

# Inactivation of Transfusion-Transmitted Vector Borne Pathogens

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## Background

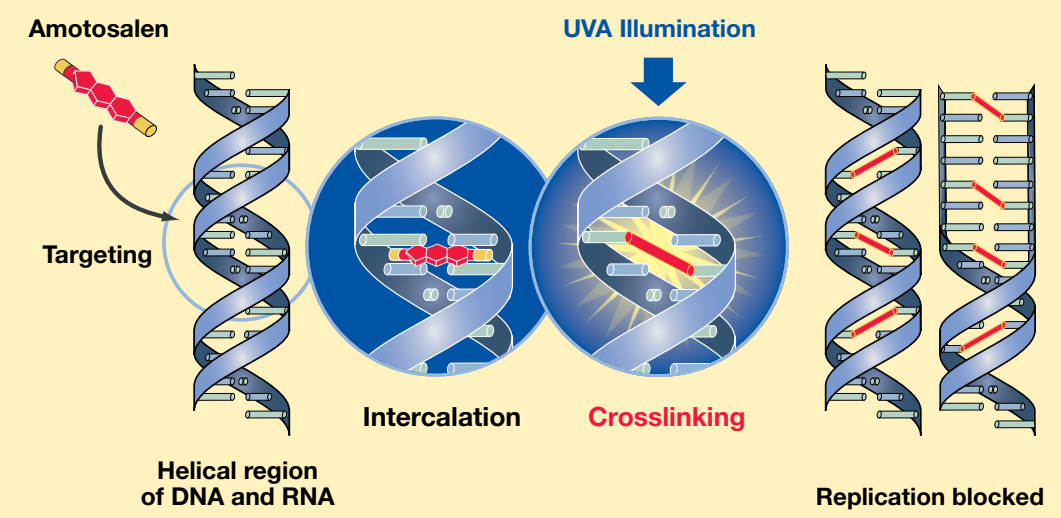
A number of blood-borne pathogens are transmitted to humans primarily by insect vectors. While the blood banking community has been aware of the risks of some vector-borne pathogens, such as malaria, for years, the awareness of risks from other vector-borne pathogens is much more recent. It has been less than ten years since West Nile virus and chikungunya virus were recognized as serious regional issues for the blood supply, and recognition of the risk of *T. cruzi* outside of South America and of *Babesia* in North America is even more recent. Vector-borne pathogens are particularly problematic for blood safety because exposure to the vectors is often frequent (e.g. mosquitoes), rendering donor screening questions insensitive, and infections are frequently asymptomatic, rendering physical findings, such as body temperature, useless.

The INTERCEPT Blood System™ for pathogen inactivation of platelet and plasma components was developed to prevent transfusion-transmitted infections. This proactive approach utilizes amotosalen HCl and UVA illumination (Figure 1) and has been demonstrated to inactivate high levels of the commonly tested pathogens (HIV, HBV, HCV, HTLV-I/II and *T. pallidum*), as well as a broad spectrum of cell-free and cell-associated, enveloped and non-enveloped viruses, gram negative and gram positive bacteria, and protozoa in platelet and plasma components (Lin 2004, Lin 2005, Pinna 2005, Singh 2006, Sawyer 2007, Sawyer 2008).

INTERCEPT™ treated components have demonstrated retention of therapeutic efficacy and safety in randomized controlled clinical trials and post marketing surveillance studies.

**Figure 1: INTERCEPT Mechanism of Action**

The INTERCEPT Blood System uses a combination of amotosalen HCl and long wavelength ultraviolet A (UVA) light. The amotosalen compound penetrates cellular and nuclear membranes and intercalates into the helical regions of DNA and RNA. Covalent crosslinks to the nucleic acid base pairs form upon exposure to UVA light, blocking DNA and RNA replication. This process inactivates leukocytes and pathogens, rendering them unable to cause disease, while retaining the function of plasma/platelets, which do not require nucleic acid replication for therapeutic efficacy.



## Aims

This study evaluated published data to determine the effectiveness of the INTERCEPT Blood System for inactivation of important vector-borne pathogens in platelet and/or plasma components.

## Methods

A review of pathogen inactivation publications was conducted to identify data pertaining to inactivation of vector-borne pathogens. Data showing inactivation of vector-borne pathogens in platelets and/or plasma by the INTERCEPT Blood System was included in this presentation.

## Results

The data identified demonstrates inactivation of high titers of 11 different pathogens by the INTERCEPT Blood System (Table 1). Pathogens identified are transmitted by 5 different types of insect vectors. Inactivation studies were conducted using full size platelet and or plasma components that were inoculated with approximately 10<sup>6</sup> viable organism/mL when possible, then treated with 150µm amotosalen and 3 J/cm<sup>2</sup> UVA treatment. Infections titers were measured before and after treatment using viability assays appropriate to each pathogen (Table 2).

**Table 2: Assay Systems for Detection and Quantification of Pathogen Viability**

Organism	Viability Assay System
West Nile virus	Plaque formation in vero cells
Chikungunya virus	Growth in vero cells
Dengue virus	Growth in vero cells
<i>Plasmodium falciparum</i>	Infectivity in red blood cells
<i>Anaplasma phagocytophilum</i>	Infectivity in mice
<i>Borrelia burgdorferi</i>	Growth in BHK-H medium
<i>Babesia microti</i>	Infectivity in mice
<i>Leishmania mexicana</i> (promastigote form)	Growth in medium 199
<i>Leishmania major</i> (amastigote form)	Growth in medium 199
<i>Trypanosoma cruzi</i>	Growth on 3T3 cells
<i>Orientia tsutsugamushi</i>	Infectivity in mice

**Table 1: Inactivation of Vector-Borne Pathogens (N=4 unless otherwise noted)**

Vector	Pathogen	Mean Log Reduction <sup>a</sup>	
		Platelets in ~65% Additive Solution / 35% Plasma	Plasma and Platelets in 100% Plasma
Mosquito	West Nile virus	>6.0 <sup>b</sup>	≥6.8 <sup>c</sup>
	Chikungunya virus	>6.4 <sup>d</sup>	≥7.6 <sup>d</sup>
	Dengue virus	>5.0 <sup>e</sup>	Not tested
	<i>Plasmodium falciparum</i>	≥6.0 <sup>f</sup>	≥6.9 <sup>f</sup>
Tick	<i>Anaplasma phagocytophilum</i>	Not tested	>4.2 <sup>d</sup>
	<i>Borrelia burgdorferi</i>	>6.8 <sup>g</sup>	>10.6 <sup>c</sup>
	<i>Babesia microti</i>	>5.3 <sup>h,f</sup>	>5.3 <sup>f</sup>
Sandfly	<i>Leishmania mexicana</i>	>5.0 <sup>i</sup>	Not tested
	<i>Leishmania major</i>	>4.3 <sup>j,l</sup>	Not tested
Reduviid bug	<i>Trypanosoma cruzi</i>	>5.3 <sup>k</sup>	>5.0 <sup>k</sup>
Mite	<i>Orientia tsutsugamushi</i>	>5.0 <sup>i</sup>	>5.5 <sup>m</sup>

a. Log reduction is calculated as log (pre-treatment titer ÷ post-treatment titer), where titer is expressed as 10<sup>x</sup> organisms/mL.

b. Lin 2005  
c. Singh 2006  
d. Sawyer 2009  
e. Lam 2007

f. Grellier 2008  
g. Lin 2004  
h. n=2  
i. Eastman 2005

j. n=1  
k. Van Voorhis 2003  
l. Rentas 2003  
m. Rentas 2004

## Conclusions

- All vector-borne viruses, bacteria and parasites tested were inactivated to or below the limit of detection by treatment with the INTERCEPT process.
- The INTERCEPT Blood System is highly effective against a spectrum of transfusion-transmitted vector-borne pathogens.

## References

Eastman, et al. 2005. Transfusion. 45:1459  
Grellier, et al. 2008. Transfusion. 48:1676  
Lam, et al. 2007. Transfusion. 47:131A  
Lin, et al. 2004. Transfusion. 44:1496

Lin, et al. 2005. Transfusion. 45:580  
Pinna, et al. 2005. Transfusion Medicine. 15:269  
Rentas, et al. 2003. Transfusion. 43:84A  
Rentas, et al. 2004. Transfusion. 44:104A

Sawyer, et al. 2008. Transfusion. 48:88A  
Sawyer, et al. 2009. Vox Sang. 96(suppl.1):233  
Sawyer, et al. 2007. Transfusion. 47:1062

Singh, et al. 2006. Transfusion. 46: 1168  
Van Voorhis, et al. 2003. Antimicrobial Agents and Chemotherapy. 47:475