FIVE PARAMETER NAT SCREENING BY TAQSCREEN MPX WITH COBAS S201 IN SWITZERLAND: VALIDATION, IMPLEMENTATION AND FIRST EXPERIENCES

A. Glauser, K. Hardegger, J. Gottschalk and B.M. Frey
Blutspende Zürich, Rütistrasse 19, 8952 Schlieren, Switzerland
www.blutspendezurich.ch

BLUTSPENDE ZÜRICH

Background

The system cobas s201/TaqScreen MPX test (Roche Diagnostics) is a fully automated multiplex nucleic acid test for blood screening for hepatitis B virus (HBV) DNA, hepatitis C virus (HCV) RNA, HIV-1 RNA (groups M and O) and HIV-2 RNA. We implemented this test system at the ZURICH Blood Transfusion Service SRC for routine analysis in April 2008 based on a head to head comparison of alternative system. We show overall performance and suitability of the system for high throughput blood donor screening aiming to improve transfusion safety.

Methods

The cobas s201 platform (Roche Diagnostics) consists of automated pooling of blood donations using Hamilton Star pipettor, automated sample preparation using cobas Ampliprep instrument and automated amplification (real time PCR) and detection using cobas TaqMan analyzer. The test cobas TaqScreen MPX for use on this platform is a CE labeled in vitro diagnostics (IVD) for detection of HBV DNA, HCV, HIV-1 and HIV-2 RNA in a multiplex assay. In reactive samples, the individual reactive parameters have to be identified using alternative testing. Limit of detection (LOD) was determined by probit analysis. Routine samples were tested in pools of six. Resolution of positive pools is performed by single donation re-testing.

Results

Table 1:
95% LOD (IU/ml) of TaqScreen MPX by Cobas s201(according to Jarvis et al., Transfusion May 2008)

	HBV			HCV			HIV-1 Group M		
Site	95% LOD IU/ml	95% Lower CI	95% Upper CI	95% LOD IU/ml	95% Lower CI	95% Upper CI	95% LOD IU/ml	95% Lower CI	95% Upper CI
Valencia	1.7	1.2	4.1	10.0	5.7	27.7	45.0	23.2	137.0
Rome	3.1	1.6	75.6	14.4	8.5	34.6	53.2	30.1	132.1
Edinburgh	1.9	1.2	4.4	9.7	5.7	23.4	47.3	26.4	118.5
Madrid	3.6	2.3	7.7	5.7	3.5	12.7	37.0	22.1	90.0
Porto	4.6	2.6	11.3	12.1	6.9	29.9	80.2	42.0	226.0
Verona	3.9	2.4	8.7	13.2	7.5	33.0	72.7	39.0	196.0
All sites combined	**3.8**	3.0	5.2	**10.8**	8.4	14.4	**56.7**	43.0	79.2
RMS (Development)	**3.7**	3.3	4.4	**10.7**	7.0	21.7	**49.0**	42.3	58.1
Zürich	**2.3**	1.4	5.4	**7.4**	4.7	15.5	**31.1**	18.6	71.8

The 95% limit of detection (LOD) for HBV, HCV and HIV were 2.3 IU/ml, 7.4 IU/ml and 31.1 IU/ml (HIV-1), respectively. These results are comparable with those of other European testing sites using the same platform and are even better than indicated in the test manual by Roche Diagnostics (table 1).

Table 2:
Results of routine testing from April 2008 to August 2009

Donations	127`108	
Batches	1`493	
Pools total	21`214	
non reactive	21`155	99.72%
invalid	566	2.67%
reactive	33	0.16%
false reactive	26	0.12%
Single donations (SD)	127`108	
non reactive	127`073	99.97%
invalid	0	0%
reactive	33	0.03%
false reactive	2	0.002%

From April 2008 to August 2009 we screened almost 130'000 single donations (SD). Rates of non reactive, reactive, invalid and false reactive results for pools of 6 and for SD are shown in table 2.

Table 3:
Reactive results from April 2008 to August 2009

Reactive results	Positive donations including control samples	Positive donors
HBs Ag confirmed positive	17	11
HCV confirmed positive	9	6
HIV-1/2 confirmed positive	5	3
Isolated NAT positive	2	1

From 33 reactive donations (including control samples), 17 (11 donors) were confirmed positive for HBV (HBsAg positive by ELISA), 9 donations (6 donors) were positive for HCV (HCV Ab positive by ELISA) and 5 donations (3 donors) were confirmed positive for HIV-1(HIV Ab and p24 Ag positive by ELISA). 2 donations (from 1 donor) were "isolated NAT reactive" (ELISA screening negative for all parameters tested). This sample could be confirmed as a true HBV windows case (HBV PCR positive, negative for HBsAg) (table 3).

Table 4:
Problems occurring from April 2008 to August 2009

Problems	N	%
Batches invalid due to instruments failures	14	0.94
Pools invalid due to instruments failures	30	0.14
Batches invalid due to handling failures	7	0.47
Pools invalid due to handling failures	2	0.009
Batches invalid due to negative positive control	10	0.67
Batches invalid due to invalid controls	3	0.20
Pools invalid (IC negative)	1	0.005
Pools false reactive	26	0.12

Problems are shown in table 4. All problems could be solved in time, meaning that there was no substantial delay in releasing blood products. Nevertheless, a backup system is advisable to ensure prompt release of blood products in case of instrument failures.
In June 2008, positive controls for HCV frequently resulted non reactive, resulting in invalid batches. This problem could clearly be traced back to reagent problems with one lot of HCV controls. Another problem are false reactive pools (reactive pools containing no reactive SD). This problem was peculiarly frequent in January 2009 with one confined lot and it could therefore also be due to reagent problems.

Conclusion

LOD of the cobas s201 TaqScreen MPX fulfills well the national and international requirements for NAT screening and proves to be most suitable for high throughput blood donor analysis. Operational consistency is excellent. The first results indicate already a net safety benefit on provision of transfusion products by application of this screening approach for Swiss blood donors.

42. Jahreskongress der Deutschen Gesellschaft für Transfusionsmedizin und Immunhämaologie (DGTI), 15.-18. September 2009, Rostock