

MEETING ABSTRACT

Open Access

# Treprostinil stimulates the engraftment of haematopoietic stem cells

Filza Hussain, Christian Bergmayr, Armam Alimohammadi, Michael Freissmuth, Eva-Maria Zebedin\*

From 17th Scientific Symposium of the Austrian Pharmacological Society (APHAR). Joint meeting with the Hungarian Society of Experimental and Clinical Pharmacology (MFT) Innsbruck, Austria. 29-30 September 2011

## Background

Successful transplantation of haematopoietic stem cells (HSC) is often limited by low transplantation efficiency. This may be enhanced by pharmacological means. In fact, HSCs require a  $G\alpha_s$ -transduced signal to re-populate the bone marrow [1]. Pretreatment with prostaglandin  $E_2$  ( $PGE_2$ ) enhances engraftment via activation of  $G\alpha_s$ -coupled  $EP_2$  and  $EP_4$  receptors [2]. Treprostinil is a stable analogue of prostacyclin/ $PGI_2$ . It predominantly acts via  $EP_2$  and  $EP_4$  receptors. Treprostinil is approved for treatment of pulmonary hypertension. Here we test the hypothesis that treprostinil may also be useful to promote engraftment of HSC.

## Methods

Generation of murine bone marrow-derived HSCs: Undifferentiated HSC (lineage-negative,  $Lin^-$  cells) were separated from bone marrow cells by MACS (magnetic-assisted cell sorting) and characterized by fluorescence-activated cell sorting (FACS) using cell surface markers. [ $^3H$ ]cAMP-accumulation assays:  $Lin^-$  cells were incubated in supplemented stem cell medium (StemSpanSFEM#09650). After 4 h at 37°C cells were stimulated with forskolin, treprostinil, other prostanoids and cholera toxin for 1 h. Bone marrow transplantation: Recipient mice were lethally irradiated.  $Lin^-$  cells were pretreated in absence/presence of 10  $\mu M$  treprostinil, treprostinil plus 30  $\mu M$  forskolin or 10  $\mu g/mL$  cholera toxin for 1 h at 37°C.  $3 \times 10^5$  cells/mouse were injected via the tail vein. Transplantation efficiency was determined by the analysis of white blood cell counts. For competition assay, equal

numbers of treated/untreated  $Lin^-$  cells, which can be distinguished according to surface expression of Ly5.1 and Ly5.2, were injected in one and the same recipient mouse.

## Results

Successful MACS-purification of  $Lin^-$  cells was documented by FACS. Next, the cAMP-response of  $Lin^-$  cells to treprostinil was tested: Treprostinil elicited a concentration-dependent accumulation of cAMP in the range of 0.1–10  $\mu M$  with an estimated  $EC_{50}$  in the range of 0.3  $\mu M$ . A beneficial effect was also observed *in vivo*: mice injected with treprostinil-pretreated  $Lin^-$  cells had significantly higher levels of circulating white blood cells when compared to those receiving vehicle-treated  $Lin^-$  cells ( $p < 0.05$ , unpaired  $t$  test). In addition, when pretreated and untreated  $Lin^-$  cells were mixed to compete for bone marrow reconstitution, the white blood cells derived from the pretreated  $Lin^-$  cell population were 1.5–3-fold more abundant than those originating from the non-treated HSC.

## Conclusions

The treprostinil-induced cAMP elevation translates into enhanced engraftment of haematopoietic stem cells. Because treprostinil is reasonably well tolerated, it may be of interest to explore its action in bone marrow transplantation in people.

Published: 5 September 2011

## References

1. Adams GB, Alley IR, Chung UI, Chabner KT, Jeanson NT, Lo Celso C, Marsters ES, Chen M, Weinstein LS, Lin CP, Kronenberg HM, Scadden DT: Haematopoietic stem cells depend on  $G\alpha_s$ -mediated signalling to engraft bone marrow. *Nature* 2009, **459**:103-107.

\* Correspondence: eva-maria.zebedin@meduniwien.ac.at  
Institute of Pharmacology, Center of Physiology and Pharmacology, Medical University of Vienna, 1090 Vienna, Austria

- Hoggatt J, Singh P, Sampath J, Pelus LM: Prostaglandin E<sub>2</sub> enhances hematopoietic stem cell homing, survival, and proliferation. *Blood* 2009, **113**:5444-5455.

doi:10.1186/1471-2210-11-S2-A6

**Cite this article as:** Hussain *et al.*: Treprostinil stimulates the engraftment of haematopoietic stem cells. *BMC Pharmacology* 2011 **11** (Suppl 2):A6.

**Submit your next manuscript to BioMed Central  
and take full advantage of:**

- Convenient online submission
- Thorough peer review
- No space constraints or color figure charges
- Immediate publication on acceptance
- Inclusion in PubMed, CAS, Scopus and Google Scholar
- Research which is freely available for redistribution

Submit your manuscript at  
[www.biomedcentral.com/submit](http://www.biomedcentral.com/submit)

