QuikRead ® Strep A test

Manufactured by

Orion Diagnostica Oy

Report from an evaluation under standardised conditions
in hospital laboratory

Organised by SKUP

SKUP i Danmark, Afdeling KKA, Odense Universitetshospital, 5000 Odense C, tlf. 65412865, www.SKUP.dk

The organisation of SKUP

SKUP Scandinavian evaluation of laboratory equipment for primary health care, SKUP, is a co-operative commitment of NOKLUS[1] in Norway, the "Afdeling for Biokemi, Farmakologi og Genetik"[2] in Odense, Denmark and EQUALIS[3] in Sweden. SKUP was established in 1997 at the initiative of laboratory medicine professionals in the three countries. SKUP is led by a Scandinavian steering committee and the secretariat is located at NOKLUS Centre in Bergen, Norway.

The goal of SKUP is to produce reliable, objective and independent information about the analytical quality and user-friendliness of laboratory equipment for primary healthcare. This information is generated by organising *SKUP evaluations*.

SKUP offers manufacturers and suppliers evaluations of equipment for primary healthcare and also of devices for self-monitoring of blood glucose. As long as the equipment is not launched onto the Scandinavian market, it is possible to have a confidential pre-marketing evaluation. The company requesting the evaluation pays the actual testing costs and in return, receives an impartial evaluation.

There are *general guidelines* for all SKUP evaluations and for each evaluation a specific *SKUP protocol* is worked out in co-operation with the manufacturer or their representative. SKUP signs *contracts* both with the requesting company and with the evaluating laboratories. A *complete evaluation* requires both one part performed by experienced laboratory personnel and one part performed by the intended users.

Each evaluation is presented in a *SKUP report* to which a unique *report code* is assigned. The code is composed of the acronym SKUP, the year and a serial number. A report code, followed by an asterisk (*), indicates a special evaluation, not complete according to the guidelines, e.g. the part performed by the intended users was not included in the protocol. If a supplier uses the SKUP name in marketing, they have to refer to www.skup.nu and to the report code in question. For this purpose the company can use a logotype from SKUP containing the report code. SKUP reports are published at www.skup.nu and www.SKUP.dk and summaries are distributed to physicians' offices, councils for laboratory medicine, laboratory instructors and healthcare authorities.

[1] NOKLUS (Norwegian Quality Improvement of Primary Care Laboratories) is an organisation attached to "Seksjon for Allmennmedisin" (Section for General Medicine) at the University of Bergen.

[2] "The SKUP-division in Denmark" is an organisation created through an agreement between the national "Fagligt Udvalg vedrørende Almen Praksis" (Professional Committee for General Practice) and the "Afdeling for Biokemi, Farmakologi og Genetik" (Department of Clinical Chemistry) at the University Hospital in Odense. "Fagligt Udvalg vedrørende Almen Praksis" is a joint committee for PLO, "Praktiserende Lægers Organisation" (General Practitioners Organisation) and "Sygesikringens Forhandlingsudvalg" (Committee for Negotiations within the General Health Insurance System).

[3] EQUALIS AB (External quality assurance in laboratory medicine in Sweden) is a limited company owned by "Sveriges Kommuner och Landsting" (Swedish Association of Local Authorities and Regions), "Svenska Läkarsällskapet" (Swedish Society of Medicine) and IBL (Swedish Institute of Biomedical Laboratory Science).

Testing of QuikRead® Strep A test

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SUMMARY

Orion Diagnostica A/S ordered a SKUP laboratory testing of QuikRead Strep A in Autumn 2006. In Scandinavia, there is no consensus on diagnosis and treatment of β-haemolytic streptococci.

Principle of the test
QuikRead Strep A test is an Immunoturbidimetric test based on microparticles coated with rabbit anti Strep A antibodies. Strep A antigen in the sample reacts with the microparticles. The turbidity of the solution change hereby. The instrument QuikRead measures the change in turbidity.

To qualify for an overall good assessment in a SKUP evaluation, the measuring system must have both satisfactory analytical quality and satisfactory user-friendliness. For analytical quality the following are evaluated: 1a) Specificity, defined by measuring other streptococci (True negative)/ (false positive + true negative). 1b) Specificity, defined by (True negative)/ (false positive + true negative). 1c) the concentration at which 50% of the tests is positive. 2) Validation of practical use: intra-person and inter-person variation. 3) Percent of tests not valid. 4) Robustness

User friendliness, parameters evaluated: user manual, time, quality control, operation of the test. The results of the evaluation are indicated as follows: not satisfactory = 0 point, less satisfactory = 1, satisfactory = 2 and very satisfactory = 3 points. Each of the 5 areas has to achieve ≥ 2 points.

Method
To decide the detection limits of QuikRead Strep A test we used serial dilutions of a known amount of *S. pyogenes* in seven different concentrations, one mix of four other streptococci-strains and a positive and a negative control.

Results, *analytical quality.*

1a) Specificity: of 20 tests 20 was negative.
1b) 120 of 120 tests with low concentrations of β-haemolytic streptococci/mL.
(from 0 to ≤ $2{,}1 \times 10^5$) were negative.
40 of 40 test of a concentration ≥ $2{,}1 \times 10^6$ β-hemolytic streptococci/mL were positive.
1c) The concentration at which 50 % of the test was positive was between $5{,}7 \times 10^5$ and $2{,}1 \times 10^6$ β-hemolytic streptococci/mL.
2) Instrumental disagreement of reading: none.
3) Invalid tests: about 2,1 %.
4) The results were repeatable.

Results*, the user friendliness* was rated 'satisfactory' for the manual, the time factors and control system. The operation was also rated 'satisfactory' even if the bottle of reagent 2 was dripping.

Conclusion
QuikRead® Strep A test does fulfil the criteria for good performing in this analysis. The user friendliness was rated as 'satisfactory'. The evaluators considered the operation of the system to be handy, especially for those who are familiar with the instrument from CRP measurements. The advantage of the instrument is that it shows the Strep A results as a positive or negative result (+ or -) and as such eliminates the subjectivity in reading the results.

The test became positive between 5,7 x 10^5 and 2,1 x 10^6 CFU/mL. The waste due to errors was 2,1%.

We do not know how QuikRead® Strep A will perform under less standardised conditions in the hands of general practitioners.

2. PLANNING of QuikRead Strep A LABORATORY TESTING

In August 2006 **Orion** Diagnostica A/S ordered a laboratory test as close as possible to the Strep A protocol performed previously. The protocol version 1.7 was changed in a few ways and was approved by the supplier: see attachment A.

Department of Clinical Microbiology (KMA), Odense University Hospital (OUH), and Professor Hans Jørn Kolmos is the comparison laboratory for the Strep A marking in Denmark.
In Scandinavia, there is no consensus of diagnosis and treatment of β-haemolytic streptococci. Denmark is homogenous compared to Norway and Sweden, because Denmark has used the methods of Statens Serum Institute (SSI) as a golden standard for diagnosis of Strep A.

This is the 12th test made by SKUP on ordinal scale. (Ordinal scale: results of qualitative and semi-qualitative measurements is often given as – or + (and some time as ++, +++ and ++++), or more correct as 0 and 1 (2,3 and 4). Normally the interpretation of the result is 'presence' or 'no presence' of the component. For this type of measurements the ordinal scale is used if one will demonstrate when 100% of the tests are positive and when 100% of the tests are negative. For all components there will be areas where some percent of the concentrations are positive and some percent are negative. These areas can not be used in quality assurance of the user; however concentrations in this area can characterise the method.

It is a demand from Danish General Practitioners that analytical quality and user friendliness are weighted equally.

The goal of the laboratory tests is to investigate the analytical performance and the user friendliness under standardised and optimal conditions. Tests with false positive or false negative results, a high variation (intra- and inter-personal or between instruments in the same concentration) or analysis too difficult can be sorted out at this point.

The laboratory testing is done in KMA and Department of Clinical Chemistry, (BFG), Odense University Hospital. Esther Jensen is responsible for the testing. The work is done by the laboratory technicians Nina Brøgger and Anette Knudsen, KMA and medical doctors Hanne Holt (KMA) and Esther Jensen (BFG)
Esther Jensen and Hanne Holt have written the protocol. The protocol is approved by SKUP and by the supplier, who signs a contract with SKUP. The supplier gives SKUP the disposal of the equipment necessary for testing. Calculating of data is done by Esther Jensen, SKUP, who also writes the

report of the testing. The report is approved by Hanne Holt, KMA. Then it is sent to the supplier and SKUP. Both get the opportunity to discuss and comment the report.
The report is published by SKUP, if the test is sold in Scandinavia.
A good laboratory test is expected to be followed by an investigation in General Practice under "real" conditions. The results in General Practice are not expected to be better than the laboratory test.

25th of September the supplier came to the laboratory to teach the test persons how to perform the test. None of the test persons were familiar with the instrument from daily day use. 10 negative and 10 positive samples were performed in the 3 instruments that were going to be used in the evaluation.

The meeting took 1-2 hours, mainly due to the testing of samples.

The testing was performed in September/October 2006.

3. CONTACT ADDRESSES

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4. **METHOD**

Qualitative detection of group A streptococcal antigen. The streptococci can be dead or alive.

Principle of the test.

QuikRead Strep A is an Immunoturbidimetric test based on microparticles coated with rabbit anti Strep A antibodies. Strep A antigen in the sample reacts with the microparticles. The turbidity of the solution change hereby. QuikRead measure the change in turbidity.

To perform the test, a throat swab specimen is collected (Dacron). The swab is placed in the empty cuvette. The Extraction Reagents 1 and 2 is added. The red colour of Extraction Reagent 2 change to yellow/orange indicating the beginning of the extraction. The swab is rotated in the solution for 30 seconds and then left in the solution for at least another 90 seconds but no longer than 15 minutes. Reaction Buffer (0,8 mL) is added to the tube on top of the swab and the two reagents using the QuikRead Dispenser. The swab is rotated vigorously and pressed against the inner wall of the cuvette to release all liquid. Then it is removed. The solution turns red again due to neutralisation.

The cuvette is closed tightly with a Strep A Reagent Cap without pressing the pink coloured inner part of the reagent cap down into the solution.

It is important not to touch the optical part of the cuvette.

The solution is stable for at least four hours but was in this evaluation measured within 15 minutes.

If a test is positive, ”positiv Strep A” is written on the display (in the Danish version).

5. PRODUCT INFORMATION

Reagents and materials supplied

Traceability of calibrators

The QuikRead® Strep A test is standardised using an antigen prepared from ATCC (American Type Culture Collection) strain 19615.

QuikRead Strep A test, Cat. No. 06152

Content: 50 tests

Strep A Reagent Caps	2x25 pcs.
Extraction Reagent 1	6 mL
Extraction Reagent 2	6 mL
Reaction Buffer	100 mL
Positive control	1 mL
Negative control	Reaction Buffer
Magnetic card	1 pc.
Cuvettes	50 pcs.
QuikRead Strep A swabs	50 individually packaged sterile Dacron-tipped swabs lot 8778. Expiration date 2010-07.
Package Insert	1 pc.

The Reagents contain sodium azide.

Extraction Reagent 1 contains acetic acid and Extraction Reagent 2 contains sodium nitrite.

See 'Warnings and precautions'.

Lot no. FO-7. Expiration date 2008-02-01

Producer: **Orion** Diagnostica Oy, PB/PL, FI-02101 Espoo, Finland, Telephone +358 10 42 61, Telefax +358 10 429 2794, www.oriondiagnostica.fi

0.8 mL Dispenser Cat. No. 06075

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Period of Investigation: September/October 2006

Writing of Report: October 2006

7. DEMANDS TO ANALYTICAL QUALITY AND TO USER FRIENDLINESS

No international (Golden) Standard for Strep A-test does exist for laboratory testing or test in General Practice.

To qualify for an overall good assessment in a SKUP evaluation, the measuring system must have both satisfactory analytical quality and satisfactory user-friendliness. Each of the areas within Analytical quality and user friendliness has to achieve 2 points.
Each area is subdivided and each subdivision has the following possible results.

0	Point	unsatisfactory
1	Point	less satisfactory
2	Points	satisfactory

Analytical quality

Parameters evaluated:

1a) Specificity, defined by measuring other streptococci (True negative)/(false positive + true negative).

1b) Specificity, defined as (True negative)/(false positive + true negative).

1c) The concentration at which 50% of the tests are positive.

2) Validation of practical use: intra-instrument and inter-instrument variation: 11 concentrations are read 20 times each by three instruments.

3) Percent of tests not valid

4) Robustness: Does the test turn positive at the expected time, as told in the insert?

Cross-reactivity tested in report: Analytical quality 1a

(Information from **Orion** Diagnostica below)

QuikRead Strep A does not cross-react with any of these organisms:

Streptococcus B	ATCC 12386
Streptococcus C	ATCC 12388
Streptococcus F	ATCC 12393
Streptococcus G	ATCC 12394
Staphylococcus aureus (Cowan)	ATCC 12598

Candida albicans	ATCC 14053
Neisseria sicca	ATCC 29259
Pseudomonas aeruginosa	ATCC 27853
Haemophilus influenzae type B	ATCC 9795
Streptococcus pneumoniae	ATCC 6303
Branhamella catarrhalis	Clinical strain

Prozone effect (Information from **Orion** Diagnostica)

No prozone effect was evident when samples with high concentrations of *Streptococcus pyogenes* antigen were tested.

Other interfering factors (Information from **Orion** Diagnostica)

If the QuikRead Instrument displays the message "Unstable sample", the sample may contain substances that interfere with the measurement. Take the cuvette out and replace it in the measurement well. If the instrument repeatedly displays "unstable sample", discard the cuvette and take a new sample. Samples containing interfering substances cannot be tested by the QuikRead Strep A method.

User friendliness. Parameters evaluated

- manual /insert
- time factors
- possibilities for quality control
- operation of the test

8. QUALITY CONTROL (Internal, external)

Internal quality assurance

1) the test is only valid if the colour of the mixtures are correct, see attachment B
2) The instrument is able to detect several errors
3) The procedure for the positive control sample was different from the procedure for the test samples and the negative control sample.

External quality assurance

1) Streptococci from agar
2) Streptococci from bouillon
3) Control materials from other manufacturers

It is recommended that

- A positiv control sample is measured when opening the box
- New users begin with a positive and a negative controlsample
- Participation in local quality assurances

Positive and Negative Quality Control material from Orion was part of the testing.

9. TEST PROCEDURES

(under standardised and optimal conditions in the laboratory)

The Strep A test samples were produced by a doctor and two laboratory technicians from KMA, OUH.

Material

For the serial dilutions the reference strain *S. pyogenes* (ATCC 19615) was used. For the mixture of different streptococci the following four rutine isolates from throat specimens were used: haemolytic streptococcus group C, haemolytic streptococcus group F, haemolytic streptococcus group G and an alfa-haemolytic streptococcus.

The grouping was done by Streptococcal Grouping Kit, Oxoid.

For dilution, a phosphate buffered saline (PBS) from SSI, art nr. 3892, was used.

5 % blood agar plates (Danish Blood Agar, Statens Serum Institute (SSI), with 5% horse blood), art nr. 677

SSI transport medium (Stuarts) art. nr. 944

Serumbouillon (bovine bouillon with defibrinogenated blood and horse serum, SSI) art nr. 1040

Method

1. Preparation of tests used in the analysis.

A tube of 10 mL broth is inoculated with *S. pyogenes* and incubated 18-24 timer at 35 °C. This culture is used for making a 10-fold dilution series in seven different concentrations: 10^6 colony forming units (cfu)/mL – 10^0 (cfu)/mL. The number of bacteria in the broth is determined by inoculating 0.1 mL of each dilution on two 5 % blood agar plates (in duplicates). After 18-24 hours incubation in 5 % CO_2, plates with 30-300 cfu are used for colony counting and calculating of the average streptococci concentration.

In the same way dilutions are made of β-haemolytic streptococci group C, G and F and α-haemolytic streptococci. The concentration of 10^7 cfu/mL is used for the preparation of a mixture with equal amounts of the 4 streptococci strains. From each of the seven concentrations of *S. pyogenes*, from the mix of streptococci and from the 100 % PBS, in total nine different concentrations, 20 samples is prepared, all together 9 x 20 = 180 tests for the investigation of the Strep A test. The positive and negative control from the box was each tested 20 times. According to the insert, the

test was positive at 5×10^5 cfu/mL. The concentration of the positive control is not mentioned in the insert. The negative control is group C streptococci.

2. Investigation of stability in SSI transport medium (Stuarts)

Due to future investigations in General Practice (to compare results from general practice with results from the department of microbiology) we investigated changes in the number of *S. pyogenes* after transportation in Stuarts transport medium in the following way:

From dilution series of *S. pyogenes* in the concentrations 10^6 cfu/mL - 10^0 cfu/mL 5 % blood agar plates are spread with charcoal swabs. The spreading is done 5 times from each dilution, n = 5. The same procedure is repeated after the charcoal swap have been stuck in the SSI´s transport medium for a few seconds (n = 5) and after 24 hours at room temperature (n=5). The concentration of *S. pyogenes* is calculated from agars with 30 to 300 cfu/mL, as described previously

The Testing

The day before the testing 10 samples in the concentration 10^5 CFU/mL and a negative test (10^0 CFU/mL) was tested to assure that the instruments and the procedure was OK. After the overnight freezing one positive and one negative sample was tested. If 10^5 CFU/mL is negative 10^0 CFU/mL is exchanged with 10^7 CFU/mL in the evaluation.
Most Strep A tests are positive between 1 x 10^5 and 1 x 10^6 CFU/mL.

The 9 different strain solutions, each divided in 20 tubes plus the negative and positive control were produced and analysed randomly. The 11 different solutions were: one pure diluent, 7 dilutions of a known amount of *S. pyogene*s, one mix of four other streptococci (β-haemolytic streptococci group C, G and F and α-haemolytic streptococci), one positive and one negative control.

The 11 Strep A concentrations were analysed 20 times each in three instruments by doctors or laboratory technicians from the departments KMA and BFG, OUH. The samples were prepared blindly by the microbiologists and analysed by the test persons from the BFG department. However the procedure for the positive control material was performed different than the procedure for the samples and the negative control material.

All readings were done according to the manual and attachment B. All tubes contain 90 % to 100 % PBS (saltdiluent).
At the time for testing the temperature in the room was 21° C.

Before the evaluation the following measurements were determined three times each: Weight of two drops from the four bottles of positive controls and from the bottles of Reagent 1 and Reagent 2. Weight of the amount of diluent absorbed by the Dacron swabs (12 swabs tested). Volume was measured for 24 drops of each.

10. RESULTS

One person measured weight and volume of the reagents belonging to 4 different test packages (Table 1).

Table 1 Variation in the size of drops and Dacron swabs used.

		2 drops of				
		Reagent 1	Reagent 2	Positive control	Dacron swabs	
		g	g	g	g	
bottle 1	1	0,08400	0,10342	0,07971	0,13408	
	2	0,08024	0,10659	0,07843	0,12075	
	3	0,08005	0,10114	0,07840	0,11165	
bottle 2	4	0,07828	0,10105	0,08210	0,12501	
	5	0,07669	0,10272	0,07966	0,11371	
	6	0,08099	0,09945	0,08280	0,12163	
bottle 3	7	0,07738	0,10127	0,07868	0,11782	
	8	0,07882	0,10297	0,07981	0,12116	
	9	0,07733	0,10260	0,07793	0,11775	
bottle 4	10	0,07878	0,09956	0,08302	0,12236	
	11	0,07855	0,10917	0,08245	0,12509	
	12	0,07369	0,10271	0,07817	0,12430	
2 drops,	mean	0,07873	0,10272	0,08010	0,12128	gram
Weight,	mean, 1 drop	0,03937	0,05136	0,04005		gram
Volume,	mean, 1 drop	38,5	44,6	40,0		µL

Three persons read randomly 220 samples of different concentrations of Strep A in three instruments.

The data are seen below in table 2.

Table 2 Random distribution of instrument used for analysing

Concentration, CFU/mL	instrument 1	instrument 2	instrument 3
PBS	6	4	10
21	10	5	5
$2{,}1 \times 10^2$	5	8	7
$2{,}1 \times 10^3$	8	5	7
$2{,}1 \times 10^4$	4	5	11
$2{,}1 \times 10^5$	7	4	9
$2{,}1 \times 10^6$	7	7	6
$2{,}1 \times 10^7$	4	11	5
buffer = negative control	10	6	4
Positive control#	6	12	2
Mix of streptococci	10	6	4
percentage of errors			

The procedure for the positive control sample was different from the procedure for the samples and the negative control sample.

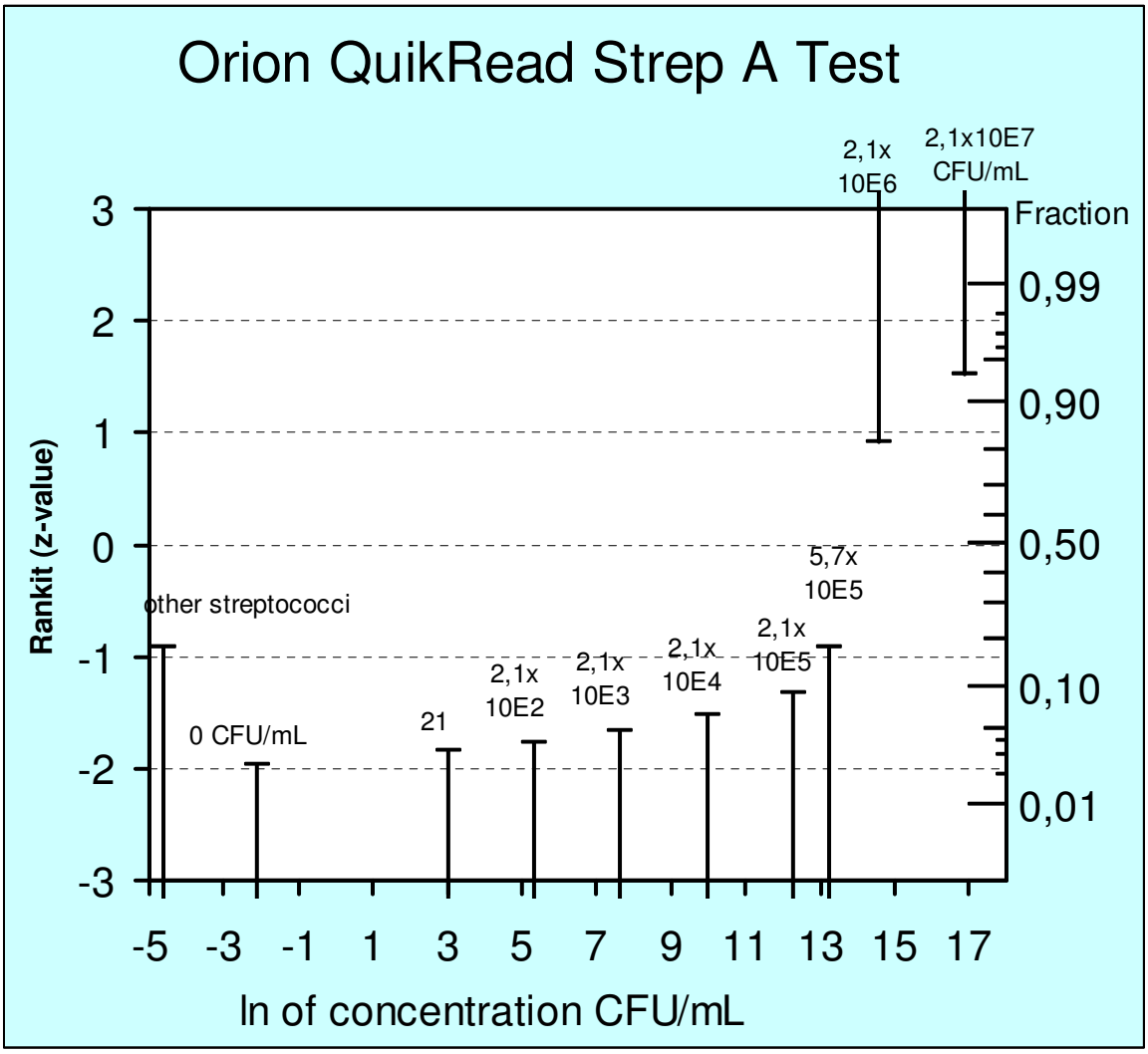
Table 3 **Results**

	Results	
Concentration, CFU/mL	positive	negative
PBS	0	20
21	0	20
$2,1 \times 10^2$	0	20
$2,1 \times 10^3$	0	20
$2,1 \times 10^4$	0	20
$2,1 \times 10^5$	0	20
$2,1 \times 10^6$	20	0
$2,1 \times 10^7$	20	0
buffer = negative control	0	20
Positive control#	18	0
Mix of streptococci	0	20

two samples were negative due to wrong procedure.

Two 'positive control' samples were negative. The procedure for the positive control was different from the procedure for the samples and the negative control sample. It was proven by difference in volume, that the two positive control samples that were negative contained 185 µl less than the other positive controls ~ the amount of two drops of the positive control, that were not dripped into the cuvette.

The results are depicted in figure 1



In Figure 1 the fractions of the positive results of a dilution series of Strep A concentrations are shown in a Rankit-plot (Rankit is a linearization of the Gaussian distribution, where z is the distance from the average in standard deviations). The corresponding fractions is indicated on the right Y-axis and the abscissa in natural logarithms (ln = log e). For each fraction the 95% confidence interval is plotted as well as the fraction 0.1, 0.5 and 0.9. The Strep A concentration is given on top of the confidence interval.

The Figure shows that QuikRead® Strep A test has all concentrations $\geq 2{,}1 \times 10^6$ β-haemolytic streptococci/mL positive and all concentrations of $\leq 5{,}7 \times 10^5$ β-haemolytic streptococci/mL negative.

Error messages

In total 6. Of the 6 errors

- 1 was caused by the lit falling too early
- 0 were exceeding the procedure time
- 5 indicated error in adding the reagent

Interference and error messages

Fingerprints before measuring of 'blank'

6 samples ($2{,}1 \times 10^4$ β-hemolytic streptococci/mL) was measured before and after fingerprints from a person that had used hand lotion were put on the low part of the tubes used for measuring. When the fingerprints were placed before the measuring of blank, it did not interfere with the results. In 3 cases the instrument came with error: 'Error in adding the reagent. Test again'

Fingerprints between the measuring of 'blank' and 'result'

6 samples ($2{,}1 \times 10^4$ hemolytic streptococci/mL) was measured before and after fingerprints from a person that had used hand lotion were put on the low part of the tubes used for measuring. When the fingerprints were placed after the measuring of 'blank' 2 of 6 results turned positive.

In four samples coal swabs were used. The error message: 'blank too high, test again' appeared.

11. EVALUATION OF ANALYTICAL QUALITY

Results, *analytical quality.*

1a) Specificity: of 20 tests 20 was negative.

1b) 120 of 120 tests with low concentrations of β-haemolytic streptococci/mL. (from 0 to $\leq 2{,}1 \times 10^5$) were negative.
40 of 40 test of a concentration $\geq 2{,}1 \times 10^6$ hemolytic streptococci/mL were positive.

1c) The concentration at which 50 % of the test was positive was between $5{,}7 \times 10^5$ and $2{,}1 \times 10^6$ haemolytic streptococci/mL.

2) Instrumental disagreement of reading: none.

3) Invalid tests: about 2 %.

4) The results were repeatable.

False positives

None

False negatives

None. Two of the positive control samples were negative. However, the reason was proven to be an error made by the test person.

User-friendliness

At the end of the evaluation period, each user filled in a questionnaire about the user-friendliness of the QuikRead Strep A. The questionnaire and expressed opinions are presented in Tables 5 — 8 below. The first column explains the evaluated properties. The second to fourth column show which alternative the evaluators could chose from. The cells that show the average or overall ratings from all evaluating sites are marked by making the frames thicker and the text bold.

Table 5. Assessment of the information in the manual / insert

Information in manual / insert about:	0 point	1 point	2 point
Well-presented, easy-to-grasp	Un-satisfactory	Less satisfactory	Satisfactory
Specimen collection	Un-satisfactory	Less satisfactory	Satisfactory
Preparations / pre-analytic/test procedure	Un-satisfactory	Less satisfactory	Satisfactory
Measurement / reading	Un-satisfactory	Less satisfactory	Satisfactory
Measurement principle	Un-satisfactory	Less satisfactory	Satisfactory
Sources of error	Un-satisfactory	Less satisfactory	Satisfactory
Fault-tracing/troubleshooting	Un-satisfactory	Less satisfactory	Satisfactory
Index	Un-satisfactory	Less satisfactory	Satisfactory
Readability / clarity of presentation	Un-satisfactory	Less satisfactory	Satisfactory
Manual in Danish, Norwegian, Swedish, English	Un-satisfactory	Less satisfactory	Danish Swedish Norwegian English
Rating for information in manual			Satisfactory

No additional notes regarding the manual were provided by the evaluators.

Table 6. Assessment of the time factors

Time factors	0 point	1 point	2 point
Preparations / pre-analytical time	>10 minutes	6 to 10 minutes	3 to 5 minutes.
Analytic time	>20 minutes	10 to 20 minutes	5 to 10 minutes
Demands to training	days	> 2 hours	½-2 hours
Stability of test, unopened, (no/package)	≤ 3 months	3 — 5 months	6 — 12 months
Storage conditions of tests, unopened	-20^{0}C	2 — 8^{0}C	15 — 30^{0}C
Rating of time factors			Satisfactory

No additional notes regarding the time factors were provided by the evaluators.

Table 7. Assessment of the quality control possibilities

Quality Control	0 point	1 point	2 point
Internal quality control	Un-satisfactory	Less satisfactory	Satisfactory
External quality control	Un-satisfactory	Less satisfactory	Satisfactory
Stability of quality control material	≤ 3 months	3 — 5 months	> 12 months
Storage conditions of control material	-20^{0}C	2 — 8^{0}C	2 — 30^{0}C
Interpretation of the Quality Control	Un-satisfactory	Less satisfactory	Satisfactory *
Rating of quality control			Satisfactory

Remarks: The procedure for the positive control sample is not the same as the procedure for the test samples and the negative control sample.

Table 8. Assessment of the operation facility

Operation facility	0 point	1 point	2 point
To prepare the test / instrument	Un-satisfactory	Less satisfactory	Satisfactory
To prepare the sample	Un-satisfactory	Less satisfactory	Satisfactory
Application of specimen	Un-satisfactory	Less satisfactory	Satisfactory
Specimen volumen	Un-satisfactory	Less satisfactory	Satisfactory
Number of procedure step	Un-satisfactory	Less satisfactory	Satisfactory
Interpretation of the test	Very difficult	Difficult	Easy
Sources of errors	Un-satisfactory	Less satisfactory	Satisfactory
Cleaning/maintenance	Un-satisfactory	Less satisfactory	Satisfactory
Hygiene, when using the test	Un-satisfactory	Less satisfactory *	Satisfactory
Environmental requirements, waste handling	Poison	Special arrangement **	Biohazard / Daily renovation
Educational requirements	Lab technician	Course	GP personal
Size and weight of package	Un-satisfactory	Less satisfactory	Satisfactory
Rating of operation			Satisfactory

* It was not possible to handle the bottle of Reagent 2 without it was dripping. The manufacturer has now changed to another bottle.
** It is recommended due to the risk of infection, that all used material should be autoclaved (1 hour at 121°C) before disposal.

Remarks: It is favourable that the individuality in reading the results is eliminated by the instrumental reading 'positive' or 'negative'.

Assessment of the user-friendliness

The overall opinion about the user-friendliness of QuikRead® Strep A test was rated 'satisfactory' for the manual, the time factors and control system. The operation was also rated 'satisfactory' even

if the bottle of reagent 2 was dripping. (The bottle was replaced after the testing; the new bottle is not evaluated). The evaluators considered the operation of the system to be handy, especially for those who are familiar with the instrument from CRP measurements. It was considered favourable that the individuality in reading the results is eliminated by the instrumental reading ‘positive’ or ‘negative’.

12. CONCLUSION

QuikRead® Strep A test does fulfil the criteria for good performing in this analysis. The user friendliness was rated as 'satisfactory'. The advantage of the instrument is that it shows the Strep A results as a positive or negative result (+ or -) and as such eliminates the subjectivity in reading the results.

The test became positive between 5,7 x 10^5 and 2,1 x 10^6 CFU/mL. The waste due to errors was 2,1%.

We do not know how QuikRead® Strep A will perform under less standardised conditions in the hands of general practitioners.

13. REFERENCES

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2) Diagnosis of Group A Streptococcal Pharyngotonsillitis in generel Practice with Five Antigen Detection Test Kits and a rapid Kit for C-Reactive Protein. Steen Hofmann og Klaus Witt. Poster c22 ASM 99th General Meeting1999 (ingen artikel, n=2078, GP's=230)

3) Diagnostik af halsbetændelse. En multipraksisundersøgelse af tre antigendetektionssæt til påvisning af gruppe A-streptokokker i svælgpodninger. Jørgen Steen Andersen, Niels Jerne Borrild og Steen Hoffmann. Ugeskrift for Læger 1994; 156:46, 6869-6872

4) Detection of group A streptococcal antigen from throat swabs with five diagnostic kits in general practice. Hoffmann S. Streptococcus Department, Staten's Serum Institut, Copenhagen, Denmark. Diagn Microbiol Infect Dis. 1990 May-Jun;13(3):209-15.

5) Detection of group A streptococcal antigen from throat swabs by use of a latex agglutination test kit in general practice. Hoffmann S, Henrichsen J. Acta Pathol Microbiol Immunol Scand [B]. 1987 Apr;95(2):89-94

6) Inserts in Danish, Norwegian, Swedish and English.

7) SKUP report nr 24. OSOM Strep A test

Attachment A

QuikRead® Strep A afprøvning

Ændringer i forhold til protokol:
Afprøvningen sker instrumentelt på QuikRead. Testen hedder QuikRead® Strep A.

Orion Diagnostica fremskaffer instrumenter, reagenser og manual til afprøvning i september/oktober måned.

Oplæring finder sted i Klinisk Biokemisk Afdeling den 25. september kl. 10.00.

Vi modtager 3 instrumenter. Instrumenterne er udstyret med detektor, så man kan følge analyseprocessen senere, hvis det skulle blive nødvendigt.

Med en analyse tid på 5 minutter (~ negativ prøve) vil 11 x 20 + 11 x 2 = 242 test vare 1210 minutter ~ 20 timer.
Prøverne analyseres uden at instrumentaflæserne kender koncentrationen af hæmolytiske streptococcer Gruppe A i prøven. Alle koncentrationer analyseres 20 gange.

Attachment B

QuikRead Strep A- afprøvning

Procedure for patientprøve

1. Tag en halspodepind som er leveret med kittet.
2. Dyp podepinden i patientprøven.
3. Sæt podepinden ned i cuvetten.
4. Tilsæt 2 dråber farveløs Extraction Reagent 1 til cuvetten med prøven.
5. Tilsæt 2 dråber rød Extraction Reagent 2. Opløsningen bliver gul/gul-orange.
6. Roter podepinden i opløsningen i 30 sek. og lad den stå i opløsningen i yderligere mindst 90 sek., men dog ikke længere end 15 min.
7. Dispenser 0,8 mL Reaction Buffer til cuvetten.
8. Roter podepinden kraftigt, løft den op af væsken og pres den mod den inderste væg af cuvetten for at frigøre al væske. Tag den op af cuvetten, vær omhyggelig ikke at spilde noget opløsning uden på cuvetten. Check at opløsningen er rød. Stop hvis opløsningen er gul (sur).
9. Luk cuvetten tæt med et strep A reagens låg. Pres ikke den inderste pinkfarvede del af låget ned. Berør ikke overfladen af den nederste del af cuvetten (den optiske del). Opløsningen er stabil i mindst 4 timer.
10. Sæt cuvetten ned i aflæsningsbrønden på instrumentet.
11. Instrumentet måler blindprøven. Dette vil max. tage 40 sek. Rør ikke cuvetten i aflæsningsbrønden under målingen
12. Tryk den inderste pinkfarvede del af låget ned. Dette vil frigøre det tørrede Strep A reagens ned i cuvetten.
13. Løft cuvetten op fra aflæsningsbrønden og bland hurtigt ved at ryste cuvetten kraftigt frem og tilbage. Opløsningen bliver mælkerødlig. Ryst mens apparatet blinker (ca. 5 sek).
14. Indsæt cuvetten tilbage i aflæsningsbrønden på instrumentet når apparatet bipper.
15. Resultatet aflæses.
16. Resultatet vises som Positiv Strep A eller Negativ Strep A. (Endepunktsmåling = 180 sek.)

Procedure for negativ kontrol

1. Tilsæt buffer til et beta-pippeglas.
2. Dyb en Strep A podepind ned i bufferen.
3. Udfør kontrolprøven som almindelig prøve, pkt. 3-16 ovenfor.

Procedure for Positiv Kontrol

1. Dryp 2 dråber af den positive kontrol som er leveret med kittet i en cuvette.
2. Dispenser 0,8 mL Reaction Buffer til cuvetten.
3. Luk cuvetten tæt med et strep A reagens låg. Pres ikke den inderste del af låget ned. Berør ikke overfladen af den nederste del af cuvetten (den optiske del). Mix opløsningen ved at ryste cuvetten let. Opløsningen er stabil i mindst 4 timer.
4. Udfør pkt. 10-16 'som almindelig prøve' ovenfor.

Attachment C

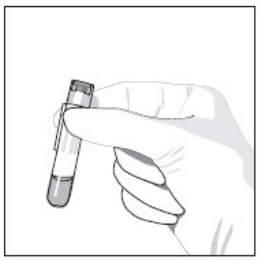
10.2 Assay procedure

The subheadings correspond to messages displayed by the instrument.

Read the card

Read the magnetic card by pulling it through the reader slot with the magnetic band facing you.

Ready for use
Strep A

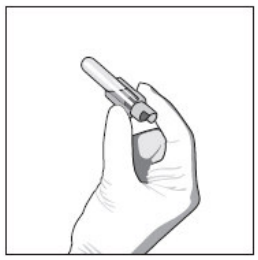


Do not proceed if the sample solution is yellow-orange.
Put the cuvette containing the previously prepared RED sample solution in the measurement well of the instrument.

Measuring blank

The instrument measures the sample blank. This will take a maximum of 40 seconds.
Do not touch the cuvette in the measurement well during the measurement.

Add reagent
Lift the cuvette



1. Press down the pink inner part of the cap (using, for example, your finger or a pen). This will release the dried Strep A reagent into the cuvette.
2. Take the cuvette from the measurement well and mix the contents very rapidly by shaking the cuvette vigorously. The solution becomes milky red.

Shake the cuvette
.....

The display shows you how long you should shake the cuvette (approx. 5 seconds). The moving dots indicate the pace of vigorous shaking. Too slow shaking may cause the error message "Faulty reagent addition. Please run again" to be displayed. In that case a new test needs to be performed.

Insert the cuvette
for measurement

Place the cuvette in the measurement well of the instrument. If the cuvette is inserted back too early, the error message "Test cancelled" or "Cuvette inserted too early" will be displayed. If the cuvette is reinserted too late (> 10 s.), the error message "Cuvette inserted too late" will be displayed. In both cases, a new test needs to be performed.

Measuring 180 s

The instrument measures the Strep A antigen concentration for three minutes. The display counts down the time from 180 seconds.
Do not remove the cuvette from the measurement well before the result is displayed and recorded. Cuvette removal will terminate the measurement immediately and erase any result displayed.

POSITIVE STREP A	*or*	***NEGATIVE STREP A***

The result appears on the display when the measurement is completed.

Performing self test ***Wait....***

When the cuvette is removed, the QuikRead 101 Instrument automatically performs a self-check. If the self-check is not successful, discard the previous test result. After a successful self-check the LCD will display:

Ready for use ***Strep A***

To begin a new test, place a cuvette with the extracted sample in the measurement well.

10.3 Brief instructions for use

1	After sampling, place the swab in a clean QuikRead cuvette.
2	Add two (2) drops of colourless Extraction Reagent 1 and two (2) drops of red Extraction Reagent 2 to the cuvette containing the swab. The solution turns yellow-orange. Incubate for two (2) minutes, swirling the swab for the first 30 seconds.
3	Dispense 0.8 ml of Reaction Buffer, making the solution turn red again. NOTE: Do not proceed if the solution remains yellow-orange (acidic), see "Warnings and precautions".
4	Swirl the swab, press to release all liquid and then remove it. Dispose of the swab. Close with QuikRead Strep A reagent cap.
5	Blank measurement for 40 seconds at most.
6	Add reagent by pressing down the pink inner part of the cap.
7	Take out the cuvette and mix thoroughly. Put the cuvette back into the measurement well.
8	The Instrument measures the Strep A antigen in the sample for 180 seconds.
9	Read the result.

10.4 Interpretation of the test results

If the instrument displays the text ***POSITIVE STREP A***, the sample contains Strep A antigens in concentrations within the measurement range.
If the instrument displays the text ***NEGATIVE STREP A***, the sample does not contain Strep A antigens in measurable concentrations.

Attachment D

Evaluations under the direction of SKUP

Summaries and complete reports from the evaluations are found at www.skup.nu and www.skup.dk

Evaluations performed in 2004 - 2006

Evaluation no.	Component	Instrument/testkit	Producer
SKUP/2006/53*	Strep A		
SKUP/2005/52*	Strep A	Clearview Exact Strep A Dipstick	Applied Biotech, Inc.
SKUP/2005/51*	Glucose[1]	FreeStyle	Abbott Laboratories
SKUP/2006/50	Glucose[1]	Glucocard X-Meter	Arkray, Inc.
SKUP/2006/48	Glucose[1]	Accu-Chek Sensor	Roche Diagnostic
SKUP/2006/47	Hematology	Chempaq XBC	Chempaq
SKUP/2005/46*	PT-INR		
SKUP/2006/45	Glucose[1]	HemoCue Monitor	HemoCue AB
SKUP/2005/44	Glucose[1]	Accu-Chek Aviva	Roche Diagnostics
SKUP/2005/43	Glucose[1]	Accu-Chek Compact Plus	Roche Diagnostics
SKUP/2005/42*	Strep A	Twister Quick-Check Strep A	ACON laboratories, Inc.
SKUP/2005/41*	HbA1c		
SKUP/2005/40	Glucose[1]	OneTouch GlucoTouch	LifeScan, Johnson & Johnson
SKUP/2005/39	Glucose[1]	OneTouch Ultra	LifeScan, Johnson & Johnson
SKUP/2004/38*	Glucose	GlucoSure Plus	Apex Biotechnology Corp.
SKUP/2004/37*	u-hCG	Quick response u-hCG	Wondsfo Biotech
SKUP/2004/36*	Strep A	Dtec Strep A testcard	UltiMed
SKUP/2004/35*	u-hCG	QuickVue u-hCG	Quidel Corporation
SKUP/2004/34*	u-hCG	RapidVue u-hCG	Quidel Corporation
SKUP/2004/33	PT-INR	Hemochron Jr. Signature	ITC International Technidyne Corp
SKUP/2004/32*	Strep A	QuickVue In-Line Strep A test	Quidel Corporation
SKUP/2004/31*	PT-INR		
SKUP/2004/30	Glucose[1]	Ascensia Contour	Bayer Healthcare
SKUP/2004/29	Haemoglobin	Hemo_Control	EKF-diagnostic

*A report code followed by an asterisk, indicates that the evaluation for instance is a pre-marketing evaluation, and thereby confidential. A pre-marketing evaluation can result in a decision by the supplier not to launch the instrument onto the Scandinavian marked. If so, the evaluation remains confidential. The asterisk can also mark evaluations at special request from the supplier or evaluations that are not complete according to SKUP guidelines, e.g. the part performed by the intended users was not included in the protocol.

[1] Including a user-evaluation among diabetic patients.

Evaluations performed in 1999 - 2003

Evaluation no.	Component	Instrument/testkit	Producer
SKUP/2003/28*	Strep A	QuickVue In-Line Strep A test	Quidel Corporation
SKUP/2003/27*	Strep A	QuickVue Dipstick Strep A test	Quidel Corporation
SKUP/2003/26*	HbA1c		
SKUP/2003/25*	HbA1c		
SKUP/2003/24*	Strep A	OSOM Strep A test	GenZyme, General Diag.
SKUP/2002/23*	Hgb, CRP	ABX Micros CRP	ABX Diagnostics
SKUP/2002/22	Glucose[1]	GlucoMen Glycó	Menarini Diagnostics
SKUP/2002/21	Glucose[1]	FreeStyle	TheraSense Inc.
SKUP/2002/20	Glucose	HemoCue 201	HemoCue AB
SKUP/2002/19*	PT-INR	Reagents and calibrators	
SKUP/2002/18	U-albumin	HemoCue	HemoCue AB
SKUP/2001/17	Haemoglobin	Biotest Hb	Biotest Medizin-technik GmbH
SKUP/2001/16*	Urin teststrip	Aution Sticks and PocketChem UA	Arkray Factory Inc.
SKUP/2001/15*	Glucose	GlucoSure	Apex Biotechnology Corp.
SKUP/2001/14	Glucose	Precision Xtra	Medisense
SKUP/2001/13	SR	Microsed SR-system	ELECTA-LAB
SKUP/2001/12	CRP	QuikRead CRP	Orion
SKUP/2000/11	PT-INR	ProTime	ITC International Technidyne Corp
SKUP/2000/10	PT-INR	AvoSure PT	Avocet Medical Inc.
SKUP/2000/9	PT-INR	Rapidpoint Coag	
SKUP/2000/8*	PT-INR	Thrombotest/Thrombotrack	Axis-Shield
SKUP/2000/7	PT-INR	CoaguChek S	Roche Diagnostics
SKUP/2000/6	Hematology	Sysmex KX-21	Sysmex Medical Electronics Co
SKUP/2000/5	Glucose	Accu-Chek Plus	Roche Diagnostics
SKUP/1999/4	HbA1c	DCA 2000	Bayer
SKUP/1999/3	HbA1c	NycoCard HbA1c	Axis-Shield PoC AS
SKUP/1999/2*	Glucose	Precision QID/Precision Plus Electrode, whole blood calibration	Medisense
SKUP/1999/1	Glucose	Precision G/Precision plus Electrode, plasma calibration	Medisense

A report code followed by an asterisk, indicates that the evaluation for instance is a pre-marketing evaluation, and thereby confidential. A pre-marketing evaluation can result in a decision by the supplier not to launch the instrument onto the Scandinavian marked. If so, the evaluation remains confidential. The asterisk can also mark evaluations at special request from the supplier or evaluations that are not complete according to SKUP guidelines, e.g. the part performed by the intended users was not included in the protocol.

[1] Including a user-evaluation among diabetic patients.

Grey area – The instrument is not in the market anymore.

Attachment E

Photos from the test procedure