2

Measurement of proteinuria

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"I had every [indication] of pre-eclampsia except for proteinuria until 38 weeks. When I finally presented with +4 protein, my BP was 198/130 and I had gained 50 lbs of water in 6 weeks."

Jenn P

SYNOPSIS

In pregnancy, there is a focus on measurement of proteinuria as it has been regarded as critical to the diagnosis of pre-eclampsia, the most dangerous of the hypertensive disorders of pregnancy. However, it is increasingly recognised that proteinuria is not essential for the diagnosis of pre-eclampsia, which can be based on other end-organ complications (such as elevated liver enzymes). Although heavy proteinuria has been linked with an increased risk of stillbirth in a 'signs and symptoms only' model of maternal risk (i.e., miniPIERS), we lack the ability to identify a level of proteinuria above which maternal and/or perinatal risk is heightened. Therefore, at present, we rely on the detection of proteinuria that exceeds what is normally excreted by healthy pregnant women. Proteinuria detection methods are also a matter of keen debate, with all available methods having advantages and disadvantages.

PHYSIOLOGICAL CHANGES OF PROTEINURIA IN PREGNANCY

During normal pregnancy, proteinuria increases through the trimesters, from 0.15 g/d outside pregnancy to 0.3 g/d during pregnancy. This is attributable to the increase in renal plasma flow and glomerular filtration rate, as well as changes in protein handling in the nephron; these changes resolve after pregnancy[1].

The proteinuria of pregnancy consists of both glomerular and tubular proteins, although the proportion of each is still a matter of debate[2].The most abundant individual protein is from the renal tubules, Tamm-Horsfall protein. Other proteins include albumin, thyroxine-binding prealbumin, immunoglobulins, α1-antitrypsin, transferrin, β-lipoprotein and low-molecular weight proteins[1].

CAUSES OF PROTEINURIA

Proteinuria screening in pregnancy is focused on the detection of pre-eclampsia, the most common cause of proteinuria in pregnancy. Pre-eclampsia affects the glomeruli, and the lesion has been termed 'glomerular endotheliosis'. This terms describes glomerular endothelial swelling and loss of the integrity of the fenestrae (i.e., sieving apparatus), leading to leakage of protein into the renal tubules and associated occlusion of the capillary lumens[3].

Proteinuria may be transient in pregnancy, although when identified, repeat testing must be

done within days to ensure that pre-eclampsia is not missed and allowed to evolve unobserved. Transient causes are associated with normal renal function and no abnormalities of urinary sediment. Causes include orthostasis (i.e., upright posture), exercise, fever or sepsis, congestive cardiac disease, or central nervous system causes such as subarachnoid or intracerebral haemorrhage, or seizures. It should be noted that orthostatic proteinuria occurs in no more than 5% of adolescents and decreases in frequency with age, being less common in those 30 years of age or older[4].

When considering the causes of persistent proteinuria in pregnancy, a full differential diagnosis should be considered. How often new proteinuria is due to causes other than pre-eclampsia is unclear, especially in under-resourced settings. In the face of this uncertainty about the cause of the proteinuria, pre-eclampsia should be regarded as the working diagnosis given the maternal and fetal risks associated with this condition. Persistent proteinuria in pregnancy may be also caused by non-pre-eclampsia glomerular disease, tubular disease, or even non-renal disease (Table 2.1). Nephrotic-range proteinuria (≥3 g/d) is suggestive of glomerular renal disease. Abnormalities of the urinary sediment (e.g., micro- or macroscopic haematuria with IgA nephropathy) may or may not be seen with renal causes of proteinuria.

Table 2.1 Causes of proteinuria (modified from Côté and Sauve[67])

Transient causes
Orthostatic (i.e., related to upright posture)
Systemic (e.g., exercise, fever or sepsis, congestive cardiac disease)
Central nervous system (e.g., subarachnoic or intracerebral haemorrhage, seizures)
Contamination (e.g., from vaginal bleeding)
Persistent
Glomerular diseases
Pre-eclampsia
Pre-gestational diabetes type 1 or type 2
Immunoglobulin A (IgA) GN
Focal and segmental glomerulosclerosis (FSGS)
Lupus nephritis
Infection-related GN (e.g., HIV, hepatitis B and C, post-streptococcal, visceral abcess, endocarditis, other)
Drug-related GN
Other glomerular disease in young women: minimal change, membranous GN, membranoproliferative GN, other rare glomerular disease (e.g., amyloidosis), Fabry, Alport)
Non-glomerular (tubulointerstitial) disease
Structural (e.g., congenital anomalies, reflux nephropathy)
Polycystic kidney disease
Interstitial nephritis
Urinary tract infection

GN, glomerulonephritis

POLICY IMPLICATION

Detecting proteinuria

- Proteinuria screening must be available wherever antenatal or postnatal care is provided
- At minimum, proteinuria testing must be performed at the first of the four WHO-recommended antenatal visits and whenever hypertension is detected[5]
- Proteinuria testing must be performed at the 6-week postpartum visit in women who developed proteinuria in pregnancy[6]

(See Appendix 2.1)

SCREENING FOR PROTEINURIA IN ANTENATAL CARE

At minimum, all pregnant women should be assessed for proteinuria in early pregnancy, to detect pre-existing renal disease and to obtain a baseline measurement in women at increased risk of pre-eclampsia[7]. Thereafter, most assessment for proteinuria occurs in women suspected of having pre-eclampsia, such as when women present with hypertension or suggestive symptoms (such as headache). The frequency of such screening is uncertain. Ideally, countries should move toward universal screening at every visit as pre-eclampsia/eclampsia may first present with isolated proteinuria[8]. In the meantime, it would seem reasonable to retest for proteinuria in response to a rising blood pressure and/or maternal symptoms or maternal/fetal signs of

pre-eclampsia. It must be emphasised that proteinuria is but one diagnostic criterion for pre-eclampsia, and the end-organ complications of pre-eclampsia may occur in the absence of proteinuria. For example, in the latest eclampsia survey in the UK, 7.5% of women had only proteinuria (and 45% had neither hypertension nor proteinuria) in the week before their eclamptic seizure[9].

As per the WHO postnatal recommendations for the care of the mother and newborn[6], proteinuria should be re-tested in women who were identified as having had proteinuria in pregnancy, (see Chapter 11 for more information about postpartum management.)

MEASUREMENT OF PROTEINURIA FOR THE DIAGNOSIS OF PRE-ECLAMPSIA

There are many options for diagnosis of proteinuria, on either random (spot) samples (such as urinary dipstick testing, heat coagulation test, urinary protein : creatinine ratio (PrCr), or urinary albumin : creatinine ratio (ACR)) and various timed urine collections (usually 24-hour). Each has advantages and disadvantages and different results for diagnostic test accuracy (Appendix 2.2).

Urine tested for proteinuria should be as 'fresh' as possible. The longer the collection to testing interval, the more likely that bacterial contamination will skew the results. Without refrigeration, urine should be tested as soon as possible after collection, and definitely within 4 hours of collection. Urine collected over a 24-hour period must be refrigerated and brought to the laboratory on the day that collection finishes.

Point-of-care urine test strips come in opaque containers that specify expiry dates. They should not be used after that time. Once the container has been opened, the lid should be replaced between strip removal so that the unused strips are kept out of sunlight.

POLICY IMPLICATION

How to screen for proteinuria

Proteinuria screening should be performed using urinary dipsticks given their ease of use and low cost, until such time that another method proves to be superior

(See Appendix 2.1)

Urinary dipstick testing for proteinuria

There are many available types of urinary dipstick testing strips for visual and automated testing, and analysers for automated dipstick analysis. As it is unclear whether a particular method has an impact on test accuracy and pregnancy outcome, it may be prudent if possible, for the health care provider to use the same type of urinary dipsticks in the clinic and to send an individual patient to the same laboratory throughout her pregnancy so that differences in test results over time are more likely to be meaningful.

Visual interpretation of urinary dipstick

Urinary dipsticks may have up to 10 chemical pads for measuring different substances in urine, including protein and albumin, although strips that restrict measurement to proteinuria or albuminuria are available. The advantage of a strip with multiple pads is that it can reveal associated urinary abnormalities that are causes of low-level proteinuria, such as haematuria or either asymptomatic bacteriuria or symptomatic urinary tract infection (both of which should be treated with antibiotics) by showing leukocytes and nitrites. The disadvantages include multiple results that may result in confusion and inappropriate further investigation; for example, leukocytes may be a completely normal finding in pregnancy given contamination of the urine by vaginal discharge.

The urinary dipstick strip should be immersed completely in a well-mixed sample of urine for a short period of time, then extracted from the container and the excess urine removed by either supporting the edge of the strip over the mouth of the container, or drying the edges of the strip on absorbent paper (Figure 2.1). The strip is then left to stand for the time necessary for the reaction to occur (usually 60 seconds, as specified by the strip manufacturer). For visual analysis, the colour on the 'proteinuria' pad is compared with the chromatic scale specific to that strip and provided by the manufacturer. For automated analyses, the machine will read out the result. Results are reported as negative, trace, 1+, 2+, 3+, or 4+ based on the concentration of proteinuria detected. Although the concentration for a given '+' may vary from one manufacturer to another (particularly at the 4+ stage), 1+ proteinuria usually reflects 0.3 g/L of proteinuria. It follows that dehydration

Figure 2.1 Accredited social health associate (ASHA) worker, India, performing urinary dipstick testing

can increase proteinuria concentration and result in a 'positive' proteinuria dipstick result.

Urinary dipstick testing for proteinuria is inexpensive, easy and widely used. In a systematic review, 1+ proteinuria by visual dipstick testing showed low sensitivity (55%, 95% CI 37–72) and reasonable specificity (84%, 95% CI 57–95) for detection of 0.3 g/d of proteinuria[10]. A threshold of 2+ proteinuria by visual dipstick testing has reported sensitivity and specificity that varies from values of 58%[11] to values of ≥80%[12–14]. How should these results be interpreted for clinical practice? Given the <90% sensitivity of dipsticks using a threshold of 1+, a negative or trace value should not be ignored in a woman with new hypertension or symptoms or signs suggestive of pre-eclampsia. Given the reasonable specificity of dipsticks (at 1+ or 2+ levels), a result ≥1+ should prompt additional investigations even when the suspicion of pre-eclampsia is low. A urinary dipstick result of ≥2+ is suggestive of 0.3 g/d or more of proteinuria by 24-hour urine collection.

Automated testing of urinary dipstick

In theory, automation has the potential to reduce errors arising from subjective interpretation of dipstick readings.

Comparisons of automated with visual-read dipsticks have used thresholds of either 1+ or 2+. Two studies have compared the diagnostic test properties of automated dipsticks for proteinuria with visual read urinary test strips for proteinuria[13,15], using a threshold of 1+. Although one study compared test strips with 24-hour urinary protein excretion (g/d)[15] and the other study used 24-hour urinary protein concentration (g/L) as the comparator[13], both studies demonstrated superior diagnostic test properties of automated (versus visual) testing, using a threshold of 1+ for proteinuria. In contrast, a more recent study failed to show superiority of automated over visual testing[16]. When a threshold of 2+ proteinuria was used, automated testing also appeared to be superior to visual testing[13], with absolute values for sensitivity by automated testing as high as >80%[13,14] but as low as 23% in another study[17].

For detection of proteinuria by 24-hour urine collection or PrCr, published sensitivities for an automated dipstick threshold of 1+ or more (41%[17], 82%[15], 90%[13] and 100%[18]) and corresponding specificities (100%, 81%, 86% and 37%) have varied widely, even when the prevalence of proteinuria in the study populations was similar (i.e., 45%[15] and 48%[17]).

The diagnostic accuracy of automated testing may depend on the choice of test strip and/or analyser. It may be premature to recommend widespread adoption of automated urine proteinuria test strip readers, although one international guideline makes such a recommendation[19].

Urinary dipstick test strips are also available for detection of albuminuria (i.e., albumin concentration) specifically. However, we are not aware of studies that have compared albuminuria dipstick testing with proteinuria dipstick testing or other methods of proteinuria testing for detection of significant proteinuria in pregnancy. Of note, albuminuria dipsticks are more expensive than are proteinuria dipsticks.

HEAT COAGULATION TEST

The heat coagulation test may be used in under-resourced settings as an alternative to dipstick testing or other methods (discussed below) that are unavailable or too costly. A test tube is filled to two-thirds with urine. A few drops of dilute acetic acid are added to make the urine sample acidic. The upper part of the test tube containing urine is heated (but not boiled) over a burner.

The presence of protein is signified by the turbidity of the urine when the tube is placed in front of a typed sheet of paper according to a pre-specified chart (Figure 2.2)[20,21]. The lower part of the tube of urine acts as a control as that urine should remain clear (Figure 2.3).

The heat coagulation test may be less sensitive than visually interpreted urinary dipsticks (at ≥1+ level) for detecting 0.3 g/d or more of urinary

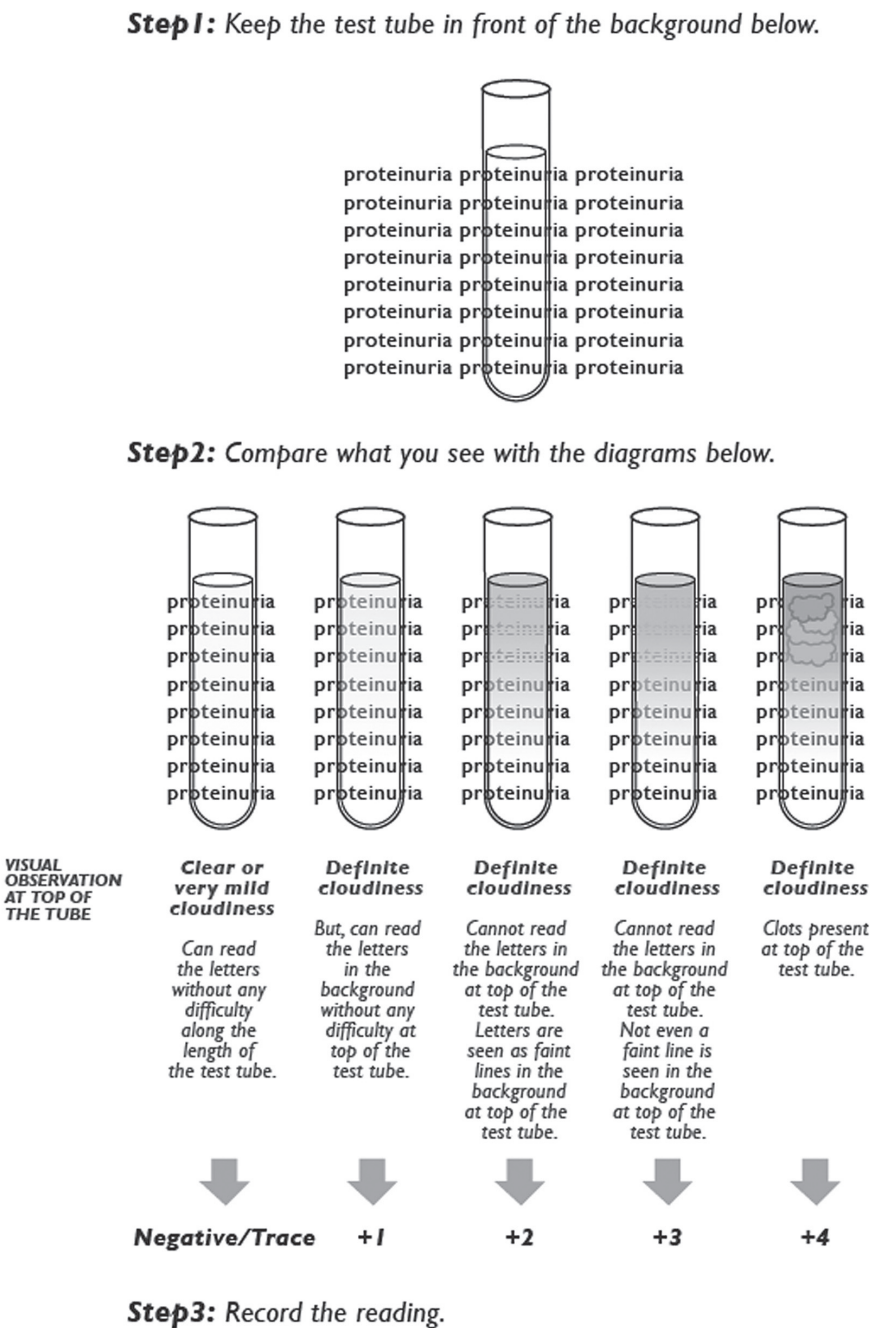

Figure 2.2 Performing the heat coagulation test and interpreting its results

protein, however, it has reported specificity that is more than 90%[20,21].

Sulfosalicylic acid testing

The sulfosalicylic acid (SSA) test is an alternative method of proteinuria testing for under-resourced settings. Ideally, the pH of urine is tested, and if >6, urine is acidified by adding one or two drops of 10% acetic acid. Then, 2 mL of 3% sulfosalicylic acid is added. After shaking the test tube, the turbidity is observed (Figure 2.4) and the tube is placed in front of a black line or bold printed fonts. The turbidity of the urine (as inferred by the ability

to see the black line or printed fonts) is used to infer the presence of proteinuria, as follows: (1) 'negative' when the black line or text is perfectly visible behind the first tube; (2) 'weakly positive' (protein concentration <0.3 g/L) when the black line or text is less visible; (3) 'positive' (protein concentration 0.3–1.0 g/L) when the black line or text is not quite visible; and (4) 'strongly positive' (protein concentration >1.0 g/L) when the black line or text is not visible at all[22].

Interest in using proteinuria testing by SSA as a screening test for proteinuria was based on the test's low cost, good specificity, feasibility and reliability.

Figure 2.3 Heat coagulation test tube showing proteinuria as turbidity at the top of the tube

In the 1980s, WHO recommended SSA testing for use in primary care centres, and two studies evaluated its test performance. Sensitivity and specificity of proteinuria testing in the field by SSA were 94.4% and 96.7% compared with dipstick testing (interpretation by laboratory staff presumed to be visual)[22], and 41.1% and 97.7%, respectively, compared with 24-hour urinary protein[23]. There are no published direct comparisons of the heat coagulation test and SSA. However, given that SSA testing is easier to perform and has similar diagnostic properties (when testing is compared with 24-hour urine testing), SSA testing would seem preferable.

Spot protein : creatinine ratio

Although point-of-care testing for spot PrCr is emerging and PrCr is easily collected by women, all PrCr ratio studies in pregnancy have had measurement of the protein and creatinine concentrations in a random urine sample performed then results calculated in the laboratory (Figure 2.5). There are many assays for proteinuria and creatinine; poor reporting of laboratory methods has prevented an analysis of the impact of various assays on PrCr results. Rapid interpretation has been further complicated by reporting of PrCr results in various units. Nevertheless, the urinary PrCr ratio has been accepted for diagnosis of proteinuria by the International, American, Australasian, Canadian and British pregnancy hypertension societies. In a systematic review, the reported cut-off varied from 17 to 57 mg/mmol (0.15–0.50 mg/mg) (median 24 mg/mmol) in nine studies (1003 hypertensive women). For a cut-off

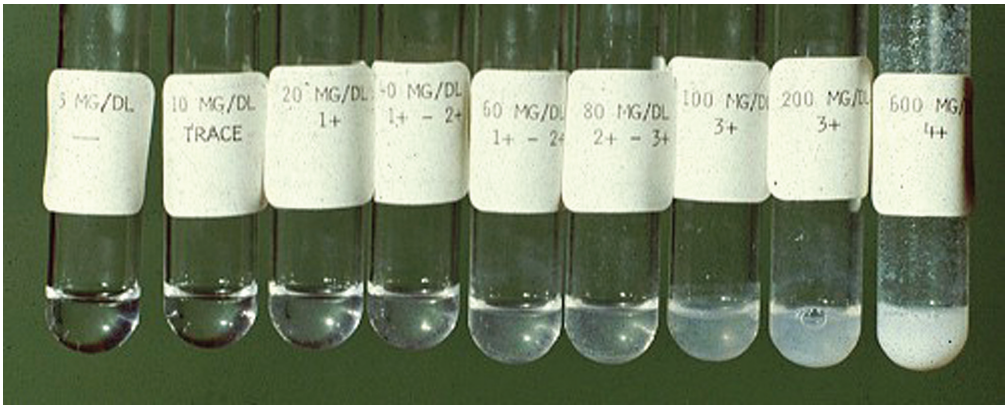

Figure 2.4 Turbidity of the urine after addition of acetic acid as part of sulfosalicylic acid (SSA) testing (from http://www.eclinpath.com/urinalysis/chemical-constituents/urine-protein-ssa/)

Figure 2.5 Woman in Nigeria preparing to collect her spot urine sample for protein : creatinine ration (PrCr) testing

of 30 mg of protein/mmol urinary creatinine, and among women with a hypertensive disorder of pregnancy specifically, the sensitivities and specificities were 83.6% (95% CI 77.5–89.7) and 76.3% (95% CI 72.6–80.0), respectively[24]. A more recent systematic review suggests that the optimum threshold for PrCr ratio to detect significant proteinuria may actually be slightly higher, at 34–40 mg/mmol (0.30–0.35 mg/mg) (summary sensitivity and specificity both >75% for 15 studies, 2790 women), although no threshold gave a sensitivity and specificity >80%[25]. A further meta-analysis of 24 studies (3186 women) endorsed a cut-off of 34 mg/mmol (0.30 mg/mg), with sensitivity and specificity >80%[26]. Four additional studies individually found sensitivity and specificities of at least 80% with optimal cut-offs of 27 mg/mmol (0.24 mg/mg)[27], 30 mg/mmol[28], 51 mg/mmol (0.45 mg/mg)[11], and 53 mg/mmol (0.47 mg/mg)[29], consistent with the previously reported range of 17–57 mg/mmol. One additional report was just outside this range (71 mg/mmol, 0.63 mg/mg)[30], and three others found that optimal cut-offs did not have both sensitivity and specificity ≥80%[31–33]. Taken together, we feel that continued use of the threshold of 30 mg/mmol is reasonable, but do recommend that proteinuria testing be viewed as only *one* aspect of the investigation of women with a hypertensive disorder of pregnancy and interpreted in the context of clinical symptoms, signs and other laboratory testing. A higher threshold may be more appropriate in twin pregnancy[34,35].

The best timing of spot urine sampling is debated. However, timing may not be critical in pregnancy[36–38] which is ideal for women with suspected pre-eclampsia who can be tested for proteinuria at the time of clinical presentation.

Spot albumin : creatinine ratio

Most clinical laboratories use immunoassays to measure urinary albumin, so there is less theoretical inter-laboratory variability for albuminuria than for proteinuria. (The remainder of labs use colourimetric methods that are less precise for low-level albuminuria.) However, there is no standardisation of method, and there are also multiple methods for measuring urinary creatinine, as stated for the PrCr. The impact of laboratory assays on albumin : creatinine ratio (ACR) results is not known.

Urinary ACR testing is available by a variety of point-of-care dipsticks. Three studies have evaluated performance in pregnancy. Two studies found the automated-read ACR dipstick to be insensitive: one used the ACR performed on a spot sample sent to the laboratory as the reference test using a cut-off of 3.4 mg/mmol (65 low risk and 43 high risk pregnancy cases)[39]. The second used 24-hour urinary protein as the reference test; reported sensitivity and specificity were 63% and 81%, respectively (163 hypertensive women)[40]. The third evaluated both visual and automated ACR dipstick performed at the bedside compared with 24-hour urinary protein (171 hypertensive women); automated ACR dipstick fared only slightly better than visual ACR dipstick with regards to sensitivity (i.e., 58% vs. 49%, respectively) and specificities were 83% for both approaches; neither ACR dipstick (visual or automated-read) in that study was better than visual *proteinuria* dipstick testing (which had a sensitivity of 51% and a specificity of 78%) for detection of 0.3 g/d or more of urinary protein in 24-hour collection[15].

Urinary ACR testing on spot urine samples is widely available in clinical laboratories in well-resourced settings. Most, but not all, studies have reported good test performance. The urinary ACR has performed well in: (1) detection of 24-hour urinary protein excretion in four prospective studies[18,41–43] (410 pregnant women), and (2) detection of 24-hour urinary albumin excretion in two other studies[44,45] (119 pregnant

women). An additional study reported that ACR correlated well with 24-hour albuminuria but not with 24-hour proteinuria[46] (31 women diagnosed with pre-eclampsia). Moreover, three different diagnostic cut-offs (of 2, 8 and 22.8 mg/mmol, equivalent to 18, 71 and 205 mg/g) have been reported for significant proteinuria[15,18].

In summary, there is insufficient information about use of ACR testing (by dipstick or through the laboratory) in pregnancy to recommend their use at the present time.

Timed urine collection

Quantification of urinary protein by 24-hour urine collection is considered to be the gold standard. However, 24-hour urine collection is time-consuming, inconvenient and often inaccurate due to inadequate 24-hour urine collection (as assessed by urinary creatinine collection of 13–18% of pre-pregnancy body weight as urinary creatinine (mmol/d))[47]. For diagnosis of proteinuria in non-pregnant populations, these logistical considerations have prompted the National Kidney Foundation and the International Society of Nephrology to abandon timed collections in favour of the spot urine samples[48,49]. However, if quantification of proteinuria is sought, then 24-hour urine collection for protein and creatinine should be used at high levels of proteinuria (i.e., spot PrCr >125 mg/mmol which is roughly equivalent to more than 1 g/d of proteinuria by 24-hour urine collection) as the spot PrCr is less reliable at high levels of proteinuria.

WHAT CONSTITUTES 'SIGNIFICANT' PROTEINURIA IN PREGNANCY?

Although 0.3 g/d of proteinuria represents the upper 95% confidence interval for proteinuria excretion in pregnancy, this threshold does not necessarily identify women at increased risk of adverse maternal and/or fetal outcomes. That threshold is not known.

A recent study reported that women who had ≥0.5 g/d were at higher risk of adverse outcomes than those with 0.3–0.5 g/d[50]. (This is discussed further in Chapter 3.)

In well-resourced settings where full maternal and fetal assessment is available, the magnitude of proteinuria once identified is not related to either short-term adverse maternal or perinatal outcomes, or long-term maternal renal prognosis. In the fullPIERS cohort, a prospective study of women admitted to hospital with pre-eclampsia, the magnitude of proteinuria (by 24-hour urine collection, visual dipstick testing, or spot PrCr) was not associated with adverse maternal or perinatal outcomes independent of routinely collected information on maternal symptoms, signs and basic blood work[51] (see Chapter 3). At least one observational study of women with pre-eclampsia failed to identify a definition of heavy proteinuria that was associated with adverse renal prognosis[52].

In contrast, in resource-poor settings where maternal symptoms and signs alone are used to guide treatment, proteinuria of ≥4+ is associated with an increased risk of stillbirth[53].

COST CONSIDERATIONS

Although visual dipstick proteinuria testing is the most widely used of the screening methods, there is no cost-effectiveness analysis of its use followed by confirmatory testing (with PrCr or 24-hour urine collection) for values ≥1+ or ≥2+.

The only health economic analyses identified were those conducted by the NICE Clinical Guideline Committee, for women with gestational hypertension who live in settings where all tests are available[54]. The Committee considered both the convenience of testing for health care providers and women, and the trade-off between the costs of a false positive test for proteinuria and the costs of missed adverse pregnancy outcomes. The analyses were highly influenced by the sensitivity of proteinuria testing methods. Assuming that sensitivity is high for both the automated dipstick and spot PrCr methods, spot PrCr may be more cost-effective than a strategy of automated dipstick testing *followed by* confirmation of ≥1+ proteinuria by either spot PrCr or 24-hour urine collection.

In low-resource referral hospital settings, limitations in central laboratory facilities will affect cost-benefit considerations.

BEST PRACTICE POINTS

(Please see Appendix 2.3 for the evaluation of the strength of the recommendation and the quality of the evidence on which they are based.)

1. All pregnant women should be assessed for proteinuria, at minimum, at their first antenatal visit.
2. Urinary dipstick testing (or SSA or heat coagulation testing if dipsticks are not available) may be used for screening for proteinuria when the suspicion of pre-eclampsia is low.
3. Significant proteinuria should be strongly suspected when urinary dipstick proteinuria is ≥2+.
4. Definitive testing for proteinuria (by urinary protein : creatinine ratio or 24-hour urine collection) is encouraged when there is a suspicion of pre-eclampsia.
5. Significant proteinuria is ≥0.3 g/d in a complete 24-hour urine collection or ≥30 mg/mmol (≥0.3 mg/mg) urinary creatinine in a random urine sample.
6. There is insufficient information to make a recommendation about the accuracy of the urinary albumin : creatinine ratio, although values <2 mg/mmol (<18 mg/g) are normal and all values ≥8 mg/mmol (≥71 mg/g) are elevated.
7. In well-resourced settings with sophisticated fetal monitoring, proteinuria testing does not need to be repeated once the significant proteinuria of pre-eclampsia has been confirmed.
8. In under-resourced settings, proteinuria testing should be repeated to detect 4+ dipstick proteinuria that is associated with an increased risk of stillbirth.

PRIORITIES FOR UNDER-RESOURCED SETTINGS

Proteinuria testing is recognised by WHO to be as a marker of high quality antenatal care[55]. In fact, proteinuria testing was recommended along with blood pressure monitoring as the original rationale for antenatal care. As such, implementation of proteinuria screening in low- and middle-income countries (LMICs) is a priority.

Demographic health survey data (2002–2008) indicate that few LMICs exceed a standard of urine testing in more than 80% of women attending antenatal care. The rate of urine testing at routine antenatal care visits is highly variable[56], particularly in sub-Saharan Africa and South/Southeast Asia where urine testing rates vary from testing in only 25% of women to testing in close to 100%. Urine testing occurs in at least 50% of women in North Africa/West Asia/Europe and at least 67% of women in Latin American/Caribbean countries. These data indicate a major failure of basic health system provision that inevitably results in avoidable large scale morbidity and mortality from hypertensive disease in pregnancy.

Table 2.2 outlines the priorities for implementation of proteinuria testing in LMICs, depending on the timing of testing (in pregnancy and postpartum) and the level of the health care system. In brief, the first priority is detection of women with pre-eclampsia (by testing for proteinuria at 20 weeks of pregnancy and beyond), followed by detection of women with underlying renal disease (by testing in the first or early second trimester, and at 6 weeks postpartum among women with proteinuria in pregnancy) who are at increased risk of pre-eclampsia.

Innovative proteinuria measurement devices are on the horizon for use in under-resourced settings and it is hoped that they will facilitate implementation of the priorities for testing outlined in Table 2.2. While the priority in high-income settings is towards laboratory-based analyses, the focus in LMICs is on point-of-care testing, particularly by community health care providers. Three active research tracks are as follows:

- *The proteinuria self-test for early detection of pre-eclampsia (the 'proteinuria pen')* was designed by graduate students at John Hopkins University,

Table 2.2 Prioritisation of urine testing for proteinuria by timing and level of health care system at which testing occurs

	Antepartum		*Postpartum*	
	Initial priority	*Ultimate goal*	*Initial priority*	*Ultimate goal*
Community				
Primary health care centre (detect and refer)	Urinary (clean-catch) dipstick testing at each visit after 20 weeks to detect pre-eclampsia	Urinary (clean-catch) dipstick testing at booking and at each visit after 20 weeks to detect both chronic renal disease and pre-eclampsia	Urinary (clean-catch) dipstick testing within 24 hours of delivery in hypertensive women to detect postpartum pre-eclampsia	Urinary (clean-catch) dipstick testing at 6 weeks after delivery for women with antenatal proteinuria to detect underlying renal disease and prompt referral
Facility				
Secondary-level facility (detect and manage/refer) Tertiary-level (referral) facility (detect and manage)	Urinary (clean-catch) dipstick testing at booking and at each visit after 20 weeks to detect both chronic renal disease and pre-eclampsia	Availability of confirmatory test for proteinuria in women with ≥1+ by urinary dipstick testing	Postpartum urinary (clean-catch) dipstick testing within 24 hours of delivery in hypertensive women to detect postpartum pre-eclampsia Postpartum urinary (clean-catch) dipstick testing at 6 weeks after delivery for women with antenatal proteinuria to detect underlying renal disease and prompt referral	

USA (http://www.appropedia.org/Proteinuria_Self-Test_Pen). Field testing is currently under the management of Jhpeigo. This felt-tip or ballpoint pen is filled with reagent that is used to mark a strip of paper. When a drop of urine is placed on the paper, if there is proteinuria, the reagent changes colour. The test is anticipated to cost <US$0.10 per use.

- *Point-of-care paper-based microfluidic diagnostic 'stamps'* have been developed by Diagnostics for All. Paper and an office printer are the equipment required to generate the postage stamp-sized paper testing tool, onto which a reagent and drop of urine are applied to indicate proteinuria (http://www.savinglivesatbirth.net/summaries/60). The test is anticipated to cost <US$0.10 per use).
- The urinary *Congo red dot test* uses a textile dye to detect elevated concentrations of misfolded urinary protein associated with pre-eclampsia[57] (http://www.usaid.gov/news-information/frontlines/open-development-development-defense/pinpointing-preeclampsia-simple-red). Testing requires the user to mix dye and urine together and put a drop on a piece of paper, where dye and any misfolded proteins in the urine combined to form a 'red dot'[57]. The test is anticipated to cost pennies per use.

WHAT INTERNATIONAL GUIDELINES SAY (APPENDIX 2.4)

Abbreviations for Clinical Practice Guidelines: ACOG (American College of Obstetricians and Gynecologists)[58], AOM (Association of Ontario Midwives), NICE (National Institutes of Clinical Excellence)[59], NVOG (National Obstetrics and Gynaecology Society, Netherlands)[60], PRECOG (Pre-eclampsia Community Guideline), PRECOG II (Pre-eclampsia Community Guideline II), QLD (Queensland, Australia)[61], SOGC (Society of Obstetricians and Gynaecologists of Canada)[62], SOMANZ (Society of Obstetric Medicine of Australia and New Zealand)[63], WHO (World Health Organization)[64].

Screening for proteinuria is advocated by five clinical practice guidelines for women with a hypertensive disorder of pregnancy (AOM[65], NICE, PRECOG[66], SOGC, SOMANZ); when performed,

testing methods should be by dipstick (visual) (PRECOG, AOM), automated (NICE), or either (SOGC), but NICE advocates using a random urine protein:creatinine ratio (PrCr) in a secondary care setting. Significant thresholds for proteinuria are: ≥1+ (PRECOG, SOGC) or ≥2+ (PRECOG II[68], QLD), with two guidelines specifying that a threshold of ≥1+ should be used only when there is associated hypertension (PRECOG II) or other manifestations of pre-eclampsia (AOM).

For quantification of proteinuria, criteria are: 'dipstick' ≥1+ (AOM), random urine PrCr ≥30 mg/mmol (PRECOG, PRECOG II, NICE, SOGC), and/or 24-hour urinary protein ≥0.3 g/d (PRECOG, PRECOG II, NICE, NVOG, ACOG SOGC) (with completeness of the urine collection emphasised by two CPGs (NICE, SOGC)).

PRIORITIES FOR FUTURE RESEARCH

- In low-resource country service settings, health systems research is needed on how to ensure consistent proteinuria screening in antenatal care, to the levels that are now being achieved for HIV testing.
- By current testing methods, what is the level of proteinuria that identifies a woman and/or fetus at increased risk of an adverse outcome?
- Are there better ways of measuring proteinuria? These should be cheaper and related to the risk of adverse pregnancy outcome. Three simple approaches, all point of care, show promise.

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