October 15, 2013

Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 5169-review.doc).

**Title:** Pharmacogenetic considerations for optimizing tacrolimus dosing in liver and kidney transplant patients

**Author:** Alessio Provenzani, Andrew Santeusanio, Erin Mathis, Monica Notarbartolo, Manuela Labbozzetta, Paola Poma, Ambra Provenzani, Carlo Polidori, Giovanni Vizzini, Piera Polidori and Natale D'Alessandro

**Name of Journal:** *World Journal of Gastroenterology*

**ESPS Manuscript NO:** 5169

The manuscript was improved according to the suggestions of reviewers. I also reduced the number of words of the title, running title and abstract. References and typesetting were corrected.

1 Reviewer n. 00503175

- In the text the word "*trough*" is always followed by the word "*level*" or "*levels*". The "t*rough level*" is the lowest concentration reached by a drug before the next dose is administered.

- In the section "Influence of genetics on tacrolimus pharmacodynamics" the repetition "*of patients*" was eliminated.

- In the table 1 the author name was corrected (*Roy* instead of *Roi*).

- In table 1 "*Hesselink et al (ref. 87)*" was used instead of "*Hesselink et al (87)*" and "*Gervasini et al (ref. 33)*" instead of "*Gervasini et al (33)*".

2 Reviewer n. 00503228

- The abstract was reduced to about 230 words and, as suggested, I inserted a comment regarding my conclusions (…*At present, research has been able to reliably show that the CYP3A5 genotype, but not the CYP3A4 or ABCB1 ones, can modify the pharmacokinetics of tacrolimus. However, it has not been possible to incontrovertibly show that the corresponding changes in the pharmacokinetic profile are linked with different patient outcomes regarding tacrolimus efficacy and toxicity. For these reasons, pharmacogenetics and individualized medicine remain a fascinating area for further study and may ultimately become the face of future medical practice and drug dosing*).

- As specified in the text it is important to consider the genetic background of the donor for liver transplant patients and of the recipient for kidney transplant patients. In the case of kidney transplant patients, the genetic background of the recipient can influence the tacrolimus level (the CYP3A is present in the liver and intestine). The presence or not of the CYP3A and ABCB1

SNPs in the graft kidney can be important in relation to the toxicity as reported in the text (…*CYP3A5 expression in the kidney may play a protective role against the development of nephrotoxicity by limiting the exposure of the organ to toxic metabolites*…in the text it is also reported… *P-glycoprotein (P-gp, also known as ABCB1), an ATP-dependent membranous transporter which helps to protect the body against toxic xenobiotics by extruding these compounds out of cells and into the intestinal lumen and bile, can limit the oral bioavailability and influence the disposition of the calcineurin inhibitors. In particular, the presence of P-gp in the intestine can limit tacrolimus absorption. Also, its presence in liver and kidney promotes tacrolimus efflux into bile and urine, respectively*…).

- In the text I discussed some studies focused on CYP3A5 expressers patients (ref. 108, 109 and 110).

Reviewer

- Regarding the relation between the angiotensin C3889 (rs4762) gene polymorphism and tacrolimus, I clarified as follows in the text: "*It is well known that tacrolimus has a negative effect on pancreatic beta islet cells and can cause glucose intolerance and diabetes mellitus. However, new studies have suggested that post-transplant diabetes mellitus can also be related to other factors and, consequently, not only to tacrolimus administration. Angiotensinogen (AGT) is the initial component of the renin-angiotensin system (RAS) and a precursor of both angiotensin I and II. In a study on 302 subjects, the authors found that the AGT gene polymorphism (rs4762) is associated with post-transplant diabetes mellitus, due to insulin resistance, in Korean renal transplant patients[124]. Molecular and genetic studies demonstrate a relationship between variants of the AGT gene, AGT gene expression and plasma AGT levels. However, the association between this gene and glucose metabolism remain controversial*".

- Regarding ABCB1, I clarified how ABCB1 SNPs can influence tacrolimus blood levels and which are the main tissues involved (…*P-glycoprotein (P-gp, also known as ABCB1), an ATP-dependent membranous transporter which helps to protect the body against toxic xenobiotics by extruding these compounds out of cells and into the intestinal lumen and bile, can limit the oral bioavailability and influence the disposition of the calcineurin inhibitors. In particular, the presence of P-gp in the intestine can limit tacrolimus absorption. Also, its presence in liver and kidney promotes tacrolimus efflux into bile and urine, respectively…).*

Thank you again for publishing our manuscript in the *World Journal of Gastroenterology*.

Sincerely yours,

Alessio Provenzani