ROLE OF PROADRENOMEDULLIN AS A BIOMARKER OF PEDIATRIC PNEUMONIA

By

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ABSTRACT

Background: Community acquired pneumonia (CAP), is an important cause of childhood mortality. Proadrenomedullin (ProADM), is a new regulatory peptide extracted from pheochromocytoma and is a precursor of adrenomedullin but a more stable form. Adult studies found that ProADM is elevated in patients with pneumonia and it is found to be correlated with severity and complications. Also other studies investigated ProADM as prognostic marker for many diseases as sepsis,dengue shock and heart failure.

Objective: The aim of this study was to measure the levels of ProADM in children with CAP and to correlate its levels with the severity and complications.

Methods: This study was done on 90 children (60 children were diagnosed as having CAP and 30 age & sex matched healthy controls). All children were subjected to history taking, clinical examination and investigations (chest x ray, Complete Blood Count, C-reactive protein, serum levels of ProADM by enzyme-linked immuno-sorbent assay ELIZA).

Modified Respiratory Distress Assessment Instrument (RDAI) score was used to classify out cases into mild, moderate and severe.

Results: Serum level of ProADM was found to be higher in CAP cases than in controls and the difference was statistically significant (Mean ± SD = 1.52±0.62 and 0.05±0.04 respectively). Also ProADM was found to be higher in complicated cases, (4 pleural effusion, 1 collapse, 1 pneumothorax) than uncomplicated cases and the difference was statistically significant (2.50±0.24 and 1.41±0.05) respectively. ProADM was increasing with increasing severity of pneumonia. Cutoff point>2.2 nmol/l was the best level to distinguish between complicated and uncomplicated cases (P=0.01 , sensitivity100%, specificity 81.5% , positive predictive value 37.5% , negative predictive value 100%).

Conclusion: level of ProADM was elevated in CAP patients than in controls and higher levels were found in severe and complicated cases.

Keywords: Proadrenomedullin, Community Acquired Pneumonia, Pediatric Pneumonia.

INTRODUCTION

Pneumonia constitutes a major proportion of the global burden of childhood disease, being responsible for around 20% of childhood deaths, the majority of which occur in developing countries. Childhood CAP accounts for between 30% and 40% of hospital admissions with associated case fatality rates of between 15% and 28%**. (1)**

ProADM directly reflects levels of active peptide adrenomedullin (ADM). ADM has multiple tissue sites of action and pluripotent function including vasodilatory, antimicrobial and anti-inflammatory activities . Two main mechanisms can explain the increase of circulating proADM in infections, including CAP. Firstly, ADM is a member of the calcitonin gene family that is extensively synthesized during infections. Secondarily, a decreased clearance by the kidneys and the lung may partly contribute to the increase in ADM plasma levels observed in CAP. **(2)**

ProADM could predict simple pneumonia versus pneumonia with complications or pleural effusion. ProADM also predicts capillary damage, leakage and risk of shock in hemorrhagic dengue and dengue shock, showing that these lesions similar to sepsis can be predicted by this marker in non-bacterial sepsis like syndromes we found that ProADM, , seems to be an interesting marker of complicated CAP, very similar to CRP in sensitivity and specificity. It could therefore help the physician with the decision to hospitalize and choose the antibiotics administration route**. (3)**

MATERIALS AND METHODS

This observasional prospective study was carried on 90 children collected from Pediatric Department, Menofiya University Hospital. Sixty patients admitted with diagnosis of CAP based on the criteria of British Thoracic Society for diagnosing pneumonia in children (**13**) and thirty age and sex matched healthy children were included as controls in the period from December 2015 to April 2016.

Inclusion criteria:

1. Age from 2 months to 5 years
2. Patients diagnosed as CAP according to clinical and radiographic data, with or without complications including Pleural effusion, Collapse and Pneumothorax).

Exclusion criteria:

1. Ages below 2 months and above 5 years.

2. Patients who had immune-deficiency, primary or acquired.
3. Patients who had been hospitalized and treated for two or more days before inclusion in the study.
4. Children with chronic heart, lung or neurological diseases.
5. Patients receiving oral or parentral antibiotic treatment for previously diagnosed community acquired pneumonia.

- All children were subjected to full history taking, complete clinical examination, PA chest x ray and complete blood count (using XT-2000i(Sysmex Corporation of America, Long Grove, Illinois, USA), c-reactive protein (Kit provided by Teco diagnosis, 1268 N, Lakeview Ave Anaheim, CA92807 USA), considered positive when the titer was>6 mg/L**(4)** and serum ProADM (using Human Pro Adreno-medullin ELISA kits, The Chongping Biospes company Ltd, china), normal range: 0.078-0.312 nmol\L **(5)**

We classified our cases according to severity, into : mild (score 0-4), moderate(score 5-8) and severe(score 9-12) according to Modified Respiratory Distress Assessment Instrument (RDAI) score .(6)

Table (1): Modified Respiratory Assessment Instrument (RADI) Score.

Clinical Parameter	Score 0	Score 1	Score 2	Score3
Respiratory Rate (per minute)	<40	40 –60	60 – 70	>70
Use of Accessory Muscles	None	1 accessory muscle used	2 accessory muscles used	>3 accessory muscles used
Color/Cyanosis	Pink in room air/no cyanosis	Cyanosed when crying	Pink with oxygen or cyanosed in room air	Cyanosed with oxygen or cardio-respiratory arrest
Auscultatory findings	Normal	Decreased air entry, no Rhonchi heard	Decreased air entry, Rhonchi heard	Silent chest

(Mansbach et al., 2009)

Ethical consideration:

1. Consent was taken from the parents/caregivers to participate in the study
2. Approval of ethical committee in the department, collegue and university were obtained before the study.
3. No conflect of interest and fund from any source.
4. The result of the study were confidential.
5. The patient has the right to withdraw from the study**.**

Statistical Analysis:

All data were analyzed using SPSS 19.0 for windows (SPSS Inc., Chicago, IL, USA) & Med Calc 13 for windows (Med Calc Software BVBA, Ostend, Belgium). Comparison between 2 groups was performed using Mann-Whitney *U*-Wilcoxon non-parametric test for continuous data and Fisher's exact test for categorical data. We constructed receiver operating characteristic (ROC)curves and determined the area under the curves (AUC).The AUC and its 95% confidence intervals were estimated for PRO-ADM and compared using anon parametric method. Sensitivity, specificity, positive and negative predictive values and cut-off point were calculated for PRO-ADM. P value less than 0.05 were considered significant.

RESULTS

Table (2): Demographic data and anthropometric measurement of studied groups

Demographic data	Pneumonia cases (N=60)		Control (N=30)		Test	p-value (Sig.)
	No.	%	No.	%		
Age (months)						
Mean ± SD	16.41± 14.81		17.76 ± 16.53		-1.217	0.224
Median (Range)	9.5 (60 – 5)		12.40 (60 – 2)			(NS)
Sex						
Male	30	50%	21	70%	3.258	0.071
Female	30	50%	9	30%		(NS)
Weight (kg)						
Mean ± SD	8.21 ± 3.44		12.36 ± 5.78		-3.473	0.001
Median (Range)	7.50 (4.50 – 15)		10 (6 – 25)			(S)
Height (cm)						
Mean ± SD	71.4 ± 14.83		83.90 ± 20.97		-2.840	0.005
Median (Range)	68 (54 – 105)		81 (55 – 128)			(S)
BMI						
Mean ± SD	15.83 ± 2.60		16.90 ± 2.88		-1.468	0.142
Median (Range)	15.90 (9.90 – 22.40)		16.30 (14 – 30)			(NS)

This table shows no statistical significant differences between cases and controls as regard to age , sex and BMI. However the weight and height of cases were lower than of controls with Statistically significant difference.

Table (3): Laboratory Data of studied groups

Hemoglobin (g/dl)				
Mean ± SD	9.83 ± 0.88	11.88 ± 0.77	-10.765	<0.001 **(HS)**
Median (Range)	10 (7.50 – 11.50)	11.90 (10.50 – 14)		
WBCs ($x10^3/mm^3$)				
Mean ± SD	12.97 ± 3.50	4.79 ± 1.10	16.506	<0.001 **(HS)**
Median (Range)	13 (5.80 – 21.50)	4.95 (3 – 7.50)		
Proadrenomedulin (nmol/L)				
Mean ± SD	1.52 ± 0.62	0.05 ± 0.04	-7.696•	<0.001 (HS)
Median (Range)	1.22 (0.10 – 2.93)	0.04 (0.03 – 0.30)		

This table shows high significant differences between studied cases and controls regarding hemoglobin, WBCs and Proadrenomedullin.

Table (4): Comparison between ProADM level in complicated and non complicated cases of pneumonia.

Serum proadrenomedulin (nmol/L)	**Pneumonia cases (N=60)**		**Test**	**p-value (Sig.)**
	Non complicated (N=54)	**Complicated (N=6)**		
Mean ± SD	1.41 ± 0.55	2.50 ± 0.24	3.574•	<0.001 **(HS)**
Median (Range)	1.20 (0.10 – 2.60)	2.45 (2.23 – 2.93)		

This table shows that serum ProADM level has high Statistically significant difference between complicated and non complicated cases.

Table (5): Comparison between cases with & without pleural effusion and serum ProADM.

Serum proadrenomedulin (nmol/L)	Pneumonia cases (N=60)		Test	p-value (Sig.)
	No pleural effusion (N=56)	Pleural effusion (N=4)		
Mean ± SD	1.46 ± 0.58	2.68 ± 0.22	-2.833•	0.005 (S)
Median (Range)	1.21 (0.10 – 2.60)	2.64 (2.47 – 2.93)		

This table shows that serum ProADM level has high Statistically significant difference between pleural effusion than non pleural effusion cases.

Table (6): Comparison between ProADM level & severity of pneumonia

Laboratory findings	Severity of pneumonia			Test	p-value (Sig.)
	Mild (N=7)	Moderate (N=39)	Severe (N=14)		
Proadrenomedulin (nmol/L)					
Mean ± SD	1.111 ± 0.219	1.396 ± 0.585	2.099 ± 0.514	12.547	0.002 (S)
Median (Range)	1.186 (0.810 – 1.420)	1.186 (0.100 – 2.650)	2.234 (1.130 – 2.930)		

This table shows Statistically significant difference in ProADM level in mild , moderate and severe cases of pneumonia, its level was increased with increasing severity of pneumonia.

Table (7): Sensitivity, Specificity, Positive Predictive Value, Negative Predictive Value, Accuracy and Area Under ROC Curve of Serum ProADM as a predictor for complication in CAP cases.

Cut-off values	SN % (95%CI)	SP % (95%CI)	PPV % (95%CI)	NPV % (95%CI)	Accuracy (95% CI)	AUROC (95%CI)
Pro-ADM >2.202	100% (54.1-100)	81.5% (68.6-90.7)	37.5% (15.2-64.6)	100% (92-100)	83.4% (67.2-91.6)	0.948 (0.8570.988)

From Receiver Operating Characteristics (ROC) curve which detect complications of pneumonia, the best cutoff point for serum ProADM

levels in the studied groups as a predictor for complications was 2.2 nmol\L with sensitivity 100.%, Specificity 81.5%, positive predictive value 37.5%, negative predictive value 100%, accuracy 83.4% and area under ROC curve 0.948.

Table (8): Sensitivity, Specificity, Positive Predictive Value, Negative Predictive Value, Accuracy and Area Under ROC Curve of Serum ProADM as a predictor for severity in CAP cases.

Cut-off values	SN % (95% CI)	SP % (95% CI)	PPV % (95% CI)	NPV % (95% CI)	Accuracy (95% CI)	AUROC (95% CI)
Pro-ADM >1.950	78.6% (49.2-95.3)	71.8% (55.1-85)	50% (27.7-72.3)	90.3% (74.2-98)	73.6% (53.5-87.7)	0.780 (0.645-0.882)

From Receiver Operating Characteristics (ROC) curve which detect severity of pneumonia, the best cutoff point for serum ProADM levels in the studied groups as a predictor for severity was 1.9 nmol\L with sensitivity 78.6%, Specificity 71.8%, positive predictive value 50%, negative predictive value 90.3%, accuracy 73.6% and area under ROC curve 0.780.

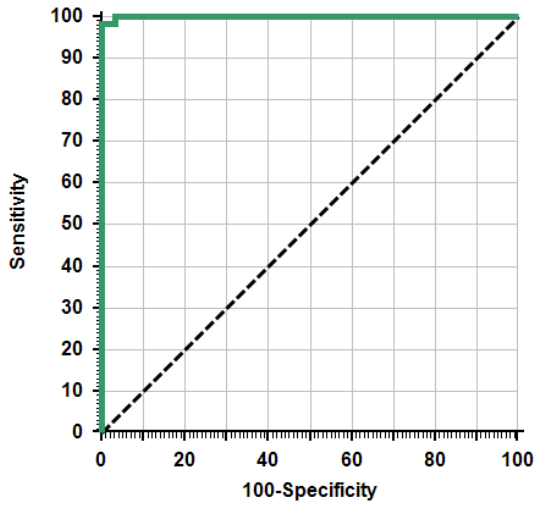

Figure (1): Receiver operating characteristic (ROC) curve of serum proadreno-medulin as a marker for complications of community-acquired pneumonia.

The cutoff point for serum ProADM levels in the studied groups as a predictor for complications was 2.2 nmol\L.

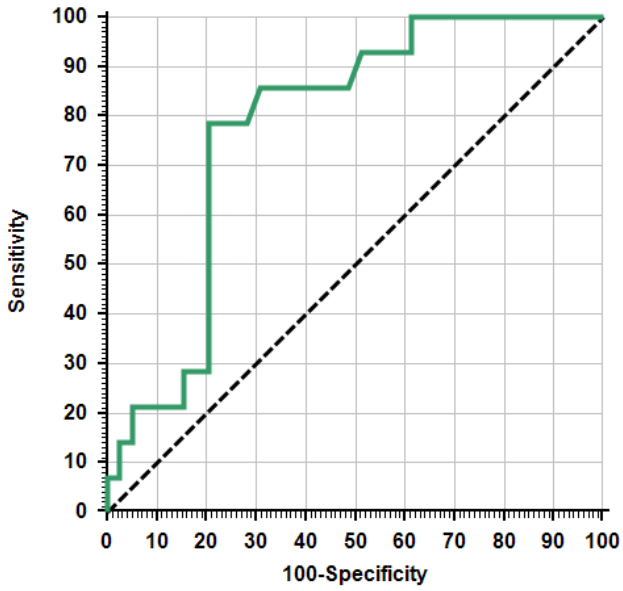

Figure (2): Receiver operating characteristic (ROC) curve of serum proadrenomedulin as a marker for differentiation between moderate community-acquired pneumonia and severe community-acquired pneumonia.

The cutoff point for serum ProADM levels in the studied groups as a predictor for severity was 1.9 nmol\L

DISCUSSION

CAP remains from the first three causes of mortality in children under five years after diarrhea and accidents. It ranges from mild affection to severe affection that requires ICU admission, due to severe complications as respiratory failure, severe sepsis or septic shock. In these patients, mortality rate is high, reaching 50% particularly in patients requiring vasopressor support. **(7)**

Many biomarkers have been used to help in diagnosis of infections like CAP. Some are used currently in clinical practice as WBC count, C-reactive protein and Pro-calcitonin. **(8)**

Regarding ProADM levels, our study showed that its level in cases was higher than in controls with mean ± SD (1.52±0.62 & 0.05±0.04) respectively.

Agustín et al., **(9)** found that, Mid Regional- ProADM levels were found to be increased in

patients with pneumonia compared with other patients with lower respiratory tract infections (acute bronchitis and acute exacerbation of asthma).

In our study, we found that there was statistically significant increase in ProADM level with increasing in severity of pneumonia **.**

Our findings is also matched with **Keuger et al ., (8)** who mentioned that there was a statistically significant increase between ProADM levels in patients with severe CAP than those with mild to moderate illness.

In our study, ProADM showed highly statistically significant increase in complicated cases than in non complicated ones.

This in agreement with ***Sanchez et al., (12)***, who concluded that ProADM is statistically significant higher in patients who developed pleural effusion or other pneumonia complications compared to uncomplicated pneumonia,($p < 0.001$).

In agreement with ***Bello et al, (10)*****,** who found that patients with complicated pneumonia had higher levels of ProADM than those who did not develop complications (2.3190 vs. 1.1758 nmol/L, $p = 0.013$)

In our study, we found that ProADM level was statistically significantly increased in patient with pleural effusion more than in patients who did not develop pleural effusion with mean (2.68 Vs 1.46 nmol/l) repectively.

This in agreement with ***Sanchez et al ., (12)***, who found that cases with pleural effusion had a median level of ProADM 2.9440 vs. 1.1373 nmol/L for the other cases not developed pleural effusion.

Our study relieved that cut-off value of ProADM to predict complications in CAP was 2.2 nmol/L with sensitivity (100%), specificity (81.5%), positive predictive value 37.5% ,negative predictive value 100%

Sanchez et al.(12) stated a cut-off point of 1.8 nmol/L (80 % sensitivity, 72% specificity).

Alcoba et al.,(11) concluded that diagnostic performance of ProADM seems excellent as accurate as classical markers as WBCs and CRP for predicting complication in CAP with sensitivity (72.7%), specificity (71.4%) and cut off point>0.16 nmol/L.

CONCLUSION

In conclusion, we found that ProADM seems to be an interesting marker to predict complication in children admitted with CAP. Studies with Large number of patients needed to confirm promising performances of ProADM in children with pneumonia.

RECOMMENDATION

We recommended that using of pro-adrenomedullin level in patients with CAP in emergency department for prediction severe and complicated cases to prevent unnecessary admission to ICU.

REFERENCES

1. **Black RE, Cousens S, Johnson HL, Lawn JE, Rudan I, Bassani DG, et al.:** Global, regional, and national causes of child mortality in 2008: a systematic analysis. Lancet.2010; 375(9730):1969–87

2. **Courtais C, Kuster N, Dupuy AM, et al.:** Proadrenomedullin, a useful tool for risk stratification in high Pneumonia Severity Index score community-acquired pneumonia. Am J Emerg Med. 2013; 31: 215-21.

3. **Müller NL, Franquet T and Lee KS.**: In: McAllister L, editor.Imaging of pulmonary infections. Philadelphia, Pa: Wolters Kluwer/Lipponcott Williams & Wilkins, 2007.

4. **ZhydkovA , Christ-Crain M , Thomann R , et al.:** Clinical Chemistry and Laboratory Medicine (CCLM). 2014 Volume 53. ; P. 559-66.

5. **Sardà Sánchez M, Joan Calzada Hernández, Susanna Hernández-Bou et al.:** pro-adrenomedullin usefulness in the management of children with community-acquired pneumonia, a preliminar prospective observational studyBMC Research Notes. 2012;5:363.

6. **Mansbach JM, Espinola JA, Macias CG, et al.**: Variability in the diagnostic labeling of nonbacterial lower respiratory tract infections: a multicenter study of children who presented to the emergency department. Pediatrics 2009; 123:e573-8

7. **Fine MJ and Aujesky D .:** The pneumonia severity index: a decade after the initial derivation and validation. Clinical infectious diseases: an official publication of the Infectious Diseases Society of America.2008 ; 47 3:S133–9.

8. **Krüger S, Ewig S, Giersdorf S,etal**: Cardiovascular and inflammatory Biomarkers to predict short- and long-term survival in community acquired pneumonia:CAPNETZ . AM J RespirCrit Care Med 2010; 182:1426 -34.

9. **Agustín Ruiz-González, Aureli Esquerda, José M. Porcel1, et al :**A Pilot Study on the Diagnostic Accuracy of Proadrenomedullin and Proatrial NatriureticPeptide in Lower Respiratory Tract Infections.*The Open Respiratory Medicine Journal,2014 , 8,* 22-27

10. **Bello S, Lasierra AB, Mincholé E, etal**: Prognostic power of proadrenomedullin in community-acquired

pneumonia is independent of aetiology 2012.

11. **Alcoba G, Manzano S, LacroixL,et al. :** Proadrenomedullin and copeptin in pediatric pneumonia: a prospective diagnostic accuracy study.BMC Infectious Diseases.2015 ; 15:347.

12. **Sardà Sánchez M, Joan Calzada Hernández, Susanna Hernández-Bou et al.:** Pro-adrenomedullin usefulness in the management of children with community-acquired pneumonia, a preliminar prospective observational study BMC Research Notes. 2012; 5:363.

13. **Harris M, Clark J, Coote N, et al**.: British Thoracic Society guidelines for the management of community acquired pneumonia in children: update. Thorax. 2011; 66 Suppl 2:ii1-ii23.

دور بروأدرينوميدالين كأحد دلالات مرض الالتهاب الرئوي في الأطفال

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نبذة مختصرة:

خلفية: الالتهاب الرئوي المكتسب من المجتمع ، هو سبب مهم من وفيات الأطفال. بروادرينوميدالين ، هو الببتيد التنظيمي الجديد المستخرج من ورم القواتم (فيوكروموسيتوما) و هو مقدمة من أدرينوميدالين ولكن بشكل أكثر استقرارا. وجدت دراسات الكبار أن بروادرينوميدالين مرتفع في المرضى الذين يعانون من الالتهاب الرئوي، ووجد أن هذا مرتبط مع شدة ومضاعفات الألتهاب الرئوي. أيضا دراسات أخرى استخدمت بروادرينوميدالين كعلامة النذير لكثير من الأمراض مثل تعفن الدم، صدمة حمى الضنك وفشل القلب.

الهدف: هدفت هذه الدراسة إلى قياس مستويات بروادرينوميدالين لدى الأطفال الذين يعانون من الالتهاب الرئوي وربط مستوياته بشدة ومضاعفات الألتهاب الرئوي

الطريقة: تم إجراء هذه الدراسة على 90 طفلا (تم تشخيص 60 طفلا بأن لديهم الالتهاب الرئوي و 30 طفلا هم الضوابط الأصحاء مع مطابقة العمر والجنس). وقد تعرض جميع الأطفال لأخذ التاريخ والفحص الإكلنيكي والفحوصات الاتية: (أشعة سينية (إكس) علي الصدر وصورة دم كاملة، والبروتين C التفاعلي، وقياس مستوي بروادرينوميدالين بطريقة الايلايزا).

واستخدمت مجموع نقاط أداة قياس الاعتلال التنفسي المعدل (رداي) لتصنيف الحالات إلى خفيفة، معتدلة وشديدة.

النتائج: تم العثور على مستوى مصل الدم من بروادرنوميدالين أعلى في حالات الالتهاب الرئوى مما كانت عليه في الضوابط الأصحاء وكان الفرق ذو دلالة إحصائية متوسط ±

انحـراف معيـارى (1,52 ± 0,62 و 0,05 ± 0,04) علـى التـوالى. كمـا وجـد أن بروادرنوميـدالين أعلـى فـي الحـالات ذوات المضـاعفات وهـم 6 حـالات (4 الانصـباب البلوري، 1 الانهيار في الرئة، 1استرواح هوائى بالصدر) من الحـالات التـى لـيس لهـا مضاعفات وكان الفرق ذو دلالـة إحصـائية متوسـط ± انحـراف معيـارى (2,50± 0,24 و1,41 ± 0.05) على التوالي. بروادرنوميدالين يتزايد مع زيـادة شـدة الالتهـاب الرئـوي. نقطـة القطـع> 2.2 نـانومول / لتـر كـان أفضـل مسـتوى للتمييـز بـين الحـالات ذوات المضـاعفات والحـالات التـي لـيس لهـا مضـاعفات (قيمـة ب=0.01، الحساسـية 100٪، النوعية 81.5٪، القيمة التنبؤية الإيجابية 37.5٪، القيمة التنبؤية السلبية 100٪).

الاسـتنتاج: مسـتوى بروادرينوميـدالين فـي مرضـى الالتهـاب الرئـوي اعلـى مـن تلـك الموجودة في الأصحاء ومستويات أعلى وجدت في الحالات الشديدة والمضاعفات

التوصيات: لقد وصينا باستخدام نسبة البروادرينوميـداللين فـى مرضـى الالتهـاب الرئـوى المكتسـب مـن المجتمـع بقسـم الطـوارئ لتوقـع الحـالات الشـديدة ذوات المضـاعفات لمنـع الحجز غير الضرورى بالعناية المركزة.

الكلمات الرئيسية: بروادرينوميدالين، الالتهاب الرئوي المكتسب من المجتمع، الالتهاب الرئوي للأطفال.