Serial serum brain-type natriuretic peptide (BNP) identifies compromised blood flow in infants with hemodynamically significant patent ductus arteriosus

Yasser Elsayed, Mary Seshia, Ronald J. Baier, Shyamala Dakshinamurti

Section of Neonatology, Department of Paediatrics and Child Health, University of Manitoba, Winnipeg, Manitoba, Canada

Abstract

Background: The physiological correlates of elevated serum brain-type natriuretic peptide (BNP) in hemodynamically significant patent ductus arteriosus (HSPDA) are unclear.

Objective: To determine if serial BNP measured at 48-72 hours of age, before and after non-steroidal anti-inflammatory drugs (NSAID) treatment of HSPDA reflect compromised blood flow indices, in infants < 31 weeks of gestational age (GA).

Design/methods: In a prospective blinded study, 70 infants < 31 weeks GA, admitted to Winnipeg NICUs from August 2010 to September 2011, had serum BNPs and echocardiograms at 48-72 hours of age, before and after medical treatment of HSPDA. All BNP and logarithm of BNP (logBNP) were correlated by linear regression with contemporaneous blood flow indices for: 1) systemic hemodynamic indices (corrected left and right ventricular outputs [LVO, RVO], RVO/LVO ratio, superior vena cava flow [SVCF], SVCF/LVO); 2) regional blood flow indices (middle cerebral artery flow [MCAF], MCAF/LVO ratio, middle cerebral artery resistive index [MCARI], middle cerebral artery pulsatility index [MCAPI]; celiac artery flow [CAF], CAF/LVO ratio, celiac artery resistive index [CARI], and celiac artery pulsatility index [CAPI]).

Results: Twenty-six of 70 infants developed HSPDA at 6 ± 2 days. Both BNP and logBNP had similar correlations with all indices, but logBNP showed better goodness of fit. The best correlation was at 48-72 hours of life. Analyzing systemic hemodynamics, logBNP best correlated with SVCF (\(\beta = -0.49, R^2 = 0.24, p < 0.0001\)), SVCF/LVO (\(\beta = -0.55, R^2 = 0.31, p < 0.0001\)), RVO/LVO (\(\beta = -0.59, R^2 = 0.35, p < 0.0001\)), LVO (\(\beta = 0.4, R^2 = 0.16, p < 0.0001\)), and RVO (\(\beta = -0.35, R^2 = 0.12, p < 0.0001\)). For regional blood flow, logBNP best correlated with MCARI (\(\beta = 0.6, R^2 = 0.35, p < 0.0001\)), MCAPI (\(\beta = 0.5, R^2 = 0.29, p < 0.0001\)), and MCAF (\(\beta = -0.34, R^2 = 0.12, p < 0.0001\)).
Conclusions: BNP correlates with blood flow indices in preterm infants with HSPDA mainly at 48-72 hours reflecting the value of pre-symptomatic physiologic prediction by BNP.

Keywords
Brain-type natriuretic peptide, PDA, preterm, blood flow, targeted neonatal echo.

Corresponding author
Yasser Elsayed, MD, PhD, Section of Neonatology, WS012 Women’s Hospital, 735 Notre Dame Ave, Winnipeg Manitoba, Canada R3E 0L8; phone: (204) 787-1853; fax: (204) 787-1587; email: yelsayed@exchange.hsc.mb.ca.

How to cite

Introduction
Patent ductus arteriosus (PDA) is the most common cardiovascular problem in preterm neonates, with an incidence as high as 33% in infants < 31 weeks gestation, and was reported to be associated with adverse outcomes. Determination of whom to treat and when to treat to minimize PDA-related morbidity remains challenging for neonatologists [1, 2].

Current studies are focusing on the acute physiologic changes and subsequent neonatal morbidity which are more likely to be related to the shunt volume across the PDA, rather than merely its patency [3, 4]. A more comprehensive and standardized assessment of the PDA shunt, including biochemical, clinical and blood flow indices, may assist in refining the early diagnosis of hemodynamically significant PDA (HSPDA), allowing for accurate estimation of its significance [5]. The likelihood of operator-dependent error from individual measurements could be minimized through this more comprehensive approach [6]. Ascertainment of the hemodynamic significance of a PDA may benefit from the inclusion of biochemical markers, together with clinical and echocardiographic evaluation [7]. A potential candidate is the brain-type natriuretic peptide (BNP), secreted by ventricular myocytes in response to volume overload of the ventricles [8]. BNP measurements have been reported useful for the diagnosis and assessment of PDA in preterm infants [9]. The half-life of BNP is 20 minutes; it causes diuresis, natriuresis, and arterial dilatation. The expected effect is a compensatory reduction of intravascular volume, and ventricular preload [10]. BNP was reported in prior studies to be useful in evaluating HSPDA and correlated with the magnitude of shunt volume, but the physiologic correlation with blood flow indices is not yet examined [9, 11]. BNP levels are variable at different time points of measurement, limiting the generalizability of this method for routine use [8, 12, 13]. We proposed to explain this phenomenon by studying the physiologic correlation of BNP levels with indices reflecting systemic and regional blood flow, considering the presence of HSPDA requiring treatment as a reference standard for a perturbed hemodynamic state [14]. We hypothesized that BNP is correlating with blood flow indices at different times of assessment in infants with HSPDA.

Methods
This was a prospective cohort study of premature neonates (< 31 weeks gestation) admitted to the Neonatal Intensive Care Units (NICUs) of Health Sciences Center and St. Boniface General Hospitals in Winnipeg (Manitoba, Canada) between August 2010 and November 2011. With institutional Research Ethics Board approval, infants were recruited, independent of clinical suspicion for PDA, and following informed parental consent. Infants with right-to-left PDA shunt or congenital heart disease (other than patent foramen ovale) were excluded. Prophylactic indomethacin was not used, and treatment given only for symptomatic PDA. In all study infants, blood flow indices were evaluated by targeted neonatal echocardiography (TNE) and Doppler techniques, which were performed after 48 and before 72 hours of age, before and within 12 hours after medical treatment of a PDA; serum BNP was measured concurrently with each TNE and Doppler assessment of blood flow indices.

Management of patent ductus arteriosus
The clinical team, blinded to blood flow indices and BNP findings, independently determined whether the PDA was symptomatic; their decision to treat was based on clinical features and independent echocardiographic confirmation of an unrestricted left-to-right shunt by the pediatric cardiologist.
The reference standard for HSPDA diagnosis was PDA diameter > 1.5 mm with a left-to-right non-restrictive shunt, as reported by the pediatric cardiologist, and subsequently received treatment for ductal closure. Clinical features considered by the clinical team for treating PDA included (but not limited to) signs of pulmonary over-circulation (pulmonary hemorrhage), extubation failure, or need for re-intubation due to increased respiratory work, persistent apnea requiring stimulation, pH < 7.25 and/or pCO₂ > 60 mmHg, and signs of systemic hypoperfusion (hypotension requiring inotropic support). The medical treatment strategy was intravenous indomethacin or ibuprofen for a maximum of 2 consecutive or separate courses together with reduced fluid intake; surgical ligation of the PDA was considered if medical treatment failed or was contraindicated.

**Measurement of serum brain-type natriuretic peptide**

Blood was drawn from an indwelling catheter or heel poke. BNP was determined in 0.3 ml whole blood by Triage® BNP bedside immunoassay (Biosite Diagnostics Inc., San Diego, CA, USA), measuring BNP in filtered serum using a murine polyclonal fluorescence-tagged anti-BNP antibody with detection range 5-5,000 pg/ml [7].

**Targeted neonatal echocardiography and Doppler techniques**

Targeted neonatal echocardiographic studies were performed using Vivid-7® (GE Healthcare, Waukesha, WI, USA) or HP SONOS® 5500 (Phillips, Andover, MA, USA) echocardiographic systems with 7-10 MHz transducers. TNE and Doppler studies were performed by a trained single operator (Y.E.) as per published guidelines [15]. Imaging data were stored digitally; off-line echocardiography analysis was performed 1-3 months after the study, to avoid recall bias of neonatal outcomes.

TNE and Doppler assessment were performed at 48-72 hours of age, irrespective of clinical symptoms. If a PDA was suspected by the clinical team, echocardiography for evaluation and management of the PDA was independently performed by the Pediatric Cardiology consultation service; in cases of PDA, TNE and Doppler assessment were independently performed by the study team before and after PDA treatment. No enrolled infant received medical treatment of the ductus before 72 hours of age.

**Measurement of systemic and regional blood flow parameters**

The aortic diameter was measured using the parasternal long axis view proximal to and just below the aortic valve annulus while aortic Doppler was performed using the apical long axis view [16]. Pulmonary valve annulus was measured while pulmonary Doppler was performed using the parasternal short axis view. Superior vena cava (SVC) diameter was measured using a high parasternal view and measured close to the entrance to the right atrium (average of maximum and minimum diameters). SVC Doppler was evaluated using the subcostal sagittal view [17]. Middle cerebral artery (MCA) diameter and Doppler were measured at the right parietal region. Celiac artery (CA) Doppler and diameter were assessed at the subcostal sagittal view just at its exit from the aorta [18-20]. Pulsatility and resistive indices (PI and RI) were calculated from peak systolic flow velocity, minimum diastolic blood flow velocity, and mean velocity [21-23]. Left ventricular output (LVO), right ventricular output (RVO), superior vena cava flow (SVCF), middle cerebral artery flow (MCAF) and celiac artery flow (CAF) were measured by the following equation [22, 24]: flow or output = velocity time integral × {π × (mean vessel diameter²/4) × heart rate}/body weight. PI was calculated according to the following equation: PI = (PSV-MDV)/MV, and RI was calculated as follows: RI = (PSV-MDV)/PSV, where PSV = peak systolic velocity, MDV = minimum diastolic velocity, MV = mean velocity (time averaged velocity).

**Features of patent ductus arteriosus**

Ductal diameter: using 2-D mode at high parasternal (ductal view), the narrowest transductal diameter was obtained, expressed as the mean of 3 measurements. Color Doppler was used to identifying flow across the PDA. The scale of the color was adjusted upwards to remove aliasing [25], according to the assessment of the PDA published by Tavera et al. [26].

Peak PDA flow velocity: the direction and peak velocity of transductal flow was assessed by pulse- and continuous-wave Doppler [3, 16].

**Statistical analysis**

SPSS® v. 21 (SPSS Inc, Chicago, IL, USA) was used to perform the statistical analysis.
Pearson correlation was used to correlate BNP with systemic and regional blood flow indices. Data are presented as mean ± SD, or median with interquartile range, as appropriate; p < 0.05 was considered significant. The logarithm of BNP (logBNP) was used for a better fit of transformed smaller number to the correlation graphs.

Results

Seventy-one of 132 eligible infants were enrolled in the study. The main reason for non-enrollment was a lack of parental consent before 72 hours of age. One infant was withdrawn from the study after enrollment due to the parental request. Thirty-seven infants of the enrolled group were males (53%). Mean ± SD of gestational age (GA) and birth weight of the enrolled group were 28 ± 1.6 weeks and 1,101 ± 280 grams respectively, the lower limit for GA was 24 weeks and for birth weight was 520 grams. A PDA was present in 56 of 70 (80%) infants at 48-72 hours of life, 31 of them were on CPAP support during the time of initial TNE study, 18 were on conventional mechanical ventilation, and 7 were on high-frequency jet ventilation support. Fourteen infants had PDA closed at 48-72 hours of life (20%), 12 of them were on CPAP support, and 2 were on conventional ventilation. Twenty-six of 70 infants developed HSPDA at 6 ± 2 days. PDA closed after one non-steroidal anti-inflammatory drug NSAID course in 8/26 (31%). Seven (27%) of neonates with clinical PDA received 2 courses of medical treatment, and after failure to close underwent surgical ligation. Infants with HSPDA were of lesser birth weight and lower gestation (Tab. 1). They also had a significantly higher BNP level. Infants with HSPDA had LVO, middle cerebral artery resistive index (MCARI), middle cerebral artery pulsatility index (MCAPI), celiac artery resistive index (CARI), and celiac artery pulsatility index (CAPI) significantly higher than infants with no HSPDA; they had significantly lower RVO, RVO:LVO ratio, SVCF, MCAF, CAF, CAF:LVO ratio; SVCF:LVO and MCAF:LVO ratios were not statistically different between both groups.

Table 1. Comparison between hemodynamically significant patent ductus arteriosus (HSPDA) and non-HSPDA groups.

<table>
<thead>
<tr>
<th></th>
<th>HSPDA n = 26</th>
<th>No HSPDA n = 44</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>GA (weeks)</td>
<td>26 (27, 28)</td>
<td>29 (29, 30)</td>
<td>0.0001</td>
</tr>
<tr>
<td>Birth weight (gm)</td>
<td>927 ± 190</td>
<td>1,191 ± 292</td>
<td>0.0001</td>
</tr>
<tr>
<td>BNP (pg/ml)</td>
<td>178 (103, 605)</td>
<td>17 (10, 33)</td>
<td>0.0001</td>
</tr>
<tr>
<td>Days on oxygen</td>
<td>71 ± 20</td>
<td>23 ± 13</td>
<td>0.001</td>
</tr>
<tr>
<td>LVO (ml/kg/min)</td>
<td>358 (300, 387)</td>
<td>295 (251, 332)</td>
<td>0.001</td>
</tr>
<tr>
<td>RVO (ml/kg/min)</td>
<td>225 (199, 269)</td>
<td>280 (259, 298)</td>
<td>0.001</td>
</tr>
<tr>
<td>RVO:LVO</td>
<td>0.68 (0.59, 0.89)</td>
<td>0.94 (0.86, 1.0)</td>
<td>0.001</td>
</tr>
<tr>
<td>SVCF (ml/kg/min)</td>
<td>87 (73, 107)</td>
<td>120 (97, 129)</td>
<td>0.001</td>
</tr>
<tr>
<td>SVCF:LVO</td>
<td>0.36 (0.33, 0.42)</td>
<td>0.4 (0.34, 0.46)</td>
<td>0.055</td>
</tr>
<tr>
<td>MCAF (ml/kg/min)</td>
<td>9.9 (8, 13.8)</td>
<td>12 (10.2, 13.5)</td>
<td>0.001</td>
</tr>
<tr>
<td>MCARI</td>
<td>0.64 (0.62, 0.81)</td>
<td>0.57 (0.45, 0.7)</td>
<td>0.001</td>
</tr>
<tr>
<td>MCAPI</td>
<td>1.3 (1.2, 2.5)</td>
<td>1.2 (0.9, 1.7)</td>
<td>&lt; 0.05</td>
</tr>
<tr>
<td>MCAF:LVO</td>
<td>0.04 (0.03, 0.058)</td>
<td>0.043 (0.03, 0.05)</td>
<td>0.48</td>
</tr>
<tr>
<td>CAF (ml/kg/min)</td>
<td>23.3 (18.5, 23.9)</td>
<td>26.9 (21.4, 32)</td>
<td>0.001</td>
</tr>
<tr>
<td>CARI</td>
<td>0.82 (0.62, 0.96)</td>
<td>0.56 (0.51, 0.63)</td>
<td>0.001</td>
</tr>
<tr>
<td>CAPI</td>
<td>1.4 (1.3, 2.2)</td>
<td>1.2 (1.1, 1.4)</td>
<td>0.01</td>
</tr>
<tr>
<td>CAF:LVO</td>
<td>0.09 (0.07, 0.11)</td>
<td>0.1 (0.07, 0.12)</td>
<td>&lt; 0.05</td>
</tr>
</tbody>
</table>

Data is presented as mean ± standard deviation or median (interquartile range); p-value is bolded where significant.

HSPDA: hemodynamically significant patent ductus arteriosus; GA: gestational age; BNP: brain-type natriuretic peptide; LVO: left ventricular output; RVO: right ventricular output; SVCF: superior vena cava flow; MCAF: middle cerebral artery flow; MCARI: middle cerebral artery resistive index; MCAPI: middle cerebral artery pulsatility index; CAF: celiac artery flow; CARI: celiac artery resistive index; CAPI: celiac artery pulsatility index.
correlation of serial BNP measurements with systemic and regional blood flow indices was at 48 to 72 hours of life (Fig. 1). Post PDA treatment logBNP also correlated with many hemodynamic parameters; BNP measured before medical treatment had little correlation with the measured parameters (Tab. 2). Analyzing systemic blood flow indices, logBNP best correlated for all the three-time points of measurements with SVCF and RVO/LVO; LVO, RVO, and SVCF:LVO ratio were significant at 48-72 hours and post treatment (Tab. 2). For regional blood flow logBNP best correlated with MCAF, MCARI, and MCAPI at all the three-time points of measurements; CAF and CARI were significant at 48-72 hours and post treatment; logBNP was correlated with CAPI and CAF:LVO only at 48-72 hours. Tab. 3 shows mean ± SD of all systemic and regional blood flow indices at 48-72 hours, pre-treatment and post-treatment; BNP was not significantly different between the 3-time points, but most of the blood flow indices were significantly different. The intra-observer variability of the TNE and Doppler measurements was good (k = 0.87, 95th CI: 0.72-94).

Discussion

In this study, we examined the correlation between BNP and both systemic and regional blood flow Doppler indices, in preterm infants < 31 weeks with HSPDA. Although there is not a universally accepted definition of HSPDA, for purposes of this study we considered a clinically symptomatic and non-restrictive PDA shunt requiring treatment as our reference definition of HSPDA [27, 28]. We recently published the pre-symptomatic prediction of morbidities related to PDA in preterm infants by BNP at 48-72 hours, but to date no studies have examined its physiologic correlation with echocardiographic blood flow indices [3]. We examined the correlation between BNP and blood flow indices at 3 different time points and found that the best correlation occurred at 48-72 hours after birth. Once the PDA is clinically symptomatic, this correlation became less significant. These data are consistent with our previous finding that BNP measured at 48 to 72 hours was a better predictor of neonatal morbidities related to PDA than other echocardiography parameters [3]. We speculate that BNP may play a temporary role in clinical compensation of systemic hypoperfusion due to HSPDA during the pre-symptomatic period [11]. This compensation may diminish at a variable period beyond 72 hours of age for unknown reasons, which would make the ductus more likely symptomatic at or beyond 48 to 72 hours of life [29]. This speculation is supported by animal studies which showed improvement in congestive heart failure with BNP infusion [30].

Table 2. Correlation of logarithm of brain-type natriuretic peptide (logBNP) with blood flow indices at the 3 time points.

<table>
<thead>
<tr>
<th></th>
<th>At 48-72 hours</th>
<th>Pre-treatment</th>
<th>Post-treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>R²</td>
<td>β</td>
<td>p-value</td>
</tr>
<tr>
<td>LVO (ml/kg/min)</td>
<td>0.16</td>
<td>0.4</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>RVO (ml/kg/min)</td>
<td>0.12</td>
<td>-0.35</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>RVO:LVO</td>
<td>0.35</td>
<td>-0.59</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>SVCF (ml/kg/min)</td>
<td>0.24</td>
<td>-0.49</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>SVCF:LVO</td>
<td>0.31</td>
<td>-0.55</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>MCARI</td>
<td>0.35</td>
<td>0.6</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>MCAPI</td>
<td>0.29</td>
<td>0.5</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>MCAF:LVO</td>
<td>0.017</td>
<td>-0.13</td>
<td>0.11</td>
</tr>
<tr>
<td>CAF (ml/kg/min)</td>
<td>0.15</td>
<td>-0.38</td>
<td>&lt; 0.0001</td>
</tr>
<tr>
<td>CARI</td>
<td>0.29</td>
<td>0.54</td>
<td>0.0001</td>
</tr>
<tr>
<td>CAPI</td>
<td>0.18</td>
<td>0.42</td>
<td>0.0001</td>
</tr>
<tr>
<td>CAF:LVO</td>
<td>0.03</td>
<td>-0.17</td>
<td>0.03</td>
</tr>
</tbody>
</table>

P-value is bolded where significant.
LVO: left ventricular output; RVO: right ventricular output; SVCF: superior vena cava flow; MCAF: middle cerebral artery flow; MCARI: middle cerebral artery resistive index; MCAPI: middle cerebral artery pulsatility index; CAF: celiac artery flow; CARI: celiac artery resistive index; CAPI: celiac artery pulsatility index.
Figure 1. Correlation between logarithm of brain-type natriuretic peptide (logBNP) and different blood flow indices at 48-72 hours of age (continues on the next page).

Please refer to Tab. 2 for R$^2$ and slope ($\beta$) values. Y axis is representing flow (ml/kg/min) for LVO, RVO, CAF, MCAF, ratio for RVO:LVO and SVC:LVO, and index (resistive or pulsatility) for the rest.

LogBNP: logarithm of brain-type natriuretic peptide; LVO: left ventricular output; RVO: right ventricular output; SVCF: superior vena cava flow; MCAF: middle cerebral artery flow; MCARI: middle cerebral artery resistive index; MCAPI: middle cerebral artery pulsatility index; CAF: celiac artery flow; CARI: celiac artery resistive index.
Pilot studies of nuclear magnetic resonance (NMR)-based urinary pharmacometabolomics can distinguish neonatal populations of ibuprofen responders from non-responders, and thus have been proposed to predict which HSPDA may be amenable to closure by medical therapy, though this approach has not yet been tested in clinical trials [31-33]. Echo-based studies cannot predict responsiveness to therapy, but can be used to answer the distinct question of urgency of need for therapy, based on an assessment of hemodynamic consequences of the PDA.

Evaluating systemic and regional blood flow indices have been described in previous studies with significant correlations in infants with HSPDA, both before and after medical treatment, but this is the first study describing quantitation of blood flow through Doppler indices of CA and MCA and correlating these indices with BNP levels during the pre-symptomatic and symptomatic periods [30]. Our data are consistent with other published studies showing variations of BNP values at different time points [8]. This makes standardization of BNP monitoring difficult in practice, and limits its reliability as a single parameter. Our study data was included in a recently published systematic review which showed the same variation between studies and at

Figure 1. Correlation between logarithm of brain-type natriuretic peptide (logBNP) and different blood flow indices at 48-72 hours of age (continues from the previous page).
Please refer to Tab. 2 for $R^2$ and slop ($\beta$) values. Y axis is representing flow (ml/kg/min) for LVO, RVO, CAF, MCAF, ratio for RVO:LVO and SVC:LVO, and index (resistive or pulsatility) for the rest.
LogBNP: logarithm of brain-type natriuretic peptide; LVO: left ventricular output; RVO: right ventricular output; SVC: superior vena cava flow; MCAF: middle cerebral artery flow; MCARI: middle cerebral artery resistive index; MCAPI: middle cerebral artery pulsatility index; CAF: celiac artery flow; CARI: celiac artery resistive index.
different time points of measuring BNP [8]. For this reason, we recommend combined integrated evaluation with TNE [34, 35]. Our data suggests a benefit of using BNP during the pre-symptomatic stage, but a lesser role for using BNP as a predictor of response to therapeutic interventions. The main limitations of our study are its small sample size, and the lack of standardization of clinical team decision-making about when to intervene by medical treatment or surgical ligation. A larger cohort would allow stratification of the study group according to gestational age cut-offs.

**Conclusions**

BNP correlates negatively with hemodynamic parameters of systemic blood flow indices in HSPDA, and positively with both MCA and CA pulsatility and resistive indices mainly at 48-72 hours, reflecting the value of pre-symptomatic physiologic prediction of BNP.

**Abbreviations**

- BNP: brain type natriuretic peptide
- CA: celiac artery
- CAF: celiac artery flow
- CAPI: celiac artery pulsatility index
- CARI: celiac artery resistive index
- HSPDA: hemodynamically significant patent ductus arteriosus
- LVO: left ventricular output
- MCA: middle cerebral artery
- MCAF: middle cerebral artery flow
- MCARI: middle cerebral artery resistive index
- MCAPI: middle cerebral artery pulsatility index
- NSAID: non-steroidal anti-inflammatory drug
- PDA: patent ductus arteriosus
- PI: pulsatility index
- PSV: peak systolic velocity
- RI: resistive index
- RVO: right ventricular output
- SVCF: superior vena cava flow
- TNE: targeted neonatal echocardiography

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**Declaration of interest**

The Authors declare no conflict of interest.

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