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# **Pathogen inactivation of non-enveloped viruses in the transfusion setting – blood components**

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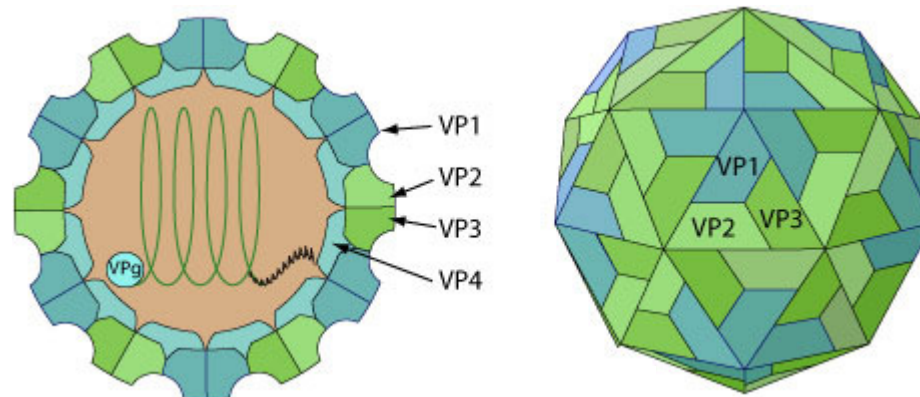
# Non-enveloped transfusion-transmissible viruses

- Most relevant:
  - Hepatitis A virus (HAV)
  - Human parvovirus B19 (B19V)
  - Hepatitis E virus (HEV)
- Current routine screening procedures for blood donations not intended for plasma pooling or fractionation do not include HAV and B19V
- Nationwide screening of blood donations for HEV RNA in some countries (e.g., GB, Netherlands, Germany from 2020)



# Non-enveloped transfusion-transmissible viruses

- Stable protein shell
- Resistant to inactivation by heat and detergents
- Tightly packed viral capsids may act as barriers against chemical inactivation agents





## Current PI technologies

| PI procedure      | Blood products |           |      |             |
|-------------------|----------------|-----------|------|-------------|
|                   | Plasma         | Platelets | RBCs | Whole blood |
| Solvent-Detergent |                |           |      |             |
| MB                |                |           |      |             |
| Amotosalen (S59)  |                |           |      |             |
| Riboflavin        |                |           | ?    |             |
| Amustaline (S303) | ?              | ?         |      | ?           |
| UVC               |                |           |      | ?           |

Procedures targeting cell membranes

Procedures targeting ribonucleic acids

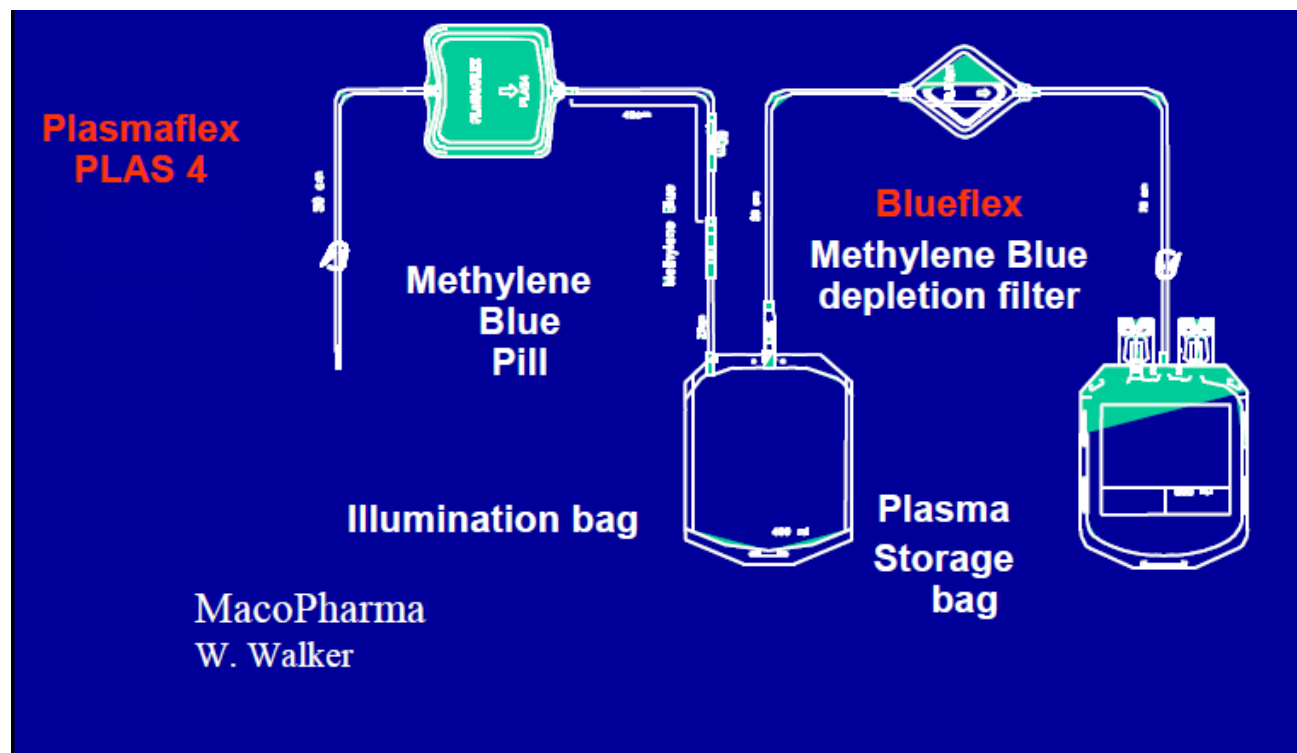
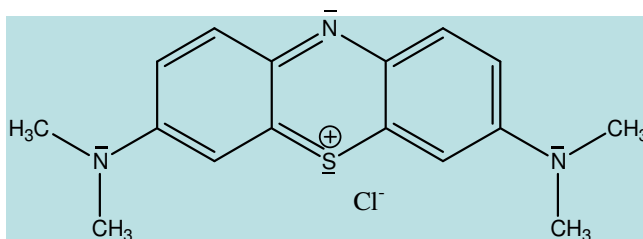


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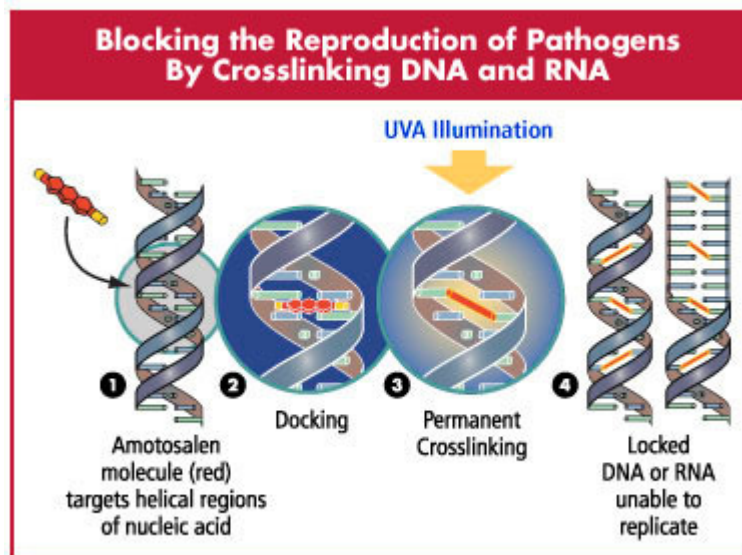
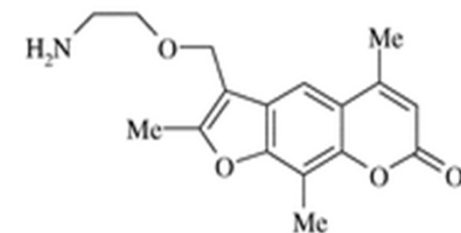
## Methylene blue/light (THERAFLEX MB-Plasma)





# UVA/amotosalen (INTERCEPT)

- Amotosalen = photosensitizer
- Photochemical procedure:
  - Crosslinking via pyrimidine bases (thymidine and cytidine) and uridine



Aminoethyloxymethyltrimethylpsoralen  
S-59, "Amotosalen"



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## UVA/amotosalen (INTERCEPT)

The INTERCEPT Blood  
System for Platelets



Step 1  
Amotosalen

Step 2  
Illumination

Step 3  
CAD

Process Complete  
Storage

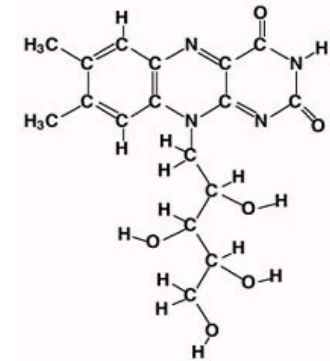
The INTERCEPT Blood  
System for Plasma



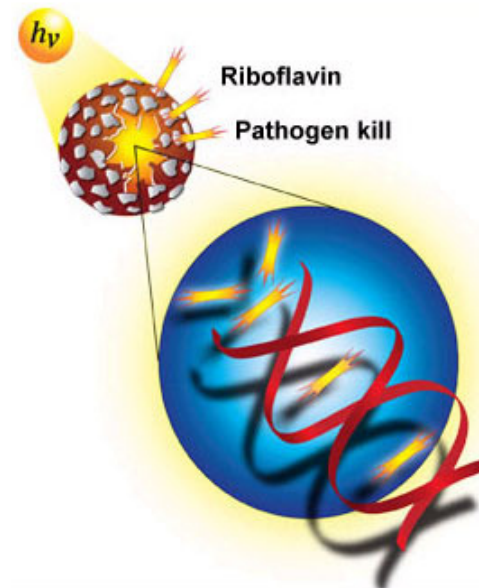


## UV/riboflavin (MIRASOL)

- Riboflavin (Vitamin B2)=photosensitizer
- Photodynamical procedure: formation of oxygen radicals (e.g.  $O_2^-$ ) und singlet  $O_2$



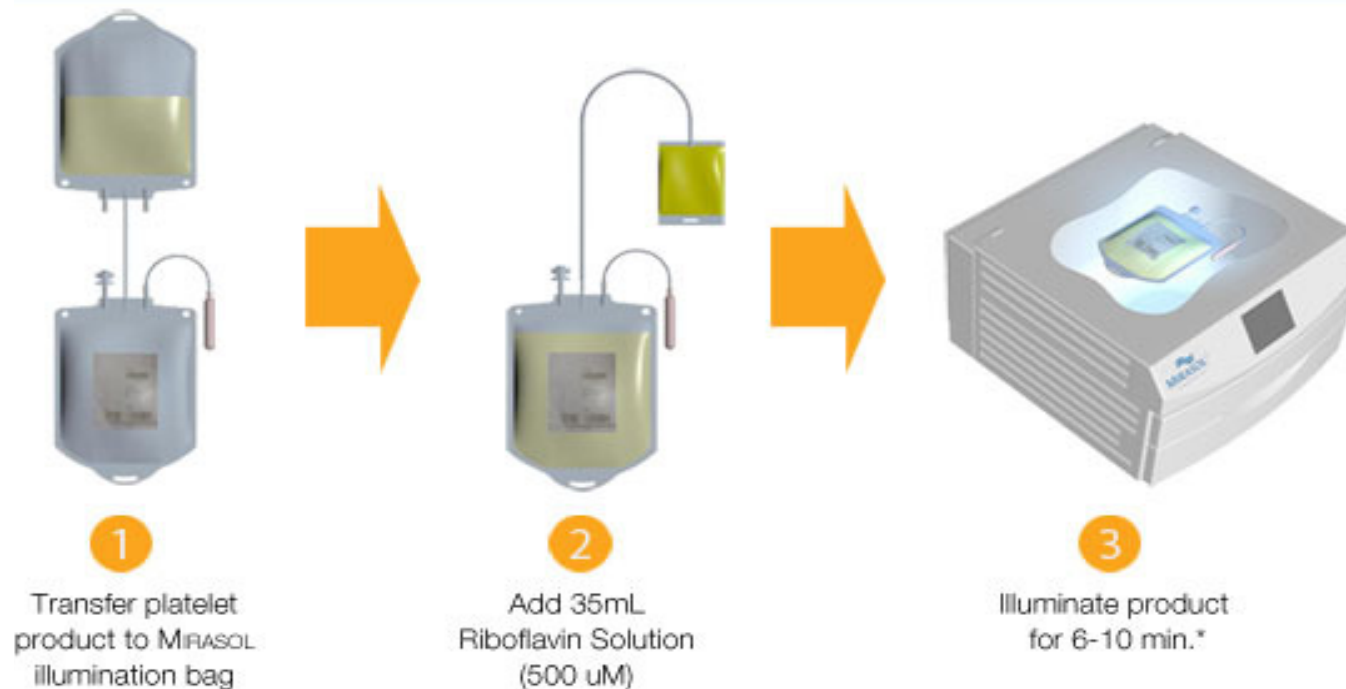
→ Photooxidation of the nucleotide guanine





## UV/riboflavin (MIRASOL)

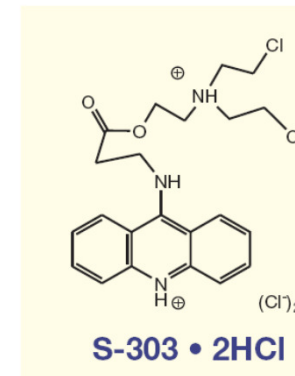
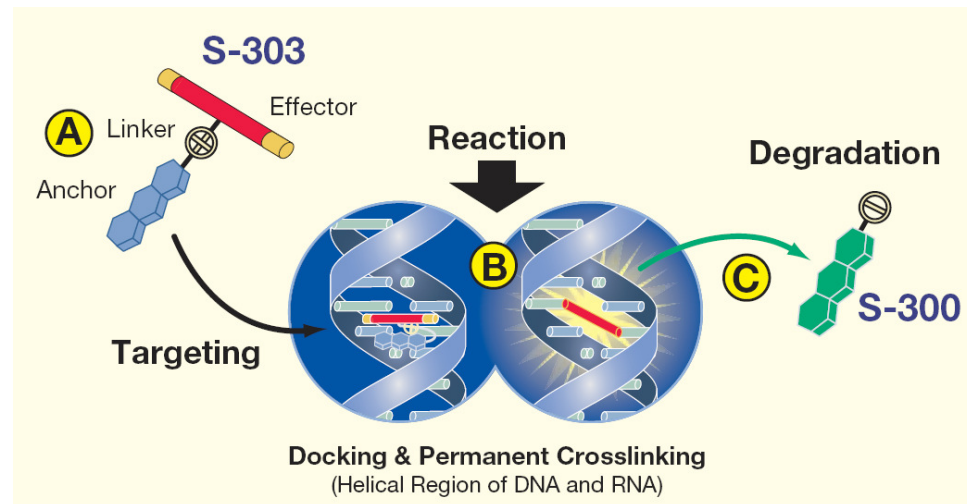
- Suited for plasma and platelets
- Evaluated in whole blood





## S303-system for RBCs

- Chemical procedure
  - Amustaline: alkylating agent
  - Glutathion



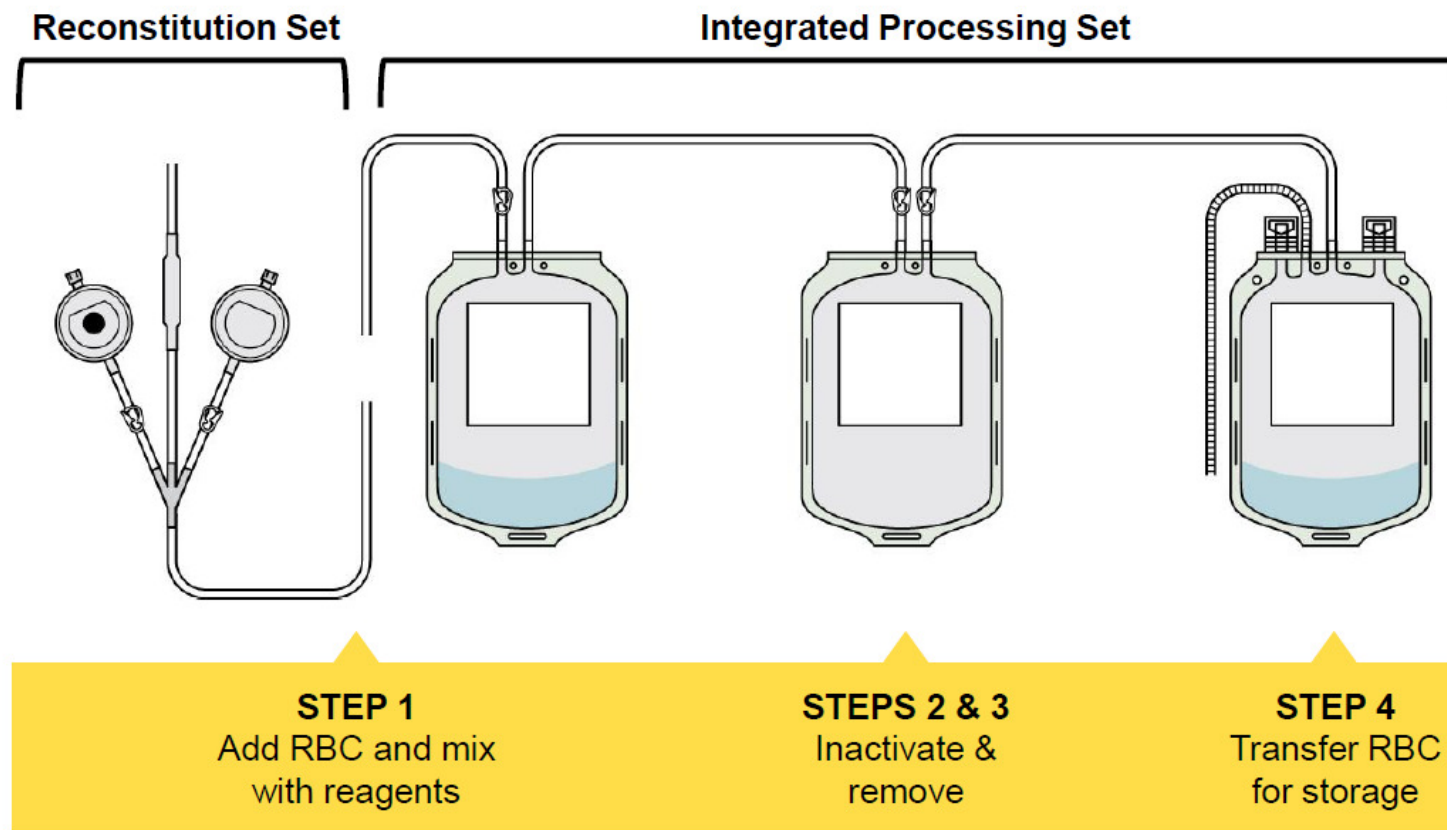


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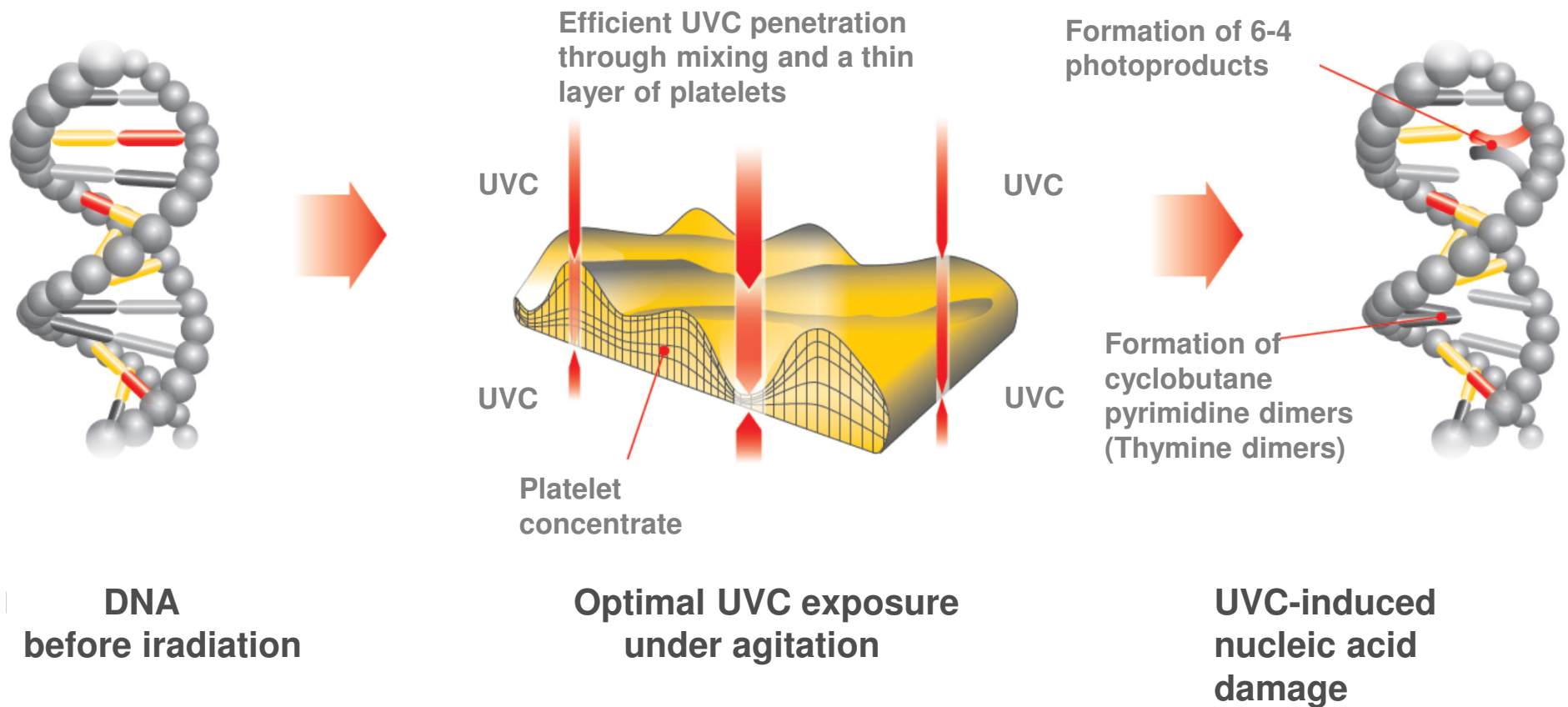
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## S303-system for RBCs





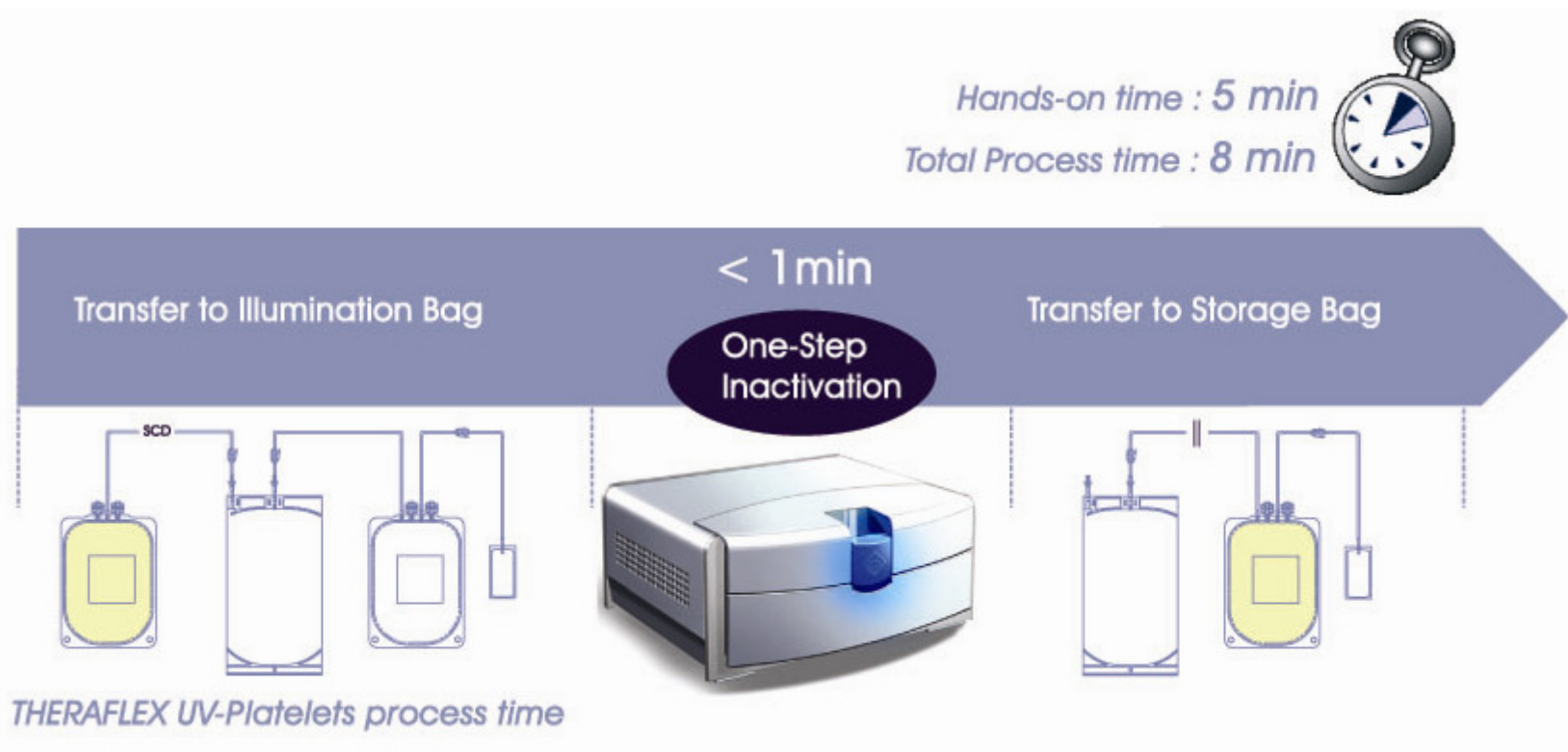
## UVC (THERAFLEX UV-Platelets)





## UVC (THERAFLEX UV-Platelets)

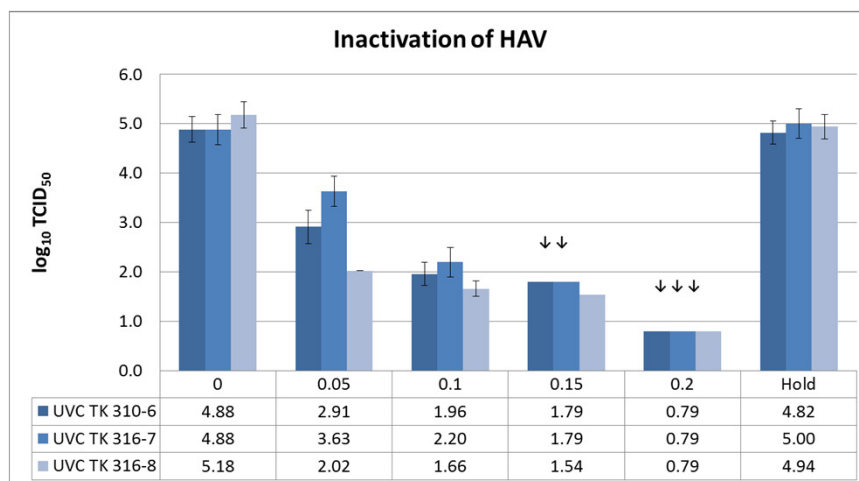
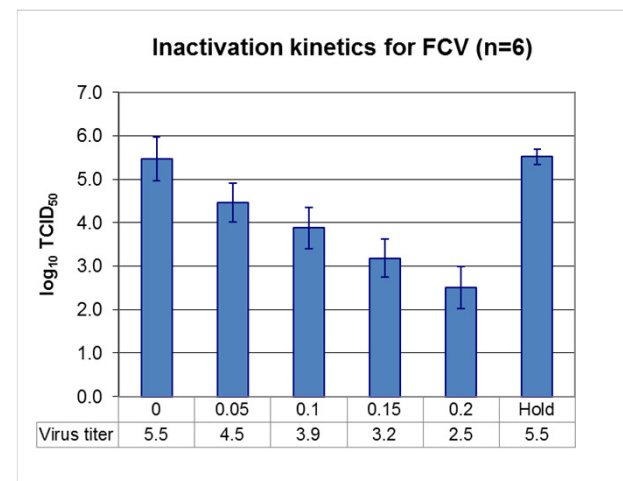
- UVC only, no exogenous component
- Suited for plasma and platelets





# Inactivation of HAV and FCV in PCs by UVC

| UVC dose<br>(cumulative) | HAV (n=3)<br>log <sub>10</sub> TCID <sub>50</sub> /mL | log <sub>10</sub> RF | FCV (n=6)<br>log <sub>10</sub> TCID <sub>50</sub> /mL | log <sub>10</sub> RF |
|--------------------------|-------------------------------------------------------|----------------------|-------------------------------------------------------|----------------------|
| Load                     | 5.0 ± 0.2                                             | 0.0                  | 5.5 ± 0.5                                             | 0.0                  |
| 0.05 J/cm <sup>2</sup>   | 2.9 ± 0.8                                             | 2.1 ± 1.0            | 4.5 ± 0.5                                             | 1.0 ± 0.1            |
| 0.10 J/cm <sup>2</sup>   | 1.9 ± 0.3                                             | 3.1 ± 0.4            | 3.9 ± 0.5                                             | 1.6 ± 0.1            |
| 0.15 J/cm <sup>2</sup>   | 1.6 ± 0.3                                             | 3.4 ± 0.4            | 3.2 ± 0.4                                             | 2.3 ± 0.2            |
| 0.20 J/cm <sup>2</sup>   | ≤ 0.8                                                 | ≥ 4.2                | 2.5 ± 0.5                                             | 3.0 ± 0.2            |
| Ref. sample              | 4.9 ± 0.1                                             | 0.1 ± 0.2            | 5.5 ± 0.2                                             | -0.1 ± 0.4           |





## Inactivation of non-enveloped viruses

| Virus type           | Log <sub>10</sub> reduction values |                |               |         |      |
|----------------------|------------------------------------|----------------|---------------|---------|------|
|                      | Methylene blue                     | UVA/amotosalen | UV/riboflavin | UVC     | S303 |
| HAV                  | 0                                  | 0.76*          | 0.6*          | ≥4.2    |      |
|                      |                                    |                | 1.8           |         |      |
| B19V                 | ≥5                                 | 3.5 - >5       |               |         |      |
|                      |                                    | 1.6 – 5.8†     |               |         |      |
| PPV (model for B19V) | 0                                  | 0.38*          | 0.28‡*        | 5.0     |      |
|                      |                                    | 0              | ≥5.0‡         |         |      |
| FCV (model for HEV)  | ≥3.9                               | 1.7-2.4        |               | 3.0     |      |
| EMCV (model for HAV) |                                    |                | 3.2           | 4.0     |      |
| Bluetongue virus     |                                    | 5.6-5.9        |               |         | ≥5.0 |
| Human adenovirus 5   | ≥5.33                              | >5.2           |               |         | >7.4 |
| Simian adenovirus 15 |                                    | 0.7-2.3        |               |         |      |
| Polio                | >1                                 |                |               |         |      |
| HEV                  |                                    |                | 2 – 3         | ongoing |      |

\*Results from a side-to-side comparison study; †The level of B19V inactivation increased with increasing time of incubation with amotosalen before UVA treatment; ‡The same strain of model virus was used; ||Quantitation of HEV RNA was performed to determine viral infectivity



## Summary I

- Tight capsid structure may exclude even low-molecular-weight compounds, such as psoralens and riboflavin, from the interior of the virus, making PI less effective against non-enveloped viruses
- Two reported cases of HEV transmission through UVA/amotosalen-treated FFP verified resistance of HEV to the INTERCEPT technology
- The THERAFLEX UV-Platelets technology
  - is a physical method
  - could be broadly effective against non-enveloped viruses
- Model viruses may not always predict susceptibility of clinical viruses
- Infectivity assays with clinical viruses should be performed, if available, to test PI capacity



# HEV load transfused resulting in posttransfusion infection or no posttransfusion infection

