Comparison of two sterile connection devices and the effect of sterile connections on blood component quality

Pieter F. van der Meer, Marjan Eijzenga, and Ruby N.I. Pietersz

BACKGROUND: Sterile connection devices (SCDs) are used to connect pieces of polyvinylchloride tubing between blood bag systems. After observing a slight decrease in inner diameter of tubing welded with the CompoDock S2 SCD, the effect of welded tubing on storage characteristics of white blood cell (WBC)-reduced red blood cells (RBCs) and platelet (PLT) concentrates was studied. Welds were made with Terumo SCD (T-SCD) or CompoDock S2, and unwelded tubing served as reference.

STUDY DESIGN AND METHODS: Three WBC-reduced RBC units or 3 PLT concentrates were pooled and divided to prevent donor-dependent differences. The units were transferred 10 times over (1) tubing with a T-SCD weld, (2) a CompoDock S2 weld, or (3) unwelded tubing. RBCs were stored for 42 days and free hemoglobin (Hb) was measured; PLT concentrates were stored for 8 days and CD62P expression was measured, as markers for blood component quality (n = 10 paired experiments).

RESULTS: WBC-reduced RBC units had similar hemolysis at the end of storage: 0.47 ± 0.28, 0.47 ± 0.35, and 0.49 ± 0.38 percent of total Hb, for tubing with a T-SCD weld, a CompoDock S2 weld, or no weld, respectively (not significant). CD62P expression of stored WBC-reduced PLT concentrates was not significantly different between the groups: 20.3 ± 5.1, 19.8 ± 5.1, and 22.3 ± 9.8 percent for tubing with a T-SCD weld, a CompoDock S2 weld, or no weld, respectively.

CONCLUSION: The quality of blood components, measured as RBC hemolysis and platelet CD62P expression, is not adversely affected by the presence of a sterile connection in the tubing, made by either the CompoDock S2 or the T-SCD.

ABBREVIATIONS: MPV = mean platelet volume; SCD(s) = sterile connection device(s).

S}terile connection devices (SCDs) are used in blood centers to connect various (blood bag) systems, for example, to pool blood components, dock on a white blood cell (WBC)-reduction filter, or connect a sample bag for quality control or bacterial screening. Currently available SCDs can weld pieces of medical polyvinylchloride (PVC) tubing and are based on either one of the following two principles. The first principle is based on heating a copper wafer to approximately 320°C. The tubing to be welded is cut by the heated wafer, and the two stubs are quickly attached to each other. The PVC cools down and the weld is complete and can be opened.

Because of the high temperature of the wafer during welding, microorganisms present at the site of the weld are killed, preventing contamination of the welded tubing. SCDs with this principle are the SCD312 (Haemonetics, Braintree, MA),1 the Terumo SCD (T-SCD; Terumo, Tokyo, Japan),2 and with a modified copper wafer and a different movement of the wafer and tubing, the total containment device (TCD, Haemonetics).3 The second principle is applied by the CompoDock (Fresenius HemoCare, Bad-Homburg, Germany).4 The pieces of tubing to be welded are first sealed, and next these seals are heated by a reusable heating element, the seal is melted, the melted seals are brought together, and the tubing is welded and can be

From the Sanquin Blood Bank North West Region, Amsterdam, the Netherlands.

Address reprint requests to: Pieter F. van der Meer, PhD, Sanquin Blood Bank North West Region, Plesmanlaan 125, 1066 CX, PO Box 9137, 1006 AC, Amsterdam, the Netherlands; e-mail: p.vandermeer@sanquin.nl.

Received for publication May 20, 2005; revision received July 1, 2005, and accepted July 19, 2005.

doi: 10.1111/j.1537-2995.2006.00738.x

opened. Because sealed tubing is welded, this is validated
to be a truly "closed" system. Both methods, however,
make structurally intact welds, and both use a very high
temperature, thus preventing the entry of bacteria into the
tubing.2,4

During an evaluation of an updated version of the
CompoDock (CompoDock Series 2; S2), we observed that
welded tubing showed a slightly decreased inner diame-
ter, with frays on the inside of the tubing. The oval shape
suggested that this was due to the fact that sealed pieces
of tubing were welded, which caused some PVC from one
side of the tubing to stick to the opposite side of the tubing
(see Fig. 1). We therefore designed a study to investigate
the effect of sterile connections in pieces of tubing on the
in vitro quality of white blood cell (WBC)-reduced red
blood cells (RBCs) and WBC-reduced platelet (PLT) con-
centrates. The blood components were transferred multi-
ple times over tubing that contained a weld made either
by the T-SCD or the CompoDock S2, and tubing without
a weld served as reference. We further measured tensile
strength and performed leak testing of tubing that was
welded by the CompoDock S2, with tubing currently used
in our blood center.

Measurement of tensile strength, leak testing, and
ease of opening

Various combinations of PVC tubing routinely used in our
blood center were used to measure the strength of welded
tubing, made by the CompoDock S2. We used tubing from
Baxter (gamma-sterilized), Fresenius HemoCare (gamma-
and steam-sterilized), and Terumo (gamma-sterilized).
The steam-sterilized tubing was filled with a liquid and is
referred to as "wet" tubing; the gamma-sterilized tubing
was not filled with a liquid and is referred to as "dry" tub-
ing. Welds were made in various combinations that may
be encountered in our blood center. Per day, 5 replicates
of each desired combination were done, and this was
repeated 5 times, resulting in 25 replicates for each tub-
ing combination. To determine the maximal strength of
the tubing, tensile strength of unwelded tubing was mea-
sured 10 times. Tensile strength was measured in welded
tubing with at least 10 cm on each side of the weld and
measured with a tensile strength measuring device (Type
122041/93, Zwick Material Testing, Ulm, Germany) as
described before.4 In short, the piece of (welded) tubing is
placed in the testing device and pulled with increasing
force. The force at which the tubing breaks is the maxi-
mum tensile strength of that piece of tubing. According to the European
pharmacopoeia,5 all welds should have a
tensile strength of greater than 20 N
(kg × m/sec²). Leak testing was per-
formed as described before,4 with a
1 bar pressure to the tubing during
30 seconds. We required that none of
the welds should leak. Ease of opening was
scored on a scale from 0 to 2, with "0"
being opening after more than
10 seconds after first try, “1” open
within
10 seconds but not at first try, and “2”
open at first try.

Whole-blood collection and
processing

Whole blood was collected in top-and-
bottom–bag systems with an in-line
RBC filter (T2988, Fresenius HemoCare
Netherlands, Emmer-Compascuum,
the Netherlands). The blood was stored
overnight at room temperature under
butane-1,4-diol cooling plates and, after
hard-spin centrifugation the following
morning, separated into plasma, a buffy
coat, and RBCs. The RBCs were immedi-
ately WBC-reduced with the in-line fil-
ter, at room temperature. After 2 hours

![Fig. 1. Example of representative welds made in PVC tubing with either the CompoDock S2 (A) or the T-SCD (B) SCDs, in front and side views.](image-url)
of resting, 5 buffy coats from identical blood group and 1 unit of plasma (from one of the donations) were connected to a pooling system (TF*FP06010M1, Terumo) with a T-SCD and centrifuged, and the PLT-rich plasma was expressed through a filter to the 1-L DnDP PVC storage container that is present in this pooling system, as previously described.  

**Pooling, dividing, processing, and storage of blood components**

From this point on, all connections between blood bag systems were made by use of either spike connections or Luer Lock connections, except in tubing intended to study the effect of a particular SCD. A schematic representation is shown in Fig. 2.

To study the effect of a sterile connection on the in vitro quality of RBCs, for each experiment, three WBC-reduced RBC units of identical ABO blood group were selected. In one of the ports of each of the RBC bags a spike-spike connector (764409, Codan, Deventer, the Netherlands; this is a spike connected to a second spike with a piece of tubing) was inserted. The second spike of this connector was inserted in the port of a 1500-mL pooling bag (Q4226, Fresenius HemoCare; this pooling bag had two spike ports and a piece of tubing with a Luer Lock connection) and the contents of each of the 3 units of RBCs was consecutively transferred into this pooling bag. The contents of the bag were mixed thoroughly, and with the Luer Lock connection, dispensed into three dual-bag systems (Q4226, Fresenius HemoCare). These dual-bag systems consisted of a 600-mL PVC RBC storage container with a piece of tubing with a Luer Lock (to connect the pooling bag) and another piece of tubing of 1000 mm that was connected to a transfer bag. In this 1000-mm tubing, a weld was made either with the T-SCD or with the CompoDock S2, while unwelded tubing served as reference. The total contents of the bag were transferred from one bag to the other by gravity through the (welded) 1000-mm tubing to study the effect of the weld on the blood component quality. (C) Transfer of pooled, divided PLT concentrates into a similar test system, but with a PLT storage container.
tubing for a total of 10 times, to have maximum effect of any deleterious consequence of the weld, when present. The transfer time (i.e., the complete transfer from the RBCs from one bag to the other) was timed with a stopwatch. The RBCs were sampled on Day 1 immediately after the above-described processing for in vitro measurement with a sample-site coupler and a needle and syringe. The RBCs were subsequently stored at 2 to 6°C and sampled on Days 28, 35, and 42. In total, units of 10 paired experiments were investigated. We required that free hemoglobin (Hb) level remained less than 5 mg per mL and that hemolysis remained less than 0.8 percent of the total amount of Hb in the unit, according to current guidelines.7

For PLT concentrates, the pooling and dividing were done as described above, but the PLT concentrates were dispensed in bag system Q4227 (Fresenius HemoCare), which consisted of a primary transfer bag with a piece of tubing with a Luer Lock, and a 1300-mL polyolefin PLT storage container that was connected to the primary container with a 1000-mm piece of tubing. Either a T-SCD weld or a CompoDock S2 weld was made in this tubing, and tubing with no weld served as reference. The PLT concentrates were transferred 11 times (to end up in the final storage container) from one bag to the other through the 1000-mm tubing by gravity. The PLT concentrates were stored in the polyolefin container, sampled on Day 1 immediately after processing with a sample-site coupler and a needle and syringe, and subsequently stored for 8 days at 20 to 24°C with gentle agitation in a temperature-controlled environment. Units of 10 paired experiments were studied. Swirl should remain present at the end of storage according to current guidelines.5 Further, we required that the CD62P expression of the units that were transferred through tubing with a weld should be no more than 20 percent higher than the CD62P expression of the reference unit.

In vitro measurements

Volumes were calculated from the net weight divided by the specific gravity of the blood component. Total blood counts and mean PLT volume (MPV) measurement were performed with a hematology analyzer (Sysmex K1000, TOA, Tokyo, Japan). WBCs in WBC-reduced components were counted with the BD Biosciences flow cytometric TOA, Tokyo, Japan). WBCs in WBC-reduced components were calculated well to our requirement of having a strength of greater than 20 N. Without a weld, the tensile strength averaged between 134 and 156 N, depending on the PD.
WBC-reduced RBCs

The pooling and dividing of the RBCs were successful, as can be concluded from the similar composition of the concentrates on Day 1 (as shown in Table 2). All WBC-reduced RBCs conformed to current guidelines with respect to RBC content and residual WBC content.

After the RBCs were transferred 10 times over welded tubing, subsequent storage did not reveal a significant effect on free Hb induced by any of the two methods of sterile connection, compared to the reference units on Day 28 (not shown), Day 35, and Day 42 (Table 2). Current guidelines specify that the amount of free Hb should be less than 5 mg per mL; all RBCs conformed on Day 28, and 9 of 10 conformed on Days 35 and 42 for all three study conditions. The unit with a higher hemolysis gave elevated values for all three conditions; therefore, it was concluded that it was not related to the method of sterile connection, but rather to a specific unit. The guidelines further specify that hemolysis rate should not exceed 0.8 percent of the total Hb content. This limit was exceeded for this particular RBC unit for all three methods of sterile connection from Day 28 onward; all others conformed.

The transfer time was 1 minute 21 seconds ± 5 seconds for the reference, 1 minute 19 seconds ± 5 seconds for tubing with a T-SCD weld, and 1 minute 25 seconds ± 4 seconds for tubing with a CompoDock S2 weld (p < 0.05 when compared to the T-SCD weld; others not significant). All RBCs were negative in the bacterial screening test at the end of the experiment.

WBC-reduced PLT concentrates

As shown in Table 3, pooling and dividing of the PLT concentrates were successfully performed among the three groups. All WBC-reduced PLT concentrates conformed to current requirements for composition and cellular content. Swirl remained present in all units until Day 8, except one unit in the reference group. This particular unit also showed a higher MPV on Day 8 compared to the other units, an observation that was again not reflected in the study units. Bacterial contamination was excluded, because all bacterial screenings were negative at the end of the experiment. CD62P expression remained below

<table>
<thead>
<tr>
<th>TABLE 2. Storage measures of WBC-reduced RBCs transferred through tubing with welds made by two different SCDs (n = 10 paired experiments)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Storage measure</td>
</tr>
<tr>
<td>------------------</td>
</tr>
<tr>
<td>Day 1</td>
</tr>
<tr>
<td>Volume (mL)</td>
</tr>
<tr>
<td>Hb (g)</td>
</tr>
<tr>
<td>WBCs (×10⁹)</td>
</tr>
<tr>
<td>PLTs (×10⁹)</td>
</tr>
<tr>
<td>Free Hb (mg/mL)</td>
</tr>
<tr>
<td>Hemolysis (%)</td>
</tr>
<tr>
<td>Day 35</td>
</tr>
<tr>
<td>Free Hb (mg/mL)</td>
</tr>
<tr>
<td>Hemolysis (%)</td>
</tr>
<tr>
<td>Day 42</td>
</tr>
<tr>
<td>Free Hb (mg/mL)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>TABLE 3. Storage measures of WBC-reduced PLT concentrates transferred through tubing with welds made by two different SCDs (n = 10 paired experiments)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Storage measure</td>
</tr>
<tr>
<td>------------------</td>
</tr>
<tr>
<td>Day 1</td>
</tr>
<tr>
<td>Volume (mL)</td>
</tr>
<tr>
<td>PLTs (×10⁹)</td>
</tr>
<tr>
<td>WBCs (×10⁹)</td>
</tr>
<tr>
<td>RBCs (×10⁹)</td>
</tr>
<tr>
<td>CD62P expression (%)</td>
</tr>
<tr>
<td>Swirl</td>
</tr>
<tr>
<td>MPV (fL)</td>
</tr>
<tr>
<td>Day 6</td>
</tr>
<tr>
<td>CD62P expression (%)</td>
</tr>
<tr>
<td>Swirl</td>
</tr>
<tr>
<td>MPV (fL)</td>
</tr>
<tr>
<td>Day 8</td>
</tr>
<tr>
<td>CD62P expression (%)</td>
</tr>
<tr>
<td>Swirl</td>
</tr>
<tr>
<td>MPV (fL)</td>
</tr>
</tbody>
</table>
20 percent on Day 6, except the aforementioned PLT concentrate in the reference group, which had an expression of 39.8 percent. The expression level increased to 45.7 percent on Day 8 for this particular unit, while all other units had CD62P expressions of on average 20 percent. These latter levels of expression reflect adequate storage conditions as used in our blood center. All welded units had a CD62P expression that conformed to our requirement of being less than 20 percent higher than the reference units. The transfer time for tubing with a CompoDock S2 weld, which was $49 \pm 4$ seconds, was significantly longer compared to tubing with a T-SCD weld ($44 \pm 2$ sec; $p < 0.01$) or no weld ($44 \pm 1$ sec; $p < 0.01$).

**DISCUSSION**

This study shows that welds made in tubing with the CompoDock S2 SCD have an oval shape, but this does not affect the physical properties of the tubing and also has no effect on the in vitro quality of RBCs and PLT concentrates.

The introduction of a weld results in a reduction of tensile strength by approximately 40 to 50 percent, but all welds made with the CompoDock S2 yielded tubing with a residual strength that exceeded the current requirement of more than 20 N by about a factor of 3. Also, all welds in this study were structurally intact, similar to earlier studies, indicating that the connection between pieces of tubing prevents entry of bacteria and thereby ensuring that an initially sterile blood component will not become bacterially contaminated. Although the term “sterile connection” is widely used, it is somewhat misleading, because it suggests that the connection is either made in a sterile environment or being sterilized. This is not true, and a more favorable term would be a “functionally closed connection” that assures the prevention of entry of bacteria in the blood component. Robert and Bégué have previously demonstrated that there is a good correlation between tensile strength and measurement of weld integrity and concluded that measurement of tensile strength could be a good indicator for correct functioning of the SCD. In other words: sterility should not be subject of testing, but rather it should be demonstrated whether or not a device can make a structurally intact connection.

The somewhat reduced inner diameter of welds made with the CompoDock S2 was reflected in the slightly longer transfer time from one container to the other compared to T-SCD welds or unwelded tubing. Despite this, the in vitro quality of both RBCs and PLT concentrates was not adversely affected by this. The RBCs showed hemolysis rates that conformed to the current guidelines, and hemolysis was similar in RBCs that were transferred over tubing with either the CompoDock S2 or the T-SCD, compared to unwelded tubing. Also for PLT concentrates, the activation marker CD62P was similar for the tubing welded with either the T-SCD or the CompoDock S2, compared to unwelded ones. In addition, no differences could be demonstrated for swirling scores and the MPV, two other commonly used quality measures.

In conclusion, the results of this study suggest that a functionally closed connection in a piece of tubing, made with either the CompoDock S2 or the T-SCD, has no deleterious effect on the quality of WBC-reduced RBCs and PLT concentrates.

**REFERENCES**

1. Yordy JR, Bettinger GE, Spencer DW. Sterility testing of Dupont’s sterile connection device for aseptically connecting plastic tubing [abstract]. Presented at the Annual Meeting of the American Society for Microbiology; 1983; New Orleans, LA.


