

European guidelines for urinalysis: a collaborative document produced by European clinical microbiologists and clinical chemists under ECLM in collaboration with ESCMID

O. Aspevall¹, H. Hallander², V. Gant³ and T. Kouri⁴

¹Department of Immunology, Microbiology, Pathology and Infectious Diseases, Karolinska Institutet, Stockholm, Sweden, ²Swedish Institute for Infectious Disease Control, Unit for Quality Assurance, Stockholm, Sweden, ³Department of Clinical Microbiology, University College Hospitals Trust, London, UK and ⁴Department of Clinical Chemistry, Center for Laboratory Medicine, Tampere University Hospital, Tampere, Finland

Accepted 8 February 2001

Urinalysis is one of the most common examinations in microbiological and chemical laboratories as well as at points of care. In addition to bacterial cultures, the term urinalysis encompasses here most common chemical tests related to diseases of the urinary tract and urine particle counting (urine microscopy).

Several existing documents can be consulted for details on the microbiological examination of urine [1–6]. While quite a few national guidelines covering aspects of urinalysis have also been published [7–9], there is no general international standard or consensus document applicable for, for example, accreditation or validation of new technology available.

Recently, a group chaired by Dr Timo Kouri, Tampere, Finland published the European Guidelines for Urinalysis [10] under the auspices of the European Confederation of Laboratory Medicine (ECLM). The complete text of this supplement is available in electronic form from Taylor & Francis at <http://www.tandf.no/sjcli>. These guidelines were prepared together with the European Society of Clinical Microbiology and Infectious Diseases (ESCMID) Working Party on Urinalysis, moderated by Dr Vanya Gant, London, UK, to guarantee the quality of the written guidelines from the microbiological point of view. Several experts from most European countries have also contributed to the review of the draft document. An introduction to the project and the recently published paper for clinical chemists is given elsewhere [11]. This paper aims to introduce the guidelines to European clinical microbiologists.

The ECLM European Urinalysis Guidelines discuss the complete process of clinical urine analysis. They embrace indications for urinalysis at several stages: diagnostic strategies;

patient preparation; specimen collection and transport; measurement procedures for chemical, morphologic and microbiological analyses; quality assurance; and transmission of information, i.e. requests and reports.

A stepwise strategy for microbiological urinalysis is also presented (Figure 1).

Diagnostic schemes are outlined for symptomatic low-risk patients, symptomatic high-risk patients, and asymptomatic bacteriuria, respectively. Acutely ill patients need an examination with high specificity to reliably demonstrate the presence of bacteria to support an immediate treatment decision, while the rest of the cases can await results from bacterial cultures. A high-performance, high-throughput screening procedure with low false-negative rates would identify true negatives and allow significant reduction in costly and unnecessary urine culture.

The guidelines supply detailed instructions for the collection, preservation and transport of specimens obtained from each patient category, to include patients with indwelling catheters and children. Practical and useful illustrations on specimen collection are presented and offered for implementation at the end of the book (also seen in the website of the project at Tampere University Hospital (<http://www.pshp.fi/labnet/EUgroup.htm>)). Collection containers, preservation and transport are also discussed.

Detailed methods are presented in four appendices. The authors have chosen to classify diagnostic measurement procedures (methods) hierarchically into four levels of performance based on accuracy of measurement: level 1, rapid procedures such as dipsticks with ordinal scale reporting; level 2, routine (quantitative) procedures in common clinical laboratory use; level 3, qualified comparison procedures; and level 4, primary reference measurement procedures (previously referred to as 'definitive methods') that are designed to give the true value of the measurand (analyzed component). In microbiology, no such primary procedures are available.

Corresponding author and reprint requests: O. Aspevall, Department of Immunology, Microbiology, Pathology and Infectious Diseases, Karolinska Hospital, Stockholm, Sweden
Tel: +46 8 58587842
Fax: +46 8 58581125
E-mail: olle.aspevall@bactlab.hs.sll.se

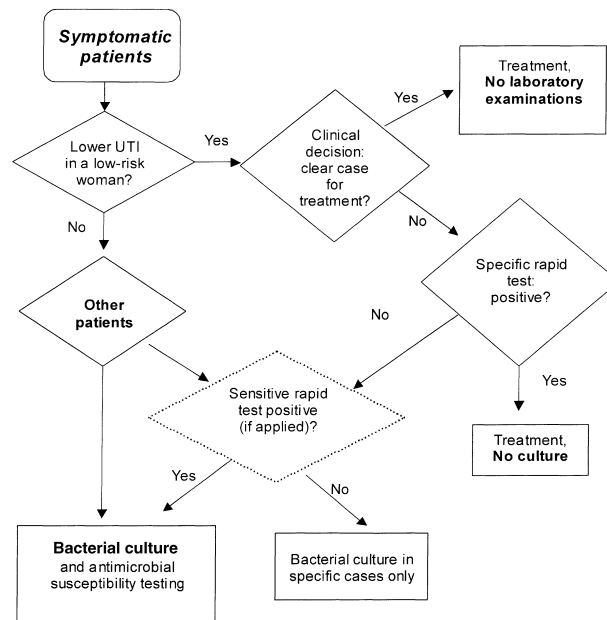


Figure 1 A sieving strategy to reduce requests for bacterial cultures. UTI, urinary tract infection. Reproduced from Kouri T, Fogazzi G, Gant V, Hallander H, Hofmann W, Guder WG, eds. ECLM. European Urinalysis Guidelines. *Scand J Clin Lab Invest* 2000; 60 (suppl 231): 38, by permission of Taylor & Francis AS.

Table 1 The pathogenicity and frequency of microorganisms in midstream urine

Pathogenicity in the urinary tract	Frequency (% of isolates)			
	A. Common (> 10%)	B. Fairly common (1–10%)	C. Uncommon (0.1–1%)	D. Rare (< 0.1%)
I. Primary pathogens	<i>E. coli</i>	<i>S. saprophyticus</i>		<i>E. coli</i> CO ₂ -dependent, <i>Salmonella</i> spp. ^a (<i>Leptospira</i> , mycobacteria)
II. Secondary pathogens		<i>Enterobacter</i> spp., <i>Enterococcus</i> spp., <i>Klebsiella</i> spp., <i>Proteus mirabilis</i> , <i>Pseudomonas aeruginosa</i>	<i>Citrobacter</i> spp., <i>Morganella morganii</i> , <i>Proteus vulgaris</i> , <i>Serratia</i> spp., <i>Staphylococcus aureus</i>	<i>Corynebacterium urealyticum</i> , <i>Haemophilus</i> spp., ^b pneumococci
III. Doubtful pathogens		GBS ^c , yeast, CNS (others) ^d	<i>Acinetobacter</i> spp., <i>Pseudomonas</i> spp., <i>Stenotrophomonas maltophilia</i>	A great number of reported cases have been published with exceptional cases of infections caused by other species
IV. Usually urethral or genital flora ^e		α-Streptococci, <i>Gardnerella vaginalis</i> , lactobacilli etc.	<i>Bifidobacterium</i> spp., 'diphtheroid' rods etc.	

^aLow concentrations are reported even if they are most likely caused by contamination during specimen collection. ^bMost often isolated from children. ^cGBS, group B streptococci (*S. agalactiae*). ^dCNS, coagulase-negative staphylococci, urease-forming isolates or isolates found in patients with indwelling catheters have increased significance. ^eNo identification and susceptibility testing (only exceptionally, if especially indicated). Reproduced from Kouri T, Fogazzi G, Gant V, Hallander H, Hofmann W, Guder WG, eds. ECLM. European Urinalysis Guidelines. *Scand J Clin Lab Invest* 2000; 60(suppl 231): 26, by permission of Taylor & Francis AS.

Procedures for bacterial culture can be outlined at three levels:

1. Level 1, screening procedure. Dipslide culture used to identify negative cultures and significant growth of *Escherichia coli* only.
2. Level 2, routine procedure. One microliter is inoculated on CLED agar (blood agar being an option) and incubated aerobically for 24 h.
3. Level 3, qualified comparison procedure. Ten microliters of urine is inoculated by pipette on CLED agar, hematin agar and blood agar aerobically, and cultured anaerobically in carbon dioxide for 48 h.

The proposal to use a liter-based unit, CFB/L (colony-forming bacteria/L), in reports of bacterial concentration in

urine instead of CFU/mL (colony-forming units/mL) is an important step in metrological standardization. The character B instead of U was chosen to avoid confusion between exponentials which might result if only mL volumes were changed to L volumes.

The authors also decided to use a novel classification system for organisms causing urinary tract infections based on criteria of pathogenicity, as developed in Sweden (Table 1).

The first group consists of *E. coli* and *Staphylococcus saprophyticus* causing urinary tract infections in individuals with normal urinary tracts. Secondary pathogenic species often occur in hospital-acquired urinary tract infections. The third and fourth groups refer to doubtful pathogens and urethral or genital flora, respectively. This classification influences the suggested limiting

Table 2 Suggested limiting concentrations of bacterial colonies justifying identification and susceptibility testing in the laboratory.

Symptoms ^a and specimens	Inoculum, minimum volume	Species type ^b	Species number	Significant colony concentration	
				CFB/L	CFU/mL
Midstream urine specimen	1 µL	I	1–2 ^c	10 ⁶	10 ³
Yes ^a		II	1	10 ⁷ (women)	10 ⁴
		II	1	10 ⁶ (men)	10 ³
		II	2	10 ⁸	10 ⁵
		III	1	10 ⁸	10 ⁵
No ^a		I–III	1	10 ⁸	10 ⁵
Yes (special)	10 µL ^d	I	1–3	10 ⁵	10 ²
Suprapubic aspiration specimen	100 µL	I–IV	1–2	10 ⁴	10 ¹
Yes or no					
Specimen from cystoscopy or single urethral catheterization	10 µL ^d	I–III	1–2	10 ⁵	10 ²
Yes or no					
Specimen from indwelling catheter	1 µL	I–III	1–3	10 ⁷	10 ⁴
Yes					
No	1 µL	I–III	1	10 ⁸	10 ⁵

^aYes = the patient has symptoms; No = no symptoms, or no information about symptoms. ^bSuggestive category based on growth characteristics (see Table 1). Species of normal urogenital flora (IV) are examined for susceptibility only if especially indicated. ^cUsually, only one species is identified and tested for antimicrobial susceptibility if 2–5 similar colonies grow (as locally agreed). Occasionally, two species may be identified for specific patient populations. Three or more species are usually reported as 'mixed culture' and considered as contaminants. Susceptibility testing of isolates from midstream urine specimens as well as other detailed strategic decisions need local clinical and microbiological consultation. ^dA 1-µL loop is practical and sufficient for routine workup. However, in specific patient groups, such as in patients with certain urologic diseases, or in the precise evaluation of patients with simple cystitis, a result at $\geq 10^5$ CFB/L (10^2 CFU/mL) and a statistically reliable culture result at 10^6 CFB/L (10^3 CFU/mL) may be clinically significant. This can only be obtained by using a 10-µL loop. This sensitized culture procedure should be especially requested to avoid extra work and costs caused by routine application of a 10-µL loop for all specimens. Reproduced from Kouri T, Fogazzi G, Gant V, Hallander H, Hofmann W, Guder WG, eds. ECLM. European Urinalysis Guidelines. *Scand J Clin Lab Invest* 2000; 60(suppl 231): 30, by permission of Taylor & Francis AS.

concentrations of bacteria justifying identification and susceptibility testing in the laboratory. Other factors taken into consideration for interpretation limits are specimen type, gender, number of isolated species and presence of symptoms. These limits allow diagnosis of urinary tract infections down to 10^6 CFB/L from midstream urine in both females and males. We give detailed recommendations on cut-offs which take the above-mentioned factors into consideration (Table 2). General screening of asymptomatic individuals should be avoided.

The procedures for bacterial culture mentioned above are described in detail in the microbiological appendix section. In addition, we provide guidance and instructions concerning the minimum criteria for identification of bacteria, serial dilution

methods for quantitation, and a level 1 'microtiter tray method' for detecting clear-cut infected urines.

Special attention is also paid to near-patient testing. Bacterial detection by non-culture methods is important in emergency diagnostics, as exemplified by test strips for nitrite and leukocyte esterase describing the performance and limits of these reactions. The technical performance of dipslide culture restricts its use to the exclusion of urinary tract infections, and identification of the primary pathogen *E. coli*.

Methods for particle analysis first define basic and advanced levels of urine particle identification for different clinical practices and laboratories (Table 3). The basic level would be mostly used in general or chemistry laboratories and for general patient

Table 3 Levels of microscopy differentiation in clinical urinalysis

Basic level	Advanced level in addition
Red blood cells	Detailed subclasses of erythrocytes: dysmorphic erythrocytes
White blood cells/granulocytes	Differentiation of leukocytes: Granulocytes, lymphocytes, macrophages (monocytes and eosinophils)
Epithelial cells Squamous epithelial cells Non-squamous = small epithelial cells	From non-squamous epithelial cells: squamous epithelial cells Renal tubular epithelial cells Transitional epithelial cells (superficial and deep) Intestinal epithelial cells (occurring after bladder surgery) Atypical cells (experienced cytopathologist)
Casts Hyaline casts Non-hyaline casts	From non-hyaline casts: Erythrocyte, granulocyte casts Renal tubular cell casts Hyaline, granular, waxy, fatty casts Bacteria and yeast-containing casts Hemoglobin and myoglobin casts Bilirubin casts
Bacteria	Gram-staining characteristics of bacteria (microbiology laboratories)
Yeasts <i>Trichomonas</i>	<i>Schistosoma haematobium</i> (in appropriate geographic locations)
Spermatozoa Artefacts (hair, paper and textile fibers, starch, glass) and mucus	Artefacts and mucus as on the left
Lipids Droplets (isolated and aggregated)	Lipids, in addition to droplets: Oval fat bodies (lipid-laden tubular cells), cholesterol crystals
Crystals Urate, oxalate (mono- and dihydrated), phosphate and cystine	Additional rare crystals: drugs, cystine, leucine, tyrosine 2,8-dihydroxyadenine, xanthine

Reproduced from Kouri T, Fogazzi G, Gant V, Hallander H, Hofmann W, Guder WG, eds. ECLM. European Urinalysis Guidelines. *Scand J Clin Lab Invest* 2000; 60(suppl 231): 21, by permission of Taylor & Francis AS.

populations. The advanced level comprises a microbiological direction (Gram staining for microbiology laboratories), a nephrologic direction (detection of renal elements, i.e. casts and renal tubular cells), or a cytopathologic direction (detection of atypical epithelial cells and malignancy).

Considerable efforts were made to address the issues of different clinical needs, and we classify routine microscopy techniques into different levels of analytic procedures. Level 1 is suggested to contain screening identification of particles, e.g. by using a microtiter tray method and ordinal scale reporting. Level 2 defines a standardized routine procedure with specific aims, such as: (a) Gram staining for classifying uropathogenic bacteria; (b) standardized urine sediment with supravital staining and/or phase contrast microscopy for precise identification of renal particles; and (c) chamber counting of uncentrifuged urine specimens for accurate counting of red and white blood cells that are partially lost during centrifugation. Level 3 should contain sensitive detection and accurate quantitation of clinically significant urine particles. If bacteria only are considered, the slide centrifugation and Gram-staining technique is recommended because of its proven performance against culture. Thus, general (chemical), microbiological and nephrologic needs for urinalysis may differ, and must be taken into account when specifying routine analytic processes.

A chapter on quality assurance gives an extensive discussion on general issues such as quality system, quality manual, and quality policy, as well as very specific matters. Analytic quality specifications are suggested for clinical microbiology laboratories, to help assessment of performance. We suggest (for example) acceptable routine levels of identification for bacteria, and acceptable turnaround times for bacterial cultures. Detail for evaluation of performance of urinalysis devices is provided as a suggestive framework.

This document will be most valuable for persons responsible for analysis of urine in clinical chemistry laboratories and in clinical microbiology laboratories. Acceptance and endorsement of the proposed procedures would be a step towards

standardization of urinalysis. This would be of great benefit to laboratories and patients alike.

REFERENCES

1. Clarridge JE, Johnson JR, Pezzlo MT. *Cumitech 2B: Laboratory diagnosis of urinary tract infections*. Washington DC: American Society for Microbiology, 1998.
2. Murray PR, Baron EJ, Pfaller MA, Tenover FC, Tenover RH, eds. *Manual of Clinical Microbiology*, 7th edn. Washington DC: American Society for Microbiology, 1999.
3. Kunin CM. *Urinary tract infections. Detection, Prevention, and Management*, 5th edn. Baltimore: Williams & Wilkins, 1997.
4. Sobel JD, Kaye D. Urinary tract infections. In: Mandell GL, Bennett JE, eds. *Mandell, Douglas and Bennett's Principles and Practice of Infectious Diseases*, 4th edn. New York: Churchill Livingstone, 1995: 662–89.
5. Aspevall O, Hallander H, eds. *Referensmetodik För Urinvägsinfektioner/Bakteriuri, I5. Reference Methodology for Urinary Tract Infections/Bacteriuria*, 2nd edn. Stockholm: Swedish Institute for Infectious Disease Control, 2000.
6. Gatermann S, Podschun R, Schmidt H et al. *Fachgruppe 'Diagnostische Verfahren in der Mikrobiologie' der Deutschen Gesellschaft für Hygiene und Mikrobiologie, DGHM. Harwegsinfektionen. Qualitätsstandards in der mikrobiologisch-infektiologischen Diagnostik*. MiQ. Berlin: Gustav Fischer, 1997.
7. NCCLS. *Urine Sediment Analysis. JCCLS Guideline GP1-P2*. Kobe: Toa Medical Electronics Ltd, 1995.
8. Koivu T, Grönroos P, Gävert J et al. Basic urinalysis and urine culture: Finnish recommendations from the working group on clean midstream specimens. *Scand J Clin Lab Invest* 1990; 50(suppl 200): 26–33.
9. National Committee for Clinical Laboratory Standards. *Urinalysis and Collection, Transportation, and Preservation of Urine Specimens*. NCCLS Document GP16-A. Wayne: NCCLS, 1995.
10. Hallander H, Hofmann W, Guder WG, eds. ECLM. European Urinalysis Guidelines. *Scand J Clin Lab Invest* 2000; 60(suppl 231): 1–96. The complete text of this supplement is also available from Taylor & Francis at <http://www.tandf.no/sjcli>.
11. Kouri TT, Gant VA, Fogazzi GB, Hofmann W, Hallander HO, Guder WG. Towards European urinalysis guidelines. Introduction of a project under European Confederation of Laboratory Medicine. *Clin Chim Acta* 2000; 297(1–2): 305–11.

APPENDIX, LIST OF CONTRIBUTORS TO THE ECLM EUROPEAN URINALYSIS GUIDELINES

Reproduced from Kouri T, Fogazzi G, Gant V, Hallander H, Hofmann W, Guder WG, eds. ECLM. European Urinalysis Guidelines. *Scand J Clin Lab Invest* 2000; 60(suppl 231): 4–5, by permission of Taylor & Francis AS.

Members of the ECLM European Urinalysis Group

Dr Timo Kouri (Chairman)
Center of Laboratory Medicine
Tampere University Hospital
PO Box 2000, FIN-33521 Tampere
Finland
Tel: +358 3 247 5484 Fax: +358 3247 5554
E-mail: Timo.Kouri@tays.fi

Dr Giovanni Fogazzi
Divisione di Nefrologia e Dialisi
Ospedale Maggiore, IRCCS
Via Commenda 15, I-20122 Milano
Italy
Tel: +39 02 5503 4557 Fax: +39 02 55034550
E-mail: croffl@polic.cilea.it

Dr Vanya Gant
Department of Clinical Microbiology
University College Hospital
Grafton Way, London WC1E 6DB
UK
Tel: +44 171 380 9913 Fax: +44 171388 8514
E-mail: v.gant@academic.uclh.nthames.nhs.uk

Dr Hans Hallander
Swedish Institute for Infectious Disease Control
Department of Quality Assurance
SE-17182 Solna
Sweden
Tel: +46 8 457 2490 Fax: +46 83025 66
E-mail: hans.hallander@smi.ki.se

Dr Walter Hofmann
Institute for Clinical Chemistry
Städtisches Krankenhaus Bogenhausen
Englschalkinger Strasse 77
D-81925 Munich
Germany
Tel: +49 89 9270 2282 Fax: +49 89 92702113
E-mail: Hofmann-Sedlmeir@t-online.de

Professor Dr Walter G. Guder
Institute for Clinical Chemistry
Städtisches Krankenhaus Bogenhausen
Englschalkinger Strasse 77
D-81925 Munich
Germany
Tel: +49 89 9270 2280 Fax: +49 89 92702113
E-mail: w.g.guder@lrz-muenchen.de

ESCMID Working Party on Urinalysis

Vanya Gant, London, UK, Moderator
Hans Hallander, Stockholm, Sweden
Francis O'Grady, Nottingham, UK
Marc Struelens, Brussels, Belgium

Other contributors

Amores C., Jaen, Spain
Anyszek Tomasz, Krakow, Poland
Arnadottir M., Reykjavik, Iceland
Aspevall O., Huddinge, Sweden
Baerheim A., Bergen, Norway/Gent, Belgium
Béné M.-C., Nancy, France
Blaseio U., Hamburg, Germany
Blondiau R., Erembodegem, Belgium
Bondarenko I., St Petersburg, Russia
Colombo J.P., Bern, Switzerland
Delanghe J., Gent, Belgium
Dybkaer R., Copenhagen, Denmark
Edel H., Munich, Germany
Emanuel V., St Petersburg, Russia
Fang-Kirchner S., Vienna, Austria
Fenili D., Romano di Lombardia, Italy
Fernandes B., Toronto, Canada

Fuellemann R., Mannheim, Germany
Garcia J.L., Sevilla, Spain
Gatermann S., Bochum, Germany
Grubb A., Lund, Sweden
Györy A.Z., Sydney, Australia
Hesse A., Bonn, Germany
Hoeiby N., Copenhagen, Denmark
Hofgärtner W., Oxford, UK
Hyltoft-Petersen Per, Odense, Denmark
Ito K., Yokohama, Japan
Itoh Y., Osaka, Japan
Ivancic M., Munich, Germany
Kermes K., Tartu, Estonia
Khorovskaya L., St Petersburg, Russia
Kimling H., Mannheim, Germany
Kokot F., Katowice, Poland
Kontiainen S., Helsinki, Finland
Kratochvila J., Nymburg, Czech Republic
Kutter D., Junglinster, Luxembourg
Lapin A., Vienna, Austria
Lipsic T., Bratislava, Slovak Republic
Malakhov V., Moscow, Russia
Naber K.G., Straubing, Germany
Naegele U., Mannheim, Germany
Newall R., Newbury, UK
Newman D.J., London, UK
Parker D., Elkhart, IN, USA
Peheim E., Bern, Switzerland
Penev M.N., Sofia, Bulgaria
Piemont Y., Strasbourg, France
Ponticelli C., Milano, Italy
Price C., London, UK
Puglia M., Elkhart, IN, USA
Rada F., Tirana, Albania
Recio F., Sevilla, Spain
Rowan R.M., Glasgow, UK
Rowlands T., Meylan, France
Salum T., Tartu, Estonia
Sandberg S., Bergen, Norway
Selgren S., Elkhart, IN, USA
Siitonen A., Helsinki, Finland
Sikirica M., Zagreb, Croatia
Steinhausen R., Mannheim, Germany
Tetz V.V., St Petersburg, Russia
Tzontcheva A., Sofia, Bulgaria
Verhaegen J., Leuven, Belgium
Zaman Z., Leuven, Belgium

Hallander 2. reference measurement procedures (previously referred to as Stockholm. at Tampere University Hospital (<http://www. analyses.IN SE ARCH OF EUROPE AN OPINION> European guidelines for urinalysis: a collaborative document produced by European clinical microbiologists and clinical chemists under ECLM in collaboration with ESCMID O. In microbiology. measure- biological and chemical laboratories as well as at points of care. ment procedures for.Â Aspevall et al European guidelines for urinalysis 175 Procedures for bacterial culture can be outlined at three urine instead of CFU/mL (colony-forming units/mL) is an levels: important step in metrological standardization. The bacteriological and clinical effects of pivmecillinam for UTIs caused by *S. saprophyticus* were high in our studies. Our findings are in concordance with Monsen et al. [19], and we agree with the authors that the breakpoints for *S. saprophyticus* (especially in UTI) need to be revised, as pivmecillinam clearly has a high effect in most cases. Thulin et al.Â Aspevall, O.; Hallander, H.; Gant, V.; Kouri, T. European guidelines for urinalysis: a collaborative document produced by European clinical microbiologists and clinical chemists under ECLM in collaboration with ESCMID. Clin. Microbiol. European Society of Clinical Microbiology and Infectious Diseases (ESCMID): treatment guidance document for *Clostridium difficile* infection (CDI). Bauer MP, Kuijper EJ, van Dissel JT; European Society of Clinical Microbiology and Infectious Diseases. Clin Microbiol Infect 2009 Dec;15:1067-79. doi: 10.1111/j.1469-0691.2009.03099.x. Abstract.Â European guidelines for urinalysis: a collaborative document produced by European clinical microbiologists and clinical chemists under ECLM in collaboration with ESCMID. Aspevall O, Hallander H, Gant V, Kouri T. Clin Microbiol Infect 2001;7:173-8. PMID:11422238.