue is a challenge frequently encountered in organ transplantation.<sup>1 2</sup>

Initial studies have shown that the recipient's T cells may recognise allogenic antigens via direct or indirect mechanisms.<sup>1</sup> In the

## Strengths and limitations of this study

- Data can be consistent, precise and reliable.
- Study findings may explain inconsistent reports concerning the effect of FTY720 in kidney transplant recipients.
- Sample size, methods for measuring outcomes and study environments may affect our study.
- The different criteria for efficacy evaluation and combination therapies may cause significant heterogeneity in this review.

case of direct recognition, intact major histocompatibility complex molecules, which usually present with the donor's self-antigens, are highly expressed on antigen-presenting cells (APCs), and may be directly recognised by the recipient's T cells without further processing by host APCs. Unlike the direct pathway, host APCs can capture and process alloantigens and present the allopeptides to self T cells in the indirect pathway.<sup>2</sup> Alloreactive responses may result in acute or chronic rejection.<sup>3</sup> Therefore, many prevention strategies have been used to boost graft survival rates. Common maintenance methods focus on the modulation or suppression of adaptive immune responses.<sup>4</sup> The most frequent procedure used to prevent graft rejection is the application of immunosuppressants such as calcineurin inhibitors (cyclosporine and tacrolimus), mTOR inhibitors (rapamycin), mycophenolic acid and corticosteroids.<sup>5</sup> Although cyclosporine and tacrolimus have been shown to effectively reduce episodes of acute graft rejection, the risk of chronic transplant rejection remains high.<sup>6</sup> Many studies have shown that FTY720 is a promising immunosuppressive drug that can be used to suppress initial and subsequent responses to renal grafts. However, the effects of the drug are not fully understood.<sup>7</sup>

FTY720 is a derivative of ISP-1 (myriocin), which is a fungal metabolite that is



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For numbered affiliations see end of article.

## Correspondence to

Professor Abbas Rezaei; rezaei@mui.ac.ir

structurally analogous to sphingosine. It is a lipid chemo-attractant found in body fluids such as blood and lymph. FTY720 is an immunomodulating drug that extends allograft survival in numerous models by inhibiting lymphocyte emigration from lymphoid organs.<sup>8</sup> The results from phase I and II clinical trials of FTY720 therapy have been promising, and phase III clinical trials are currently ongoing.<sup>9</sup> The potential mechanism of action of FTY720 is the downregulation of sphingosine-1-phosphate receptor (S1PR). Activated lymphocytes express high levels of S1PR, which binds to sphingosine-1-phosphate (S1P) and causes egression of lymphocytes from the lymph nodes and spleen. FTY720 binds to S1PR1 and reduces its cell surface expression. Thus, it blocks T cell egression from lymphoid organs and acts as an immunosuppressive drug.<sup>10</sup>

A large number of preclinical studies using animal models have demonstrated the efficacy of FTY720 in solid organ allograft survival prolongation alone or in combination with cyclosporine.<sup>11 12</sup>

In the last decade, many de novo renal transplant recipients have received FTY720 at doses of 2.5 or 5.0 mg/day. The doses were mostly combined with corticosteroids and cyclosporine or mycophenolate mofetil (MMF). In most studies, the recipients were followed for more than 12 months after transplantation.<sup>13–15</sup>

Phase I clinical studies in humans have revealed that FTY720 at doses of 1.0 mg/day or higher significantly reduced peripheral blood lymphocyte counts.<sup>16</sup> Phase II clinical trials have demonstrated the efficacy of FTY720 in the prevention of acute rejection in renal transplant recipients.<sup>15</sup> In Europe and Australasia, phase III randomised controlled trials (over 1 year) showed 2.5 mg FTY720 with MMF significantly prevented renal acute rejection. However, more instances of antibody-mediated rejection and more episodes of biopsy-proven chronic rejection were reported with 2.5 mg FTY720 than with 2 g MMF.<sup>17</sup>

In a multicentre, randomised, open-label renal transplantation study for the evaluation of 2.5 mg FTY720 versus MMF in a combination regimen with tacrolimus and corticosteroids, the incidence of treated biopsy-proven acute rejection was 22.9% with FTY720 and 18.5% with MMF. These results showed that FTY720 combined with tacrolimus and steroids did not have a significant therapeutic advantage over MMF for the prevention of acute rejection in de novo renal transplant recipients over a 1-year period.<sup>13</sup> Comparison of data from other phase II and phase III trials in another multicentre study showed that FTY720-based regimens have no beneficial effects on the preservation of renal function in recipients at high risk for delayed graft function (DGF) and no advantage in preventing acute rejection.<sup>18</sup>

However, clinical trials have shown that FTY720 has promising efficacy in preventing acute rejection in de novo renal transplant recipients.<sup>15 17</sup> In contrast, some studies have reported that treatment with FTY720 alone or in combination with conventional

immunosuppressants and steroids did not result in a significant therapeutic advantage in preventing acute rejection in renal transplant recipients. In some cases, the use of FTY720 led to an increase in acute antibody-mediated rejection.<sup>14 18</sup> These studies suggest that our knowledge of the effect of FTY720 in preventing allograft rejection and supporting renal graft survival is incomplete and so a comprehensive and precise review is required.

## METHODS AND ANALYSIS

This protocol was written in accordance with the PRISMA-P guidelines for systematic reviews.<sup>19</sup>

### Inclusion criteria for study selection

#### Types of studies

All RCTs in which patients in either intervention or control groups received FTY720 alone or in combination with other immunomodulators were included without restriction regarding time, language or publication type.

#### Types of participants

Adults and children who received a renal transplant from a living or brain-dead donor and were prescribed FTY720 alone or in combination with other immunomodulators were included. Recipients of simultaneous transplants were excluded from this review.

#### Types of interventions

Studies reporting any type of FTY720 administration (oral, injection, etc), alone or in combination with any other immunosuppressants and at any doses in renal transplantation, were included in the review. Comparative interventions, including placebo controls and other conventional treatments, were also included.

#### Types of outcome measures

##### Primary outcomes

- ▶ Death
- ▶ Loss of function
- ▶ Incidence of acute kidney rejection (diagnosed by clinical assessment, biopsy evaluation and steroid resistance).

##### Secondary outcomes

- ▶ Assessment of renal graft function by glomerular filtration rate, cystatin C, neutrophil gelatinase-associated lipocalin (NGAL) and other biomarkers of kidney function
- ▶ Incidence of treatment-related adverse reactions (biochemical, neurological, haematological, gastrointestinal, etc)
- ▶ Incidence of onset of new diseases such as diabetes
- ▶ Incidence of malignancy
- ▶ Incidence of DGF
- ▶ Incidence of infection.

## Search methods for identifying studies

### Electronic searches

We identified relevant RCTs, in any language, by conducting a systematic search of the Web of Sciences, MEDLINE, Scopus, and Cochrane databases, the Cochrane Central Registry of Controlled Trials and the Cochrane Renal Group's specialised register (table 1).

### Searching other resources

Hand searching to identify relevant clinical studies mentioned in reports of transplant meetings, conference proceedings and abstracts was performed. We also scanned the bibliographies of all retrieved trials and other relevant publications, including reviews and meta-analysis paper references, for additional relevant articles.

## Data collection and analysis

### Selection of studies

Eligible studies were presented according to the PRISMA 2009 Flow Diagram. Our group worked in pairs. Two authors (RG and RF) independently screened

and reviewed articles to identify titles and available abstracts. They also acquired the full text of any article judged to be potentially eligible. The same review team independently applied eligibility criteria to the full text of potentially eligible articles. The reviewers resolved disagreement by consensus; any remaining differences were resolved through discussion with NT and RA.

### Data extraction and management

Using standard extraction forms, two authors (RG and RF) extracted data concerning participants, sample sizes, duration of studies and of follow-up, study environment, interventions, outcomes, results and methods to measure outcomes. Some heterogeneity may have occurred because of potential variations in sample sizes, duration of study, interventions and methods used to measure outcomes; these variations were discussed among the authors. The authors of the trial were contacted if data were missing or more information was required. Studies reported in non-English language journals were translated before assessment.

## Assessment of risk of bias in the included studies

### Measuring the effect of treatment

Continuous data were expressed as mean differences with 95% CIs. For dichotomous data, the risk ratio with corresponding 95% CIs was used. For data measured using similar scales with similar units, weighted mean differences were used. Otherwise, the standardised mean difference was used to analyse the extracted data.

### Dealing with missing data

We attempted to contact the relevant authors if data were missing. If we were unable to obtain missing data or information, the study was omitted from data synthesis.

### Assessment of heterogeneity

We estimated the level of heterogeneity across the studies using STATA software (V.12), the  $\chi^2$  test for statistical heterogeneity, and the  $I^2$  statistic in forest plots. Values of  $I^2 < 25\%$  were classified as low heterogeneity, 25–50% as moderate heterogeneity and  $> 50\%$  as large heterogeneity (grades A, B and C, respectively).

### Assessment of reporting biases

We used funnel plots to detect potential reporting bias and small-study effects. Asymmetries for more than 10 studies were included in the meta-analysis using the Egger method.<sup>20</sup>

## Data synthesis

If the extracted data were suitable for meta-analysis, STATA software was used to combine the relative risks for dichotomous outcomes, and the mean differences for continuous outcomes with 95% CIs were measured. If we found no evidence of heterogeneity, we used the fixed effect model; otherwise, we applied a random effects model. If significant heterogeneity between

**Table 1** Search strategy used for the PubMed database\*

No.	Search items
1	Fingolimod
2	FTY720
3	FTY-720
4	FTY 720
5	Gilenya
6	fingolimod hydrochloride
7	2-amino-2-(2-(4-octylphenyl)ethyl)-1,3-propanediol hydrochloride
8	sphingosine-1-phosphate receptor agonist
9	sphingosine-1-phosphate receptor modulator
10	sphingosine-1-phosphate receptor
11	1 OR 2–10
12	systematic
13	11 AND 12
14	Kidney transplantation
15	Renal transplantation
16	Renal transplantations
17	Transplantation, renal
18	Grafting, kidney
19	Kidney grafting
20	Transplantation, kidney
21	Kidney/transplantation
22	14 OR 15–21
23	11 AND 22
24	intervention
25	effectiveness
26	comparative
27	clinical trial
28	controlled
29	placebo
30	24 OR 25–29
31	23 AND 30

\*This search strategy will be suitable for other electronic databases.

studies was found, we used subgroup analysis to identify possible clinical or methodological causes and offered appropriate explanations for the differences.

### Subgroup analysis

We performed subgroup analysis for heterogeneity caused by main factors such as sample sizes, methods used to measure outcomes, study environments and types of intervention. For example, types of interventions were classified based on the use of FTY720 alone or in combination with other immunomodulators.

### Sensitivity analysis

We conducted a sensitivity analysis to validate the study design, conclusions and sample size and to investigate suspected funnel plot asymmetry.

### Ethics and dissemination

Ethics approval was not required as we did not use patient data in this systematic review. The resulting review article was submitted for publication in a peer-reviewed journal.

## DISCUSSION

The results of some clinical trials have demonstrated that FTY720 has promising efficacy for preventing acute rejection in de novo renal transplant patients.<sup>15 21 22</sup> The first phase IIA study of de novo renal transplantation showed that the incidence of biopsy-confirmed acute rejection at 3 months after kidney transplantation was dose dependent. The rate of acute rejection at a dose of 0.25 mg was twofold higher than that at 2.5 mg. In addition, the incidence of the composite endpoint (biopsy-confirmed acute rejection, graft loss or death) was lower with 2.5 mg FTY720 compared to lower doses.<sup>15</sup> In another study, an FTY720 blood concentration of 4 ng/mL in the early weeks after transplantation was associated with a decrease in the maximal lymphocyte count. This decrease was correlated with evidence of rejection prophylaxis.<sup>21</sup> The results of a 6-month, double-blind study indicated that the composite endpoint occurred within 6 months in 24% of FTY720 patients and 39% of MMF patients. In addition, FTY720 was associated with lower creatinine clearance but a higher incidence of bradycardia than MMF.<sup>22</sup> On the other hand, some studies have demonstrated that FTY720 alone or in combination with conventional immunosuppressants and steroids did not decrease acute rejection in renal transplant recipients.<sup>17 18</sup> Indeed, in some cases, FTY720 caused an increase in acute antibody-mediated rejection. An exploratory, 1-year, multicentre study in de novo renal transplant patients at risk for DGF suggested that there were no apparent benefits with FTY720-based regimens (2.5 mg) for the prevention of acute rejection and preservation of renal function in renal transplant recipients at high risk for DGF.<sup>18</sup>

A 1-year, randomised controlled trial in Europe and Australasia reported that 2.5 mg FTY720 was associated with more antibody-mediated rejection and more episodes of biopsy-proven chronic rejection than MMF. These unfavourable outcomes indicate a limited effect of FTY720 on B cells.<sup>17</sup> Given these ambiguous and inconclusive results, a comprehensive, objective and systematic review of the effects of FTY720 on graft survival in renal transplant recipients was required. Therefore, the main purpose of this review was to estimate the relative and absolute effects of FTY720 therapy in kidney transplant recipients and determine its beneficial and harmful effects on renal tissue graft outcome and survival. A minor objectives of this review was to determine which conventional immunosuppressants have the greatest synergy with FTY720 for the prevention of acute rejection and renal tissue graft survival.

### Author affiliations

<sup>1</sup>Department of Immunology, Isfahan University of Medical Sciences, Isfahan, Iran

<sup>2</sup>Immunology Research Center, Iran University of Medical Sciences, Tehran, Iran

<sup>3</sup>Department of Immunology, School of Medicine, Iran University of Medical Sciences, Tehran, Iran

<sup>4</sup>Department of Endocrinology and Female Infertility, Royan institute for Reproductive Biomedicine, ACECR, Tehran, Iran

<sup>5</sup>Department of Health Sciences Education Development, School of Public Health, Tehran University of Medical Sciences, Tehran, Iran

**Contributors** RG contributed to the conception of the study and drafted the manuscript protocol. RF revised the protocol draft. The search strategy was designed by all the authors and performed by RG and RF, who screened the potential studies independently and extracted data from the included studies. NT and RA resolved any discrepancies. AAK and AR assessed the risk of bias and completed the data synthesis.

**Competing interests** None declared.

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**Data sharing statement** All data generated during the project will be made freely available via the bmjopen website. DOIs for these data will be provided (as part of the DataCite programme) and cited in any published articles using these data and any other data generated in the project. Any data relevant to a published article will be made available alongside the article when published.

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## REFERENCES

- Land WG. Emerging role of innate immunity in organ transplantation: part I: evolution of innate immunity and oxidative allograft injury. *Transplant Rev (Orlando)* 2012;26:60–72.
- Chai JG, Ratnasothy K, Bucy RP, *et al.* Allospecific CD4+ T cells retain effector function and are actively regulated by Treg cells in the context of transplantation tolerance. *Eur J Immunol* 2015;45:2017–27.
- Kreisel D, Krupnick AS, Gelman AE, *et al.* Non-hematopoietic allograft cells directly activate CD8+ T cells and trigger acute rejection: an alternative mechanism of allorecognition. *Nat Med* 2002;8:233–9.
- Webster AC, Pankhurst T, Rinaldi F, *et al.* Monoclonal and polyclonal antibody therapy for treating acute rejection in kidney transplant recipients: a systematic review of randomized trial data. *Transplantation* 2006;81:953–65.



5. Ciancio G, Burke GW, Gaynor JJ, *et al.* A randomized long-term trial of tacrolimus and sirolimus versus tacrolimus and mycophenolate mofetil versus cyclosporine (NEORAL) and sirolimus in renal transplantation. I. Drug interactions and rejection at one year. *Transplantation* 2004;77:244–51.
6. Kunzendorf U, Ziegler E, Kabelitz D. FTY720—the first compound of a new promising class of immunosuppressive drugs. *Nephrol Dial Transplant* 2004;19:1677–81.
7. Chueh S-CJ, Kahan BD. Update on FTY720: review of mechanisms and clinical results. *Curr Opin Organ Transplant* 2003;8:288–98.
8. Budde K, Schütz M, Glander P, *et al.* FTY720 (fingolimod) in renal transplantation. *Clin Transplant* 2006;20(Suppl 17):17–24.
9. Chun J, Hartung HP. Mechanism of action of oral fingolimod (FTY720) in multiple sclerosis. *Clin Neuropharmacol* 2010;33:91.
10. Lee CW, Choi JW, Chun J. Neurological S1P signaling as an emerging mechanism of action of oral FTY720 (fingolimod) in multiple sclerosis. *Arch Pharm Res* 2010;33:1567–74.
11. Brinkmann V, Lynch KR. FTY720: targeting G-protein-coupled receptors for sphingosine 1-phosphate in transplantation and autoimmunity. *Curr Opin Immunol* 2002;14:569–75.
12. Brinkmann V, Pinschewer DD, Feng L, *et al.* FTY720: altered lymphocyte traffic results in allograft protection. *Transplantation* 2001;72:764–9.
13. Hoitsma AJ, Woodle ES, Abramowicz D, *et al.* FTY720 combined with tacrolimus in de novo renal transplantation: 1-year, multicenter, open-label randomized study. *Nephrol Dial Transplant* 2011;26:3802–5.
14. Mulgaonkar S, Tedesco H, Oppenheimer F, *et al.* FTY720/cyclosporine regimens in de novo renal transplantation: a 1 year dose finding study. *Am J Transplant* 2006;6:1848–57.
15. Tedesco-Silva H, Mourad G, Kahan BD, *et al.* FTY720, a novel immunomodulator: efficacy and safety results from the first phase 2A study in de novo renal transplantation. *Transplantation* 2005;79:1553–60.
16. Kahan BD, Karlix JL, Ferguson RM, *et al.* Pharmacodynamics, pharmacokinetics, and safety of multiple doses of FTY720 in stable renal transplant patients: a multicenter, randomized, placebo-controlled, phase I study. *Transplantation* 2003;76:1079–84.
17. Salvadori M, Budde K, Charpentier B, *et al.* FTY720 versus MMF with cyclosporine in de novo renal transplantation: a 1-year, randomized controlled trial in Europe and Australasia. *Am J Transplant* 2006;6:2912–21.
18. Tedesco-Silva H, Lorber MI, Foster CE, *et al.* FTY720 and everolimus in de novo renal transplant patients at risk for delayed graft function: results of an exploratory one-yr multicenter study. *Clin Transplant* 2009;23:589–99.
19. Moher D, Shamseer L, Clarke M, *et al.* Preferred reporting items for systematic review and meta-analysis protocols (PRISMA-P) 2015 statement. *Syst Rev* 2015;4:1.
20. Higgins JP, Altman DG, Gøtzsche PC, *et al.* The Cochrane Collaboration's tool for assessing risk of bias in randomised trials. *BMJ* 2011;343:d5928.
21. Ferguson R, Mulgaonkar S, Tedesco H, *et al.* High efficacy of FTY720 with reduced cyclosporine dose in preventing rejection in renal transplantation: 12-month preliminary results. *Am J Transplant* 2003;3(Suppl 5):311.
22. Tedesco-Silva H, Szakaly P, Shoker A, *et al.* FTY720 versus mycophenolate mofetil in de novo renal transplantation: six-month results of a double-blind study. *Transplantation* 2007;84:885–92.