

# Copeptina = ADHD?

SMC 22/02/2020

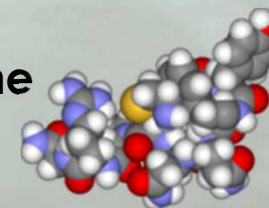
Ferrara Aula Magna S. Anna (Cona)

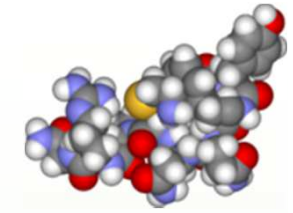
Relatore: Dott. Alessio Cariani

Biologo Clinico

Laboratorio Analisi Chimico Cliniche e Microbiologiche

S. Anna (Cona)

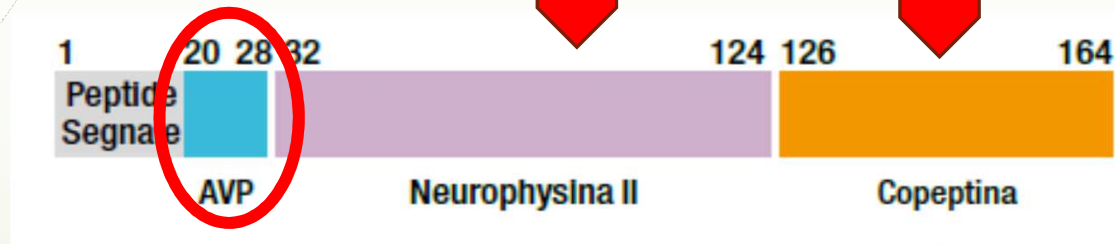
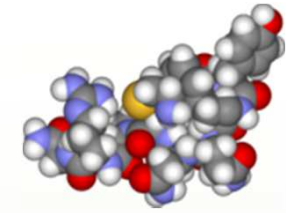




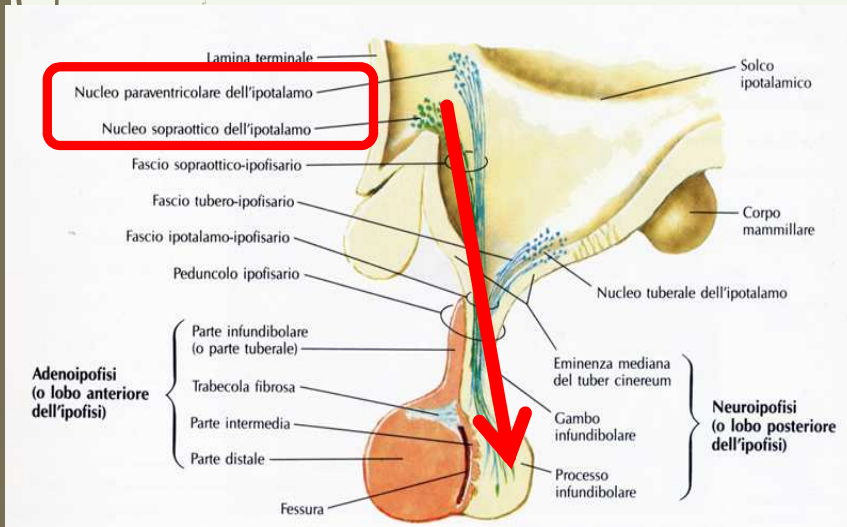
**Perché ci poniamo questa domanda?**



# ADH/Arginina Vaso Pressina Biochimica



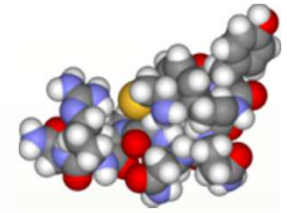
- La vasopressina è un peptide sintetizzato nei nuclei sopraottico e paraventricolare dell'ipotalamo (neuroni magnocellulari) e trasportata per via assonale nella neuroipofisi dove viene immagazzinato in granuli.
- La AVP viene sintetizzata sotto forma di preormone di 164 AA (**pre-proAVP**) che durante le fasi di trasporto assonale, intra vescicolare, dall'ipotalamo all'ipofisi viene **clivato** da 4 enzimi fino ad ottenere la struttura biologicamente attiva dell'AVP costituita da **9 aminoacidi**, contemporaneamente alla formazione di una molecola di Neurofisina II ed una di Copeptina.



Holwerda DA . A glycopeptide from the posterior lob of pig pituitaries. Eur J Biochem 1972;28:334-501.

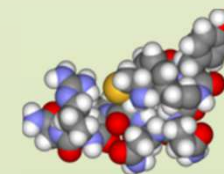
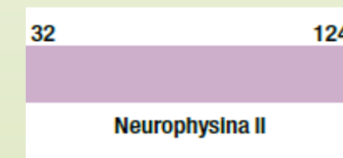
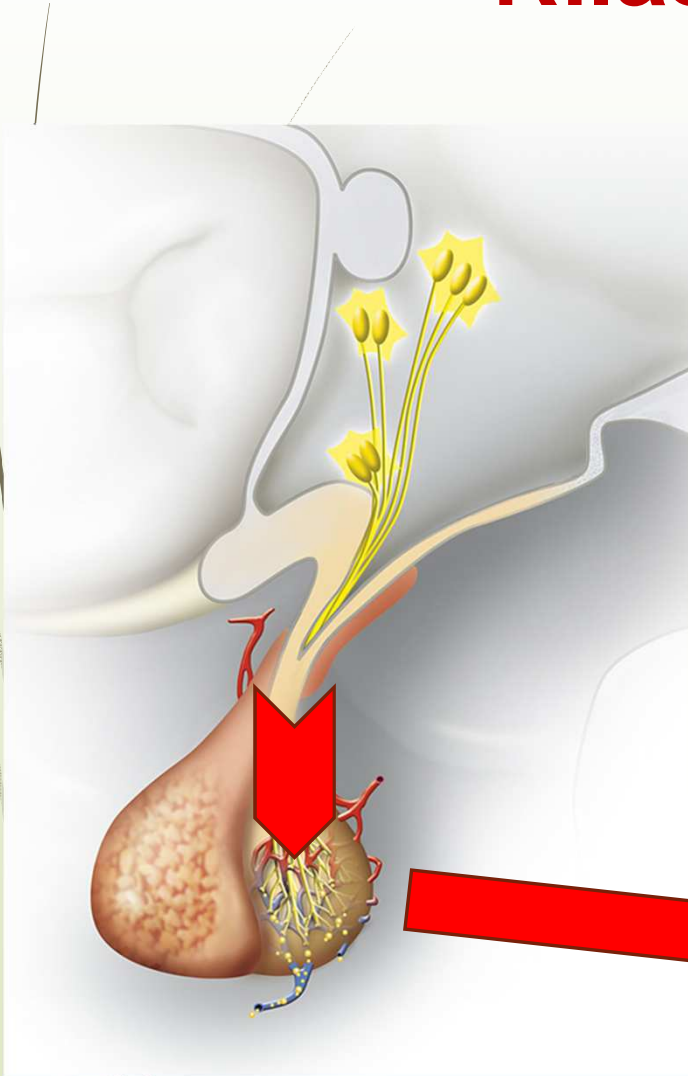
# AVP

## Rilascio in circolo



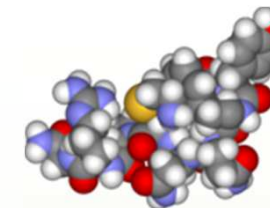
- Aumento dell'osmolalità plasmatica
- Riduzione volume arterioso circolante

**- Ipotensione (Shock,..)**



# Vasopressina

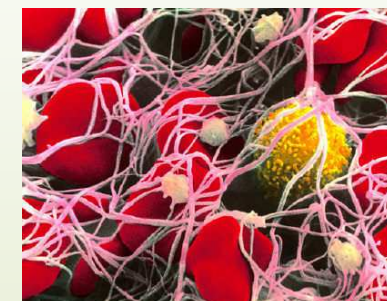
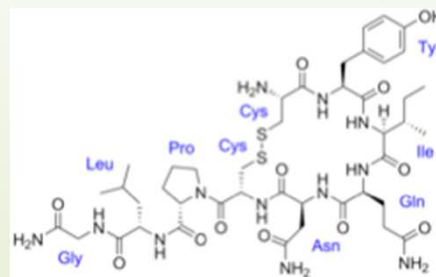
## Altri Effetti!?



**Antidiuresi**

**Emostasi**

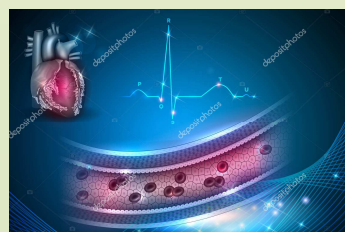
**Glicogenolisi epatica**



**Rilascio Corticotropine "STRESS"**

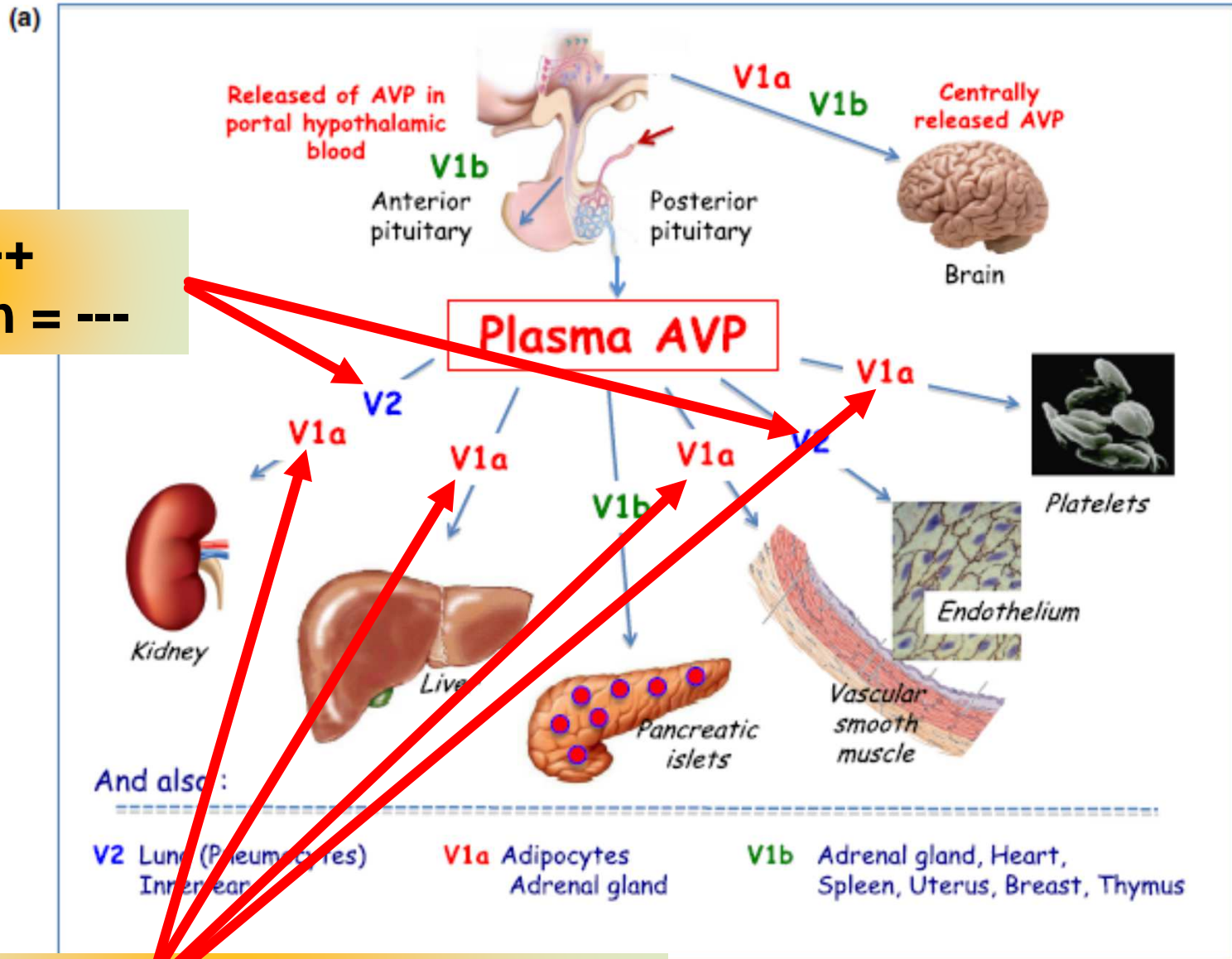
**Effetti centrali (comportamento)**

**Vasocostrizione**



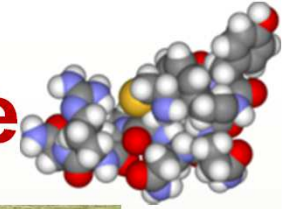


**DAVP = ++**  
**Tolvaptan = ---**



**Terlipressina = ++ (Glipressina)**

# Perchè non possiamo continuare a misurare la AVP?



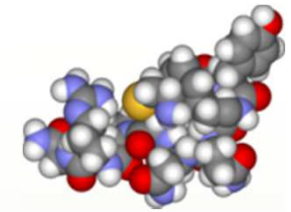
- >90% di AVP circolante è legato alle piastrine e necessita di una complessa fase preanalitica di «distacco»
- La sua emivita è < 20 min (clearance renale-epatica; endo/aminopeptidasi endoteliali e circolanti)
- AVP è molto instabile nel plasma a RT
- AVP è instabile nel plasma anche a -20°

- Servono 1-4 ml di plasma
- AVP, per le sue piccole dimensioni (1,080 kD), è quantificabile solo con metodica RIA competitiva
- TAT di laboratorio è minimo di 48-72h (sedute mensili !!)

***Fase preanalitica che introduce un errore non accettabile ed una fase analitica che impone dei tempi di risposta altrettanto non accettabili.***

# RIA

Robertson GL, Mahr EA, Athar S, Sinha T. Development and clinical application of a new method for the radioimmunoassay of arginine vasopressin in human plasma. *J Clin Invest* **1973**;52: 2340-52.



Radioattività

*The End*

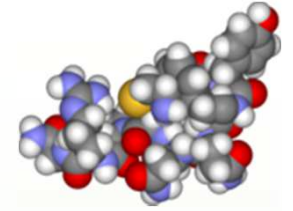
Standardizzazione

Calibration

This assay is calibrated against the first international  
WHO standard 77/501



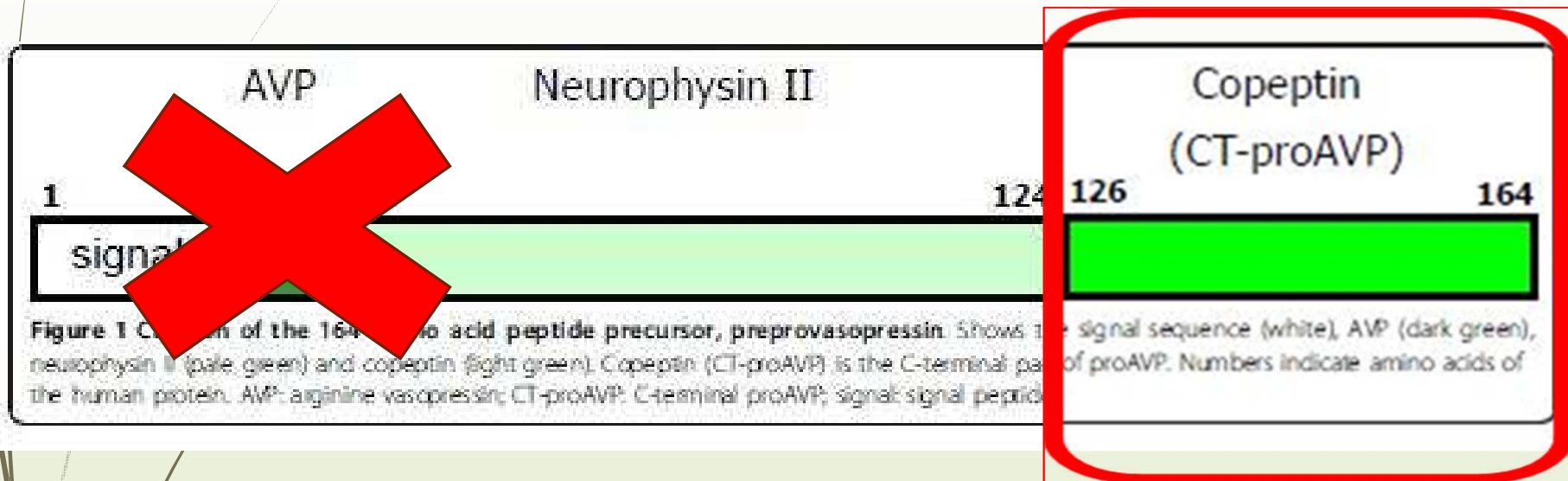
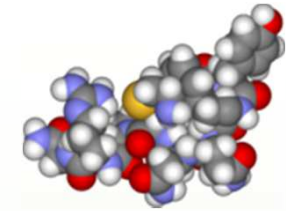
**La soluzione?**



***La misurazione diretta  
della Copeptina?***



# Copeptina (Ct-ProAVP)


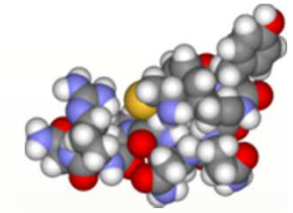


**COPEPTINA:** glicoproteina di 39 AA, 5kD, funzioni ignote

(maturazione proteolitica, ripiegamento della provasopressina?; interazione con calnexina-calreticolina??)

**Neurofisina** = carrier per ripiegamento e trasporto assonale

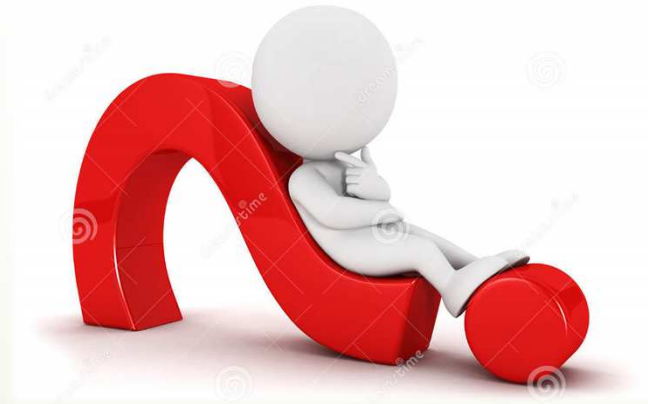
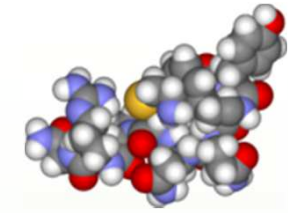




La Copeptina  
è un marker surrogato  
della  
Vasopressina ?

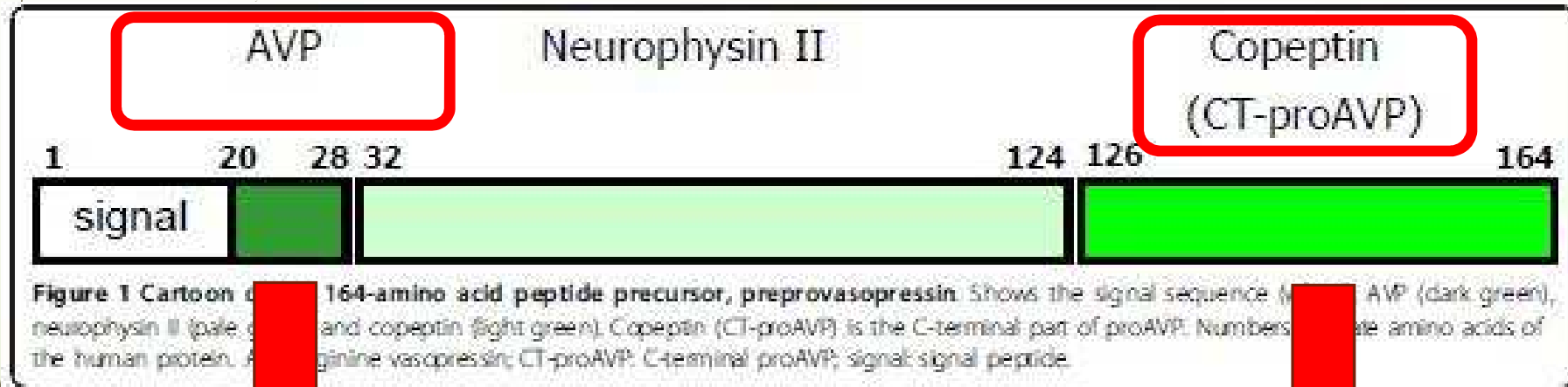
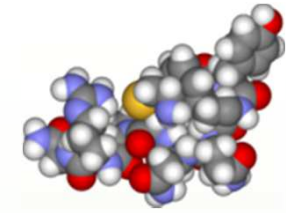
La misura analitica  
della Copeptina è  
affidabile e «fruibile»  
clinicamente?

La Copeptina può, allo  
stato attuale, essere  
usata nella routine  
clinica?



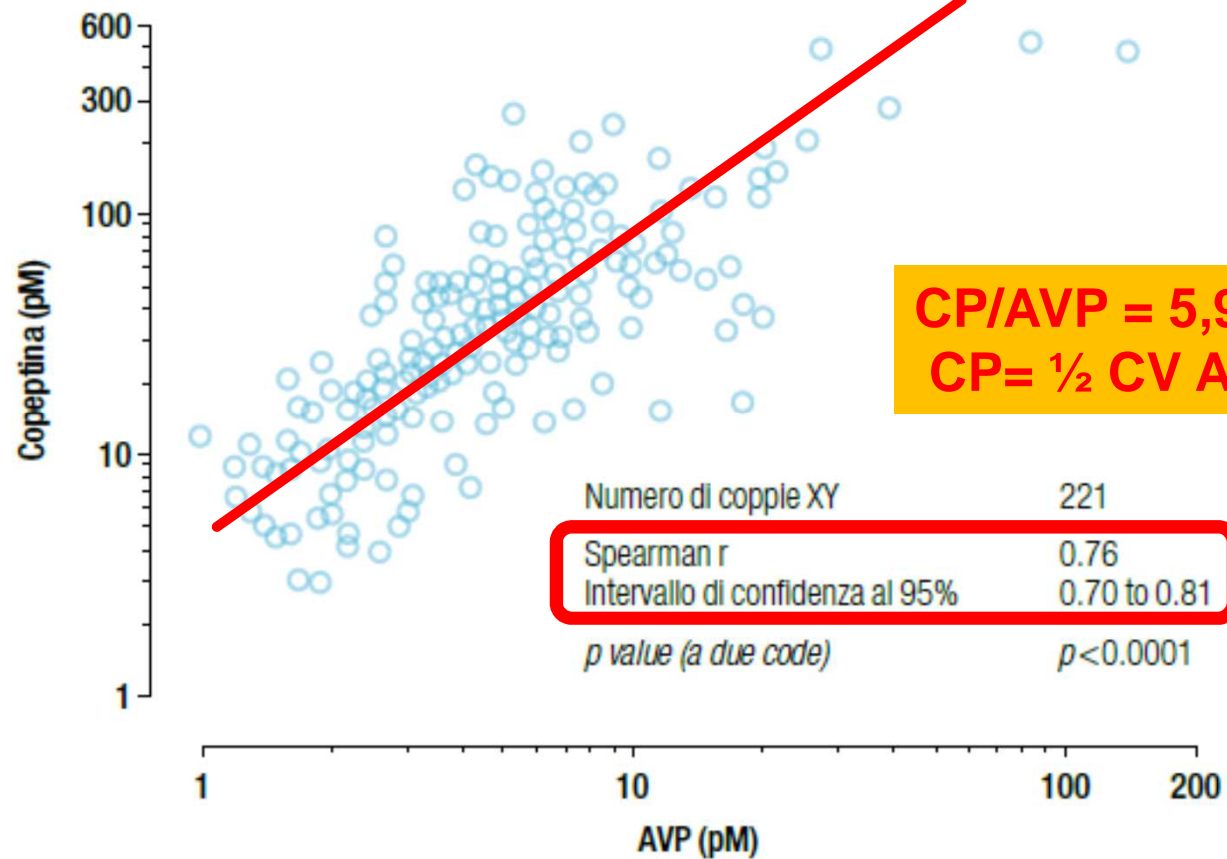
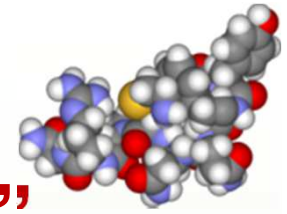
La Copeptina  
è un marker surrogato  
della  
Vasopressina ?

# Copeptina surrogato AVP? Presupposti biochimici



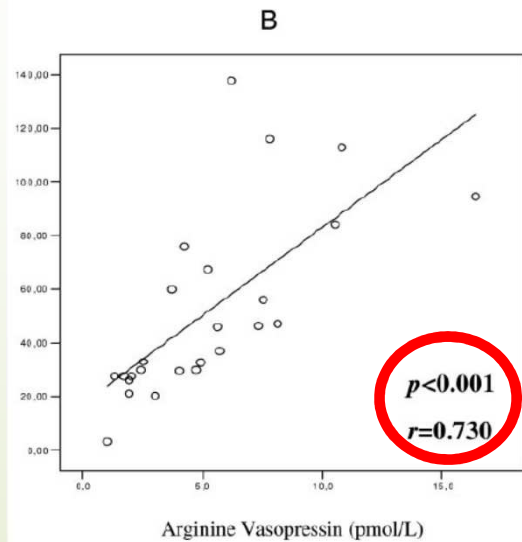
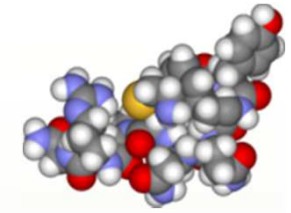
1. AVP, Neurofisina II e Copeptina sono rilasciate in un rapporto molare di 1:1
2. CP >> emivita in vivo e +++ stabile ex vivo
3. CP = glicopeptide di 39 AA/5 kD dotato di >> antigenicità rispetto AVP

# Correlazione Copeptina vs Vasopressina in soggetti "sani"



13. Jochberger S et al., Shock 2009; 31 (2): 132-8

# Correlazione Copeptina Vasopressina in soggetti "patologici"



**SEPSI**

**n 25**

**CP/AVP = 12 +/- 7.4**

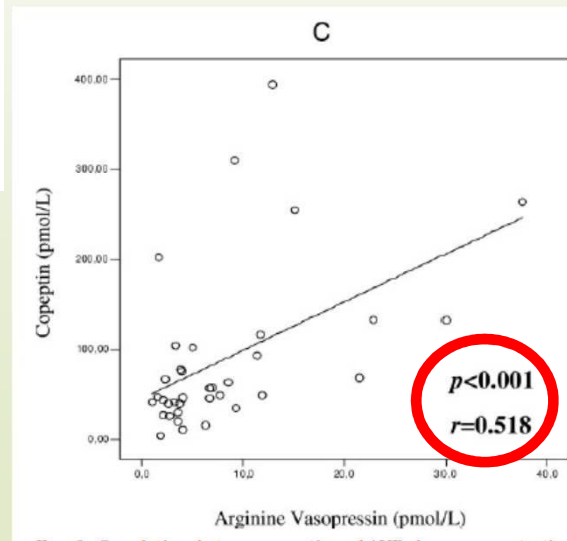
0021-972X/06/\$15.00/0  
Printed in U.S.A.

**2006**

The Journal of Clinical Endocrinology & Metabolism 91(11):4381-4386  
Copyright © 2006 by The Endocrine Society  
doi: 10.1210/jc.2005-2830

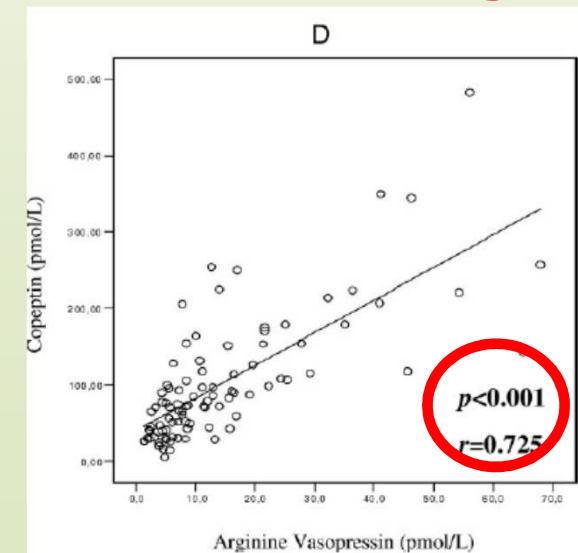
## Copeptin and Arginine Vasopressin Concentrations in Critically Ill Patients

Stefan Jochberger, Nils G. Morgenthaler, Viktoria D. Mayr, Günter Luckner, Volker Wenzel, Hanno Ulmer, Siegfried Schwarz, Walter R. Hasibeder, Barbara E. Friesenecker, and Martin W. Dünser

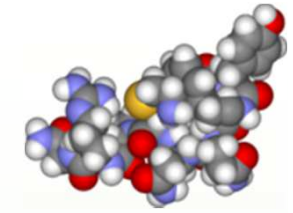


**SIRS**

**cardiochirurgia**



# Copeptina..... di genere?



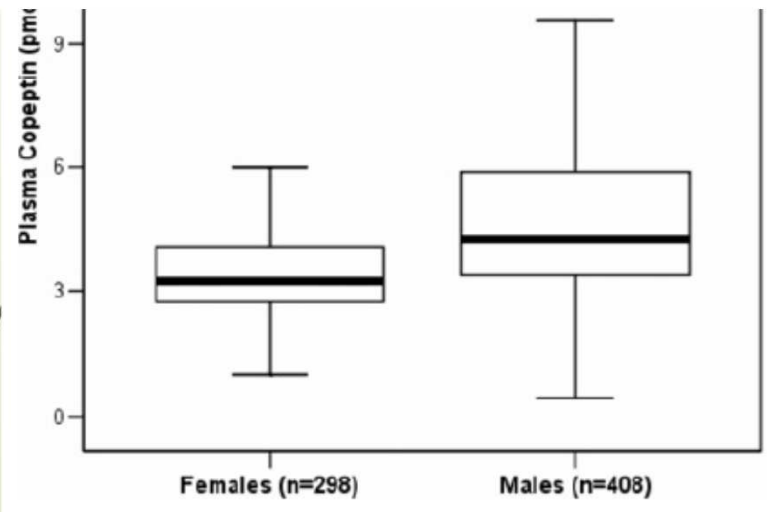
2009

Clinical Science (2009) 116, 257–263 (Printed in Great Britain) doi:10.1042/CS20080140

Gender and renal function influence plasma levels of copeptin in healthy individuals

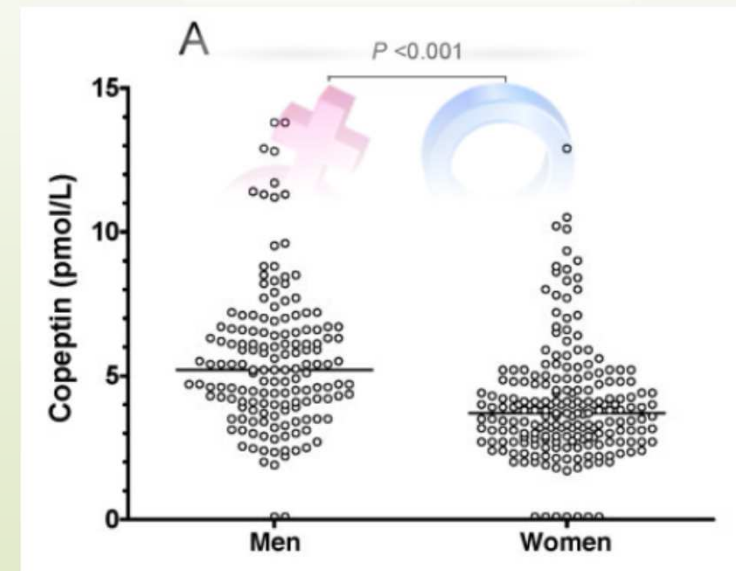
Sanjay S. BHANDARI\*, Ian LOKE\*, Joan E. DAVIES\*, Ian B. SQUIRE\*, Joachim STRUCK† and Leong L. NG\*

\*Department of Cardiovascular Sciences, University of Leicester, Leicester LE2 7LX, U.K., and †B.R.A.H.M.S. Aktiengesellschaft, Hennigsdorf, 16761 Berlin, Germany



n 298  
Mediana 3,2  
pmol/L (1,0-14,8)

n 408  
Mediana 4,3  
pmol/L (0,4-44,3)



Clinical Chemistry 52:1  
112–119 (2006)

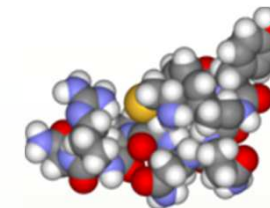
2006

Endocrinology and  
Metabolism

Assay for the Measurement of Copeptin, a Stable Peptide Derived from the Precursor of Vasopressin

NILS G. MORGENTHAUER,\* JOACHIM STRUCK, CHRISTINE ALONSO, and ANDREAS BERGMANN

# Copeptina e funzione renale. (eGFR-MDRD)



KI REPORTS  
KIReports.org

CLINICAL RESEARCH

2017

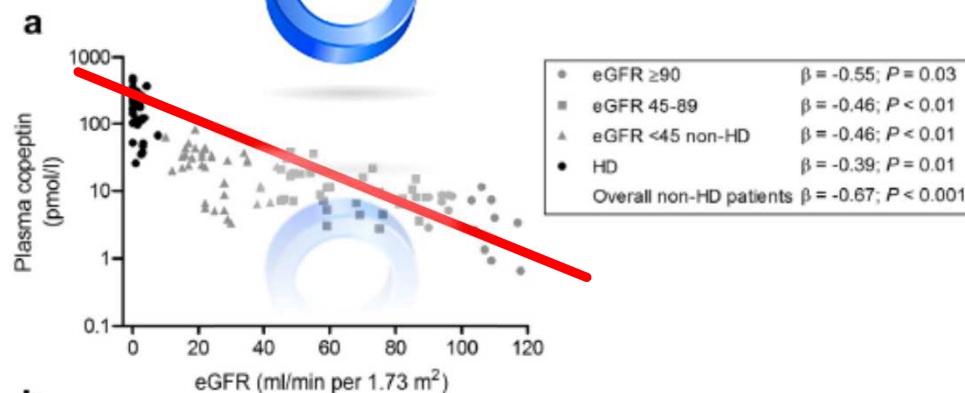


## The Effect of Renal Function and Hemodialysis Treatment on Plasma Vasopressin and Copeptin Levels

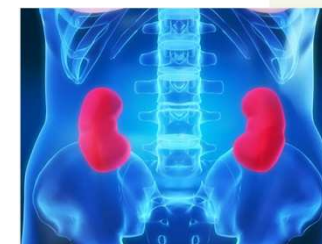
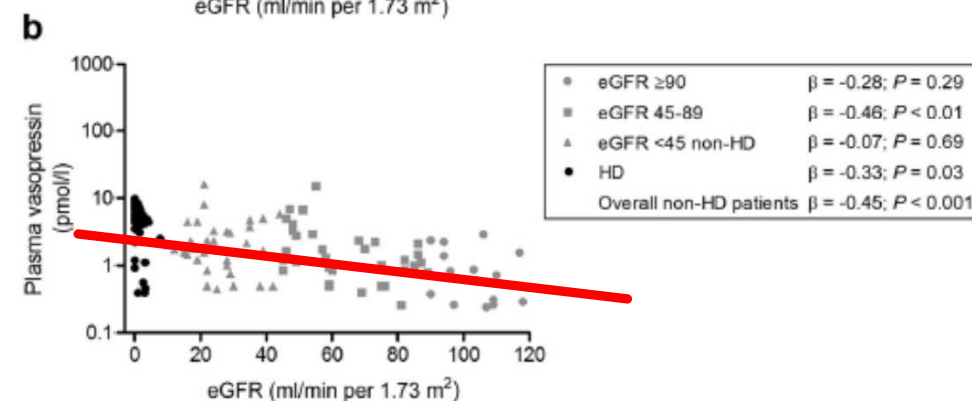
Esmée M. Ettema<sup>1</sup>, Judith Heida<sup>1</sup>, Niek F. Casteleijn<sup>1</sup>, Lianne Boesten<sup>2</sup>, Ralf Westerhuis<sup>3</sup>, Carlo A.J.M. Gaillard<sup>1</sup>, Ron T. Gansevoort<sup>1</sup>, Casper F.M. Franssen<sup>1</sup> and Debbie Zitterma<sup>1</sup>

<sup>1</sup>Department of Internal Medicine, Division of Nephrology, University Medical Center Groningen, University of Groningen, Groningen, the Netherlands; <sup>2</sup>Department of Laboratory Medicine, Usselland Hospital, Capelle aan den IJssel, the Netherlands; and <sup>3</sup>Dialysis Center Groningen, Groningen, the Netherlands

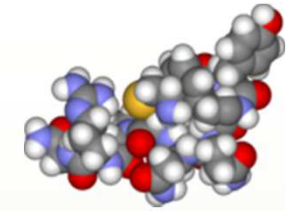
CP



VP



**Figure 2.** Associations of renal function with (a) copeptin and (b) vasopressin. Gray symbols indicate healthy participants and chronic kidney disease patients not on dialysis. Black symbols indicate hemodialysis (HD) patients. eGFR, estimated glomerular filtration rate.



# La Copeptina «non ha età»

Clinical Chemistry 52:1  
112-119 (2006)

2006

Endocrinology and  
Metabolism

## Assay for the Measurement of Copeptin, a Stable Peptide Derived from the Precursor of Vasopressin

NILS G. MORGENTHAUER,<sup>\*</sup> JOACHIM STRUCK, CHRISTINE ALONSO, and ANDREAS BERGMANN



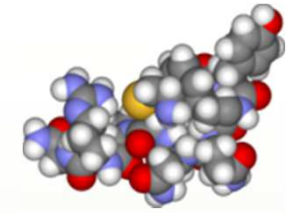
Table 1. Copeptin in 359 healthy blood donors stratified by age.<sup>a</sup>

	Blood donor age, years					
	18-24	25-34	35-44	45-54	55-64	65-80
n	46	65	76	48	84	40
Copeptin, pmol/L						
Mean (SD)	5.1 (2.4)	4.1 (1.8)	4.6 (2.0)	4.8 (2.4)	4.5 (2.6)	5.0 (2.0)
Median	4.9	3.9	4.1	4.1	4.1	4.7
Range	1.8-13.8	1.0-8.8	1.0-11.4	1.0-12.9	1.0-13.8	2.4-11.3

<sup>a</sup> All age groups showed gaussian distribution (by Kolmogorov-Smirnov test). Differences in mean or median copeptin values between groups were not significant (by Kruskal-Wallis ANOVA).

# Copeptina

## Ritmo circadiano?



Research Article

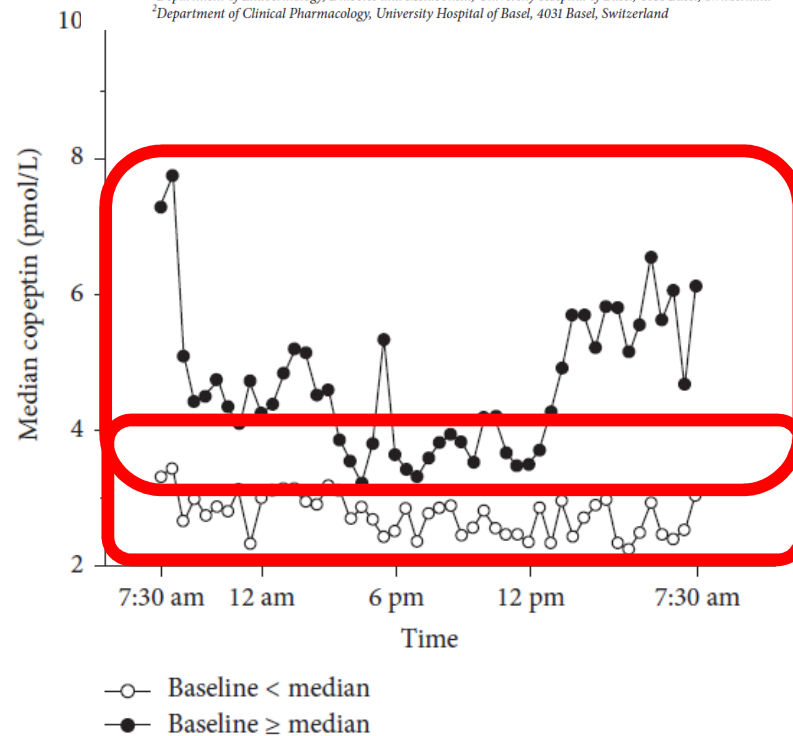
**The Circadian Rhythm of Copeptin, the C-Terminal Portion of Arginine Vasopressin**

**2017**

Svetlana Beglinger,<sup>1</sup> Jürgen Drewe,<sup>2</sup> and Mirjam Christ-Crain<sup>1</sup>

<sup>1</sup>Department of Endocrinology, Diabetes and Metabolism, University Hospital of Basel, 4031 Basel, Switzerland

<sup>2</sup>Department of Clinical Pharmacology, University Hospital of Basel, 4031 Basel, Switzerland

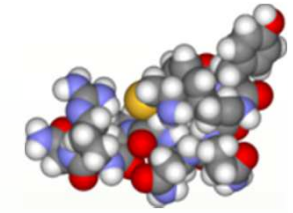


.....la copeptina mostra un ritmo circadiano «significativo» soltanto negli individui che hanno delle concentrazioni superiori alla mediana di 4,2 pmol/L...

.....potrebbe essere misurata in qualsiasi orario.....!!!!

# Copeptina

## Altri fattori



Clinical Chemistry 52:1  
112-119 (2006)

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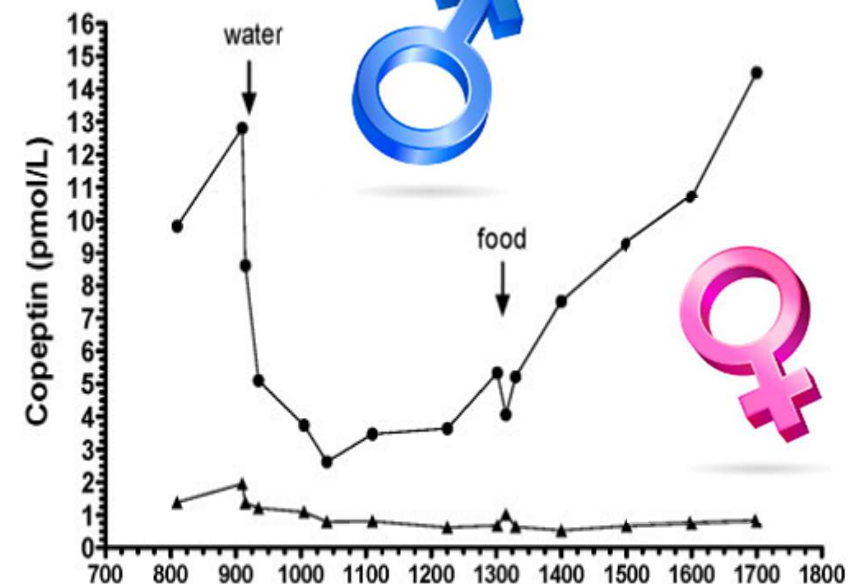
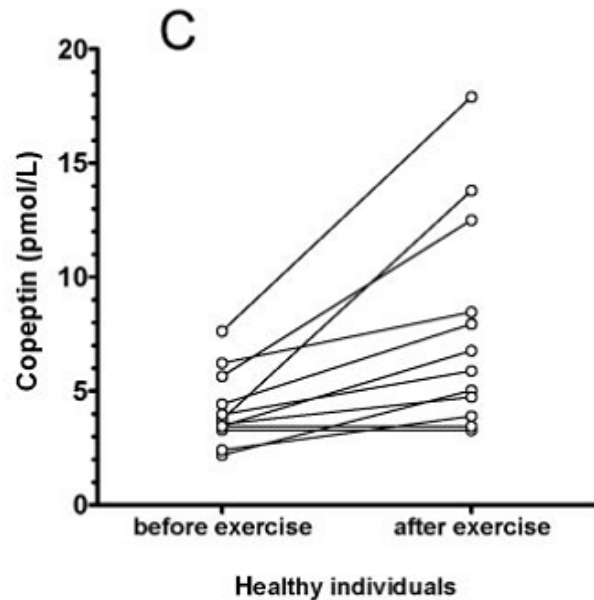
Assay for the Measurement of Copeptin, a Stable Peptide Derived from the Precursor of Vasopressin

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### Esercizio fisico

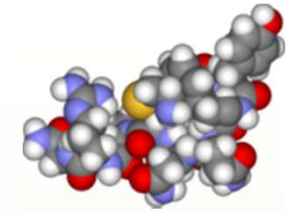


### Cibo e Acqua



# Copeptina

## Valori... «normali»? ? ? ! ! !



### Review

Davide Bolignano, Aderville Cabassi, Enrico Fiaccadori, Ezio Ghigo, Renato Pasquali, Andrea Peracino\*, Alessandro Peri, Mario Plebani, Antonio Santoro, Fabio Settanni and Carmine Zoccali

**Copeptin (CTproAVP), a new tool for understanding the role of vasopressin in pathophysiology**

**4-7 pmol/L (varianza x100;x1000)**

Clinical Chemistry 52:1  
112-119 (2006)

Endocrinology and  
Metabolism

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**Mediana: 4,2 pmol/L**  
**Range : 1-13,8 pmol/L**

Uno studio su una popolazione apparentemente sana (N= 297, entrambi i sessi) ha mostrato una mediana di 3,9 pmol/l e la seguente distribuzione, come mostrato nel seguente istogramma.

Fattore di conversione..... 1 pmol/L = 4,02 pg/mL

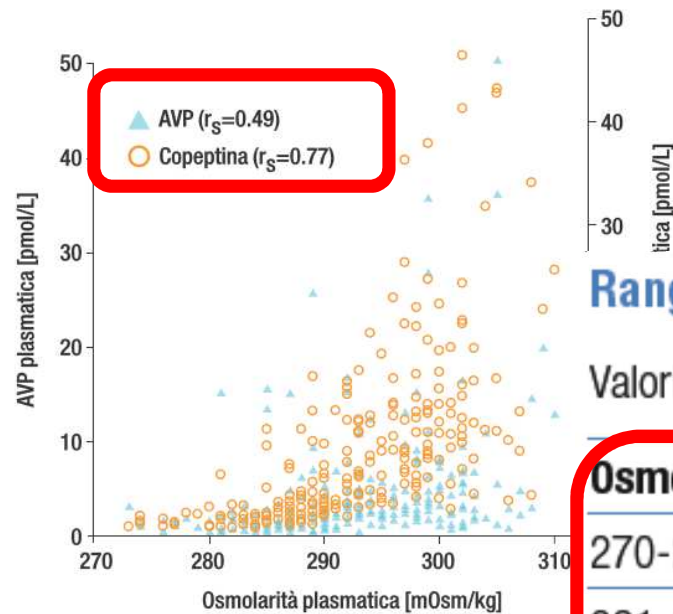
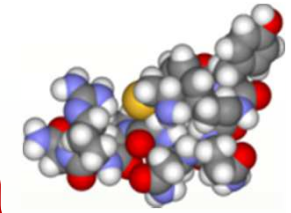


**Nota: Si raccomanda a ogni laboratorio di stabilire i propri range di riferimento basati su un numero rappresentativo di pazienti e/o di testare la validità dei dati del kit del produttore. I valori sono stati stabiliti sul siero e sul plasma**

**eGFR (MDRD)? Stato di idratazione? Etnia? .....**

# Copeptina

## Valori di riferimento e Osmolarità



Rispetto alla vasopressina, la Copeptina ha una migliore correlazione con l'osmolarità plasmatica

### Range di riferimento

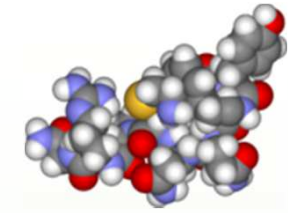
Valori di Copeptina in rapporto all'osmolarità plasmatica:

Osmolarità [mosmol/kg]	Copeptina [pmol/L]
270-280	0.81-11.6
281-285	1.0-13.7
286-290	1.5-15.3
291-295	2.3-24.5
296-300	2.4-28.2



La misura della  
Copeptina è affidabile  
e «fruibile»  
clinicamente?

# Misurazione Copeptina Matrice/i...?



Clinical Chemistry 52:1  
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I colori rispettano la normativa ISO 6710 ed EN 14120 per l'identificazione delle provette.



Azzurro

Coagulazione



Nero

VES



Rosso

Siero



Giallo con

Separatore di siero



Verde

Ecografia



Viola

EDTA



Grigio

Inibitori di fibrina

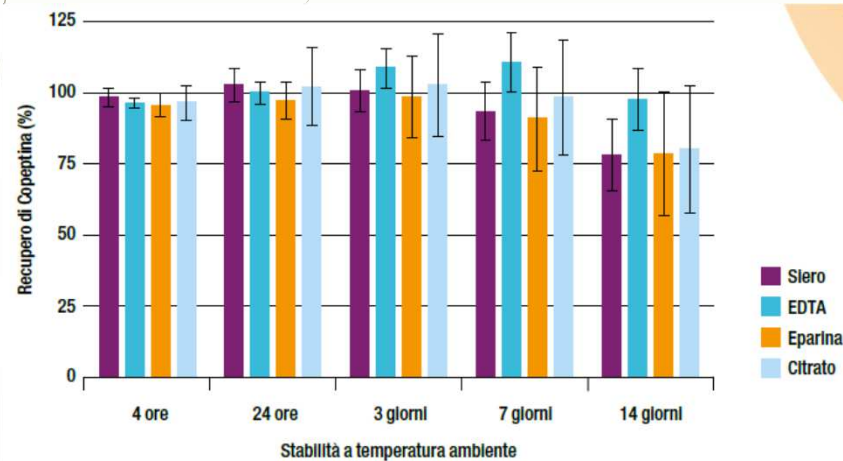
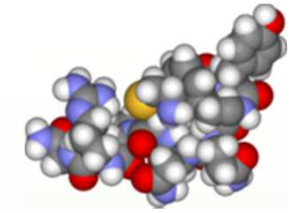
Mediana  
4,3 pmol/L  
NO!!!!!!

Mediana  
4,1 pmol/L

Mediana  
4,3 pmol/L

Mediana  
4,0 pmol/L

# Copeptina Stabilità



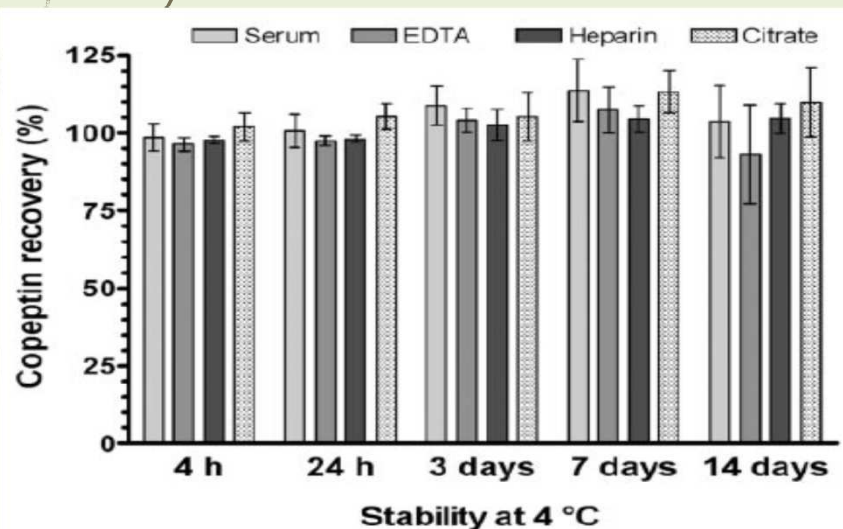
Clinical Chemistry 52:1  
112-119 (2006)

2006

Endocrinology and  
Metabolism

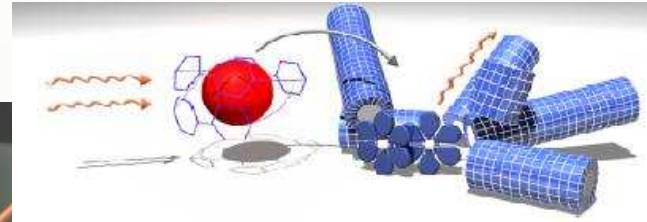
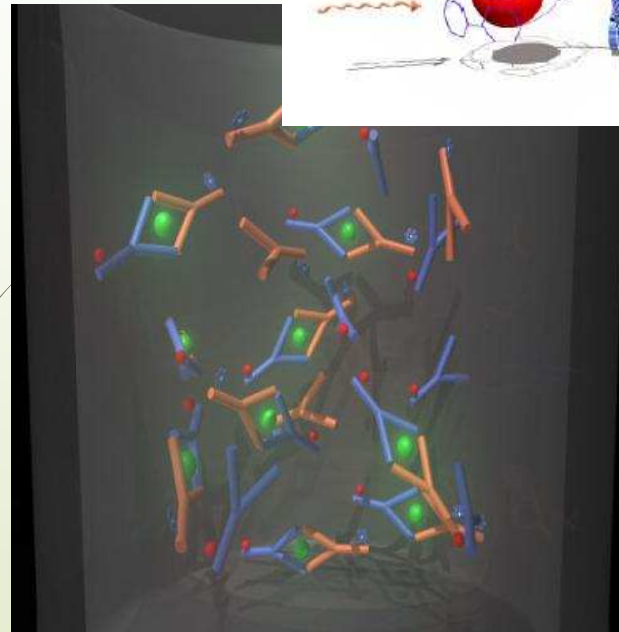
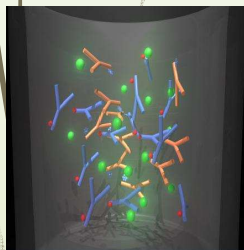
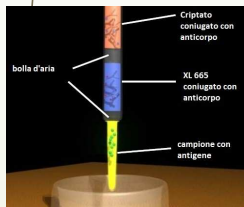
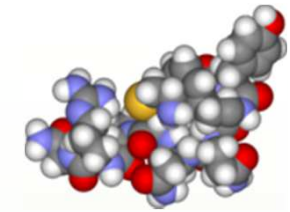
## Assay for the Measurement of Copeptin, a Stable Peptide Derived from the Precursor of Vasopressin

NILS G. MORGENTHALER,\* JOACHIM STRUCK, CHRISTINE ALONSO, and ANDREAS BERGMANN



- Stabile a temperatura ambiente per oltre 7 giorni
- Stabile a 4° C per oltre 2 settimane
- Stabile indefinitamente a -20° C
- Stabile fino a 4 cicli di congelamento e scongelamento

# Time Resolved Amplified Cryptate Emission<sup>®</sup>



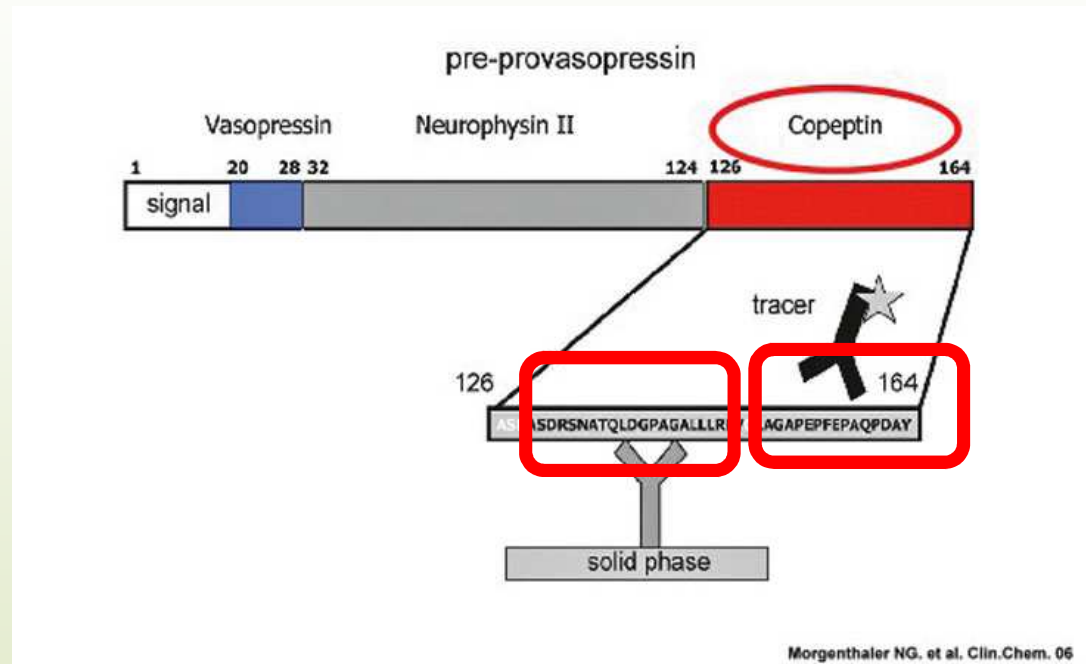
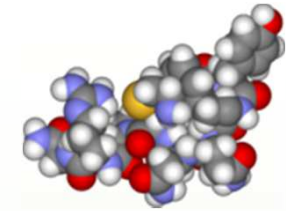
1. Eccitazione dell'immunocomplesso con un laser all'azoto a 337 nm
2. Trasferimento di energia in fluorescenza a 620 nm (millisecondi) da una molecola donatrice (Europio trisbipiridina criptato) a una molecola recettrice XL 665 (Alloficocianina)
3. Emissione (da XL 665) di un segnale fluorescente a 707 nm (nanosecondi).

L'intensità del segnale a 707 nm è direttamente proporzionale alla quantità di coceptina e può essere misurato nell'ordine dei **microsecondi**

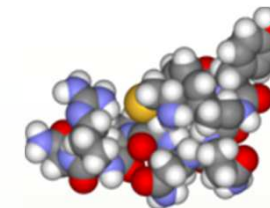
# Copeptina

## Specificità Tecnologia TRACE

### Bersagli anticorpali



- Ab pecora policlonale coniugato a donatore criptato con un atomo terbio/europio per **seq. AA132-147 (PATV17)**
- Ab murino monoclonale coniugato ad accettore XL **seq. AA149-164 (PLAY 17)**



# Copeptina T.R.A.C.E

## Prestazioni piattaforma automatizzata



Tecnologia	Time Resolved Amplified Cryptate Emission (TRACE)
------------	---

Misurazione diretta	0.7...500 pmol/L
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Range di misurazione con diluizione automatica	0.7...2000 pmol/L
--	-------------------

Sensibilità funzionale di dosaggio	<1,08 pmol/L
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**LOQ = 1,23**

Limite di rilevabilità	0.69 pmol/L	<b>LOD</b>
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Tempo di incubazione	14 minuti
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Volume del campione	50 $\mu$ L
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Tipo di campione	Siero, plasma (EDTA, eparina)
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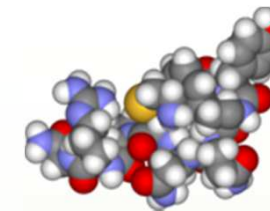
Determinazioni	50
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- Nessuna procedura di estrazione (AVP legato a piastrine per il 90%)
- Maggiore precisione analitica generale (CV interlab < 20%)

# TRACE

## Precisione/Riproducibilità

### CLSI EP5-2A



Range di concentrazione	% CV intra-dosaggio
2,0 – 4,0 pmol/L	< 15,0 %
4,0 – 15 pmol/L	< 8,0 %
15 – 50 pmol/L	< 4,0 %
> 50 pmol/L	< 3,0 %
Campioni fuori range (> 500 pmol/L)	< 4,0 %

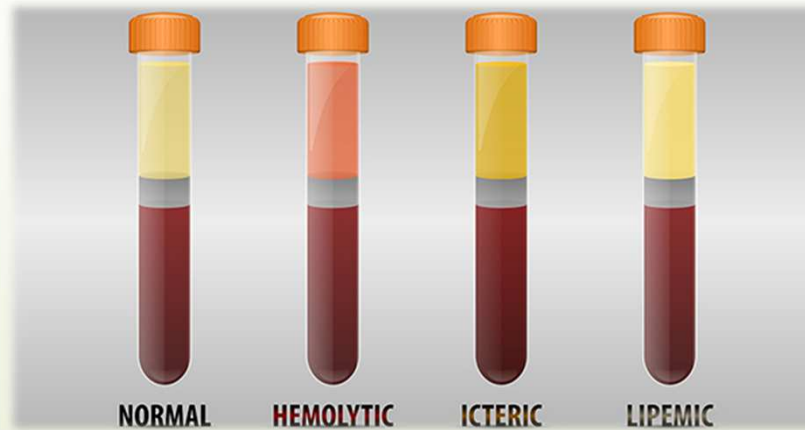
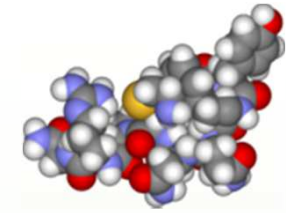


Range di concentrazione	% CV inter-dosaggio
2,0 – 4,0 pmol/L	< 18,0 %
4,0 – 15 pmol/L	< 10,0 %
15 – 50 pmol/L	< 5,0 %
> 50 pmol/L	< 5,0 %
Campioni fuori range (> 500 pmol/L)	< 6,0 %

# TRACE

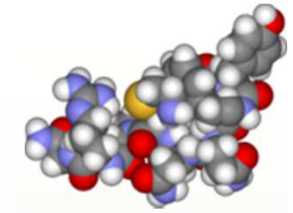
## Fattori di disturbo?

### CLSI-EP7A2



Fattore	Descrizione
Bilirubina	Nessun effetto significativo fino a 5 mg/dL
Emoglobina	Nessun effetto significativo fino a 500 mg/dL
Albumina sierica umana	Nessun effetto significativo fino a 5 g/dL
Trigliceridi	Nessun effetto significativo fino a 5 g/L

# Interferenze iatrogene CLSI-EP7A2

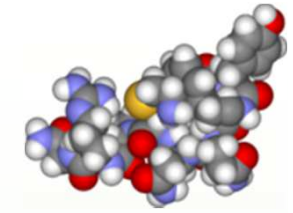


Farmaco	Concentrazione fino a
Abciximab	20 µg/ml
Paracetamolo	250 µg/ml
Acido acetilsalicilico	600 µg/ml
Allopurinolo	40 µg/ml
Ambroxol	18 µg/ml
Ampicillina	50 µg/ml
Acido ascorbico	40 µg/ml

Atenololo
Atorvastatina
Caffeina
Captopril
Cloranfenicolo

Ossitetraciclina	5 µg/ml
Fenitonina	60 µg/ml
Propranololo	5 µg/ml
Quinidina	20 µg/ml
Ramipril	12 µg/ml
Acido salicilico	600 µg/ml
Simvastatina	16 µg/ml
Teofillina	60 µg/ml
Tirofiban	1,7 µg/ml
Trimetoprima	40 µg/ml
Verapamil	2 µg/ml
Warfarin	30 µg/ml

Farmaco	Concentrazione fino a
Cinnarizina	2,7 µg/ml
Bisolfato clopidogrel	2,5 µg/ml
Cocaina	1 µg/ml
	50 µg/ml
	7,5 ng/ml
	900 ng/ml
	7 µg/ml
	60 µg/ml
	12 µg/ml
	400 µg/ml
	8 U/ml
	500 µg/ml
	20 µg/ml
	5 U/ml
	15 µg/ml
	5 µg/ml
	2 mg/dl
Nifedipina	600 ng/ml
Nitrofurantonina	4 µg/ml
Nitroglicerina	0,5 µg/ml
Nistatina	7,5 µg/ml

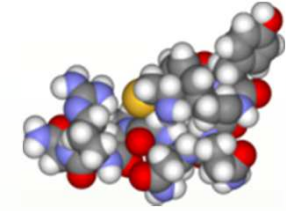



## Quadri clinici «interferenti»... .....o indicazione diagnostica?

- SIRS, sepsi, sepsi grave, shock settico
- Malattie cardiovascolari quali l'insufficienza cardiaca
- Infezioni delle basse vie respiratorie e pneumopatia cronico ostruttiva
- Terapie antagoniste del recettore AVP (Tolvaptan) ed altri quadri in cui l'AVP ha un ruolo fisiopatologico importante.



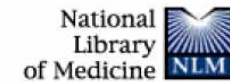
# Copeptina Riassumendo



- 
- **Massimo sfruttamento del potenziale diagnostico della vasopressina**, anche in caso di somministrazione terapeutica di AVP ed analoghi.
  - Migliore prestazione analitica in termini di **sensibilità e affidabilità dei risultati**
  - Ottimizzazione della **diagnosi differenziale** delle patologie endocrine
  - Miglioramento della **gestione del paziente** e del **flusso di lavoro** per **ridotti tempi analitici**

- 
- Studiare meglio l'asse **vasopressina-sete-concentrazione delle urine**
  - **Ridisegnare il profilo di azione della vasopressina** (clonaggio recettori, anticorpi recettore specifici, farmaci agonisti ed antagonisti specifici)
  - Costruire uno **standard di riferimento internazionale** per la copeptina
  - **Nuovi studi con i nuovi metodi analitici** per **identificare meglio i diversi valori di riferimento «patologici»** nei diversi quadri clinici

# Molto interesse, buone potenzialità ma dati ancora da consolidare!!!



## Diabetes and Metabolism

Hindawi Publishing Corporation  
Disease Markers  
Volume 2015, Article ID 614145, 9 pages  
<http://dx.doi.org/10.1155/2015/614145>

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Review Article

Sofia  
Gunn

### Copeptin Testing in Acute Myocardial Infarction: Ready for Routine Use?

Review

Sebastian Johannes Reinstadler, Gert Klug, Hans-Josef Feistritzer, Bernhard Metzler, and Johannes Mair

EB 201

Department of Internal Medicine III, Cardiology and Angiology, Medical University of Innsbruck, Anichstrasse 35, 6020 Innsbruck, Austria

Vasoq

Ray El Boustany

Inserm UMRS 1138, Vascular Complications of Diabetes, Paris, France

This article is based on the presentation for the *European Journal of Endocrinology*, European Congress of Endocrinology (ECE) 2019 held at Lyon, France

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BioMed Research International  
Volume 2016, Article ID 3624730, 7 pages  
<http://dx.doi.org/10.1155/2016/3624730>

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DOI 10

Research Article

### Assessment of Diagnostic and Prognostic Role of Copeptin in the Clinical Setting of Sepsis

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di

Stefania Battista,<sup>1</sup> Umberto Audisio,<sup>1</sup> Claudia Galluzzo,<sup>1</sup> Matteo Maggiorotto,<sup>1</sup> Monica Masoero,<sup>1</sup> Daniela Forno,<sup>1</sup> Elisa Pizzolato,<sup>1</sup> Marco Ulla,<sup>1</sup> Manuela Lucchiari,<sup>2</sup> Annarita Vitale,<sup>2</sup> Corrado Moiraghi,<sup>1</sup> Enrico Lupia,<sup>1</sup> Fabio Settanni,<sup>2</sup> and Giulio Mengozzi<sup>2</sup>

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Baris

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REVIEW  
published: 07 August 2016  
doi: 10.3389/fped.2016.00075



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Published online: June 20, 2018

### Arginine Vasopressin and Copeptin in Perinatology

Katrina Suzanne Evers and Svon Wollmann\*

Division of Neonatology, University of Basel Children's Hospital (UNGB), Basel, Switzerland

monia:

## Copeptin in acute coronary syndromes and heart failure management: State of the art and future directions

*Place de la copeptine dans la prise en charge du syndrome coronaire aigu et de l'insuffisance cardiaque : état des connaissances et perspectives futures*

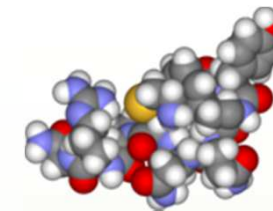
Guillaume Schurtz<sup>a,b</sup>, Nicolas Lamblin<sup>a,b,c</sup>,  
Christophe Bauters<sup>a,b,c</sup>, Patrick Goldstein<sup>d</sup>,  
Gilles Lemesle<sup>a,b,c,\*</sup>

### pathophysiological insights and practical implications

Kenan Yalta, Orkide Palabiyik, Muhammet Gurdogan and Yekta Gurlertop

Leong L. Ng  
Institutes, University of Leicester, Leicester LE2 7LX, U.K., and †B.R.A.H.M.S. Aktiengesellschaft, iny

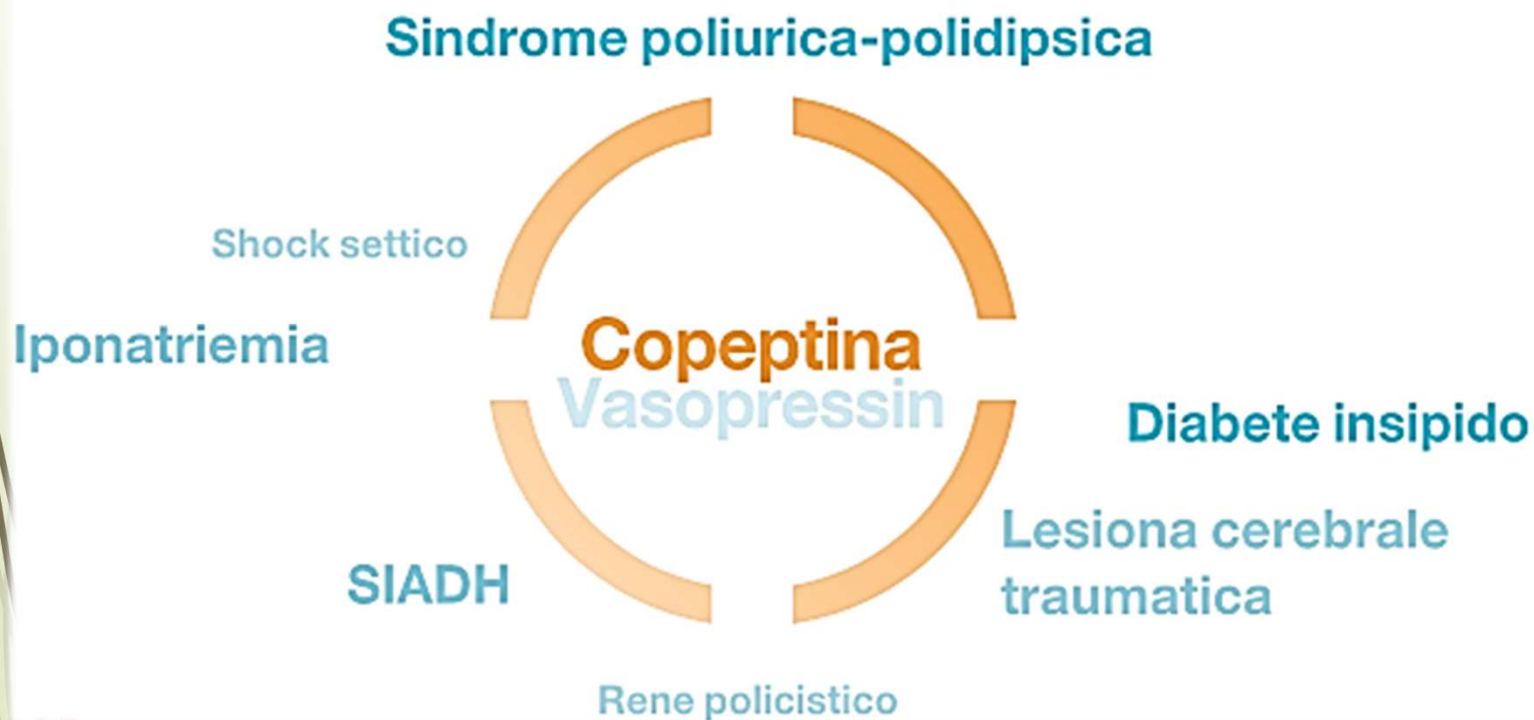
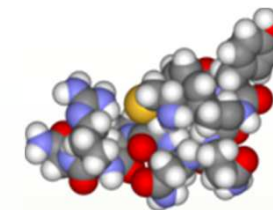
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als



La Copeptina può,  
allo stato attuale,  
essere usata nella  
routine clinica?

# Copeptina

## Applicazioni cliniche consolidate e potenziali



***Grazie...***



***.....per l'attenzione***