Research

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Nappy pad urine samples for investigation and treatment of UTI in young children:

the 'DUTY' prospective diagnostic cohort study

Abstract

Background
The added diagnostic utility of nappy pad urine samples and the proportion that are contaminated is unknown.

Aim
To develop a clinical prediction rule for the diagnosis of urinary tract infection (UTI) based on sampling using the nappy pad method.

Design and setting
Acutely unwell children <5 years presenting to 233 UK primary care sites.

Method
Logistic regression to identify independent associations of symptoms, signs, and urine dipstick test results with UTI; diagnostic utility quantified as area under the receiver operator curves (AUROC). Nappy pad rule characteristics, AUROC, and contamination, compared with findings from clean-catch samples.

Results
Nappy pad samples were obtained from 3205 children (82% aged <2 years; 48% female), culture results were available for 2277 (71.0%) and 30 (1.3%) had a UTI on culture. Female sex, smelly urine, darker urine, and the absence of nappy rash were independently associated with a UTI, with an internally-validated, coefficient model AUROC of 0.81 (0.87 for clean-catch), which increased to 0.87 (0.90 for clean-catch) with the addition of dipstick results. GPs' 'working diagnosis' had an AUROC 0.63 (95% confidence intervals [CI] = 0.53 to 0.72). A total of 12.2% of nappy pad and 1.8% of clean-catch samples were 'frankly contaminated' (risk ratio 6.66; 95% CI = 4.95 to 8.96; $P<0.001$).

Conclusion
Nappy pad urine culture results, with features that can be reported by parents and dipstick tests, can be clinically useful, but are less accurate and more often contaminated compared with clean-catch urine culture.

Keywords
antibacterial agents; diagnosis; infant; paediatrics; primary health care; urinary tract infections.

INTRODUCTION

Urinary tract infection (UTI) may be missed in up to 80% of children presenting to primary care.[1,2] Accurate diagnosis of UTI is essential to avoid over- or undertreatment with antibiotics and to appropriately target burdensome and expensive investigations.[3] This is especially important in younger, pre-verbal children who are not yet toilet-trained and who often present with non-specific symptoms, making the decision about which children to investigate for UTI difficult.[3] Obtaining a urine sample can be time consuming and especially challenging in primary care, where most children first present.[4] The nappy pad sampling method in young children in nappies (diapers), when a clean-catch sample cannot be obtained, has been recommended by the National Institute for Health and Care Excellence (NICE).[3] Urine sampling needs to be simple, reliable, and acceptable, and parents find nappy pads easy to use, comfortable, for their children and prefer them to the clean-catch method.[5] Nappy pad sampling is used in everyday care,[1] and GPs report using nappy pad urine collection in over 40% of infants.[6] Many parents feel that the clean-catch method is messy and time consuming and give up trying.[3,5] However, the clinical utility of the information obtained from the nappy pad method of urine sampling is unclear, contamination rates may be higher than other sampling methods, and

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How this fits in

Up to 80% of urinary tract infections (UTI) in young children presenting to primary care are missed. Timely and accurate diagnosis is essential to avoid over- or undertreatment and investigation. This is especially difficult in pre-verbal children who are not toilet trained, and present with undifferentiated symptoms. GPs use, and parents prefer, nappy pads for collecting urine from children who are still in nappies, but the clinical utility of data derived from nappy pad samples, the added value of dipstick testing, and the proportion of contaminated samples is not known. It was found that culture results from urine obtained using nappy pads, together with features that can be reported by parents, can be clinically useful in identifying acutely unwell pre-school children presenting to primary care who have a UTI, but with less accuracy compared to clean-catch sampling. However, contamination rates are nearly seven times higher in nappy pads than in clean-catch samples. Clean-catch urine sampling in children in primary care should therefore be prioritised over the nappy pad method, but if urine sampling is done using nappy pads, then the addition of dipstick testing significantly improves diagnostic accuracy.

children in nappies present differently to older children who are more able to describe symptoms and in whom clean-catch sampling is easier. Obtaining urine samples by more invasive methods such as suprapubic aspiration or catheterisation is neither feasible nor acceptable in most primary care settings.

The aim of the study therefore was to develop a clinical prediction rule for the diagnosis of UTI based on sampling using the nappy pad method, and compare its diagnostic utility to a similar rule based on 'clean-catch' urine samples.[7] In addition, the added diagnostic value of dipstick testing once a nappy pad sample had been obtained was estimated, and contamination rates were compared by the sampling method.

METHOD
Participants
The Diagnosis of Urinary Tract infection in Young children (DUTY) study was a multicentre, prospective, diagnostic cohort study that recruited children aged <5 years, in primary care.[8] Children were eligible if presenting with any acute (<28 days), undifferentiated illness (even when the clinician was confident of the diagnosis,

such as a child with bronchiolitis), and/or new urinary symptoms.

Index tests and urine collection
Following consent, 107 index test items were recorded. Parent-reported items included the child's medical history and symptoms. Clinician-assessed items came from a full clinical examination, including their global impression of illness severity (rated 0–10), their rating of the likelihood of UTI, and urine dipstick results (performed after rating UTI likelihood). Index test items were derived from a literature review and input from the co-investigator group.

The NICE recommended 'Newcastle Nappy Pads' were used for those children who wore nappies (diapers) and for those in whom the parent/guardian did not think clean-catch would be successful.[3,8,9] First, the parent was asked to clean the nappy area using water or wipes (the wipes being supplied by the study). A nappy pad was inserted inside a clean nappy, and the nappy refastened. The nappy pad was removed as soon as the child urinated, in order to reduce the risk of contamination. The perineum was cleaned again and a fresh pad inserted every 30 minutes until micturition, or immediately if the pad became contaminated with faeces. Once the child had urinated, the research nurse or clinical study officer (RN/CSO), wearing disposable gloves, removed the pad and urine was extracted into a sterile container as per the manufacturer's instructions (Newcastle Urine Collection Pack; Ontex Ltd, Corby, UK; NHS Supplies). If it was not possible to obtain a sample before the child left the primary care site, the parent was given the necessary equipment and advice on obtaining a urine sample at home. The parent was advised to store the sample in the fridge and return it to their primary care site as soon as possible, ideally within 24 hours. The RN/CSO telephoned parents the next day to remind them to return the sample. Where feasible, the RN/CSO offered to collect the urine sample from the child's home.

At the primary care site, urine samples were dipstick tested (using Siemens/Bayer Multistix 8SG) for blood, protein, glucose, ketones, nitrite, leukocyte esterase, pH, and specific gravity (eight dipstick index tests). All index tests and the clinician's working diagnosis ('clinical diagnosis') were measured blind to the reference standard.

When there was at least 1 mL of urine leftover after the priority sample was sent to a NHS laboratory, it was sent, in boric acid monovettes, using first class Royal Mail Safeboxes™, to the Public Health Wales

Microbiology NHS Laboratory in Cardiff (a research laboratory). Results from the research laboratory are used in the current analyses.

Reference standard

The research laboratory spiral-plated (Don Whitley Scientific, Shipley, UK) 50 µL onto chromogenic agar and standard blood agar. A full description of the methods used in the research laboratory, and how these differed from the range of standard operating procedures in local NHS laboratories, is forthcoming.[10] Quantitative total counts were recorded for up to six organisms and the presence of antimicrobial substances measured. Samples were processed using a single, standardised procedure. Uropathogens were defined as members of the *Enterobacteriaceae* group. The microbiological definition of UTI used was the presence of $\geq 10^5$ colony-forming units (CFU)/mL of a single uropathogen ('pure growth'), or $\geq 10^5$ CFU/mL of a uropathogen with a ≥ 3 $\log_{10}$ (1000-fold) difference between the growth of this and the next species ('predominant growth'). Aside from the children's dates of birth, laboratory staff were blind to index tests. As there is no single accepted definition of contamination, three definitions were considered: growth of >2 organisms of $>10^5$ CFU/mL ('frank contamination');[11] growth of ≥ 2 organisms at $>10^5$ CFU/mL ('heavy contamination' according to Rao *et al*[12] and Feasey[13]), and growth of >2 organisms at $>10^4$ CFU/mL ('probable contamination' according to Jackson *et al*[1]), or 'frank contamination' according to Bekeris *et al*[14]).

Statistical analysis

The frequency of symptom and sign categories were examined, blind to their associations with urine culture results, and the least frequent categories were merged before analyses. Logistic regression was used to estimate associations of index tests with urine culture positivity. *P*-values were derived using likelihood ratio tests. For ordinal variables, both heterogeneity and trend *P*-values were derived. Continuous variables were divided into quintiles and trend *P*-values were derived using the median within categories. Plots of the log odds of culture positivity were examined against the median within quintiles for evidence of non-linearity.

Two methods for dealing with missing data were used, including the response 'don´t know' to questions about the presence of symptoms such as pain or crying, when passing urine. In both univariable and multivariable analyses' missing data were coded as the modal value, usually as absence of the symptom. Multivariable analyses were repeated using the chained equations approach to multiple imputation: estimates and Wald *P*-values[15] based on 50 imputed datasets derived using Rubin's rules.[16]

Prediction rules were derived in three stages. First, symptoms and signs were selected, with either trend or heterogeneity univariable *P*-values <0.01. Second, models were derived from selected symptoms and signs using backwards stepwise selection and an exclusion criterion of heterogeneity *P*-value >0.1. Third, backwards stepwise selection was used, with the same exclusion *P*-value for models in which dipstick results were added. The effect of using more liberal *P*-value thresholds of 0.1 and 0.2 at the first stage was investigated, and no important differences were found in the final models (results available from the authors on request).

Diagnostic accuracy was quantified as the area under the receiver operating characteristic (AUROC) curve (also known as the 'c-statistic'). AUROC values for clinical judgement of UTI were also estimated. Internal validation of the models was conducted using the bootstrap procedure described by Steyerberg[17] and a calibration slope (shrinkage factor) was calculated, by which model coefficients were multiplied, in order to derive internally-validated odds ratios (OR). For each model, cut-points corresponding to a range of values for sensitivity were selected, and then the corresponding specificity, negative and positive predictive values, and the proportion of children classified positive, were calculated. These were compared against 'clinical diagnosis' of UTI (where clinicians considered UTI to be 'fairly' or 'very' certain). Models were re-run, leaving out predictive features that could lack face validity.

Sample size calculation

The sample size calculation assumed a candidate predictor with 10% prevalence and UTI prevalence of 2%. With 80% power and a two-sided α of 5%, 3000 urine sample results were required to detect an OR of 2.4, while 3100 results would give a 95% confidence interval [CI] with width 10% for an algorithm with 80% sensitivity. It was originally proposed to recruit 4000 children, combining analyses for children with both clean-catch and nappy pad samples, anticipating recovery of urine samples from 77.5% for algorithm derivation, and a

Table 1. Nappy pad samples: children's characteristics and crude odds ratios for index tests associated with UTI

Demographics/index tests	N (%)[a]	UTI (%)[b]	Crude OR[c]	95% CI
Total	2277	30 (1.3)		
Age of child				
<6 months	369 (16.2)	5 (1.4)	1.72	0.54 to 5.46
6 to <12 months	603 (26.5)	11 (1.8)	2.33	0.90 to 6.04
1 to <2 years	884 (38.8)	7 (0.8)	1	ref
2 to <3 years	353 (15.5)	7 (2.0)	2.53	0.88 to 7.28
3 to <4 years	58 (2.5)	0 (0.0)	n/a	
≥4 years	10 (0.4)	0 (0.0)	n/a	
Time from index tests to taking urine sample				
Sample before recruitment	120 (5.3)	2 (1.7)	1.33	0.31 to 5.67
<24 hours	1982 (87.0)	25 (1.3)	1	ref
24 to <48 hours	109 (4.8)	3 (2.8)	2.22	0.66 to 7.45
48 to <72 hours	18 (0.8)	0 (0.0)	n/a	
≥72 hours	48 (2.1)	0 (0.0)	n/a	
Clinician diagnosis prior to dipstick				
No UTI certain to very certain	1033 (45.4)	8 (0.8)	0.52	0.22 to 1.19
Uncertain or no UTI fairly certain	1201 (52.7)	18 (1.5)	1	ref
UTI fairly to very certain	38 (1.7)	4 (10.5)	7.76	2.49 to 24.18
Missing	5 (0.2)	0 (0.0)		
Sex				
Male	1183 (52.0)	9 (0.8)	1	ref
Female	1094 (48.0)	21 (1.9)	2.55	1.16 to 5.60
Smelly urine				
No problem	1518 (66.7)	12 (0.8)	1	ref
Slight problem	206 (9.0)	4 (1.9)	2.20	0.73 to 6.61
Moderate problem	138 (6.1)	5 (3.6)	4.18	1.52 to 11.50
Severe problem	26 (1.1)	4 (15.4)	20.21	6.29 to 64.97
Missing/not known	389 (17.1)	5 (1.3)		
Darker urine				
No problem	1764 (77.5)	19 (1.1)	1	ref
Slight problem	83 (3.6)	2 (2.4)	2.19	0.51 to 9.43
Moderate or severe problem	41 (1.8)	4 (9.8)	9.59	3.17 to 29.02
Missing/not known	389 (17.1)	5 (1.3)		
Nappy rash[d]				
No problem	1715 (75.3)	29 (1.7)	1	ref
Slight to severe problem	560 (24.6)	1 (0.2)	0.10	0.01 to 0.77
Missing	2 (0.1)	0 (0.0)		
Dipstick: leucocytes				
Negative	1759 (77.3)	13 (0.7)	1	ref
Trace	125 (5.5)	1 (0.8)	1.09	0.14 to 8.38
+	119 (5.2)	4 (3.4)	4.69	1.50 to 14.61
++	177 (7.8)	4 (2.3)	3.12	1.01 to 9.66
+++	91 (4.0)	8 (8.8)	12.99	5.24 to 32.20
Missing	6 (0.3)	0 (0.0)		
Dipstick: nitrites				
Negative	1916 (84.1)	13 (0.7)	1	ref
Positive	355 (15.6)	17 (4.8)	7.39	3.55 to 15.35
Missing	6 (0.3)	0 (0.0)		

OR = odds ratio. ref = reference. UTI = urinary tract infection. [a]Percentage relative to the total number of observations (N = 2277). [b]Percentage relative to the total number of observations within that category. [c]Crude ORs calculated using modal imputation.

nappy pad results to derive the model, and to conduct internal bootstrap validation instead of external validation.

RESULTS

Participants

Participants from 233 primary care sites (225 GP practices, four walk-in centres and four children's emergency departments [EDs]) across England and Wales between April 2010 and April 2012, were recruited. Of 10 138 children who screened eligible, 1276 (12.6%) declined participation, 1684 (16.6%) could not be recruited for other reasons, and 15 (0.15%) withdrew.

Of the 233 primary care sites taking part, 198 sites (85%) completed and returned at least one screening log to the study centre. These showed that 7350 children were screened but not recruited because they declined (n = 1276), were not eligible (n = 4390), or for other reasons (n = 1684), which included: left the primary care site prior to invitation (n = 811); did not give consent (n = 214); or there was a language barrier (n = 112) and an appropriate translator was not available at the time of recruitment. There were slightly more males (mean difference of 5.2%; 95% = CI 2.2 to 8.2%) among those for whom participation was declined (n = 1276) compared with those who did agree to participate in the DUTY study (n = 7163). The mean age in the declined sample was 24.06 months versus 26.88 months among participants (mean difference 2.04 months, 95% CI = 1.08 to 3.00 months). Clinical information on those who declined was not collected.

Urine was collected from a total of 6241 children, 3205 using nappy pads. A total of 3164 (98.7%) nappy pad samples were sent to NHS laboratories, and 2363 (73.7%) to the research laboratory. The number of reference standard results available from the research laboratory (the final analytical sample) was 2277 (71.0%). A total of 82% of children providing nappy pad samples were aged <2 years (mean 1.3 years, SD 0.8), and the mean illness severity score was 2.3 points (SD 1.5); 48% of children were female; and overall, 1.3% had a UTI (Table 1). The clean-catch sample (n = 2740) had a mean age of 3.5 years (SD 1.0), mean illness severity score of 2.2 points (SD 1.6), and 53.8% were female.

In total, 2102 (92.3%) samples were provided within 24 hours of the index test measurement, and there was no relationship between UTI and time from urine collection to laboratory arrival. Antimicrobial substances, which can arise from the use of both systemic antibiotics and

further 2000 children for external validation. However, the need to stratify and report separate analyses by the urine collection method was not anticipated. For the present analysis, it was decided to use all available

Table 2. Nappy pad samples: models based on symptoms and signs; and on symptoms, signs, and dipstick results, including results based on multiple imputation

Index tests	Symptom and sign model				Symptom sign and dipstick model			
	Adjusted OR[a]	95% CI[a]	MI adjusted OR	95% CI	Adjusted OR	95% CI[a]	MI adjusted OR[a]	95% CI
Sex								
Male	1	ref	1	ref	1	ref	1	ref
Female	1.95	1.11 to 3.41	1.96	1.06 to 3.61	1.41	0.80 to 2.48	1.45	0.78 to 2.72
Smelly urine								
No problem	1	ref	1	ref	1	ref	1	ref
Slight problem	1.61	0.73 to 3.54	1.97	0.82 to 4.71	1.44	0.67 to 3.11	1.79	0.76 to 4.23
Moderate problem	2.51	1.14 to 5.51	3.39	1.46 to 7.89	2.15	0.98 to 4.68	2.96	1.26 to 6.97
Severe problem	7.40	2.98 to 18.36	10.14	3.85 to 26.69	3.97	1.58 to 9.96	6.13	2.28 to 16.47
Darker urine								
No problem	1	ref	1	ref	1	ref	1	ref
Slight problem	1.89	0.66 to 5.46	1.99	0.65 to 6.12	1.81	0.65 to 5.07	1.92	0.63 to 5.88
Moderate or severe problem	2.46	0.98 to 6.21	2.26	0.85 to 6.01	2.29	0.93 to 5.62	2.27	0.87 to 5.93
Nappy rash								
No problem	1	ref	1	ref	1	ref	1	ref
Slight to severe problem	0.16	0.04 to 0.66	0.13	0.03 to 0.61	0.19	0.05 to 0.71	0.16	0.04 to 0.66
Dipstick: leukocytes								
Negative					1	ref	1	ref
Trace					0.87	0.23 to 3.31	0.81	0.18 to 3.61
+					2.06	0.92 to 4.63	2.18	0.88 to 5.43
++					1.63	0.73 to 3.62	1.78	0.73 to 4.30
+++					3.27	1.66 to 6.41	3.35	1.57 to 7.15
Dipstick: nitrites								
Negative					1	ref	1	ref
Positive					3.16	1.91 to 5.24	3.70	2.10 to 6.52
Receiver operating characteristic curve								
ROC	0.769	0.68 to 0.85	0.805	0.72 to 0.89	0.858	0.79 to 0.93	0.870	0.80 to 0.94
Validated ROC[b]	0.744		0.778		0.799		0.821	
Δ ROC[c]					0.089	0.02 to 0.16	0.065	0.00 to 0.13
Δ ROC[c] P-value					0.012		0.036	
Calibration slope	0.695		0.749		0.647		0.708	

OR = odds ratio. MI = multiple imputation. ref = reference. ROC = receiver operating characteristic. [a]Missing values coded to modal category. [b]Internal validation using the bootstrap procedure. [c]The difference in ROC between symptom and sign model and symptom, sign and dipstick model. OR calculated using shrunken estimates from the bootstrap internal validation calibration slope.

Figure 1. Receiver operating characteristic curves for symptoms and signs model (solid line), and symptoms, signs and dipstick model (dotted line) for nappy pad urine samples for diagnosing UTI in young children.
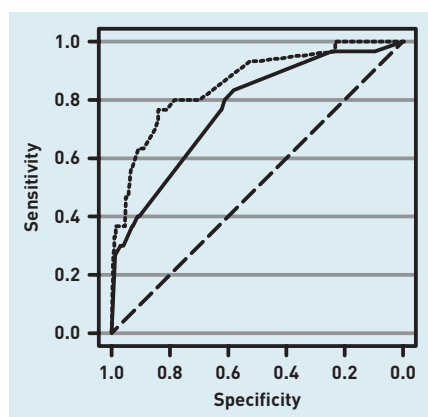
locally applied cleaning agents, were found in 6.6% of nappy pad samples, and were more likely to be present in children with, than without, UTI. GPs' 'working diagnosis' would have correctly identified four (13.3%) of the 30 UTIs, with 97.0% specificity and an AUROC 0.63 (95% CI = 0.53 to 0.72).

Nappy pad model
Table 2 shows adjusted ORs for the index tests (Table 1) selected for the nappy pad model. Parent-reported smelly urine, darker urine, female sex, and the absence of a nappy rash, were independently associated with UTI: for the first two, there was evidence of graded associations. No clinical examination findings were independently associated with UTI. The presence of leukocytes and nitrites from dipstick urine testing were independently associated with UTI. The symptoms and signs model had reasonable diagnostic accuracy (validated AUROC for the multiple imputed model was 0.78 and diagnostic accuracy increased [P = 0.036] with addition of dipstick findings [validated AUROC 0.82]). Figure 1 shows the multiple imputed receiver operating characteristic curves for the models with and without dipstick urinalysis.

The multiple imputation analysis was re-run, excluding the nappy rash variable, and it was found that there was a reduction of 0.07 in validated AUROC in the symptom and sign model (from 0.78 to 0.71), and a reduction of 0.03 in the validated AUROC in the symptom, sign, and dipstick model (from 0.82 to 0.80). The association between antimicrobial substances in the urine and nappy rash was checked, and no association was found (P = 0.82).

Table 3. Numbers of contaminated samples using different definitions of contamination

Study	Contamination definition	Clean-catch contaminated, n (%)	Nappy pad contaminated, n (%)	Risk difference, 95% CI	Risk ratio, 95% CI	P-value
Jackson *et al*, 2005[11] frank contamination	>10^5 >2 organisms	50/2740 (1.8)	277/2277 (12.2)	0.103 (0.089 to 0.118)	6.666 (4.959 to 8.963)	<0.001
Feasey, 1999[13] or Rao *et al*, 2004[12] heavy contamination	>10^5 ≥2 organisms	78/2740 (2.8)	426/2277 (18.7)	0.159 (0.141 to 0.176)	6.572 (5.196 to 8.312)	<0.001
Jackson *et al*, 2005[11] or Bekeris *et al*, 2008[14] probable or frank contamination	>10^4 >2 organisms	175/2740 (6.4)	599/2277 (26.3)	0.199 (0.179 to 0.219)	4.119 (3.513 to 4.829)	<0.001

Frank contamination is the definition of contamination that was used in the subsequent analyses.

Comparison of findings using nappy pads and catch samples

The validated AUROC for the nappy pad model was inferior to the model derived using clean-catch samples, which was 0.87 for symptoms and signs, increasing to 0.90 with dipstick results (to be reported fully in a future publication). Table 3 provides the proportion of nappy pad and clean-catch samples considered to be contaminated according to three published definitions.[11–14] 'Frankly contaminated' urine was found in 12.2% of nappy pad and 1.8% of clean-catch samples, risk ratio 6.66 (95% CI = 4.95 to 8.96; P<0.001).

DISCUSSION

Summary

Four features (female sex, smelly urine, darker urine, and absence of nappy rash) that could be reported by parents, and no clinical signs, were associated with a microbiological diagnosis of UTI in children sampled using nappy pads. These features were substantially more predictive of a microbiological diagnosis of a UTI than clinicians' 'working' or clinical diagnoses, but less predictive than data obtained for older children sampled using the clean-catch methods. More than 10% of samples obtained by nappy pads were 'frankly' contaminated, compared with <2% of samples obtained by clean-catch from predominantly older children. The addition of dipstick testing improved diagnostic accuracy of nappy pad samples.

Strengths and limitations

The DUTY study is the largest primary care diagnostic accuracy study of clinical symptoms, signs, and dipstick tests for diagnosing UTI in young children, and it achieved high levels of data completeness. Clinicians were asked to obtain a clean-catch urine sample whenever possible, but ultimately, the decision whether to sample by clean-catch or the nappy pad method was up to the parents, who generally used the nappy pad method in younger children. Children sampled using the nappy pad method were therefore younger and may have differed in other ways as well, for example being more unwell or distressed. Parents were asked to replace pads at regular intervals until a sample was obtained. However, alarms were not used to trigger a scheduled replacement of pad, and this may have led to increased contamination rates.[12]

There was a relatively small number of UTIs diagnosed microbiologically in this study, with fewer diagnosed from the fraction of the urine samples sent to the research laboratory compared to the fraction of the samples sent to the NHS laboratories, perhaps because of more intensive methods used in the research laboratory resulting in fewer false-positives. It is plausible that the nappy pad contamination masked the presence of UTI leading to underdiagnosis in comparison with clean-catch samples, in which lower contamination and higher UTI rates were observed. The conservative criteria for a microbiological UTI diagnosis may have also contributed.

This study reference standard defined 'uropathogens' as members of the *Enterobacteriaceae* family at the UK guidelines'[19,20] threshold of a pure/predominant growth of ≥10^5 CFU/mL. It was decided not to use a lower threshold, as this carries an increased risk of false-positives, although there are recommendations that a lower threshold should be used.[3,21,22] The diagnosis of UTI is a clinical one, taking microbiological analysis into account, and a lower microbiological threshold in the presence of high clinical suspicion would be acceptable for the purposes of clinical care

as opposed to this diagnostic study. This study used a rigorous criterion (minimum 3-log difference between the predominant and next most concentrated organism) for defining predominance. This definition could have reduced estimated prevalence if some UTIs were incorrectly classified as contamination. In addition, a small proportion of the positive cultures may have been false-positives due to asymptomatic bacteriuria or contamination.

Collecting an uncontaminated urine specimen is most difficult in the youngest children, and no method has yet been found to reliably distinguish pathogen from contaminant, especially when they coexist. The study's definition of UTI excluded atypical bacteria causing UTIs, which are also thought to be more common in younger children, potentially reducing this estimated UTI prevalence.[19,23]

Comparison with existing literature

The NICE guidelines found 'insufficient data to draw conclusions about urine collection bags and urine collection pads', but recommended their use when a clean-catch sample cannot be obtained.[3] The authors have been unable to identify further studies addressing nappy pads since the NICE recommendations were published.[24] A systematic review of the accuracy of specimens obtained from nappy pads included three studies that compared sampling by nappy pad to sampling by urine bag, and one study that compared nappy pad specimens to specimens obtained by suprapubic aspiration. The latter study found 100% sensitivity and 94% specificity between the two methods. A randomised trial found that replacing pads every 30 minutes until a sample was obtained, reduced contamination.[12]

The authors found a 1.3% prevalence of microbiological diagnoses of UTI. The only other UK primary care study found a 6% prevalence when urine samples were analysed in NHS laboratories.[2] A similar UTI prevalence of 5.6% for the DUTY study urine samples overall in NHS laboratories was found.[18] Fever was not an inclusion criterion in that study or in the present study. However, a systematic review of 10 studies, eight of which were conducted in a hospital ED, with one in a clinic and ED setting, and one in a clinic setting, and all conducted in the US apart from a clinic study in Taiwan, found a UTI prevalence of 7% among infants presenting with fever.[25] An Australian study (with incomplete urine sampling) found a prevalence of 3.4% children presenting with fever to EDs.[26]

The authors' systematic search identified one systematic review that included eight primary studies of 7892 children aged <5 years[24] and three further primary studies[26–28] that included 17 462 children, that assessed associations between clinical features symptoms and signs and a UTI diagnosis. The data found showed that no individual symptom or sign or combination of symptoms or signs was sufficient to rule in a diagnosis of UTI. Among the remaining studies, largely conducted in hospital EDs, abdominal pain, back pain, dysuria, frequency, and new-onset urinary incontinence increased the likelihood of a UTI.[29] Stridor, audible wheeze, circumcision, temperature <39°C with a source, abnormal chest sounds, chest crackles, age ≤3 years, not feeling hot, and breathing difficulty decreased the likelihood of UTIs. The largest study, which included almost 16 000 children aged <5 years presenting to EDs in Australia,[26] derived a diagnostic model based on a combination of 27 symptoms and signs. However, this study did not involve systematic urine sampling, and most children did not have urine sampled. This model was found to have an AUROC of 0.80 (95% CI = 0.78 to 0.82), which is similar to findings from this DUTY study for children sampled with nappy pads.

Previous investigation of malodorous urine has shown conflicting results,[30] but the present study strongly supports its diagnostic value. The authors investigated, but did not find evidence for, a number of non-specific symptoms (including fever, vomiting, lethargy, irritability, and poor feeding) previously found to be associated with UTI[24] and recommended for clinical use by NICE.[3] It remains possible that such symptoms are of use in the secondary care settings in which studies reporting their utility were conducted, or in children with a different illness spectrum. This finding underlines the importance of including a wide range of illness presentation in studies of predictors of diagnoses, especially when symptoms and signs are notoriously non-specific. Studies that include only children with symptoms and signs previously found to be associated with the diagnosis risks missing previously unidentified predictors and 'research circularity' (looking for, and finding, symptoms and signs in children included in studies if they have those symptoms and signs).

The reduction in the risk of UTI associated with presence of a nappy rash should be interpreted with caution. The inverse association may arise through lower likelihood of a UTI when there is a

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plausible alternative diagnosis (conditioning on the common effect of primary care attendance).[31] Alternative explanations are that rash may be a risk factor for contamination of urine, and this masks the presence of a UTI, or that skin products used to treat nappy rash could render the urine sterile. However, the authors found no evidence of an association between nappy rash and contamination, nor the presence of antimicrobial substances in the urine and nappy rash. An increased likelihood of contamination of nappy pad samples could also explain the more modest associations of symptoms and dipstick test results with UTI than were found in clean-catch samples.[10] The models were re-run excluding nappy rash and modest reductions were found in the symptoms and signs, and symptoms, signs, and dipstick model AUROCs (0.07 and 0.03, respectively).

Implications for research and practice
Nappy pad urine sample culture results, together with symptoms that can be reported by parents can be clinically useful in identifying acutely unwell pre-school children presenting to primary care who have a UTI, but with less accuracy, and with increased contamination compared with clean-catch sampling. Clean-catch urine sampling in children in primary care should be prioritised, especially in children with nappy rash. However, if sampling is done using nappy pads, then the addition of dipstick testing significantly improves diagnostic accuracy.

Further research is needed to distinguish pathogens from contaminants, and to establish the cost-effectiveness of different sensitivity and specificity cut points using routine health service laboratory results. It is not known precisely how results from clean-catch sampling compare with nappy pad sampling in younger children; contamination may vary by age as well as by sampling method. Randomising children to the sampling method could shed light on this.

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