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Circulating Biologically Active Adrenomedullin (bio-ADM) Predicts Hemodynamic Support Requirement and Mortality During Sepsis

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BACKGROUND: The biological role of adrenomedullin (ADM), a hormone involved in hemodynamic homeostasis, is controversial in sepsis because administration of either the peptide or an antibody against it may be beneficial.

METHODS: Plasma biologically active ADM (bio-ADM) was assessed on days 1, 2, and 7 after randomization of 956 patients with sepsis or septic shock to albumin or crystalloids for fluid resuscitation in the multicenter Albumin Italian Outcome Sepsis trial. We tested the association of bio-ADM and its time-dependent variation with fluid therapy, vasopressor administration, organ failures, and mortality.

RESULTS: Plasma bio-ADM on day 1 (median [Q1-Q3], 110 [59-198] pg/mL) was higher in patients with septic shock, associated with 90-day mortality, multiple organ failures and the average extent of hemodynamic support therapy (fluids and vasopressors), and serum lactate time course over the first week. Moreover, it predicted incident cardiovascular dysfunction in patients without shock at enrollment (OR [95% CI], 1.9 [1.4-2.5]; $P < .0001$, for an increase of 1 interquartile range of bio-ADM concentration). bio-ADM trajectory during the first week of treatment clearly predicted 90-day mortality after adjustment for clinically relevant covariates (hazard ratio [95% CI], 1.3 [1.2-1.4]; $P < .0001$), and its reduction below 110 pg/mL at day 7 was associated with a marked reduction in 90-day mortality. Changes over the first 7 days of bio-ADM concentrations were not dependent on albumin treatment.

CONCLUSIONS: In patients with sepsis, the circulating, biologically active form of ADM may help individualizing hemodynamic support therapy, while avoiding harmful effects. Its possible pathophysiologic role makes bio-ADM a potential candidate for future targeted therapies.

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ABBREVIATIONS: ADM = adrenomedullin; ALBIOS = Albumin Italian Outcome Sepsis; bio-ADM = bioactive adrenomedullin; HR = hazard ratio; SOFA = sequential organ failure assessment

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Since its discovery in 1993,[1] the 52-amino acid peptide adrenomedullin (ADM) has been implicated in the pathobiology of several diseases, including cardiovascular disorders.[2] ADM is indeed up-regulated in different tissues as well as in several conditions, in association, among others, with myocardial injury, systemic inflammatory response syndrome, shock, cellular hypoxia, and oxidative stress. In addition, remarkably high circulating levels of ADM have been reported in sepsis,[3] where its role remains controversial.[4] On one hand, during sepsis, ADM can exert beneficial effects by reducing endothelial hyperpermeability and vascular leakage, as shown in cell culture models.[5-7] Indeed, exogenous administration of ADM appears to be protective in animal models of endotoxin[8] and α-toxin infusion,[6] or ARDS.[9] On the other hand, it is well known that higher circulating levels of ADM are invariably associated with worse outcomes.[10-13] Moreover, administration of anti-ADM antibodies can attenuate sepsis-induced multiple organ failures in murine models,[14-16] and a humanized monoclonal anti-ADM antibody is under clinical development.[17] Several factors may be at the interplay of such apparent paradox, such as differences in the circulating levels of ADM, the specific type of experimental model used, the stage of the disease, and the underlying co-morbidities and co-interventions in clinical settings.

The interpretation of ADM as a biomarker is difficult because several fragments of the precursor prohormone circulate in blood and do not necessarily reflect the activity of ADM. A sandwich immunoassay for specifically measuring the biologically active (amidated) form of ADM (named bioactive ADM or bio-ADM) has been developed.[18] By using these reagents, bio-ADM levels in patients being evaluated at the ED for acute heart failure were shown to be predictive of clinically relevant 30-day outcomes.[19] In a monocentric study performed on 101 consecutive patients presenting at the ED with the suspicion of sepsis, Marino et al[18] observed that bio-ADM levels were associated with short-term mortality and vasopressor requirement.

Here we aimed at testing, for the first time, this biomarker in a large and well-defined cohort of patients with severe sepsis or septic shock, enrolled in the biomarker substudy of the Albumin Italian Outcome Sepsis (ALBIOS) trial.[20] Given the biological role of ADM in fluid homeostasis and systemic circulation, and the profound pathophysiologic perturbations seen in the early phase of sepsis and during its treatment, we were particularly interested in describing the relation between bio-ADM and hemodynamic support requirements during the first days of treatment. In addition, we described bio-ADM trajectory over time and its prognostic value for 90-day mortality.

Materials and Methods

Study Design

ALBIOS was a multicenter, pragmatic, open-label, randomized trial that enrolled 1,818 patients with severe sepsis or septic shock admitted to 100 ICUs. Study design, inclusion and exclusion criteria, and main results have been published elsewhere.[20]

The study complied with the 1975 Declaration of Helsinki as revised in 2008 and was approved by the institutional review board of each center. Written informed consent or deferred consent was obtained from each participant, according to the Italian legislation.

A detailed description of data collection procedures, clinical definitions and statistical methods can be found in e-Appendix 1.

Sample Collection and Circulating Biomarker Measurements

In a subset of 956 patients recruited from 40 centers participating in a predefined biomarker substudy, venous blood samples were serially collected 1, 2, and 7 days after enrolment (or at ICU discharge, whichever came first) and centrifuged; plasma was then shipped on dry ice to a central repository and stored at −70°C until assayed. The bio-ADM was measured using a chemiluminescence immunoassay provided by Sphingotec GmbH.[18] The assay sensitivity is 2 pg/mL. In prior work,[18] the median bio-ADM concentration of 200 healthy adults equaled 20.7 pg/mL, with a 99th percentile of 43.0 pg/mL.

Results

Variables Associated With bio-ADM Levels

Median concentration of bio-ADM on day 1 was 110 pg/mL (interquartile range, 59-198) (n = 956). Table 1

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TABLE 1] Patient Demographic and Baseline Physiologic Characteristics According to Tertiles of bio-ADM Concentrations on Day 1

Characteristics	T1 (<73 pg/mL)	T2 (73-153 pg/mL)	T3 (≥154 pg/mL)	*P* Value
No. (%)	319 (33.4)	318 (33.3)	319 (33.4)	
Age (y)	68 (53-76)	71 (60-78)	71 (62-78)	.004
Men, No. (%)	194 (60.8)	193 (60.7)	174 (54.5)	.19
BMI (kg/m^2)	24.8 (23.2-27.7)	25.5 (23.2-28.6)	26.1 (23.8-29.4)	.004
Reason for admission to ICU, No. (%)				< .0001
Elective surgery	28 (8.8)	16 (5)	23 (7.2)	
Emergency surgery	86 (27)	115 (36.2)	150 (47)	
Medical	205 (64.3)	187 (58.8)	146 (45.8)	
Pre-existing conditions, No. (%)				
Liver disease	3 (0.9)	5 (1.6)	6 (1.9)	.60
COPD	37 (11.6)	29 (9.1)	52 (16.3)	.02
Chronic renal failure	10 (3.1)	11 (3.5)	21 (6.6)	.06
Immunodeficiency	27 (8.5)	40 (12.6)	57 (17.9)	.002
Congestive or ischemic heart disease	53 (16.6)	50 (15.7)	61 (19.1)	.50
Any previous conditions	105 (32.9)	110 (34.6)	150 (47)	< .0001
SAPS II score	38 (30-50)	45 (35-55)	53 (43-63)	< .0001
SOFA score	5 (3-7)	7 (5-10)	10 (8-12)	< .0001
Physiological variables				
Heart rate (beats/min)	90 (76-101.5)	95 (80-110)	104 (89-117)	< .0001
Mean arterial pressure (mm Hg)	85 (77-93)	80 (72-88)	75 (67-83)	< .0001
Central venous pressure (mm Hg)	10 (7-12)	11 (8-13.75]	12 (9-15)	< .0001
Central venous oxygen saturation (%)	75 (70-80)	75 (70-80)	76 (70-80)	.886
PaO_2/FIO_2	218 (160-284)	228 (163-284)	200 (138-272)	.008
Urinary output (mL)[a]	2,050 (1,310-3,003)	1,970 (1,115-3,015)	1,285 (568-2,200)	< .0001
Laboratory variables				
Serum albumin (g/L)	28 (24-31)	27 (23-31)	25 (21-29)	< .0001
Serum lactate (mmol/L)	1.3 (0.9-1.8)	1.6 (1.2-2.42)	2.3 (1.6-4.2)	< .0001
Hemoglobin (g/dL)	10.6 (9.6-11.8)	10.5 (9.5-11.6)	10.5 (9.5-11.5)	.47
Platelet count (10^9/L)	178.0 (119.5-240.5)	140.0 (80.0-218.5)	113.5 (63.0-185.0)	< .0001
Serum creatinine (mg/dL)	0.9 (0.6-1.3)	1.3 (0.9-2.2)	2.0 (1.3-3.2)	< .0001
White blood cells (10^3/mm^3)	12.1 (8.6-16.7)	12.5 (7.5-19.8)	11.7 (6.9-17.5)	.26
Serum bilirubin (mg/dL)	0.7 (0.4-1.1)	0.9 (0.5-1.6)	1.3 (0.7-2.4)	< .0001
Positive blood culture, no. events/no. patients (%)	59/280 (21.1)	100/288 (34.7)	112/282 (39.1)	< .0001
Septic shock, No. (%)	139 (43.6)	194 (61)	206 (64.6)	< .0001

(Continued)

TABLE 1] *(Continued)*

Characteristics	T1 (<73 pg/mL)	T2 (73-153 pg/mL)	T3 (≥154 pg/mL)	*P* Value
Randomized to albumin, No. (%)	161 (51)	159 (50)	153 (48)	.80
Antibiotics at randomization, No. (%)	294 (92.2)	300 (94.3)	302 (94.7)	.37
Antibiotics 6 h after randomization, No. (%)	319 (100)	315 (99.1)	318 (99.7)	.17
90-d mortality, No. events /No. patients (%)	87/312 (27.9)	117/316 (37.0)	165/315 (52.4)	< .0001

SAPS II = Simplified Acute Physiology Score II; SOFA = Sequential Organ Failure Assessment.
[a]Urinary output indicates total urinary output from the time of enrollment to day 1.

shows patients' demographic and baseline characteristics according to tertiles of bio-ADM concentrations on the same day. By multiple linear regression analysis, the variables most strongly associated with higher bio-ADM concentrations, were, by decreasing order of r^2, higher sequential organ failure assessment (SOFA) score ($F = 31$, $P < .0001$), ICU admission after emergency surgery ($F = 15$, $P < .0001$), higher heart rate ($F = 21$, $P < .0001$), higher serum creatinine levels ($F = 17$, $p<0.0001$), higher central venous pressure ($F = 16$, $P = .0001$), lower serum albumin concentration ($F = 14$, $P = .0002$), positive blood culture ($F = 7$, $P = .001$), lower mean arterial pressure ($F = 13$, $P = .0003$), higher BMI ($F = 12$, $P = .0005$), and immunodeficiency ($F = 11$, $P = .001$).

bio-ADM, Hemodynamic Goals, and Support Requirements

Higher concentrations of bio-ADM on day 1 were associated with less frequent achievement of hemodynamic goals during the first 6 to 24 hours after randomization in terms of central venous pressure and oxygen saturation, mean arterial pressure, and serum lactate concentrations (e-Table 1). Patients in the upper tertile of bio-ADM more frequently received two or more vasoactive drugs and showed higher values of inotropic score and vasopressor dependency index compared with the middle and the lower tertiles (e-Table 1). As shown in Figure 1, the inotropic score over the first 7 days of treatment in patients alive at that time was greater in those with higher levels of bio-ADM at day 1 compared with those with lower levels. In parallel, serum lactate levels were significantly higher in patients in the upper tertile (Fig 1). Moreover, patients in the upper tertile of bio-ADM also showed greater need of fluids during the first week of treatment, as shown by the average fluid administered, the average amount of albumin or crystalloids received, and the average and the cumulative fluid balances observed over the first 7 days (e-Table 1).

bio-ADM and Organ Failures

Prevalent and incident organ failures, defined as organ-specific SOFA scores of 3 or 4 for the respiratory, coagulation, and hepatic and renal systems, were more frequent at higher concentrations of bio-ADM on day 1 (e-Fig 1). Accounting for multiple organ failures and death, we observed elevated bio-ADM levels in patients with multiple prevalent or multiple incident organ failures and in those who died during the follow-up period.

bio-ADM and Septic Shock

Patients presenting with septic shock at the time of enrollment had significantly higher concentrations of bio-ADM than those with severe sepsis only ($P < .0001$) and showed a greater reduction over the first 7 days compared with patients without shock (P for interaction time × shock < .0001; e-Fig 2).

Among patients without shock at study entry, there was a progressive increase in the likelihood of having incident shock with higher concentrations of bio-ADM (e-Fig 3), which remained significant after adjustments for the Simplified Acute Physiology Score II or for clinically relevant variables (e-Table 2).

bio-ADM and 90-Day Mortality

During 90 days of follow-up, 369 patients (38.6%) died. Main clinical characteristics of survivors and nonsurvivors are shown in e-Table 3. e-Figure 4 shows the Kaplan-Meier mortality at 90 days by tertiles of

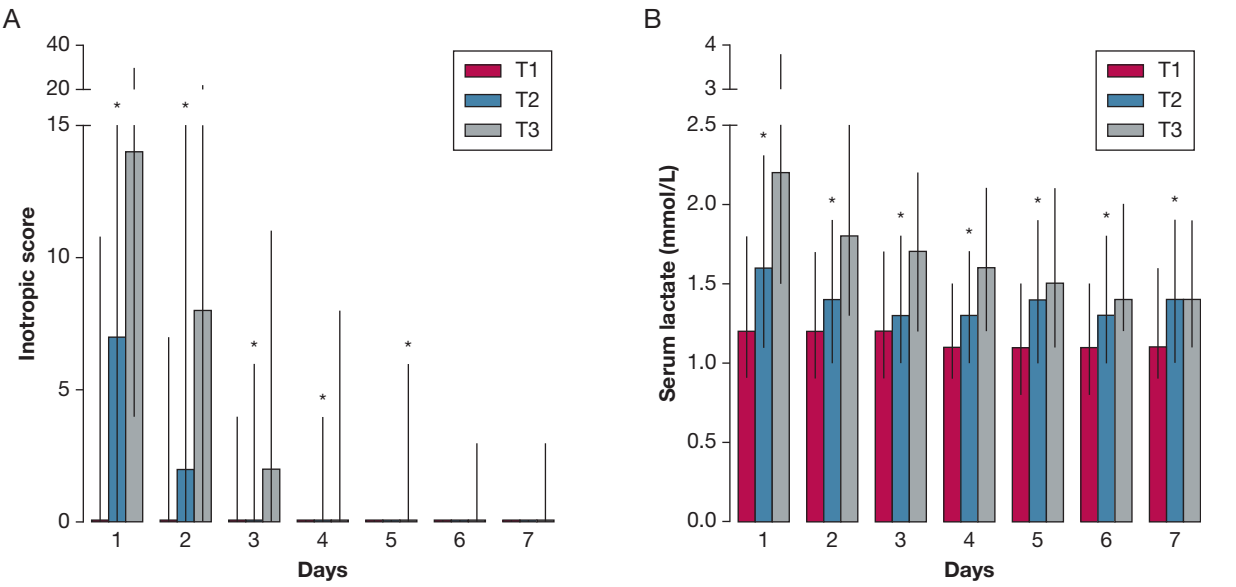


Figure 1 – *Time course of inotropic score and serum lactate concentration according to early bio-ADM levels. Evolution of the inotropic score (A) and serum lactate concentrations (B) over the first 7 days after enrollment in patients alive at 7 days, according to tertiles of bio-ADM concentrations on day 1. The inotropic score was calculated each day as follows (30): (dopamine dose × 1) + (adrenaline dose × 100) + (noradrenaline dose × 100), wherein all doses are expressed as μg/kg/min. See Methods for further details. *P < .01 across tertiles of bio-ADM by linear regression analysis. After adjustment for the simplified acute physiologic score II score, P < .05 at day 1 and day 2 for inotropic score; P ≤ .01 from day 1 to day 7 for serum lactate. bio-ADM = bioactive adrenomedullin.*

bio-ADM concentrations on day 1, with a clear, stepwise increment of the risk of mortality with increasing bio-ADM levels. The progressive relationship between bio-ADM concentrations on day 1 and 90-day mortality (with a 1.9-fold relative increase in event rate from the lower to the upper tertile), was confirmed by restrictive spline curves (e-Fig 5).

In Cox proportional hazard models adjusted for clinically relevant covariates, bio-ADM concentrations on day 1 remained independently associated with 90-day mortality in the overall cohort (Table 2).

Time Course of bio-ADM and 90-Day Mortality

To evaluate the association between the variation over time of bio-ADM and short-term mortality, we analyzed bio-ADM time course over the first 7 days by mortality after this period. bio-ADM decreased over the first 7 days after randomization in the subgroup of patients surviving the first week of treatment, whereas it remained stable in those dying (*P* for interaction between time and mortality < .0001, e-Fig 6).

Figure 2 shows the Kaplan-Meier survival curves for 90-day mortality according to the evolution of bio-ADM levels from day 1 to day 7, stratified according to the median concentration of bio-ADM on day 1 (110 pg/mL). Patients with bio-ADM concentrations decreasing from above the median value on day 1 to below the median value on day 7 showed a 90-day mortality rate close to those with bio-ADM remaining below the median value at both time points (33.3 vs 26.7%, Fig 2). In contrast, outcome was markedly worse and comparable both in patients with increasing bio-ADM concentrations over time (66.7%) and in those with higher bio-ADM levels at both time points (64.6%). In time-dependent Cox regression models adjusted for clinically relevant covariates, evolution of bio-ADM concentrations over the first 7 days was independently associated with 90-day mortality (Fig 3).

bio-ADM and Other Clinical Outcomes

Finally, higher concentrations of bio-ADM on day 1 were associated with further clinically relevant outcomes (e-Table 4). Of note, after adjustments, patients with higher levels of bio-ADM at day 1 showed longer length of hospital stay and a higher incidence of renal replacement therapy during the study period (*P* <.01, for both) compared with patients with lower levels of bio-ADM.

Randomized treatment (albumin or crystalloids vs crystalloids alone) had no effect on bio-ADM concentrations over time (e-Table 5).

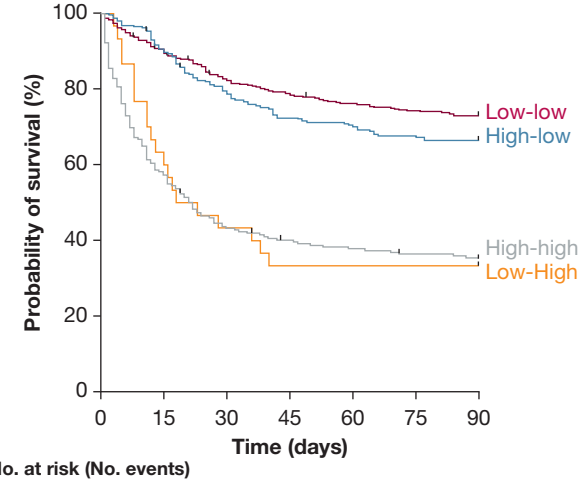
TABLE 2] Relation of bio-ADM with 90-Day Mortality

	No. Events/No. Patients at Risk	$\chi^2/\Delta\ \chi^2$	*P* Value	HR (95% CI)
All patients				
Univariate (bio-ADM)	369/956	64.9	< .0001	1.38 (1.29-1.47)
Bivariate (bio-ADM and SAPS II)	369/956	18.6	< .0001	1.21 (1.12-1.31)
Multivariable	290/788	5.9	.015	1.14 (1.03-1.27)
Patients with severe sepsis				
Univariate (bio-ADM)	135/417	27.9	< .0001	1.50 (1.31-1.71)
Bivariate (bio-ADM and SAPS II)	135/417	9.5	.0021	1.29 (1.11-1.50)
Multivariable	106/342	6.0	.01	1.28 (1.06-1.53)
Patients with septic shock				
Univariate (bio-ADM)	234/539	34.0	< .0001	1.35 (1.24-1.47)
Bivariate (bio-ADM and SAPS II)	234/539	10.1	.0015	1.19 (1.08-1.32)
Multivariable	184/446	2.0	.16	1.10 (0.97-1.25)

bio-ADM concentration on day 1 was entered as continuous variable. The multivariable model includes bio-ADM, age, SOFA score, serum lactate concentration, and central venous oxygen saturation at baseline.[20] HR interval for an increase of 1 interquartile range of bio-ADM concentration on day 1. $\Delta\chi^2$ refers to an incremental value of bio-ADM compared with the model with clinical variables alone. bio-ADM = bioactive adrenomedullin; HR = hazard ratio. See Table 1 legend for expansion of other abbreviation.

Discussion

In our study, we showed that (1) plasma concentration of bio-ADM was high, especially in patients with septic shock; (2) higher bio-ADM on day 1 predicted the intensity of the hemodynamic support requirement over the first week of treatment; (3) bio-ADM predicted incident organ-specific failure and shock; and (4) bio-ADM's trajectory over the first week was



No. at risk (No. events)			
Low-low	448	348 (96)	321 (120)
High-low	255	183 (70)	168 (84)
Low-High	30	10 (20)	10 (20)
High-High	223	87 (133)	76 (144)

Figure 2 – *Mortality according to the evolution of bio-ADM concentrations over 7 days. Patients were divided into four groups according to bio-ADM concentrations on day 1 and day 7, according to the median value of bio-ADM on day 1 (110 pg/mL). Kaplan-Meier survival curves at 90 days are shown for these four groups. See Figure 1 legend for expansion of abbreviation.*

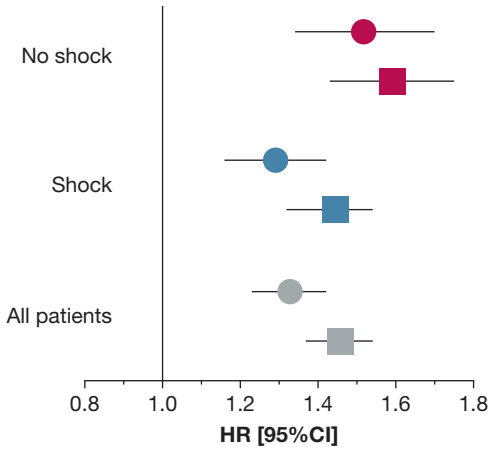


Figure 3 – *Time-dependent bio-ADM concentration predicts 90-day mortality. Association between time-dependent bio-ADM concentration from day 1 to day 7 and 90-day mortality in the entire study population and by the presence or absence of shock at the time of enrollment. Time-dependent bio-ADM concentration from day 1 to day 7 was entered as a linear variable in univariate (square) or multivariable (circle) models. The model included time-dependent bio-ADM, age, sequential organ failure assessment score, serum lactate concentration, and central venous oxygen saturation at baseline.[20] HR interval for an increase of 1 interquartile range of time-dependent bio-ADM concentration. HR, hazard ratio. See Figure 1 legend for expansion of other abbreviation.*

strongly and independently associated with 90-day mortality.

ADM exerts multiple effects with possible clinical importance, such as vasodilation, diuresis, and natriuresis.[21] Despite its importance in the pathophysiology of sepsis,[22] experimental findings seem controversial: two apparently opposite interventions, exogenous administration of the peptide or modulation with antibodies directed against the same peptide, produce benefits in preclinical sepsis models.[4] The design of the present study is not ideal for providing mechanistic insights into the role of ADM in sepsis. We confirm that the biologically active ADM circulates in elevated concentrations during severe sepsis and septic shock. In previous studies, ADM levels have been estimated either with a competitive radioimmunoassay with technical limitations,[10] with an assay of unknown analytical performance,[13] or by measuring an inactive, stable fragment of pro-ADM.[11,12] Because the activity of ADM depends on the conversion of the C-terminal glycine residue to an amide and because amidation rate differs among pathological states,[23,24] the levels measured in the present study are very likely to better reflect the biological relevance of ADM in sepsis.

Aggressive fluid therapy and hemodynamic support to restore peripheral organ perfusion based upon predefined hemodynamic targets are the cornerstone of the management of patients with sepsis.[25] Despite these standardized procedures, several experimental and clinical evidence suggest that this approach may have harmful effects, and should be individualized.[26-30] Because instrumental and laboratory measurements may have incomplete value in guiding individual hemodynamic support requirement,[31-34] we sought to investigate whether a simple, objective measurement of a circulating biomarker early in the clinical course of sepsis could predict the subsequent need for fluids or vasopressors administration. We indeed observed that higher bio-ADM levels on day 1 predicted more positive fluid balance and higher doses of vasopressor agents administered during the first 7 days of treatment, independently on the overall patient severity. In the same cohort of patients, we previously showed that plasma concentrations of a natriuretic peptide were extremely elevated, up to levels normally encountered during acute heart failure.[35] We speculate that these levels may reflect excessive fluid administration in the early phase of the treatment of sepsis, resulting in high atrial and ventricular filling pressures, which are the main determinants of cardiac release of natriuretic peptides.[36] Taken together, our findings show a clear association between the up-regulation of a biomarker of an endocrine system controlling fluid homeostasis and vasodilation with fluid overloading as well as the intensity of hemodynamic support applied during severe sepsis and septic shock. It remains to be formally demonstrated, however, in appropriately designed, prospective clinical studies, that bio-ADM levels may be targeted by adjusting fluid therapy or vasopressors in sepsis, with a favorable effect on outcomes.

Higher levels of bio-ADM on day 1 predicted incident shock. Moreover, patients with early elevated bio-ADM had a higher proportion of organ dysfunction or failures. Of note, after correction for competing risks between the copresence of multiorgan failures or the subsequent development of death, we found an independent association between bio-ADM and multiple prevalent or incident organ failures and with the subsequent development of death during follow-up. Taken together, these results highlight the role of bio-ADM as an early indicator of an increased risk of sepsis-related morbidity and mortality. Our findings also support two clinically relevant aspects that merit further discussion. First, early bio-ADM monitoring may guide toward a timely implementation and monitoring of an adequate hemodynamic support in those patients who are likely to develop subsequently overt cardiovascular dysfunction. Indeed, similar to what has been observed for the initiation of antimicrobial therapy,[37,38] evidence has shown an increased risk of death for each hour delay in the initiation of vasopressor therapy during septic shock.[39] Second, because of the key role of the cardiovascular system in the pathophysiology of sepsis and septic shock, bio-ADM may appear as a novel candidate for a targeted therapy of sepsis, aimed at blunting its excessive effects.

The trajectory of bio-ADM over the first 7 days after the diagnosis of severe sepsis or septic shock was a strong indicator of subsequent mortality. We also confirm the prognostic value of repeated measurements of bio-ADM, even at 70 pg/mL, a different cutoff previously identified in an independent cohort of 101 consecutive patients admitted to the ED with suspected sepsis (e-Fig 7).[18] The association between time-dependent bio-ADM variation and outcome was independent of the more robust risk factors commonly observed in patients with sepsis, and already tested in the original ALBIOS trial[20] (ie, age, total SOFA score, serum lactate concentration, and central venous oxygen saturation). Consequently, bio-ADM may be considered a good candidate for early risk stratification in patients with sepsis or septic shock. Moreover, our data suggest that

the sequential assessment of bio-ADM may be important to monitor the efficacy of the hemodynamic treatment applied, which may be targeted to obtain a reduction of bio-ADM levels (ie, to a level lower than 110 pg/mL).

The present study has some limitations. First, ALBIOS was a pragmatic trial, and circulating biomarkers were assessed first on the morning after enrollment; therefore, bio-ADM levels on day 1 might have been affected by early study treatment. Nonetheless, the availability of plasma samples at later time points (over the first week of treatment) allows the evaluation of the time course of the biomarkers with a reasonable accuracy during the first critical phase of the treatment of sepsis. Second, because only a relative small percentage of patients initiated vasopressor therapy after day 1, the present study can only provide indication on the potential of bio-ADM as an early predictor of fluid requirement, or as an indicator of the response to vasopressor administration. Further studies in large cohorts of patients with the suspicion of sepsis and without shock are necessary to further elucidate this hypothesis.

Conclusions

We showed in a large and representative cohort of acutely ill patients with severe sepsis or septic shock that the circulating, biologically active form of adrenomedullin is extremely elevated. The strict association between early bio-ADM levels and subsequent development of multiple organ dysfunctions and death, in addition to hemodynamic support requirement, denotes an important role of bio-ADM in the pathophysiology of sepsis and septic shock. It remains to be demonstrated whether it will be valuable for clinical monitoring of fluid and vasopressor requirements to individualize an adequate hemodynamic support therapy and avoid potentially harmful effects.

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Author contributions: P. C. and S. M. had full access to all of the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis. P. C., R. L., and S. M. have contributed substantially to the conception and design, acquisition of data, analysis and interpretation of data, and have drafted the submitted article. G. M. and M. C. have contributed to the acquisition of data and have revised the submitted article critically for important intellectual content. O. H. has done the statistical analysis, contributed to the interpretation of data, and has revised the submitted article critically for important intellectual content. J. S., A. B., G. T., A. P., and L. G. have contributed to the conception and design of the study and have revised the submitted article critically for important intellectual content. All authors undertook final approval of the manuscript.

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Collaborators: A complete list of centers and investigators participating in the Albumin Italian Outcome Sepsis substudy on biomarkers has been published elsewhere (Masson S, Caironi P, Spanuth E, et al; ALBIOS Study Investigators. Presepsin (soluble CD14 subtype) and procalcitonin levels for mortality prediction in sepsis: data from the Albumin Italian Outcome Sepsis trial. *Crit Care.* 2014;18(1):R6.).

Additional information: The e-Appendix, e-Figures, and the e-Tables can be found in the Supplemental Materials section of the online article.

References

1. Kitamura K, Kangawa K, Kawamoto M, et al. Adrenomedullin: a novel hypotensive peptide isolated from human pheochromocytoma. 1993. *Biochem Biophys Res Commun*. 2012;425(3):548-555.
2. Ishimitsu T, Ono H, Minami J, Matsuoka H. Pathophysiologic and therapeutic implications of adrenomedullin in cardiovascular disorders. *Pharmacol Ther*. 2006;111(3):909-927.
3. Wang P. Adrenomedullin in sepsis and septic shock. *Shock*. 1998;10(5):383-384.
4. Kox M, Pickkers P. Adrenomedullin: its double-edged sword during sepsis slices yet again. *Intensive Care Med Exp*. 2014;2(1):1.
5. Hippenstiel S, Witzenrath M, Schmeck B, et al. Adrenomedullin reduces endothelial hyperpermeability. *Circ Res*. 2002;91(7):618-625.
6. Temmesfeld-Wollbruck B, Brell B, David I, et al. Adrenomedullin reduces vascular hyperpermeability and improves survival in rat septic shock. *Intensive Care Med*. 2007;33(4):703-710.
7. Temmesfeld-Wollbruck B, Brell B, zu Dohna C, et al. Adrenomedullin reduces intestinal epithelial permeability in vivo and in vitro. *Am J Physiol Gastrointest Liver Physiol*. 2009;297(1):G43-G51.
8. Ertmer C, Morelli A, Rehberg S, et al. Exogenous adrenomedullin prevents and reverses hypodynamic circulation and pulmonary hypertension in ovine endotoxaemia. *Br J Anaesth*. 2007;99(6):830-836.
9. Muller-Redetzky HC, Will D, Hellwig K, et al. Mechanical ventilation drives pneumococcal pneumonia into lung injury and sepsis in mice: protection by adrenomedullin. *Crit Care*. 2014;18(2):R73.
10. Ueda S, Nishio K, Minamino N, et al. Increased plasma levels of adrenomedullin in patients with systemic inflammatory response syndrome. *Am J Respir Crit Care Med*. 1999;160(1):132-136.
11. Christ-Crain M, Morgenthaler NG, Struck J, Harbarth S, Bergmann A, Müller B. Mid-regional pro-adrenomedullin as a prognostic marker in sepsis: an observational study. *Crit Care*. 2005;9(6):R816-R824.
12. Guignant C, Voirin N, Venet F, et al. Assessment of pro-vasopressin and pro-adrenomedullin as predictors of 28-day mortality in septic shock patients. *Intensive Care Med*. 2009;35(11):1859-1867.
13. Chen YX, Li CS. Prognostic value of adrenomedullin in septic patients in the ED. *Am J Emerg Med*. 2013;31(7):1017-1021.
14. Wang P, Ba ZF, Cioffi WG, Bland KI, Chaudry IH. The pivotal role of adrenomedullin in producing hyperdynamic circulation during the early stage of sepsis. *Arch Surg*. 1998;133(12):1298-1304.
15. Struck J, Hein F, Karasch S, Bergmann A. Epitope specificity of anti-adrenomedullin antibodies determines efficacy of mortality reduction in a cecal ligation and puncture mouse model. *Intensive Care Med Exp*. 2013;1(1):22.

16. Wagner K, Wachter U, Vogt JA, et al. Adrenomedullin binding improves catecholamine responsiveness and kidney function in resuscitated murine septic shock. *Intensive Care Med Exp*. 2013;1(1):21.

17. Blet A, Sadoune M, Polidano E, et al. Hemodynamic effects of adrecizumab in sepsis rat [abstract]. *Intensive Care Med Exp*. 2015;3:A618.

18. Marino R, Struck J, Maisel AS, Magrini L, Bergmann A, Di Somma S. Plasma adrenomedullin is associated with short-term mortality and vasopressor requirement in patients admitted with sepsis. *Crit Care*. 2014;18(1):R34.

19. Self WH, Storrow AB, Hartmann O, et al. Plasma bioactive adrenomedullin as a prognostic biomarker in acute heart failure. *Am J Emerg Med*. 2016;34(2):257-262.

20. Caironi P, Tognoni G, Masson S, et al. Albumin replacement in patients with severe sepsis or septic shock. *N Engl J Med*. 2014;370(15):1412-1421.

21. Hinson JP, Kapas S, Smith DM. Adrenomedullin, a multifunctional regulatory peptide. *Endocr Rev*. 2000;21(2):138-167.

22. Pugin J. Adrenomedullin: a vasodilator to treat sepsis? *Crit Care*. 2014;18(3):152.

23. Bunton DC, Petrie MC, Hillier C, Johnston F, McMurray JJ. The clinical relevance of adrenomedullin: a promising profile? *Pharmacol Ther*. 2004;103(3):179-201.

24. Ohta H, Tsuji T, Asai S, Tanizaki S, et al. A simple immunoradiometric assay for measuring the entire molecules of adrenomedullin in human plasma. *Clin Chim Acta*. 1999;287(1-2):131-143.

25. Dellinger RP, Levy MM, Rhodes A, et al. Surviving sepsis campaign: international guidelines for management of severe sepsis and septic shock: 2012. *Crit Care Med*. 2013;41(2):580-637.

26. Marik P, Bellomo R. A rational approach to fluid therapy in sepsis. *Br J Anaesth*. 2016;116(3):339-349.

27. Genga K, Russell JA. Early liberal fluids for sepsis patients are harmful. [published online April 7, 2016]. *Crit Care*. http://dx.doi.org/10.1097/CCM.0000000000001829

28. Sharif S, Owen JJ, Upadhye S. The end of early-goal directed therapy? *Am J Emerg Med*. 2016;34(2):292-294.

29. Alsous F, Khamiees M, DeGirolamo A, Amoateng-Adjepong Y, Manthous CA. Negative fluid balance predicts survival in patients with septic shock: a retrospective pilot study. *Chest*. 2000;117(6):1749-1754.

30. Boyd JH, Forbes J, Nakada TA, Walley KR, Russell JA. Fluid resuscitation in septic shock: a positive fluid balance and elevated central venous pressure are associated with increased mortality. *Crit Care Med*. 2011;39(2):259-265.

31. Saugel B, Ringmaier S, Holzapfel K, et al. Physical examination, central venous pressure, and chest radiography for the prediction of transpulmonary thermodilution-derived hemodynamic parameters in critically ill patients: a prospective trial. *J Crit Care*. 2011;26(4):402-410.

32. Marik PE, Cavallazzi R. Does the central venous pressure predict fluid responsiveness? An updated meta-analysis and a plea for some common sense. *Crit Care Med*. 2013;41(7):1774-1781.

33. Wetterslev M, Haase N, Johansen RR, Perner A. Predicting fluid responsiveness with transthoracic echocardiography is not yet evidence based. *Acta Anaesthesiol Scand*. 2013;57(6):692-697.

34. Eskesen TG, Wetterslev M, Perner A. Systematic review including re-analyses of 1148 individual data sets of central venous pressure as a predictor of fluid responsiveness. *Intensive Care Med*. 2016;42(3):324-332.

35. Masson S, Caironi P, Fanizza C, et al. Sequential N-terminal pro-B-type natriuretic peptide and high-sensitivity cardiac troponin measurements during albumin replacement in patients with severe sepsis or septic shock. *Crit Care Med*. 2016;44(4):707-716.

36. Latini R, Caironi P, Masson S. Cardiac dysfunction and circulating cardiac markers during sepsis. *Minerva Anestesiol*. 2016;82(6):697-710.

37. Kumar A, Roberts D, Wood KE, et al. Duration of hypotension before initiation of effective antimicrobial therapy is the critical determinant of survival in human septic shock. *Crit Care Med*. 2006;34(6):1589-1596.

38. Ferrer R, Martin-Loeches I, Phillips G, et al. Empiric antibiotic treatment reduces mortality in severe sepsis and septic shock from the first hour: results from a guideline-based performance improvement program. *Crit Care Med*. 2014;42(8):1749-1755.

39. Bai X, Yu W, Ji W, et al. Early versus delayed administration of norepinephrine in patients with septic shock. *Crit Care*. 2014;18(5):532.