Diagnosing Significant PDA Using Natriuretic Peptides in Preterm Neonates: A Systematic Review

Madhulika Kulkarni, MD; Ganga Gokulakrishnan, MD; Jack Price, MD; Caraciolo J. Fernandes, MD; Mariska Leeflang, PhD; Mohan Pammi, MD, PhD

abstract

BACKGROUND AND OBJECTIVES: Echocardiogram is the gold standard for the diagnosis of hemodynamically significant patent ductus arteriosus (hsPDA) in preterm neonates. A simple blood assay for brain natriuretic peptide (BNP) or amino-terminal pro-B-type natriuretic peptide (NT-proBNP) may be useful in the diagnosis and management of hsPDA. Our objectives were to determine the diagnostic accuracy of BNP and NT-proBNP for hsPDA in preterm neonates and to explore heterogeneity by analyzing subgroups.

METHODS: The systematic review was performed as recommended by the Cochrane Diagnostic Test Accuracy Working Group. Electronic databases, conference abstracts, and cross-references were searched. We included studies that evaluated BNP or NT-proBNP (index test) in preterm neonates with suspected hsPDA (participants) in comparison with echocardiogram (reference standard). A bivariate random effects model was used for meta-analysis, and summary receiver operating characteristic curves were generated.

RESULTS: Ten BNP and 11 NT-proBNP studies were included. Studies varied by methodological quality, type of commercial assay, thresholds, age at testing, gestational age, and whether the assay was used to initiate medical or surgical therapy. Sensitivity and specificity for BNP at summary point were 88% and 92%, respectively, and for NT-proBNP they were 90% and 84%, respectively.

CONCLUSIONS: The studies evaluating the diagnostic accuracy of BNP and NT-proBNP for hsPDA varied widely by assay characteristics (assay kit and threshold) and patient characteristics (gestational and chronological age); therefore, generalizability between centers is not possible. We recommend that BNP or NT-proBNP assays be locally validated for specific patient population and outcomes, to initiate therapy or follow response to therapy.
Brain natriuretic peptide (BNP) and amino-terminal pro-B-type natriuretic peptide (NT-proBNP) are synthesized and released into the circulation by the cardiac ventricular myocytes in response to pressure overload, volume expansion, and increase in myocardial wall stress. Within the myocytes, the precursor pro-BNP (108 amino acids) is converted to the biologically active form, BNP (77–108 amino acids) and the inactive NT-proBNP fragment (1–76 amino acids). BNP improves myocardial relaxation and regulates responses to acute increases in ventricular volume by opposing the vasoconstriction, sodium retention, and antidiuretic effects of the activated renin-angiotensin-aldosterone system. Plasma BNP and NT-proBNP are cleared by the kidneys and hence elevated in patients with renal failure. NT-proBNP has a longer half-life (60–120 minutes vs 22 minutes) and is more stable in vitro. BNP and NT-proBNP are well-established markers of heart failure in adults and children. Many commercial kits are available for assessment of BNP and NT-proBNP, and levels reported vary with chronological age, gestational age (GA), PDA, and renal function. The normative values of BNP and NT-proBNP in neonates from various studies are presented in Supplemental Table 3. BNP and NT-proBNP values are higher in the first 3 days of life and decline progressively thereafter. Levels in preterm neonates are higher than in full-term neonates and are similar by 1 month of life. NT-proBNP values are 6 to 20 times higher than BNP values, and the ratio between NT-proBNP and BNP is influenced by age.

BNP and NT-proBNP assays have been reported in the management of multiple neonatal conditions including hsPDA, BNP and NT-proBNP have been used in preterm neonates both for diagnosis and to initiate medical or surgical treatment of hsPDA. The diagnostic accuracy of BNP and NT-proBNP in the management of hsPDA in neonates has not been systematically reviewed.

Our primary objective was to determine the diagnostic accuracy of cardiac biomarkers BNP and NT-proBNP in the diagnosis of hsPDA in preterm neonates.

The secondary objective was to explore heterogeneity among studies evaluating BNP and NT-proBNP by analyzing the following subgroups: commercial assay, test threshold, age of the patient at testing, GA at birth, and whether the test was used to initiate medical or surgical treatment.

METHODS
The method recommended by the Cochrane Diagnostic Test Accuracy Working Group was followed (http://srtda.cochrane.org/). The title has been registered with the Cochrane Neonatal Review Group.

Criteria for Consideration of Studies for Review
Prospective and retrospective studies that evaluated blood BNP or NT-proBNP (index tests) in the diagnosis of PDA (target condition) in preterm neonates in conjunction with an echocardiogram (reference standard) were eligible for inclusion. Studies were excluded in which a threshold was not reported or could not be obtained from the authors. There is no consensus for echocardiographic criteria for hsPDA. We chose 2 echocardiographic criteria to define hsPDA in preterm infants based on the best available evidence: LA/Ao ratio >1.5 and ductal diameter >1.5 mm (as measured by color Doppler), which are early predictors of hsPDA. LA/Ao ratio is estimated from the diameter of the left atrium (LA) and from the aortic root diameter (Ao) in the parasternal long-axis view at the level of the aortic valve.

Search Strategy for Identification of Studies
We searched the following sources in April 2014 without any language restriction:

1. Electronic bibliographic databases were searched: Medline (1966 to present) and Cumulative Index to Nursing and Allied Health Literature (CINAHL) (1982 to present).
2. Abstracts of conferences: proceedings of pediatric academic societies (American Pediatric Society, Society for Pediatric Research, and European Society for Pediatric Research) were searched from 1990 from the journal Pediatric Research and abstracts2view.com online.
3. The Web of Science was searched.
4. PubMed’s related citations feature and relevant identified articles were searched.
5. The Science Citation Index was used to identify relevant articles by using previously identified articles.
6. Additional searches were made from reference lists in identified studies.

The search strategy is described in Supplemental Appendix 1.

Data Collection and Analyses
Selection of Studies
All titles and abstracts identified by our search strategy were screened for relevance by authors M.K. and M.P. together. All identified articles that were relevant to the review were retrieved in full and evaluated for...
inclusion eligibility by M.P. and M.K. independently. The results were compared and disagreements were resolved by mutual discussion.

**Data Extraction and Management**

Data extracted included author, year of publication and journal, study design, study population, reference standard and performance of the reference standard, index tests and performance of the index tests, information about quality assessment items based on Quality Assessment of Diagnostic-Accuracy Studies (QUADAS-2), and data for 2×2 tables. Additional information to clarify the study design and data was sought from the authors via e-mail for ≥3 attempts. All the data were entered electronically on Microsoft Excel (Microsoft Corp, Redmond, WA) spreadsheets by M.P. and M.K. independently. The data extracted by each author were compared, and any discrepancies were resolved by mutual discussion and input from author C.J.F.

**Assessment of Methodological Quality**

The methodological quality of each study was assessed as recommended by the Cochrane Diagnostic Test Accuracy Working Group adapting from the QUADAS-2 by authors G.G. and M.K. The 4 domains assessed for risk of bias were patient selection, index test, reference test, and flow and timing. Applicability concerns were assessed in the first 3 domains, as described in Supplemental Appendix 2. The data were then entered into Review Manager Software (RevMan 5.3; The Cochrane Collaboration, Oxford, England) for meta-analysis and to generate figures.

**Statistical Analyses and Data Synthesis**

We constructed 2×2 tables for all studies with the reference standard and index tests and enumerated true-positives, false-positives, false-negatives, and true-negatives for all the reported thresholds. For the studies that reported sensitivity and specificity at defined thresholds, reverse calculation was done to generate a 2×2 table. Data were entered in RevMan 5.3, and forest plots with 95% confidence intervals (CIs) for sensitivity and specificity for each study were created. Results were also plotted in the receiver operating characteristic (ROC) space with 95% confidence estimates, summary point, and the summary curve. The meta-analyses by bivariate random effects model were performed by using the metandi package in the statistical software Stata 11 (Stata Corp, College Station, TX). The following covariates were evaluated: test assay kit, thresholds (as continuous), GA, chronological age at the time of the test, and whether used to initiate medical or surgical therapy.

**Investigations of Heterogeneity Among Studies**

We investigated heterogeneity through subgroup analyses if data were available from ≥4 studies:

1. Type of commercial test assay used for BNP and NT-proBNP, because measurements may vary with each commercial assay.
2. Effects of age at testing: BNP and NT-proBNP levels are higher at birth and then decline over time.
3. Effects of GA: BNP and NT-proBNP levels are higher in preterm infants than in term infants.

**FIGURE 1**

Inclusion process for the systematic review based on Preferred Reporting Items for Systematic Reviews and Meta-Analyses. a Review articles, animal studies, and studies that did not report outcomes of interest (congenital heart disease, intraventricular hemorrhage, transient tachypnea of the newborn, hypoxic-ischemic encephalopathy, respiratory distress syndrome) or did not evaluate BNP or NT-proBNP. b Reasons for exclusion are listed in Table 2.
### TABLE 1 Characteristics of Included Studies

<table>
<thead>
<tr>
<th>Study, Country</th>
<th>Study Design</th>
<th>Inclusion Criteria for Participants’ and Summary Statistic: GA (wk) or BW (g)</th>
<th>Index Test, Assay Kit, Threshold Cutoffs, and Timing of the Assay</th>
<th>Reference Standard Echocardiogram</th>
<th>Area Under the Curve</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 Lee et al 2013, Korea</td>
<td>Retrospective</td>
<td>BW &lt;1000 g</td>
<td>BNP Mean ± SD Test kit: Triage, San Diego Control group</td>
<td>Multiple: 864 pg/mL*, 100 pg/mL, 150 pg/mL, 200 pg/mL, 400 pg/mL*, 600 pg/mL, 900 pg/mL</td>
<td>Symptomatic PDA: presence of 2 of the following 5 signs with the confirmation of a large L → R ductal flow by echocardiogram</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>GA: 27.3 ± 2.3 BW: 855 ± 112 hsPDA group: GA: 27.1 ± 2.2 BW: 819 ± 123</td>
<td></td>
<td>Clinical criteria</td>
</tr>
<tr>
<td>2 Mine et al 2013, Japan</td>
<td>Retrospective</td>
<td>GA &lt;33 wk, BW &lt;1500 g</td>
<td>BNP Median and interquartile range Test kit: Shionospot, Osaka, Japan Control group</td>
<td>250 pg/mL* for indomethacin treatment GA: 28.1 (25.5–29.2) wk BW: 950 (799–1181) g hsPDA group GA: 28.0 (735–1137) g</td>
<td>Echocardiogram at the time of admission and every 12 h</td>
</tr>
<tr>
<td>3 Kim and Shim 2012, Korea</td>
<td>Prospective</td>
<td>GA &lt;37 wk</td>
<td>BNP Mean ± SD Test kit: Abbott, IL 412 pg/mL Control group</td>
<td>GA: 32.83 ± 2.22 wk BW: 1830 ± 480 g hsPDA group GA: 32.18 ± 1.67 wk BW: 1850 ± 480 g</td>
<td>Echocardiographic criteria</td>
</tr>
<tr>
<td></td>
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<td></td>
<td>GA: 412 pg/mL Timing: day 4 of life</td>
<td></td>
<td>LA/Ao &gt;1.4 Diastolic turbulence of pulmonary artery on Doppler Ductal diameter of PDA &gt;1.4 mm PLUS 3/6 clinical signs Systolic/continuous murmur Increased precordial pulse Bounding peripheral pulses Resting tachycardia Unexplained deterioration of respiratory status and increased vascular marking, heart enlargement, or pulmonary edema on chest radiograph</td>
</tr>
<tr>
<td>4 Elsayed et al 2012, Canada</td>
<td>Prospective</td>
<td>GA &lt;31 wk</td>
<td>BNP Mean ± SD Test kit: unknown 90 pg/mL No PDA GA: 29.3 ± 1.0 wk BW: 1325 ± 261 g Non-hsPDA GA: 28.2 ± 1.7 wk BW: 1160 ± 257 g hsPDA GA: 26.9 ± 1.4 wk BW: 925 ± 209 g</td>
<td>Echocardiogram</td>
<td></td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>BW: 1325 ± 261 g Non-hsPDA GA: 28.2 ± 1.7 wk BW: 1160 ± 257 g hsPDA GA: 26.9 ± 1.4 wk BW: 925 ± 209 g</td>
<td></td>
<td>PDA diameter &gt;1.5 mm L → R nonrestrictive shunt</td>
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<tr>
<td>Elsayed et al 2011,57 Canada</td>
<td>Prospective GA &lt;31 wk Mean (±SD) Non-hsPDA group GA: 28.6 ± 1.1 wk BW: 1270 ± 27 g hsPDA group GA: 26.7 ± 1.2 wk BW: 887 ± 16 g</td>
<td>BNP and PDA score Test kit: unknown 90 pg/mL and PDA score &gt;7 Timing: 48–72 h of age</td>
<td>Echocardiogram • PDA diameter &gt;1.5 mm • L → R nonrestrictive shunt</td>
<td>NA</td>
<td></td>
</tr>
<tr>
<td>Kalra et al 2011,18 USA</td>
<td>Prospective BW &lt;1250 g, GA &lt;34 wk with normal renal function</td>
<td>BNP</td>
<td>Did not describe the demographics of the patient population</td>
<td>Test kit: Triage, San Diego PDA was reported as no, small, moderate, or large based on ductal diameter at the pulmonary side, absolute size of the PDA in comparison with the branch pulmonary arteries. Secondary considerations were length of color flow jet into the main pulmonary artery, presence of diastolic reversal of flow in the descending aorta, and the presence of left atrial and ventricular dilatation including LA/Ao ratio.</td>
<td>Echocardiogram</td>
</tr>
<tr>
<td>Chen et al 2010,22 USA</td>
<td>Retrospective GA 24–32 wk with ≥1 BNP–echocardiogram pair (done on same day) Mean ± SD GA: 27.3 ± 2.1 wk BW: 980 ± 276 g</td>
<td>BNP</td>
<td>Test kit: Triage, San Diego Multiple cutoffs: 40 pg/mL, 90 pg/mL, 90 pg/mL, 100 pg/mL, 200 pg/mL, and 300 pg/mL for moderate–large PDA and large PDA Timing: variable 123 ng/L or pg/mL LA/Ao &gt;1.5</td>
<td>Echocardiogram • Ductus diameter &gt;1.5 mm</td>
<td>0.85</td>
</tr>
<tr>
<td>Czernik et al 2008,24 Germany</td>
<td>Prospective GA &lt;28 wk Median (interquartile range) Control group GA: 26 (25–27) wk BW: 948 (720–1100) g hsPDA group GA: 25 (24–26) wk BW: 737 (650–827) g</td>
<td>BNP</td>
<td>Test kit: ADVIA Centaur, Germany Multiple cutoffs: 550 pg/mL predict ductus intervention. Others were 380 pg/mL, 450 pg/mL, 500 pg/mL, 600 pg/mL, and 650 pg/mL Timing: second day of life Need for ventilatory support</td>
<td>Echocardiogram showing • Narrowest ductal diameter &gt;2 mm • PDA with L → R shunt</td>
<td>0.88</td>
</tr>
<tr>
<td>Study, Country</td>
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<tr>
<td>9 Sanjeev et al 2005,25 USA</td>
<td>Prospective</td>
<td>BW 500–1500 g Mean ± SD GA: 26 ± 2 wk BW: 873.5 ± 247 g</td>
<td>BNP Test kit: Triage, San Diego, CA Multiple cutoffs: 70 pg/mL, 50 pg/mL, 80 pg/mL, 90 pg/mL*, 100 pg/mL Timing: variable</td>
<td>Echocardiogram</td>
<td>0.91</td>
</tr>
<tr>
<td>10 Choi et al 2005,26 Korea</td>
<td>Prospective GA 25–34 wk</td>
<td></td>
<td>BNP Mean ± SD Test kit: Triage, San Diego, CA LA/Ao.1.4, LV/Ao.2.1</td>
<td>Symptomatic PDA: presence of 2 of the following 5 signs with the confirmation of a large L → R ductal flow by echocardiogram</td>
<td>0.997</td>
</tr>
<tr>
<td>1 Occhipinti et al 2014,74 Italy</td>
<td>Prospective GA 23–32 wk Mean ± SD</td>
<td>NT-proBNP Test kit: Roche Elecsys, Germany 9854 pg/mL Timing: 6–24 h of life</td>
<td>• Ductal diameter/birth wt ratio &gt;1.4 or • LA/Ao ratio &gt;1.4 or • Pulsatile flow pattern</td>
<td></td>
<td>0.86</td>
</tr>
<tr>
<td>2 Buddhe et al 2012,60 USA</td>
<td>Prospective Mean ± SD GA: 28.0 (±2.4) wk BW: 1053 (±278) g</td>
<td>NT-proBNP Test kit: VITROS NT-proBNP reagent pack 5900 pg/mL Timing: day 3–5 of life</td>
<td>PDA size ≥1 mm with ≥2 additional features of PDA • Continuous murmur • Pulse pressure &gt;25 mm Hg • Worsening respiratory status • LA/Ao ratio &gt;1.4 • Resistive index of SMA calculated as peak systolic velocity — end diastolic velocity/time averaged mean velocity &gt;6 • Base excess ≤–5</td>
<td></td>
<td>0.98</td>
</tr>
<tr>
<td>3 Cambonie et al 2012,27 France</td>
<td>Prospective GA: 26.6 (±2.7) wk BW: 829 (±275) g</td>
<td>NT-proBNP Test kit: Rd. Dimension, Siemens, Newark, NJ 8500 pg/mL Timing: 24–72 h of life</td>
<td>Echocardiogram showing PDA and ≥2 of the following criteria: • LA/Ao ratio &gt;1.48 • Retrograde or absent diastolic flow in the ACA or SMA • Growing or pulsatile pattern flow in the DA • End-diastolic flow velocity in the LPA &gt;0.20 m/s Requirement of invasive ventilation with fraction of inspired oxygen &gt;0.3 after surfactant treatment or invasive ventilation plus catecholamine use for severe hypotension</td>
<td></td>
<td>0.87</td>
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<td>Study, Country</td>
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</tbody>
</table>
| 4 Letzner et al 2012,38 Switzerland | Prospective GA< 32 wk Median (5%-95% range)  
Control group  
GA: 30.7 (28.2–31.9) wk  
BW: 1400 (806–20111) g  
hsPDA group  
GA: 28 (24.7–31.6) wk  
BW: 960 (573–1787) g | NT-proBNP  
Test kit: BRAHMS KRYPTOR, Germany  
2316 pg/mL on day 0  
10 253 pg/mL on day 2–3 | • Presence of PDA with L→R shunting  
• Narrowest ductal diameter ≥1.5 mm and  
• LA/Ao ratio ≥1.5 and  
• Diastolic retrograde flow in the postductal descending aorta | 0.64 |
| 5 Letshwiti et al 2011,61 Ireland | Prospective BW<1500 g Mean ± SD  
GA: 28.3 (±2.5) wk  
BW: 1115 (±255) g | NT-proBNP  
Test kit: Roche Elecsys, Germany  
3587 ng/L = 3587 pg/mL | • Ductal diameter >2 mm | 0.93 |
| 6 Martinovici et al 2011,28 Belgium | Prospective GA<32 wk or BW<1500 g Mean ± SD  
Control group  
GA: 29.8 ± 2.2 wk  
BW: 1290 ± 360 g  
hsPDA group  
GA: 28.0 ± 1.7 wk  
BW: 1080 ± 350 g | NT-proBNP  
Test kit: Roche Elecsys, Germany  
10 000 pg/mL on day 2  
5000 pg/mL * on day 4 | • Ratio of ductal diameter to birth wt (DA/kg in mm/kg) >1.4 and  
• LA/Ao >1.4 | 0.92 |
| 7 Deorari et al 2011,59 India | Prospective GA<32 wk and BW<1500 g Mean ± SD  
GA: 50.3 ± 1.6 wk  
BW: 1050 ± 237 g | NT-proBNP  
Test kit: Roche Elecsys, Germany  
17 984 pg/mL  
Timing: day 7 of life  
NT-proBNP | • Duct size ≥1.5 mm along with one of the following criteria:  
• LA/Ao ratio ≥1.5 or  
• Absent or retrograde flow in descending aorta | 0.8 |
| 8 Ramakrishnan et al 2009,30 UK | Prospective GA 23–34 wk Mean ± SD  
GA: 28.2 ± 2.8 wk | NT-proBNP  
Test kit: Roche Elecsys, Germany  
Multiple cutoffs:  
2850 pmol/L = 24 102 pg/mL *,  
1280 pmol/L = 10 825 pg/mL,  
5180 pmol/L = 43 638 pg/mL | • Duct diameter >1.5 mm | 0.887 |
| 9 Nuntnarumit et al 2009,10 Thailand | Prospective GA<33 wk Median (interquartile range)  
Control group  
GA: 31 (28–35) wk  
BW: 1360 (730–1830) g  
hsPDA group  
GA: 29 (27–31) wk  
BW: 1250 (925–1540) g | NT-proBNP  
Test kit: Roche Elecsys, Germany  
10 180 pg/mL  
Timing: day 2 of life | • Heart murmur  
• Persistent tachycardia (heart rate >160/min)  
• Hyperactive precordium  
• Bounding pulse, pulse pressure >25 mm Hg  
• Hepatomegaly  
• Pulmonary hemorrhage  
• Increasing respiratory support by 20% increase in oxygen supplementation or in pressure support and  
• Radiographic evidence of cardiomegaly or pulmonary congestion | 0.964 |
4. We determined whether the results of the test were used to initiate medical or surgical treatment of PDA.

5. Assay threshold cutoffs vary with the commercial assay and patient characteristics such as GA and chronological age.

RESULTS

We identified 82 records through database searches and 21 additional articles from conference abstracts and cross-referencing. The inclusion process is detailed in the Preferred Reporting Items for Systematic Reviews and Meta-Analyses flow diagram (Fig 1). Ten studies that evaluated BNP and 11 studies that evaluated NT-proBNP in the diagnosis of hsPDA met our inclusion criteria (Table 1). Excluded studies and reasons for exclusion are described in Table 2.

Methodological assessment of included studies revealed study deficiencies in the following domains (Fig 2, Supplemental Figs 5 and 6):

1. Patient selection: Exclusion criteria in the individual studies were variable and were not defined in some studies, which may have introduced bias. Some studies were retrospective in design. Inclusion criteria differed, and all studies except had restrictive inclusion criteria by birth weight (BW) and GA. Additional inclusion criteria included presence of respiratory distress necessitating mechanical ventilation and surfactant performed on the same day, which might have excluded some eligible patients with hsPDA.

2. Index test: No study had a predefined threshold, and some studies did not blind the clinician. Elsayed et al used BNP in addition to a clinical PDA score as the index test. The PDA score incorporated echocardiographic parameters reflective of both volume and pressure overload (maximum score 15), and clinical, radiologic, and laboratory features of both pulmonary overcirculation and systemic hypoperfusion (maximum score 13).

3. Reference standard: All studies used an acceptable reference standard, the echocardiogram, but echocardiogram criteria to define hsPDA were variable, which may introduce bias. There was absence of blinding of the cardiologists to the BNP assay in a few studies.

4. Flow and timing: We noted longer or unclear time intervals between the blood test (index test) and echocardiogram (reference standard) in some studies, notably in the study by Kim and Shim, where time interval between the index and reference standard was as long as 48 hours.

Estimated summary sensitivity for studies evaluating BNP was 0.88 (95% CI, 0.76–0.95) and for

<table>
<thead>
<tr>
<th>Study, Country</th>
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<th>Inclusion Criteria for Participants and Summary Statistic: GA (wk) or BW (g)</th>
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<th>Reference Standard Echocardiogram Area Under the Curve</th>
</tr>
</thead>
<tbody>
<tr>
<td>10 Farombi-Oghuvbu et al 2008, Ireland</td>
<td>Prospective</td>
<td>GA &lt;34 wk and BW &lt;2.0 kg Median (range) GA: 30 (24–33) wk BW: 1220 (550–1950) g Median</td>
<td>NT proBNP Test kit: Roche Elecsys, Germany 11 395 pg/mL Timing: day 3 of life</td>
<td>Echocardiogram on days 1, 3, 5, and 10 of life • Large ductal flow with L → R shunt 0.978</td>
</tr>
<tr>
<td>11 El-Khuffash et al 2007, Ireland</td>
<td>Prospective</td>
<td>BW &lt;1500 g Median (interquartile range) GA: 28 (26.1–29.5) wk BW: 1121 (948–1253) g</td>
<td>NT-proBNP: at 12 h and day 3 of life Test kit: Roche Elecsys, Germany 5000 pmol/L = 42 285 pg/mL Timing: day 3 of life</td>
<td>Echocardiogram at 12 h, day 3, days 5–6, and after PDA treatment • Ductal diameter &gt;1.5 mm 0.866</td>
</tr>
</tbody>
</table>

ACA, anterior cerebral artery; DA, ductus arteriosus; LPA, left pulmonary artery; SMA, superior mesenteric artery.

* Threshold level used in the Forest plots and ROC analysis when using only one value per study;

† Threshold level chosen by the authors for individual ROC analysis.

A"
TABLE 2 Characteristics of Excluded Studies

<table>
<thead>
<tr>
<th>Excluded Study</th>
<th>Assay</th>
<th>Reason for Exclusion</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 Attridge et al 2006</td>
<td>BNP</td>
<td>BNP to optimize indomethacin treatment and not for the diagnosis of PDA</td>
</tr>
<tr>
<td>2 da Graca et al 2006</td>
<td>BNP</td>
<td>No cutoff threshold reported</td>
</tr>
<tr>
<td>3 Elsayed et al 2013</td>
<td>BNP</td>
<td>Included patients with PDA only and used BNP to predict failure of medical therapy</td>
</tr>
<tr>
<td>4 Elsayed et al 2013</td>
<td>BNP</td>
<td>Correlation of BNP and regional blood flow in PDA and for the diagnosis of PDA</td>
</tr>
<tr>
<td>5 Flynn et al 2005</td>
<td>BNP</td>
<td>Insufficient data for 2×2 table; BNP &gt;300 pg/mL predicted significant PDA</td>
</tr>
<tr>
<td>6 Hammerman et al 2012</td>
<td>BNP and NT-proBNP</td>
<td>Review article</td>
</tr>
<tr>
<td>7 Hollinger et al 2011</td>
<td>BNP</td>
<td>No cutoff threshold reported</td>
</tr>
<tr>
<td>8 Holmström et al 2001</td>
<td>BNP</td>
<td>Insufficient data for 2×2 table</td>
</tr>
<tr>
<td>9 Hsu et al 2010</td>
<td>BNP</td>
<td>Included patients with PDA and used BNP to predict indomethacin responsiveness</td>
</tr>
<tr>
<td>10 Jeevananthan et al</td>
<td>BNP</td>
<td>Insufficient data for 2×2 table</td>
</tr>
<tr>
<td>11 Kazanci et al 2012</td>
<td>BNP</td>
<td>No cutoff threshold reported</td>
</tr>
<tr>
<td>12 Mannarino et al 2010</td>
<td>BNP</td>
<td>Compared BNP in term versus preterm infants</td>
</tr>
<tr>
<td>13 Perugu et al 2011</td>
<td>BNP</td>
<td>BNP and other markers for ventricular function in extremely low birth weight infants and not for the diagnosis of PDA</td>
</tr>
<tr>
<td>14 Puddy et al 2002</td>
<td>BNP</td>
<td>No cutoff threshold reported</td>
</tr>
<tr>
<td>15 Tauber et al 2013</td>
<td>BNP</td>
<td>No cutoff threshold reported</td>
</tr>
<tr>
<td>16 Bagñoli et al 2010</td>
<td>NT-proBNP</td>
<td>No cutoff threshold reported</td>
</tr>
<tr>
<td>17 Celik et al 2012</td>
<td>NT-proBNP</td>
<td>Urine levels of the natriuretic peptide</td>
</tr>
<tr>
<td>18 Czernek et al 2015</td>
<td>NT-proBNP</td>
<td>Urine levels of the natriuretic peptide</td>
</tr>
<tr>
<td>19 El Khuffash et al</td>
<td>NT-proBNP</td>
<td>Used to predict which infants with PDA will suffer death or intraventricular hemorrhage</td>
</tr>
<tr>
<td>20 El Khuffash et al</td>
<td>NT-proBNP</td>
<td>No outcome of interest reported</td>
</tr>
<tr>
<td>21 Hammerman et al 2010</td>
<td>NT-proBNP</td>
<td>Included patients with hsPDA only. used percentage drop in NT-proBNP to predict response to treatment</td>
</tr>
<tr>
<td>22 Nuntnarumit et al</td>
<td>NT-proBNP</td>
<td>Used a predefined cutoff from previous study for early targeted treatment with indomethacin</td>
</tr>
<tr>
<td>23 Sellmer et al 2011</td>
<td>NT-proBNP</td>
<td>No cutoff threshold reported</td>
</tr>
<tr>
<td>24 Tosse et al 2012</td>
<td>NT-proBNP</td>
<td>Urine levels of the natriuretic peptide</td>
</tr>
<tr>
<td>25 Yildirim et al 2010</td>
<td>NT-proBNP</td>
<td>No cutoff threshold reported</td>
</tr>
</tbody>
</table>

NT-proBNP was 0.90 (95% CI, 0.79–0.96). Summary specificity for studies evaluating BNP was 0.92 (95% CI, 0.81–0.97) and for NT-proBNP 0.84 (95% CI, 0.77–0.90). Forest plots based on a single threshold from each study show that sensitivity across studies ranged from 0.60 to 1.0 for BNP and 0.58 to 1.0 for NT-proBNP, and specificity ranged from 0.6 to 1.0 for BNP and 0.57 to 1.0 for NT-proBNP (Fig 3). We also plotted the included studies in the ROC space to give a sense of the distribution of the sensitivity and specificity of the studies (Fig 4). We report the summary ROC curve, summary estimates of specificity and sensitivity, and the 95% confidence regions.

We analyzed subgroups of studies evaluating BNP and NT-proBNP based on the type of test kit, age at testing, and whether used to initiate medical or surgical treatment, and summary estimates are shown in Supplemental Table 4 and Supplemental Figs 7 and 8. Summary estimates by meta-analysis were possible only if ≥4 studies were available in the subgroup. Only 1 test kit and only day 3 of testing had ≥4 studies for both BNP and NT-proBNP. Only 2 studies evaluated BNP for surgical closure of the ductus and none for NT-proBNP. We could not explore heterogeneity associated with gestational age or threshold cutoffs because of insufficient data and the large variability among studies, respectively.

DISCUSSION

We synthesized data from 21 studies by meta-analysis; 10 evaluated BNP and 1 evaluated NT-proBNP in the diagnosis of hsPDA in preterm neonates. Estimated summary sensitivity for BNP was 0.88 (95% CI, 0.76–0.95), and summary specificity was 0.92 (95% CI, 0.81–0.97).

However, there was wide variation in sensitivity and specificity, and 2 of the 10 studies, at the reported thresholds, reported sensitivity <0.720,21 Mine et al21 described 2 thresholds in their study: 250 pg/mL for indomethacin treatment with sensitivity of 0.60 (95% CI, 0.36–0.81) and 2000 pg/mL for the surgical treatment of PDA with sensitivity of 1.00 (95% CI, 0.4–1.00), both with wide 95% CIs. Conversely, Lee et al20 described sensitivity and specificity at a range of thresholds and, at a chosen threshold of 864 pg/mL, had a high specificity 0.95 (95% CI, 0.84–0.99) but low sensitivity at 0.55 (95% CI, 0.36–0.73). One outlier was the threshold given by Mine et al for indomethacin unresponsiveness and surgical indication with 100% sensitivity at a high level of 2000 pg/mL.21

The summary sensitivity for NT-proBNP was 0.90 (95% CI, 0.79–0.96), and summary specificity was 0.84 (95% CI, 0.77–0.90). In the 11 included studies that evaluated NT-proBNP, there was less variation in sensitivity and specificity of NT-proBNP when compared with BNP in the detection of hsPDA. In 2 of the 11 studies, sensitivity was <0.723,28 because they used a higher threshold >40 000 pg/mL. Letshwiti et al61 described a much lower threshold (3587 pg/mL) than El Khuffash and Molloy34 (42 285 pg/mL) or Ramakrishnan et al20 (24 102 pg/mL) but had 100% sensitivity with narrow
confidence intervals (95% CI, 0.83–1.00). The cutoffs in this study may be lower because of evaluation on day 7, when a physiologic decline in NT-proBNP levels is to be expected.6,7,62,63

We assessed the methodological quality of studies by using the 4 domains of the QUADAS-2 checklist: patient selection, index test, reference standard, and flow and timing. Overall, most studies scored as low or unclear for risk of bias and applicability concerns. All studies enrolled preterm neonates, but some of them were more restrictive than others. BNP levels vary by GA, and this could affect threshold values used to diagnose hsPDA. Some of the studies have additional inclusion criteria, such as ventilator support and surfactant,27 and these criteria might introduce applicability concerns. Failure to report exclusion criteria or variable exclusion criteria may add bias.30,57–59 Three studies had a retrospective design but had comparable inclusion and exclusion criteria and definition for hsPDA.20–22

As described by Zonnenberg and de Waal,64 there were wide variations in the definition of hsPDA in the included studies. This raises applicability concerns because a neonate with hsPDA in 1 study may be labeled as non-hsPDA in another. All the included studies evaluating NT-proBNP were prospective in design and more uniform in respect to inclusion criteria based on GA compared with studies evaluating BNP. One of the included studies (Kim and Shim19) reported the time interval of up to 48 hours between the index test and the reference standard. Because the half-life of BNP is 22 minutes and that of NT-proBNP is 60 to 120 minutes, changes in the physiologic status at such long time intervals may decrease the comparability of the index and reference tests.2,3

Elsayed et al57,58 used a clinical PDA score in addition to BNP levels for an ROC analysis. The clinical PDA score incorporated echocardiographic parameters reflective of both volume and...
and pressure overload (maximum score 15) and clinical, radiologic, and laboratory features of both pulmonary overcirculation and systemic hypoperfusion. In addition, only 34 of 90 eligible neonates were recruited in the study. A sensitivity analysis excluding the 2 studies by Elsayed et al with unknown test kits, unknown exclusion criteria, PDA scores, and unaccountable total patients did not change our summary estimates of sensitivity and specificity significantly (summary sensitivity [0.87 vs 0.88] and specificity [0.89 vs 0.92]).

We attempted to explore heterogeneity on the basis of type of assay kit, age at testing, gestational age, threshold cutoffs, and whether indicated for medical or surgical closure. Data were insufficient to make any meaningful comparisons. There were wide variations in the threshold cutoffs in studies on BNP as well as NT-proBNP, possibly because of differences in assay characteristics or patient characteristics, which preclude recommendation of a specific threshold value of BNP or NT-proBNP for the diagnosis of hsPDA.

BNP and NT-proBNP assays are widely available and used in children and adults, both in hospitals and in the community, to diagnose or monitor cardiac failure in at-risk patients. Many studies of BNP and NT-proBNP have been reported from the developing countries, in resource-limited settings. The easy availability and its potential to complement echocardiography in the diagnosis of hsPDA or cardiac dysfunction in patients with bronchopulmonary dysplasia is likely to increase its usage in neonatal units, especially in resource-limited areas. In adults, serial NT-proBNP estimations to guide therapy decrease mortality and hospitalization and decrease health care costs. However, studies that evaluated cost-effectiveness of BNP or NT-proBNP have not been reported in the neonatal population.

**Strength and Weaknesses of the Review**

**Strengths**

Our systematic review follows the method recommended by the Cochrane Diagnostic Test Accuracy Working Group. We searched comprehensively for all eligible studies by using clinically relevant inclusion criteria. We used the bivariate random effects model for meta-analyses of the included studies and strove to explain the sources of heterogeneity by subgroup analyses based on type of commercial assay, test threshold, age of the patient at testing, gestational age, and whether used for medical or surgical treatment of the PDA.

**Weaknesses**

Unlike meta-analyses of randomized control trials, heterogeneity is a well-recognized problem in reviews of diagnostic test accuracy. Despite our extensive search strategy, we may have missed potential studies, because diagnostic accuracy studies are poorly tagged in electronic databases. Publication bias in studies reporting diagnostic test accuracy has been poorly studied. Poor reporting of study design, method of enrollment, and patient characteristics may hamper methodological assessment and external validity of the studies.

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**FIGURE 4**

Studies of BNP and NT-proBNP are plotted in the ROC space. BNP studies are represented in blue ovals, NT-proBNP in red diamonds, and the size of the study symbols weighted for sample size. The summary curves (black line for BNP and red for NT-proBNP), summary points (black dot for BNP and red for NT-proBNP), and the 95% confidence region (dotted line) were derived from meta-analyses. Summary estimates at the summary point are as follows: BNP, sensitivity 88% and specificity 92%; NT-proBNP, sensitivity 90% and specificity 84%.
Another limitation of our review might be that echocardiogram (reference standard) parameters to diagnose hsPDA were not consistent between studies. A systematic review of the definition of hsPDA by echocardiogram highlights this issue in detail.64 An ideal reference standard in the diagnosis of hsPDA may be a composite of echocardiographic, clinical, and radiographic findings, which must be evaluated and validated.

Applicability of Findings to Clinical Practice and Policy

New diagnostic tests can assume the following roles in a diagnostic pathway: replacement of the existing test, triage, or add-on to an existing test. In the context of hsPDA, it is not reasonable to replace the echocardiogram with BNP or NT-proBNP testing. The standard of care is to confirm the presence of hsPDA and rule out ductal-dependent lesions with an echocardiogram before initiating therapy. BNP and NT-proBNP testing may be useful to triage cases of suspected hsPDA to decrease the need for echocardiograms, especially in resource-poor settings. A sensitivity of >85% would be preferable in this setting, where echocardiogram would be indicated if the BNP or NT-proBNP assay exceeds the threshold cutoff. In 100 preterm neonates with hsPDA, we would miss 15 neonates after BNP or NT-proBNP assay. Serial estimations dictated by the clinical condition may diagnose hsPDA in the missed neonates. BNP or NT-proBNP levels do not decline with the worsening or persistence of hsPDA, and echocardiogram can be performed later. A decrease in the need for echocardiograms will decrease use of resources, including personnel and equipment, in addition to avoiding patient discomfort. This decrease can have a huge impact on the health care costs because the cost of 1 echocardiogram is >10 times the cost of a BNP or NT-proBNP test. BNP can also be used as an add-on test to echocardiogram and has the advantage of serial testing for trends before or after initiating medical therapy to guide management without the need for serial echocardiograms.10,23,26,28,30 Threshold cutoffs should be based on the normative values at different gestational and chronological ages, validated locally for the type of commercial assay used and the patient population being investigated. Costs of the BNP and NT-proBNP assays must be balanced with its ability to affect clinical outcomes before widespread acceptance in clinical practice.

CONCLUSIONS

We found wide variability in the diagnostic accuracy of BNP and NT-proBNP in the diagnosis of hsPDA in neonates. Heterogeneity in test results may result from both the characteristics of the assay (type of assay or thresholds used) and patient characteristics (gestational and chronological age), and therefore generalizability is limited. We recommend that the type of assay should be locally validated in the specified population for the specified outcome (to initiate therapy or follow response after therapy) for diagnostic accuracy before use to guide clinical decisions.

Future studies should be designed satisfying the methodological quality items expounded in the QUADAS-2 evaluation system, so that studies are of high methodological quality with minimal bias. Studies reporting diagnostic test accuracy should explicitly state the method of enrollment (prospective or retrospective), characteristics of the population assessed (eg, gestational age, birth weight, comorbidity), blinding of reference standard and index tests, and explanation of withdrawals. There is a real need for international consensus on defining hsPDA based on echocardiographic and objective clinical parameters. Studies should explicitly state the details of the clinical setting and patient characteristics so that clinicians can determine the generalizability of the diagnostic test to their patient population. A composite scoring system that includes gestational and chronological age-specific BNP values and clinical parameters may improve diagnostic accuracy and generalizability but needs evaluation.

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REFERENCES


12. Costello-Boerrigter LC. Cardiac natriuretic peptides: contributors to cardiac cachexia or possible anti-obesity agents or both? Diabetes. 2012;61(10): 2403–2404


32. El-Khuffash A, Molloy EJ. Are B-type natriuretic peptide (BNP) and N-terminal pro-BNP useful in neonates? Arch Dis Child Fetal Neonatal Ed. 2007; 92(4):f320–f324


64. Zonnenberg I, de Waal K. The definition of a haemodynamic significant duct in

65. Elsayed YN. NICU Hemodynamic Group. PDA score at 48 to 72 hours of age (Manitoba score) predicts a hemodynamically significant PDA. *Arch Dis Child.* 2012;97(suppl 2):a228


83. El-Khuffash AF, Slevin M, McNamara PJ, Molloy EJ. Troponin T, N-terminal pro natriuretic peptide and a patent ductus arteriosus scoring system predict death before discharge or neurodevelopmental outcome at 2 years in preterm infants. *Arch Dis Child Fetal Neonatal Ed.* 2011;96(2):113–117


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