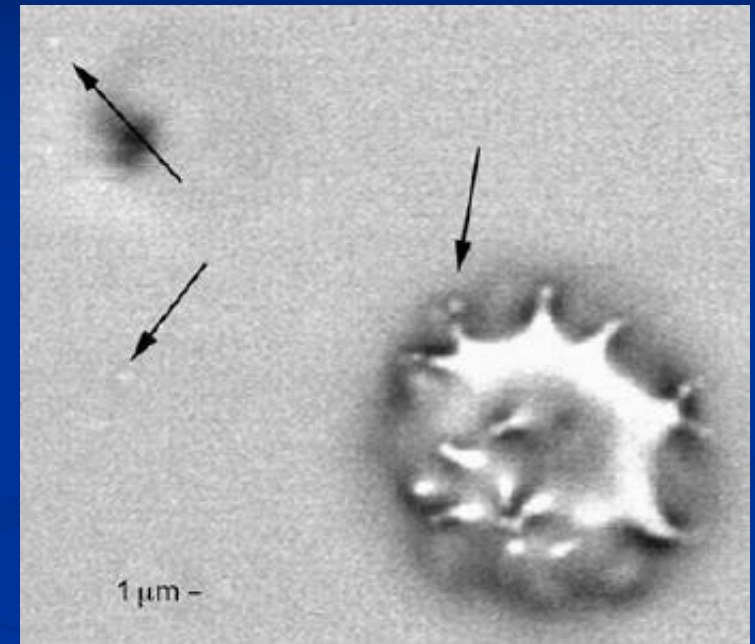


Inflammatory mediators unrelated to leucocytes: microvesicles and lipids

- Echinocytic changes after 2-3 weeks.
- The tip of echinotic spicules shed RBC microvesicles in supernatant.
 - Integral part of RBC ageing process.
 - Accelerated in stored RBC units.
- These microvesicles (ectosomes) contain lipids that can amplify an inflammation.
- These microvesicles also bind annexin V.
 - A procoagulant state results, which facilitates generation of thrombin (transfusion-induced facilitation of thrombin or TRIFT effect).
 - This might increase risk of thrombosis.
 - No procoagulant state if storage < 21 days.

Sweeney et al. Transfusion 2009;49:1569-79.



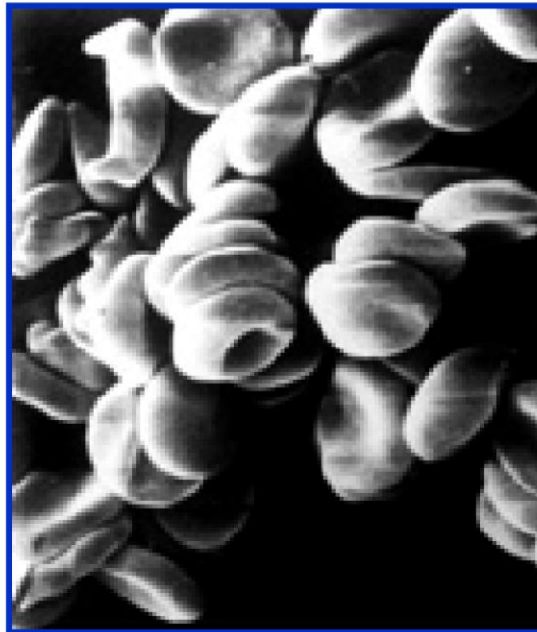
By confocal differential interference contrast microscopy (x 4000), it is possible to observe forming microparticles (arrows) from spicules of echinocytes (Tissot et al. Curr Opin Hematol 2010;17:571-7).

Systemic and regional DO_2 and VO_2

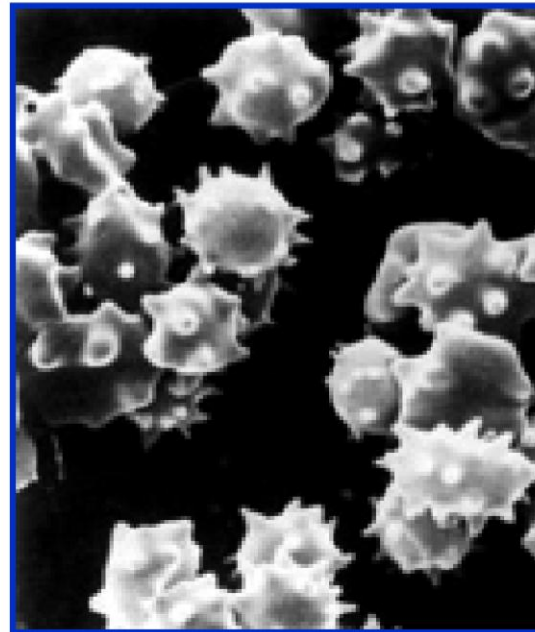
- A RBC transfusion always increase global O_2 delivery (DO_2), but it does not always increase local DO_2 and VO_2 ; why?
- Four mechanisms may explain why a RBC transfusion does not increase regional DO_2 :
 - RBC plugging:
 - Decreased RBC deformability
 - Increased adherence
 - Increased aggregation
 - Vasoconstriction of microvasculature.

Echinocytes, deformability

- RBC diameter: 7.2 microns.
- Capillary diameter: 3-8 microns.



Day 1



Day 21



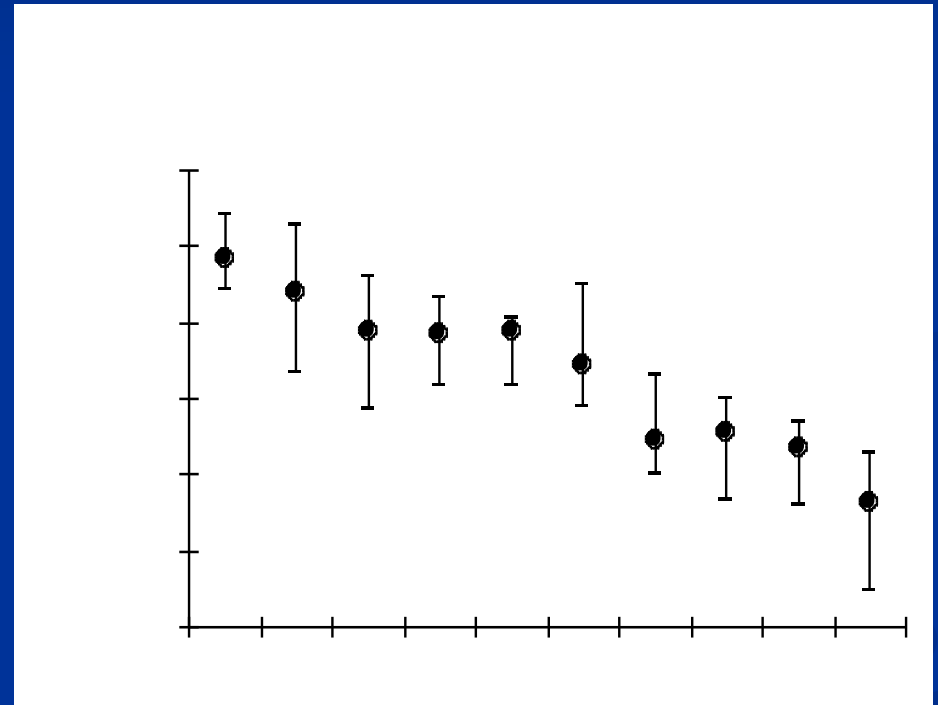
Day 35

Scanning electron micrographs of red blood cells isolated from stored blood on Day 1, Day 21, and Day 35. During storage, the shape of RBCs changed gradually from normal discoid to echinocytes (dented or shriveled red cells).

Reproduced with permission from: Hovav et al. *Transfusion*. 1999;39:277-281.

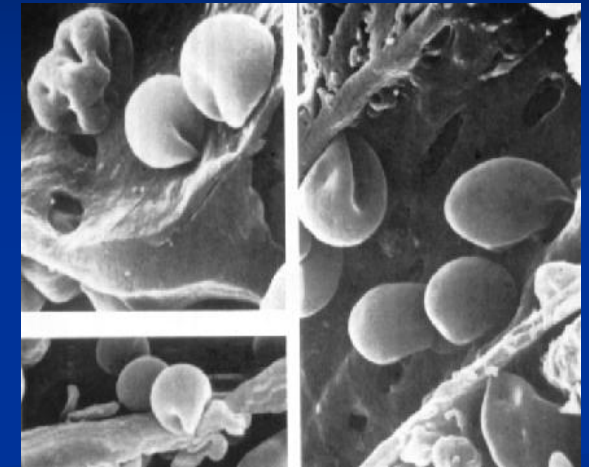
Length of storage and RBC deformability (not reversible)

- Progressive lost of RBC deformability while stored.
 - Bennett-Guerrero E, Veldman TH, Doctor A, et al. PNAS 2007;104:17063-8.
- This is **not reversible**.
 - Bosman et al. Transf Med 2008;18:335-47.



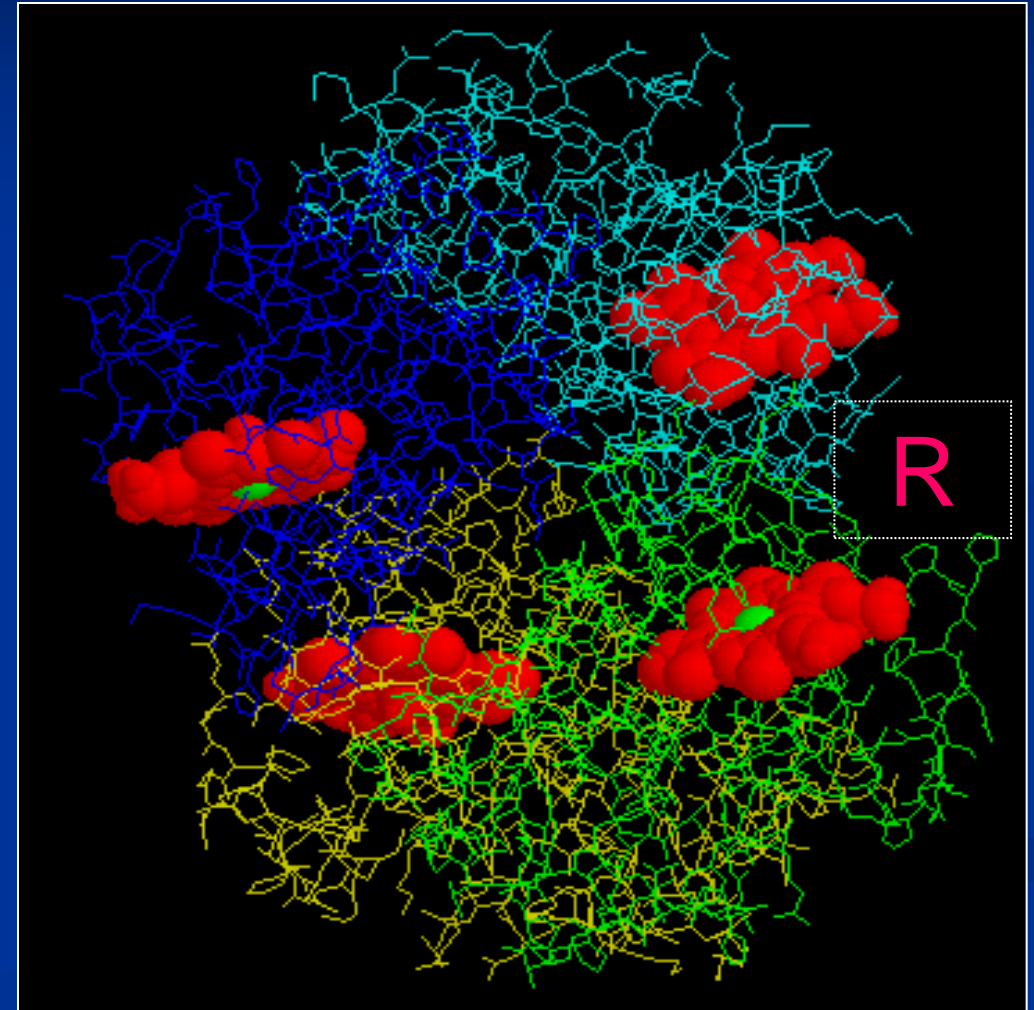
Increased RBC adherence and aggregation

- Normally, RBC adherence to endothelial cells is insignificant.
- In stored RBC units, translocation of phosphatidyl-serine (PS) to RBC membrane is observed over time.
 - This is associated with **increased adherence** of RBC.
 - It is also associated with **more aggregation**: RBC surface PS has been reported to cause platelet activation and aggregation, which can be prothrombotic (Koshkaryev et al. Transfusion 2009;49:2136-43)
- NB: prestorage leuco-reduction abolished this increased adherence of stored RBC (Chin-Yee et al. Transfusion 2009;49:2304-10).



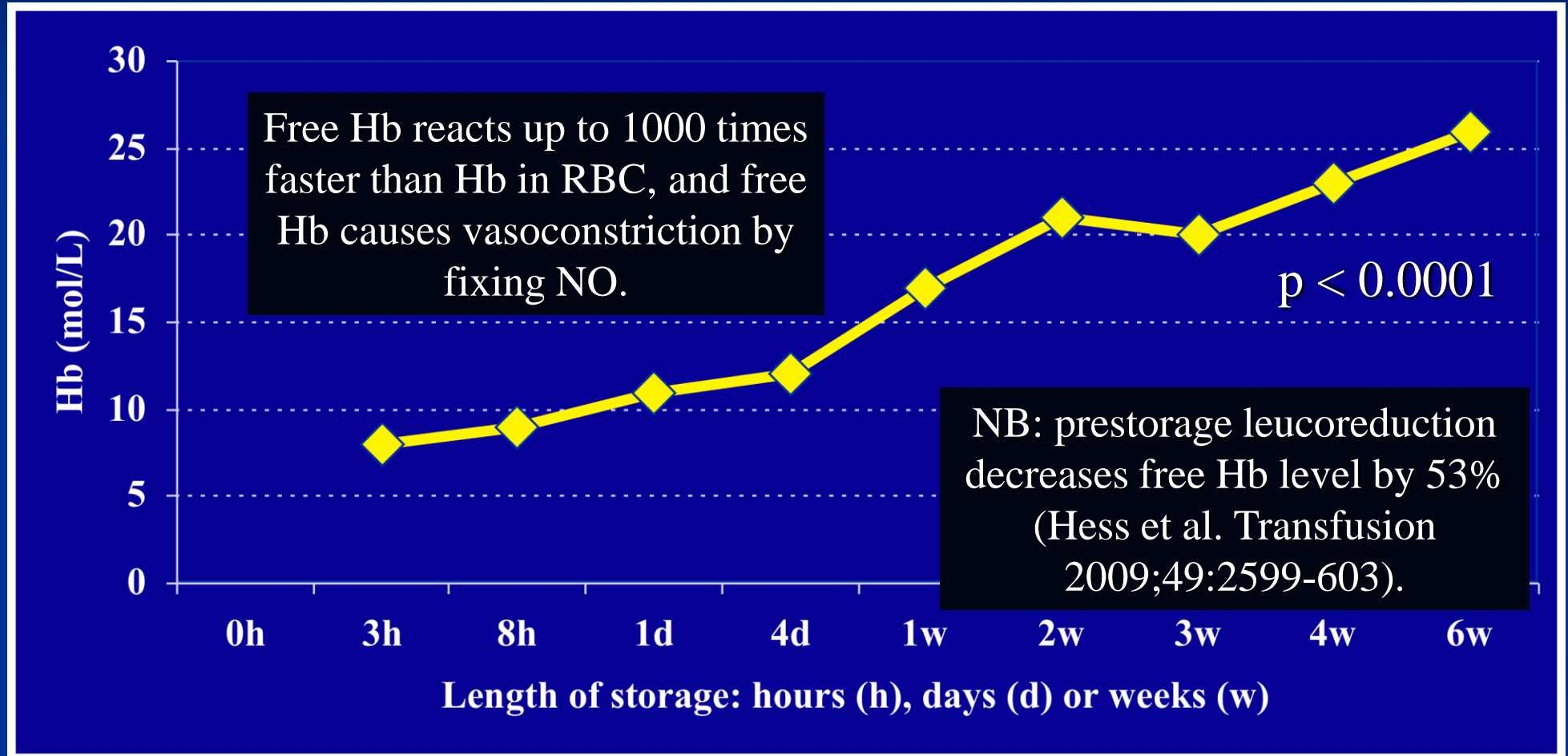
Hemoglobin (Hb), NO and regulation of vascular tone

- Hemoglobin molecules interacts not only with O_2 and CO_2 , but also with nitric oxide (NO).
- Hb in RBC regulates small vessels tone and regional blood flow.
 - RBC react almost immediately to local cellular hypoxia by releasing NO: local vasodilatation results.
 - Vice-versa, RBC catch more NO if local cellular VO_2 seems adequate: this causes local vasoconstriction.
- This function is almost immediately disturbed by storage (< 3 hours of storage).



Thanks to Dr Alan Doctor

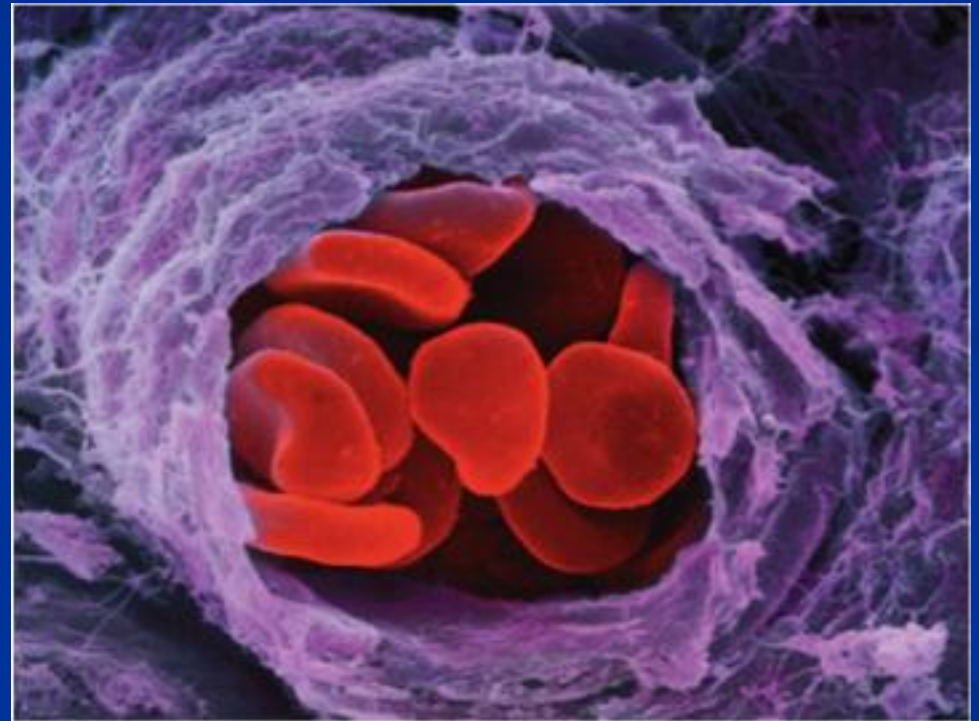
Length of storage and free hemoglobin (Hb) in supernatant



Tiré de: Bennett-Guerrero E, Veldman TH, Doctor A, et al. Evolution of adverse changes in stored RBCs. PNAS 2007;104:17063-8.

Transfused RBC transfusion and regional DO_2 and VO_2

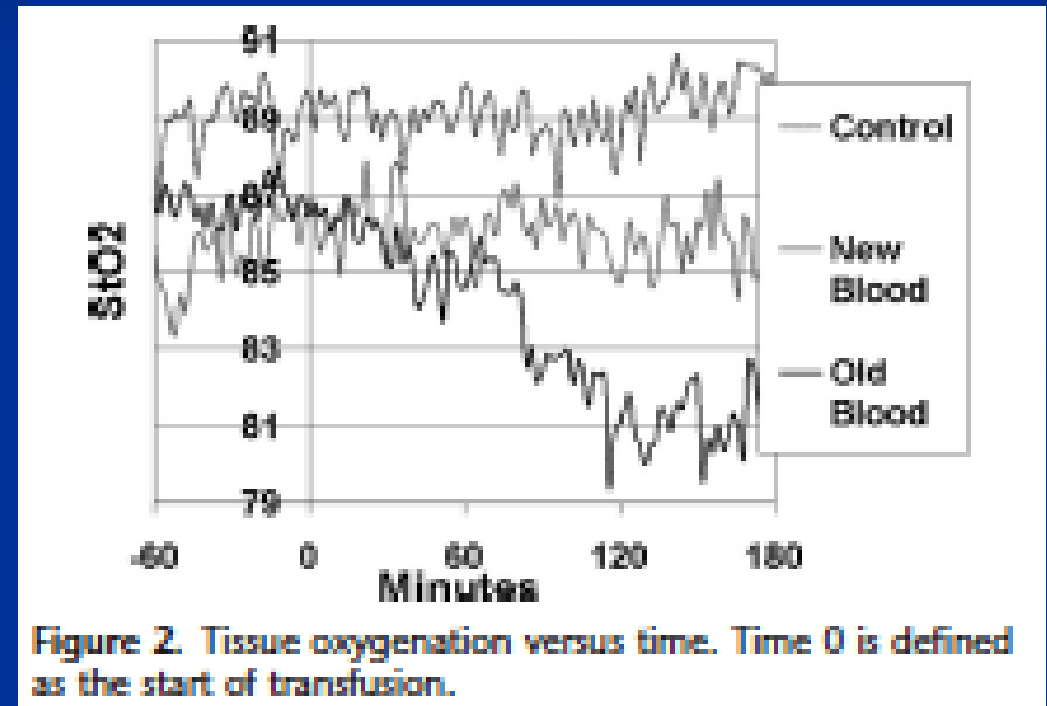
- RBC transfusions may impair regional blood flow and cellular VO_2 by many mechanisms:
 - RBC plugging:
 - Decreased deformability.
 - Increased adherence.
 - Increased aggregation.
 - Vasoconstriction (NO-Hb, free Hb).
 - Moreover, low 2,3-DPG may alter O_2 release.
- RBC transfusions improve global DO_2 , but this does not always result in better local DO_2 & VO_2 .



Thanks to Dr Alan Doctor

Effect of length of storage on O₂ tissular saturation (StO₂) in trauma

- Control group: no transfusion.
 - Number of patients = 16
 - No decline in StO₂ comparing baseline to transfusion period.
- New blood: storage < 21 days.
 - 15 patients
 - No decline in StO₂.
- Old blood: storage > 21 days.
 - 17 patients
 - Significant decline in StO₂ (p < 0.05).



Kiraly LN, et al. J Trauma
2009;67:29-32

**Clinical evidence that
there can be an
association between
length of storage and
outcome in ICU:
observational studies**

Health problems possibly attributable to older RBC units



- More deaths?
- Transfusion-related immunomodulation (TRIM)?
 - More inflammation.
 - More multiple organ dysfunction syndrome (MODS)?
 - Decreased immune capacity against infections?
 - More nosocomial infections?
- Other health problems:
 - More thrombosis & ischemic lesions?

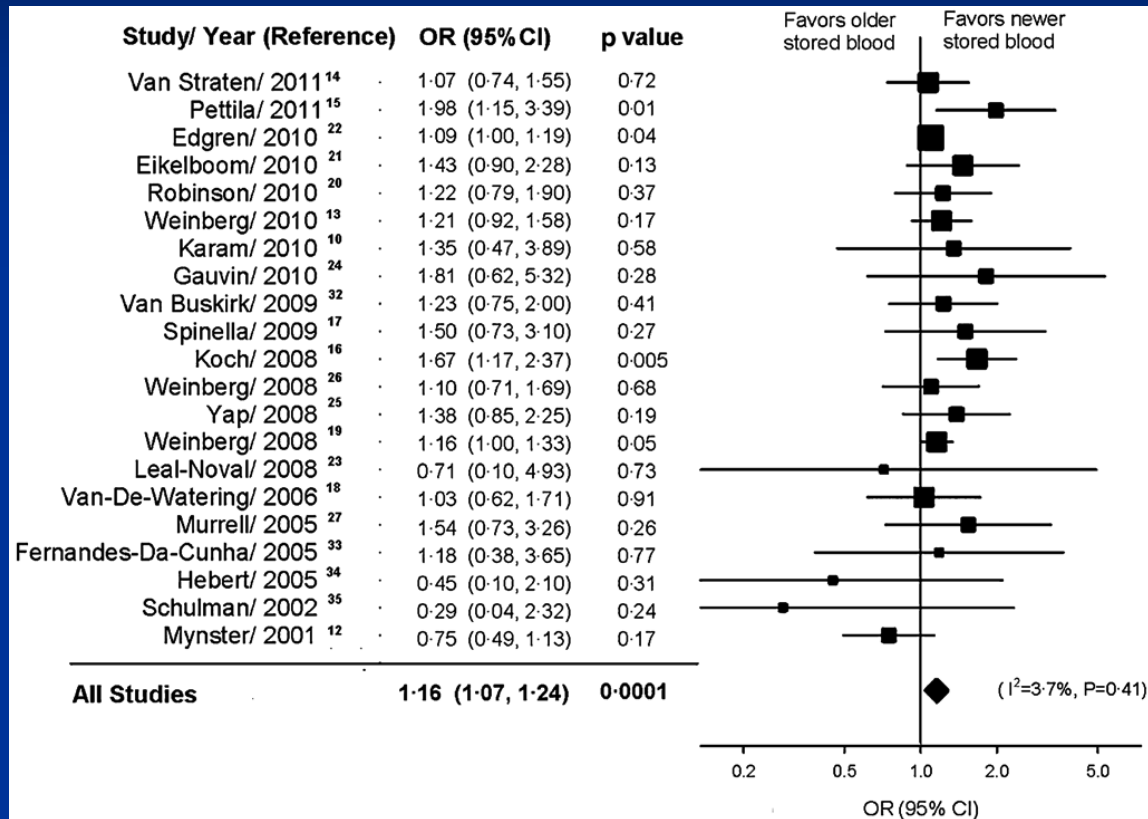
Clinical evidence that there can be an association between length of storage and outcome in ICU

Mortality

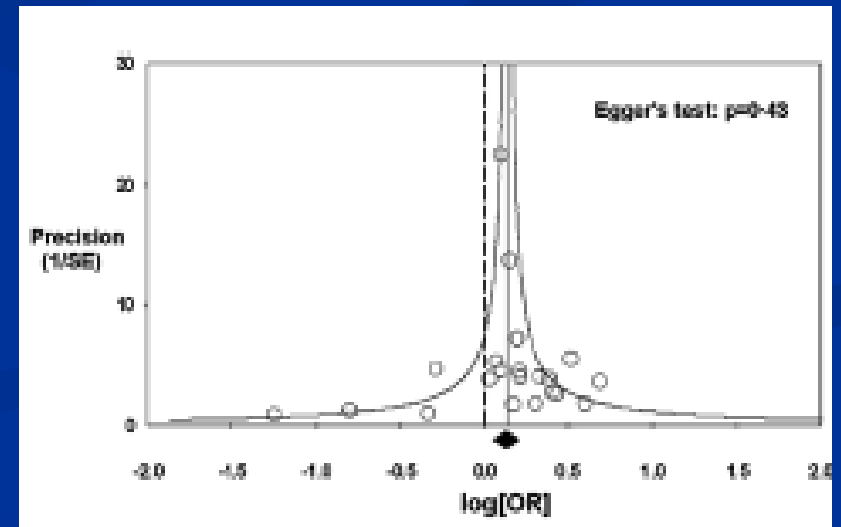
Thrombosis and ischemic lesions

Other outcomes (MODS, etc)

Older RBC units and mortality

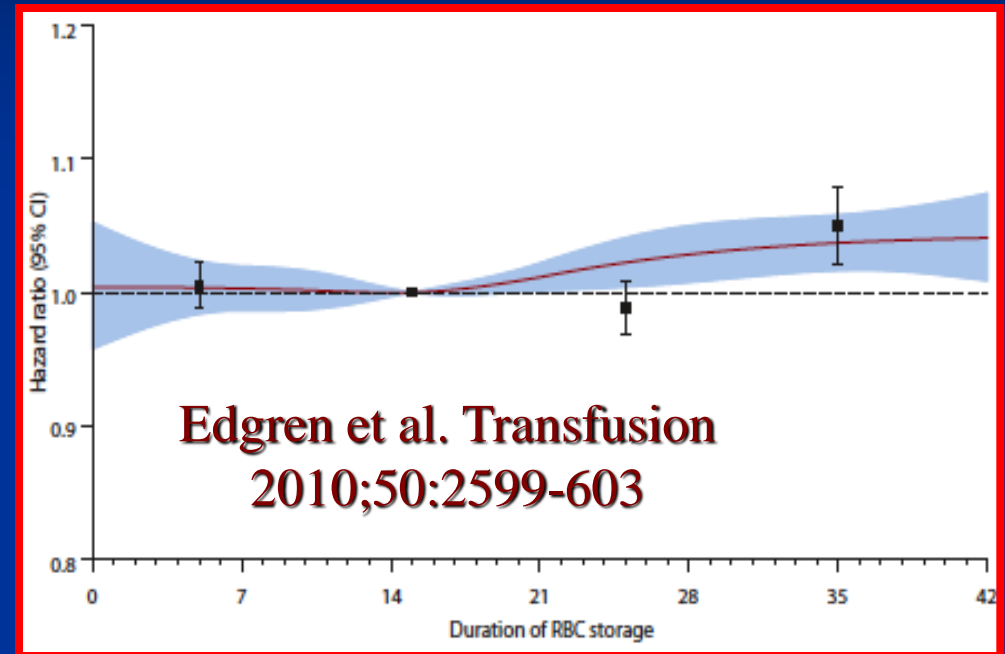
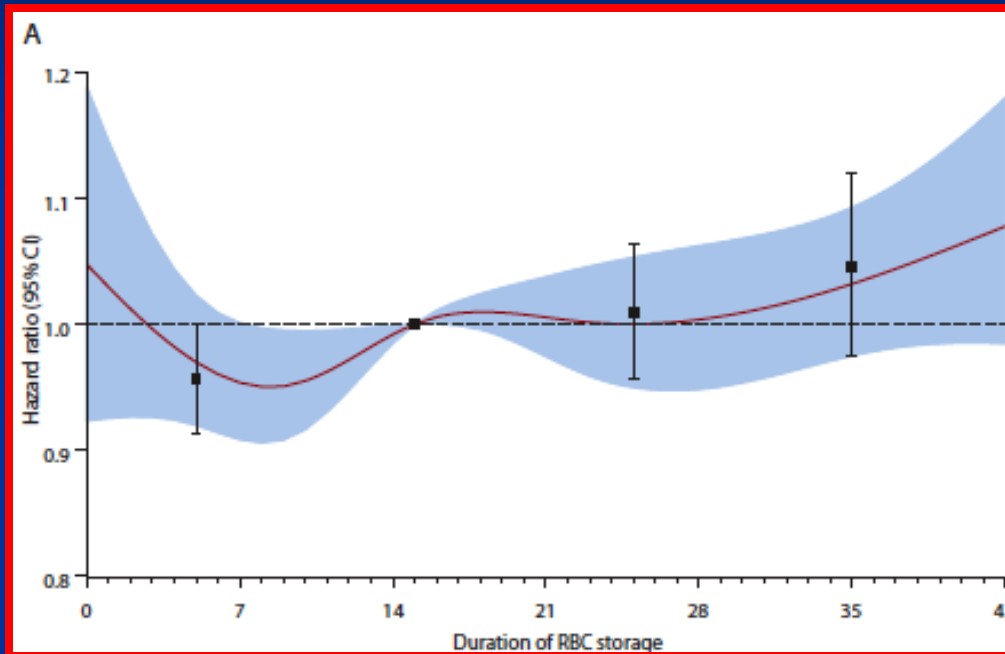


- 21 studies (3 RCTs).
- OR: 1.16 (95%_{CI}: 1.07-1.24).
- NNT: 97 patients (95%_{CI}: 63-199) need to be treated with 1 RBC unit to save 1 life.
- No publication bias (funnel plot). Egger test: $p = 0.43$.



Wang et al. Transfusion 2012;52:in press

Older RBC units and mortality in a whole population (not only ICU)



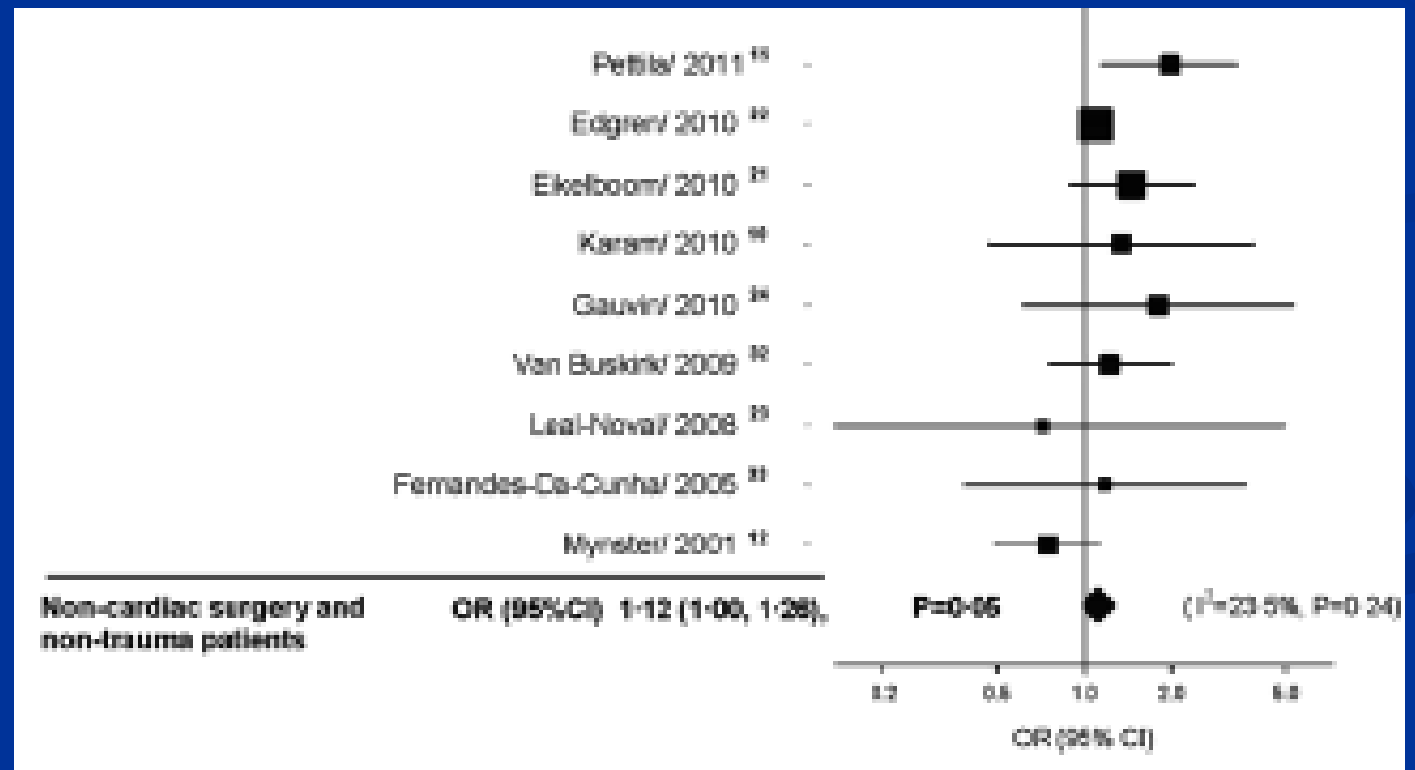
If older RBC units is really a risk factor of mortality, this happens only with AOB ≥ 30 days.

With 2 years follow up, the hazard ratio for mortality was 1.05 (95% CI: 1.02-1.08) among recipients of blood stored 30-42 days vs stored 10-19 days.

Association between older RBC units and mortality in non-cardiac adults

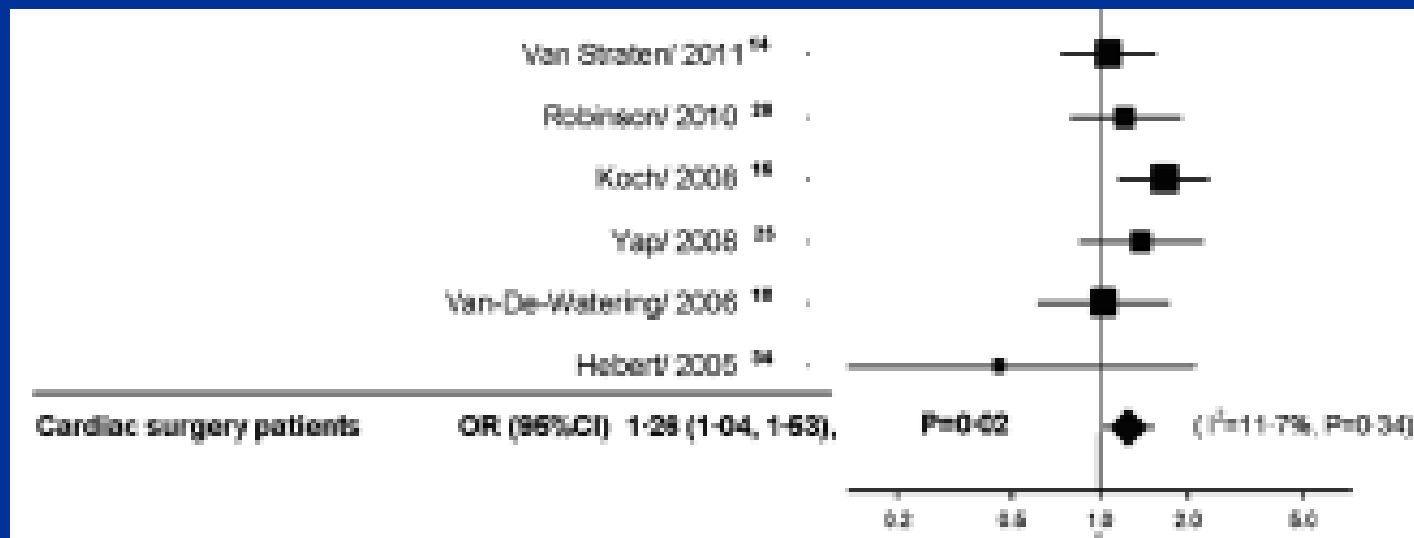
- 9 studies.
- OR: 1.12 (95%CI: 1.12-1.26).

Wang et al.
Transfusion
2012;52: in
press



Association between older RBC units and mortality in cardiac adults

- 6 studies.
- OR: 1.26 (95%CI: 1.04-1.53).



Wang et al. Transfusion 2012;52: in press