

Initial evaluation of an image analysis device for the screening of cultures from clinical specimens



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OBJECTIVES

In recent years there has been an increase in the application of image analysis technologies within the clinical laboratory, particularly within the cell-based fields of haematology, anatomical pathology and cytopathology.

The agar plate remains an important diagnostic tool within the clinical laboratory and has not yet been replaced by alternate technologies. Agar plates continue to provide a reliable microbial detection system, even though they are associated with requirements for highly trained staff, laboratory space and a minimal access to automated handling.

The Automated Plate Assessment System (APAS) is an image analysis system dedicated to screening agar plates following incubation. The system detects colony growth, enumerates the various colony types present and applies conventional microbiological logic to sort each plate into categories suitable for further processing.

The aim of this study was to evaluate the abilities of this image device to screen three routinely used agars following inoculation with clinical specimens.

METHODS

One hundred and one stool samples were inoculated onto Xylose Lysine Desoxycholate agar and one hundred and seventy urines onto Horse Blood Agar and Brilliance™ UTI Clarity Agar bi-plates (Thermo Fisher Scientific, Thebarton, Australia).

Following incubation, the plates were read by experienced microbiologists and then analysed by a prototype Automated Plate Assessment System APAS® (LBT Innovations Ltd, Adelaide, Australia). The results for colony detection, enumeration and preliminary colony identification were then compared.

Results generated from the two-plate urine protocol by the APAS® Decision Support System were also compared with those produced by the microbiologists.

RESULTS

FAECES ON XLD

When read by the microbiologists, 60 of the 101 XLD plates were found to contain target colonies that would require further identification while APAS® determined that 69 plates contained colonies requiring identification (Table 1).

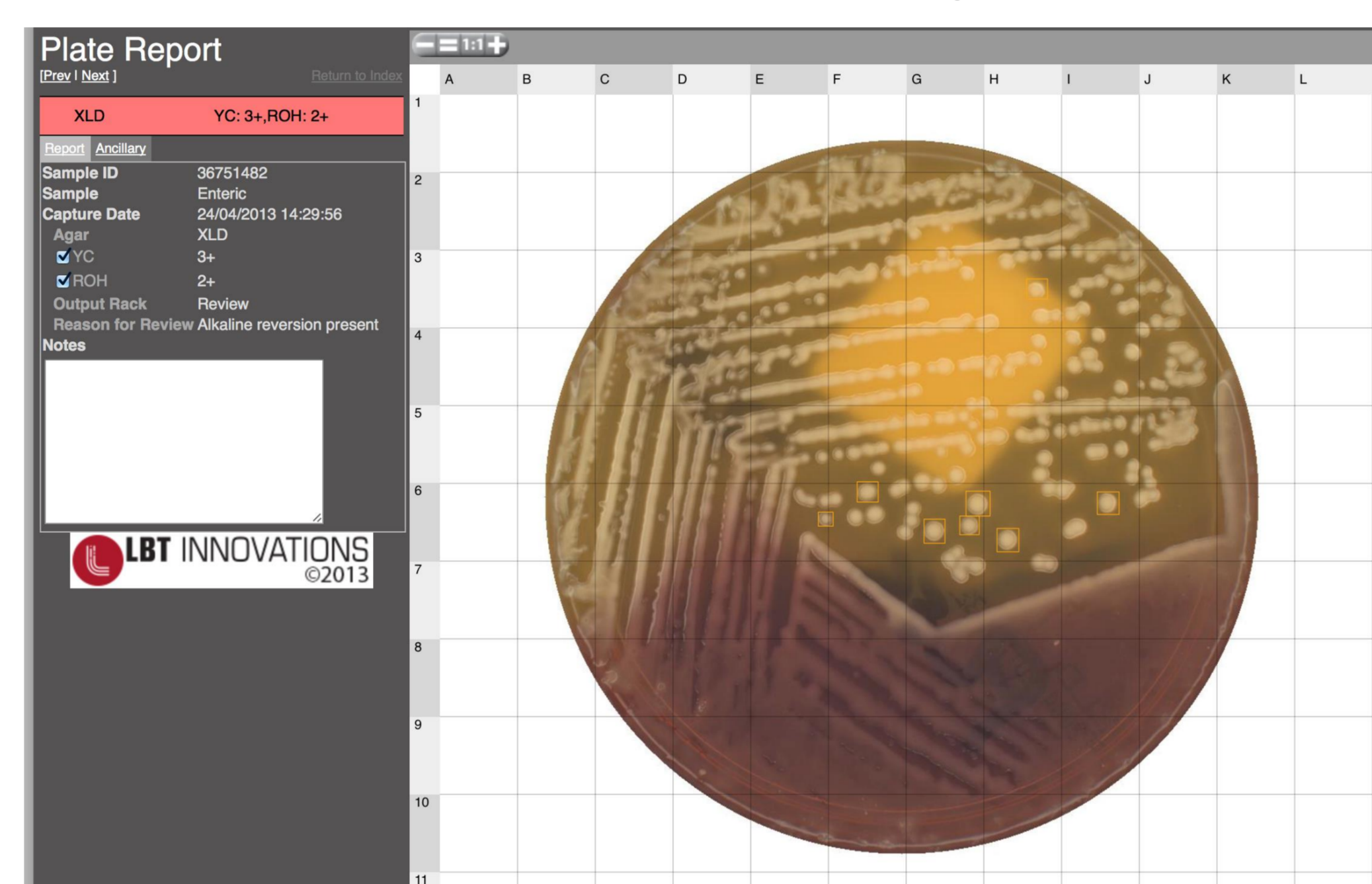
Table 1. Comparison of APAS® findings with manually read XLD agar plates

Plates with target colonies on XLD as determined by microbiologists	Plates with target colonies on XLD as determined by APAS®
60/101	69/101

Of the 9/101 (9%) false positives found, 7 contained heavy growths of coliform bacilli showing alkaline reversion. APAS® was generally able to differentiate between red-pink target colonies and alkaline reversion (Fig. 1) but on this occasion reported more cautiously on these cases.

Where potentially pathogenic colonies were detected, enumeration using a semi-quantitative recording method (1+ to 4+) showed agreement with the manual assessment in 98% of cases within a 1+ variation.

Fig 1. The APAS® Plate Report review screen alerting the operator to the presence of alkaline reversion on XLD agar



URINE

With the urine cultures, 82 cases showed growth at $\geq 10^5$ CFU/ml, 36 at 10^4 CFU/ml and the remaining 52 at 10^3 CFU/ml or no growth (Table 2). APAS® was therefore shown to accurately group this group of cases into growth categories in 96% of cases.

In each case, APAS® provided an option for the operators to review the plate images (Fig. 2).

Fig 2. The APAS® Plate Report review screen for urine

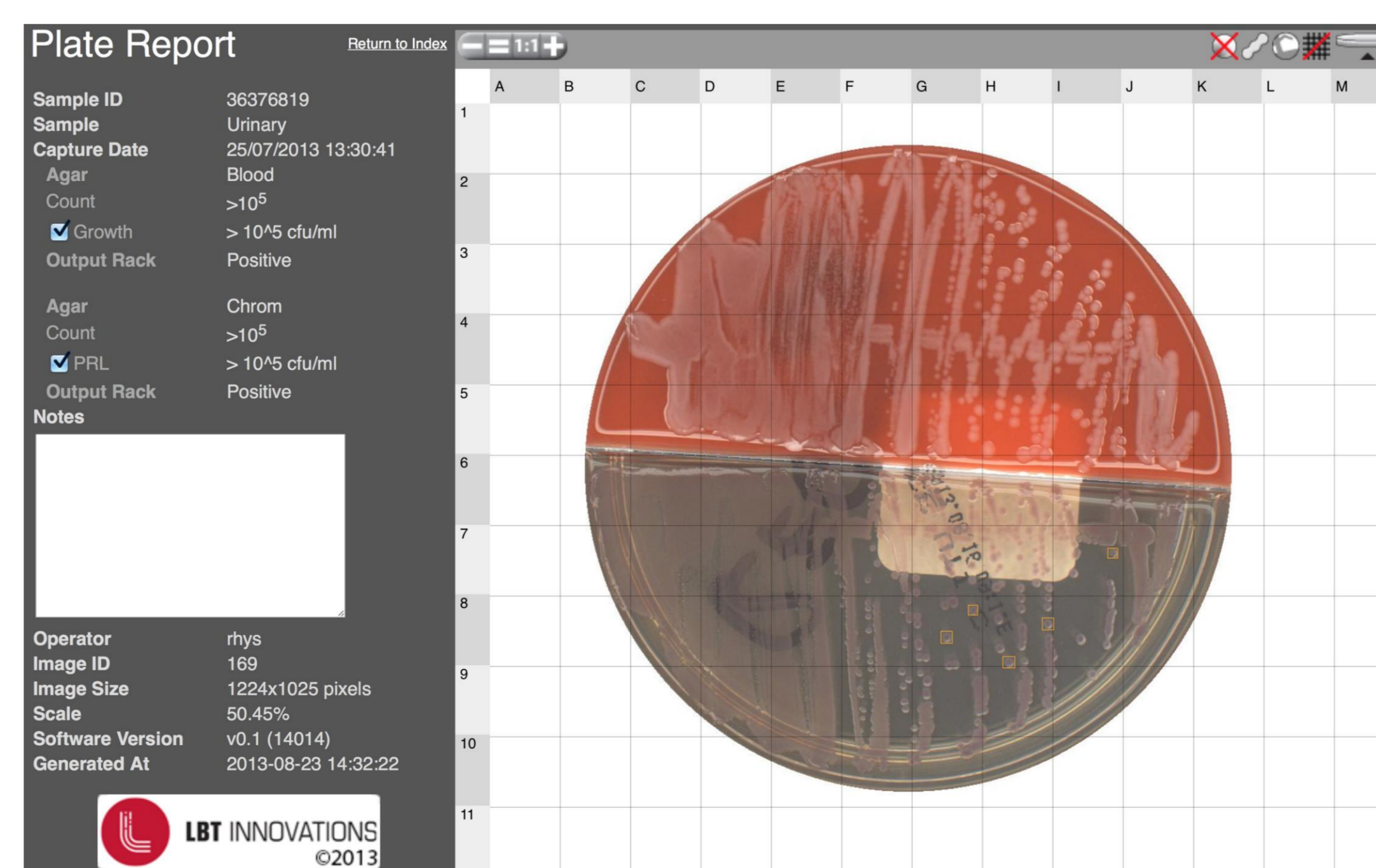


Table 1. Comparison of APAS® urine colony counts with manually read plates

Colony Count (CFU/ml)	Counts determined by Microbiologists	Counts determined by APAS®
0 – 10^3	52	47
10^4	36	42
10^5	82	81
Total	170	170

Using results from both the HBA and Brilliance™ UTI agars, APAS® determined a correct preliminary identification for the primary pathogen in urines with $\geq 10^5$ CFU/ml in 95% of cases (See Table 3).

It also alerted the operator to important features such as the presence of beta haemolysis in 4 cases and the potential presence of *Staphylococcus saprophyticus* in 3 cases. The identification of swarming *Proteus spp.* was also used by APAS® to alert the operators to the need for a review of the colony count as this organism has the potential to confuse the colony count.

Table 3. Preliminary ID comparison of primary isolates between APAS® and microbiologists

Colony Description	Manual	APAS
Straw/beige colonies, brown halo e.g. <i>Proteus spp.</i>	8	9
Pink colonies e.g. <i>E. coli</i>	25	24
Blue colonies e.g. Coliforms, <i>Klebsiella spp.</i>	7	6
Small, white/cream colonies e.g. <i>Staphylococcus spp.</i>	12	10
Small blue-green colonies e.g. <i>Enterococcus spp.</i>	10	10
Small pink/white colonies e.g. <i>S. saprophyticus</i>	3	3
Brown-green colonies e.g. <i>Pseudomonas spp.</i>	5	6
Small colony beta haemolytic e.g. <i>Streptococcus spp.</i>	4	4
Small colony non haemolytic e.g. <i>Streptococcus spp.</i>	3	5
Straw colonies e.g. Chromogenic Negative <i>E. coli</i>	5	5
Total	82	82

CONCLUSIONS

The image analysis technology used by APAS® demonstrated an ability to detect target colonies on XLD agar and differentiated them from colonies of non-pathogenic species. No potential pathogens were missed by APAS® in this series of cases.

As a device for screening a multi-agar protocol such as urine, APAS® also was able to perform colony counts and provide a preliminary identification for the primary isolates.

The incorporation of a Decision Support System with plate interpretation logic and standard reporting rules further enhanced the usefulness of the system.

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