AMILINA:
sola o in compagnia

Salvatore Piro
Università di Catania

Disclosure

Il sottoscritto **Salvatore Piro**

ai sensi dell'art. 3.3 sul Conflitto di Interessi, pag. 17 del Reg. Applicativo dell'Accordo Stato - Regione del 5 novembre 2009

dichiara

di non avere conflitti di interessi riguardo la seguente presentazione

dichiara

di aver effettuato una presentazione scientifica per Abbot

Disclosure 2

Titolo simposio: **OK GLP-1 RA……**

V SESSIONE

OK per GLP-1 RA. Ma insieme a chi?

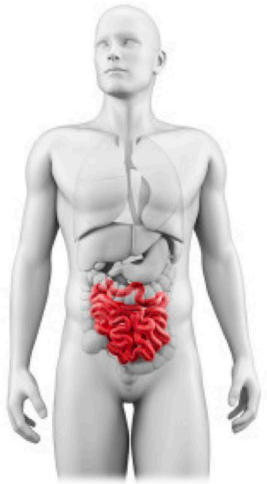
Moderatori: Katherine Esposito, Gian Paolo Fadini

09.30	GIP (e antiGIP) *Francesco Giorgino*	→ **+ GIP**
09.50	Discussione	
10.00	Glucagone *Riccardo C. Bonadonna*	→ **+ Glucagone**
10.20	Discussione	
10.30	Amilina *Salvatore Piro*	→ **+ Amilina**
10.50	Discussione	

Siamo così certi che il **GLP-1** possa essere considerato un **punto di partenza stabile** per le nostre prospettive future**?**

RIVOLUZIONE CULTURALE

GLP-1 ⇌ **Diabete**



ELIXA
Lixisenatide in Patients with Type
and Acute Coronary Syndr

The NEW ENGLAND JOURNAL of MEDICINE
LEADER
Liraglutide and Cardiovascular Outcomes in Type 2 Diabetes

SUSTAIN-6
Semaglutide and Cardiovascular Outcomes in Patients with Type 2 Diabetes

EXSCEL
Effects of Once-Weekly Exenatide on Cardiovascular Outcomes in Type 2 Diabetes

HARMONY
Albiglutide and cardiovascular outcomes in patients with type 2 diabetes and cardiovascular disease (Harmony Outcomes): a double-blind, randomised placebo-controlled trial

PIONEER 6
Oral Semaglutide and Cardiovascular Outcomes in Patients with Type 2 Diabetes

Articles
REWIND
Dulaglutide and cardiovascular outcomes in type 2 diabetes (REWIND): a double-blind, randomised placebo-controlled trial

SOUL
WILEY
Effects of oral semaglutide on cardiovascular outcomes in individuals with type 2 diabetes and established atherosclerotic cardiovascular disease and/or chronic kidney disease: Design and baseline characteristics of SOUL, a randomized trial

SELECT
The NEW ENGLAND JOURNAL of MEDICINE
Semaglutide and Cardiovascular Outcomes in Obesity without Diabetes

Fase 2.0

SURPASS program
Diabetes Ther
https://doi.org/10.1007/s13300-020-00981-0
REVIEW
The Role of Tirzepatide, Dual GIP and GLP-1 Receptor Agonist, in the Management of Type 2 Diabetes: The SURPASS Clinical Trials
Thinzar Min · Stephen C. Bain

Guardando nel dettaglio

Giusto per una visione realistica…..

Il GLP-1

Concentrazioni fisiologiche **0-15** **pmol/L** a digiuno

Concentrazioni fisiologiche **20-45** **pmol/L** post-prandiali

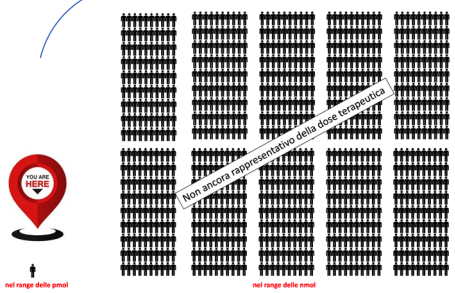
- Baggio LL, Drucker DJ (May 2007). "Biology of incretins: GLP-1 and GIP". Gastroenterology. 132 (6): 2131–57. doi:10.1053/j.gastro.2007.03.054. PMID 17498508
- Marathe CS, Rayner CK, Jones KL, Horowitz M (June 2013). "Glucagon-like peptides 1 and 2 in health and disease: a review". Peptides. 44: 75–86. doi:10.1016/j.peptides.2013.01.014. PMID 23523778. S2CID 22641629
- Holst JJ (October 2007). "The physiology of glucagon-like peptide 1". Physiological Reviews. 87 (4): 1409–39. doi:10.1152/physrev.00034.2006. PMID 17928588
- Deacon CF, Holst JJ (August 2009). "Immunoassays for the incretin hormones GIP and GLP-1". Best Practice & Research. Clinical Endocrinology & Metabolism. 23 (4): 425–32. doi:10.1016/j.beem.2009.03.006. PMID 19748060
- Muller TD, CF, Finan B. eta l (September 2019). »Glucagon like peptide 1 (GLP-1) ". Molecular Metabolism https://doi.org/10.1016/j.molmet.2019.09.010

In corso di trattamento farmacologico

variabile **[concentrazione]**

nel range delle micromol

nel range delle nmol



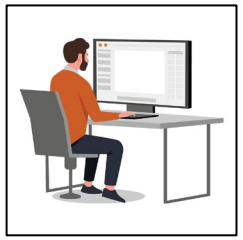
X 1.000

X 10.000

Considerando la variabile **[tempo]**

10.000 minuti compresi in una settimana

1:10.000

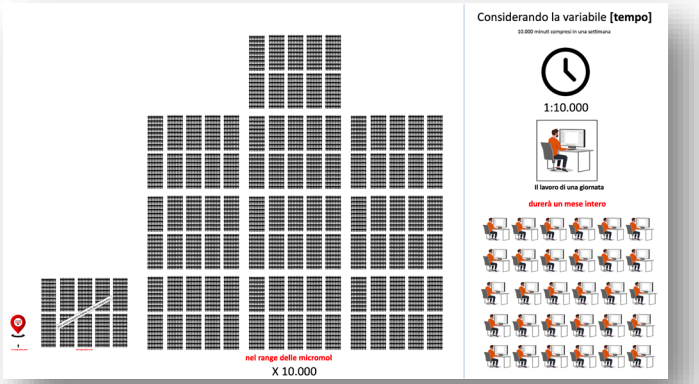


Il lavoro di una giornata

durerà oltre un mese intero

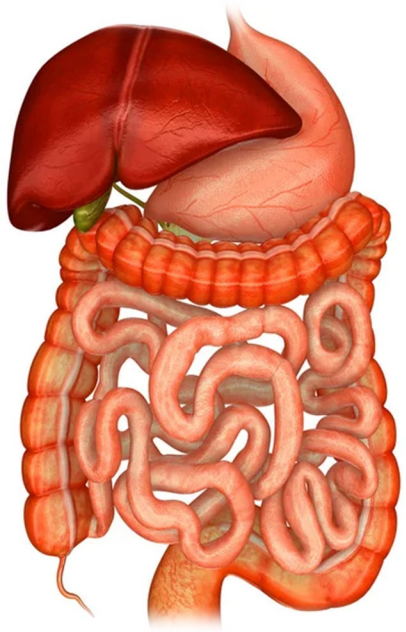
…inoltre

…le interazioni



INTESTINO COME GHIANDOLA ENDOCRINA

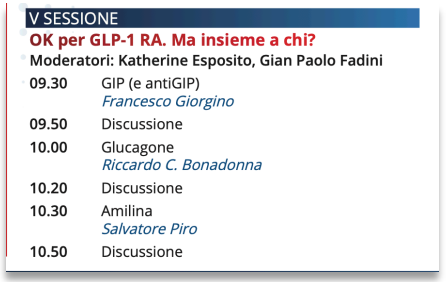
(noto dal 1904)



Sono noti ad oggi oltre **20 ormoni** prodotti dall'intestino che agiscono come MODULATORI localmente, perifericamente ed a livello centrale

Rehfeld J.F. Horm Metab Res 2004

OK GLP-1 RA……



V SESSIONE

OK per GLP-1 RA. Ma insieme a chi?

Moderatori: Katherine Esposito, Gian Paolo Fadini

09.30	GIP (e antiGIP) *Francesco Giorgino*
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10.20	Discussione
10.30	Amilina *Salvatore Piro*
10.50	Discussione

20 differenti ormoni….

…che interagiscono tra loro

20!

In statistica ed in matematica…

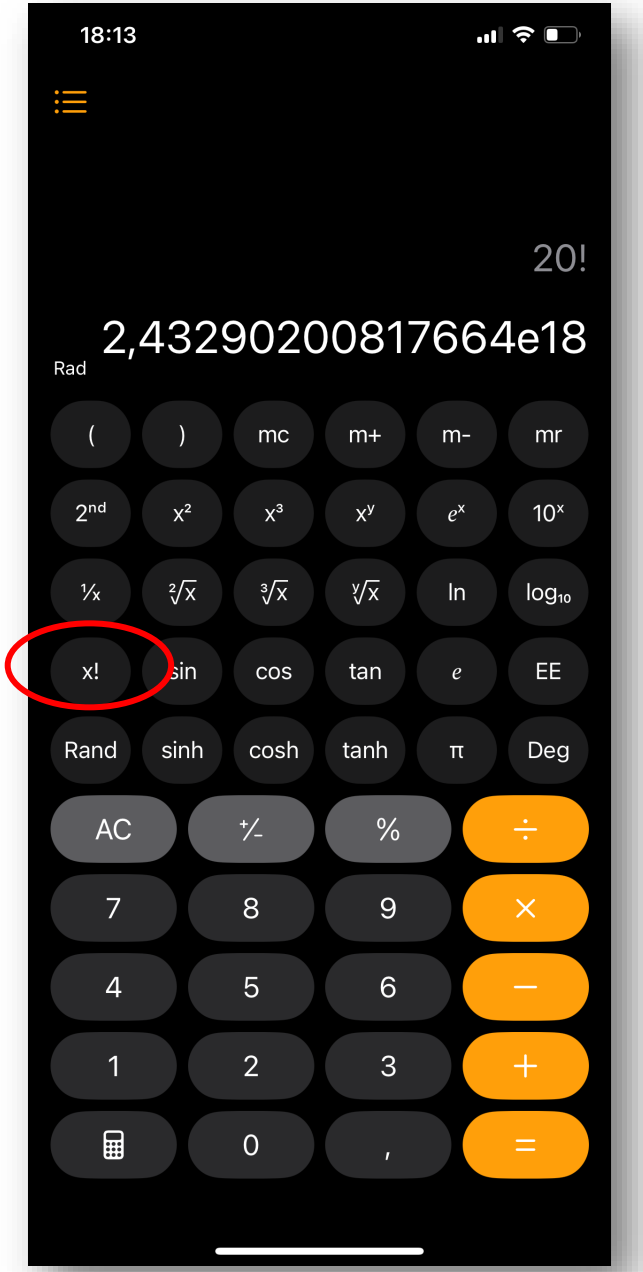
I numeri fattoriali

20!

1x2x3x4x5x6……..x20

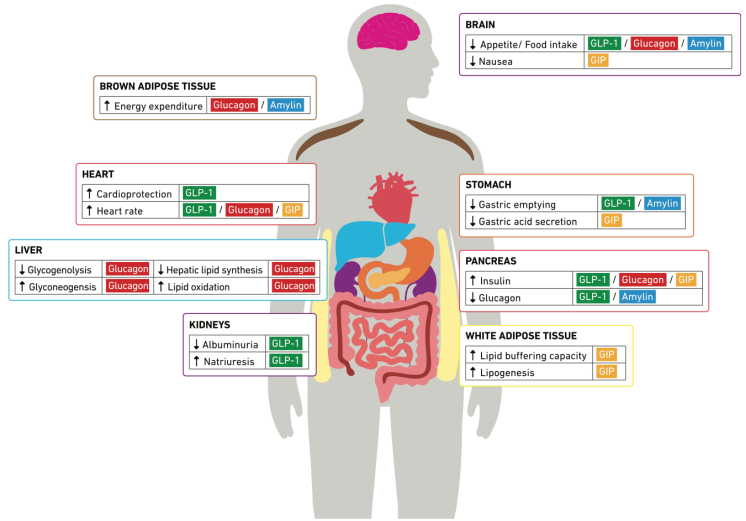
«il prodotto di tutti i numeri naturali da 1 a *n*.»

«Nel calcolo infinitesimale,
misura statistica **per misurare interazioni tra cose**…»



Numero di possibili interazioni
tra i 20 fattori combinati tra loro

Differente espressione recettoriale



https://www.treccani.it/enciclopedia/fattoriale_%28Enciclopedia-della-Matematica%29/

EXPERT OPINION ON PHARMACOTHERAPY
2024, VOL. 25, NO. 7, 801–818
https://doi.org/10.1080/14656566.2024.2356254

OK GLP-1 RA……insieme a chi?

V SESSIONE

OK per GLP-1 RA. Ma insieme a chi?

Moderatori: Katherine Esposito, Gian Paolo Fadini

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Amilina

Un altro dei 20

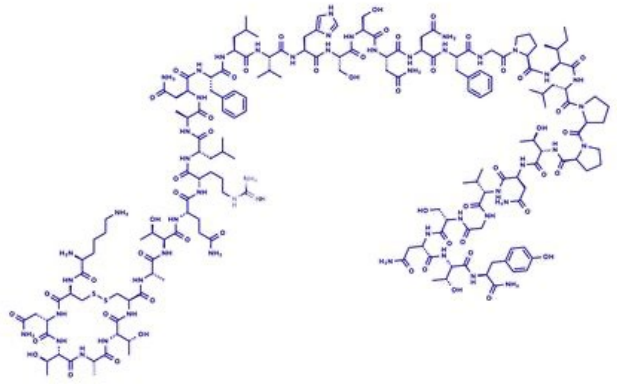
AMILINA (nota come **IAPP**: Islet Amyloid Poly-Peptide): peptide di 37 aminoacidi

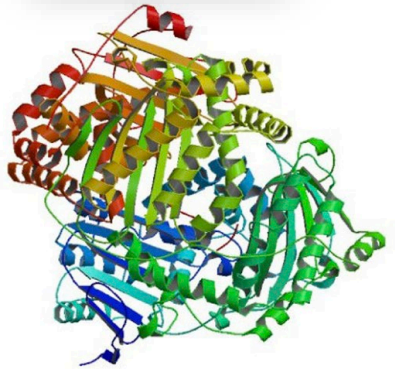
Prodotto prevalentemente da **cellule beta** dell'isola;

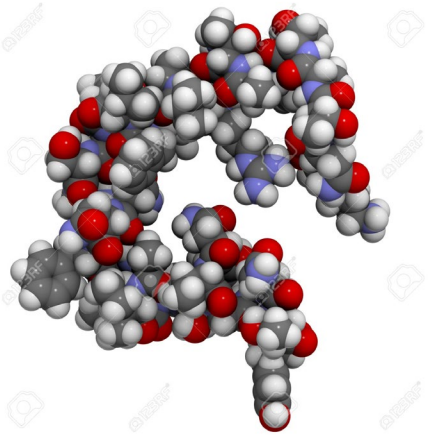
Prodotto anche da **stomaco**, **gangli spinali e cervello**;

Sembra possedere caratteristiche utili per l'omeostasi glicemica ed il controllo del peso

Compreso nella lista dei 20!







https://doi.org/10.1016/j.appet.2022.105965

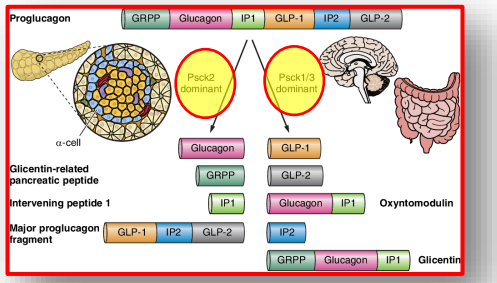
Immagini da: https://www.google.com/search?sca_esv=b95f46454ff89c88&sxsrf=ADLYWIKf4oWbqH9TIUxQxAhzQmpQj_rqaw:1728313502270&q=amilina&udm=2&fbs=AEQNm0Be9hsxO5zOUoY5v2srYNPRIvTz_02aG-_CVE5t-hWDE-o4NknGVSyJJ2oODmr6AzzAs2HXn22T5ul0WKsZVMmJpVctjfuP_7P6Q0vzDgg0UR2OBy8kQja4_o4EYJ19-Z4htN5rihC5e5nWN0tX3m-NShysbeSfMs-yIqyBGYs3SN_Nt8&sa=X&sqi=2&ved=2ahUKEwin1PPCxfyIAxWB87sIHXWCCbsQtKgLegQIGRAB&biw=2268&bih=995&dpr=2#vhid=cIqIxt4ZhciinM&vssid=mosaic

AMILINA prodotta come pre-pro-ormone (precursore di 89 aa)

dal gene della calcitonina (*calcitonin gene peptide superfamily*)

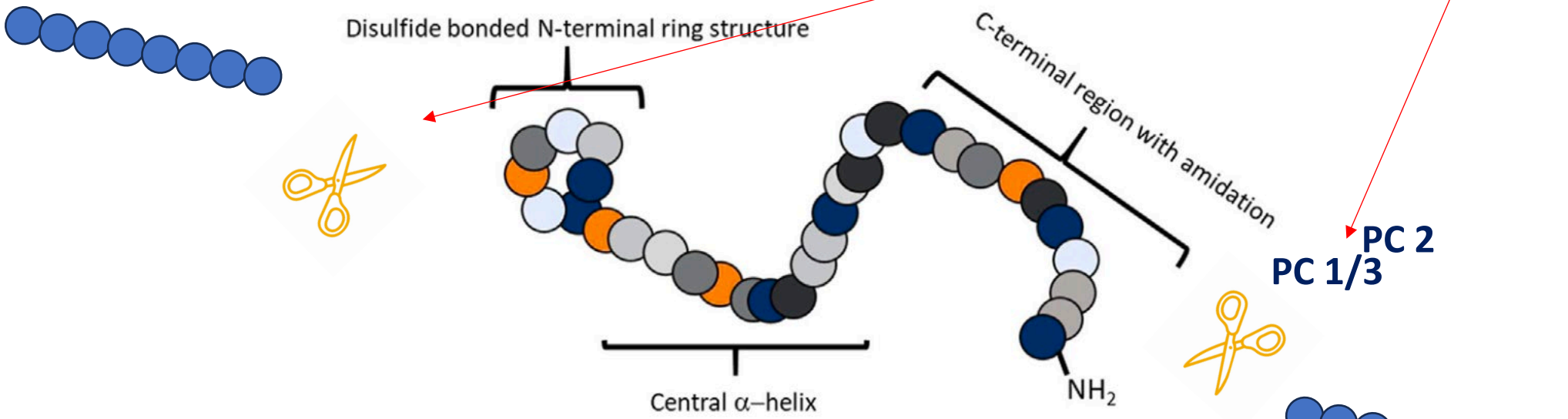
https://doi.org/ 10.1016/0006-291X(86)90708-4.

Similitudini con i 20!



DOI:10.1152/physrev.00013.2014

Peptide maturo 37 aa



PC 2

PC 1/3

Stessi enzimi di clivaggio che tagliano glucagone e GLP-1 dal proglucagone

pM 4KDa

https://doi.org/10.1016/j.molmet.2020.101109

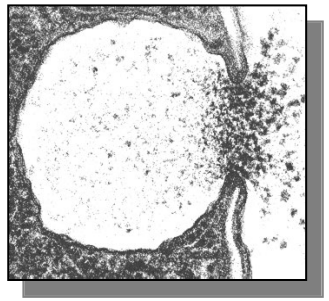
Review

Cell PRESS

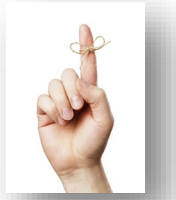
The insulin secretory granule as a signaling hub

Jakob Suckale[1] **and Michele Solimena**[1,2]

[1] Molecular Diabetology, Paul Langerhans Institute Dresden, School of Medicine and University Clinic 'Carl Gustav Carus', Dresden University of Technology, Dresden 01307, Germany
[2] Max Planck Institute of Molecular Cell Biology and Genetics, Dresden 01307, Germany

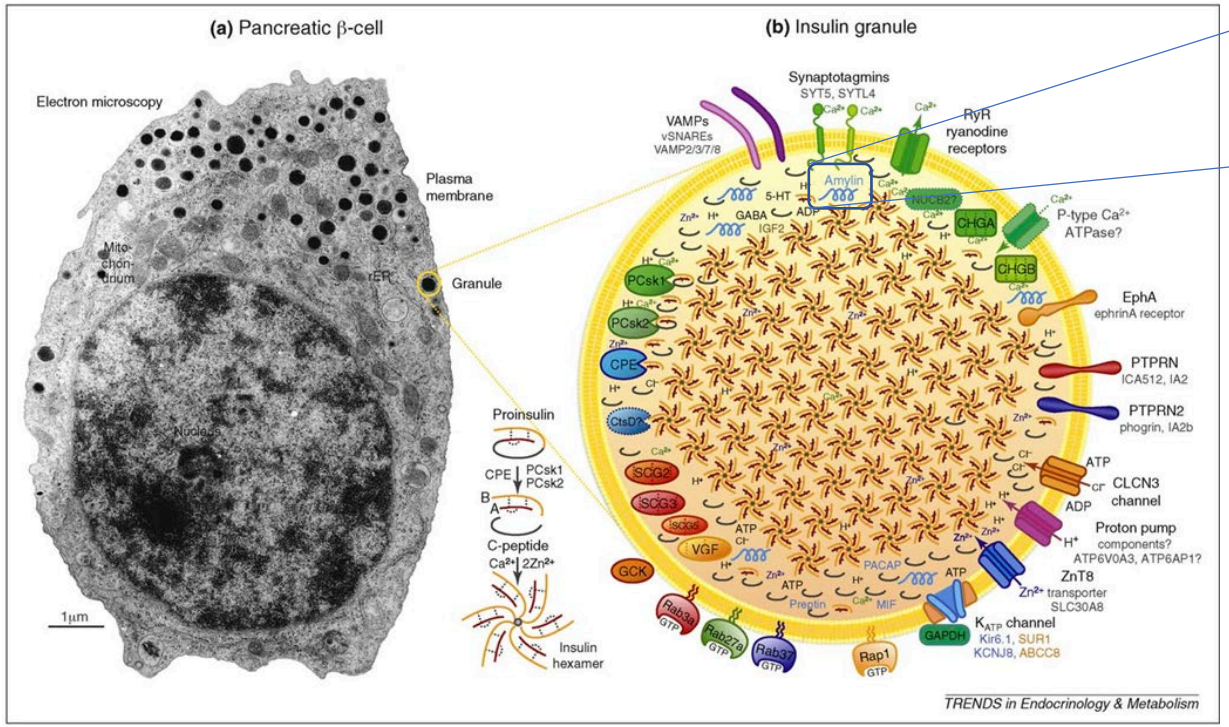


Nel DMT1



no granulo=no amilina

Koda J.E. Fineman M. et al. (1992) (339) Lancet



Amylin

Fisiologicamente è **co-secreta** con l'**insulina** rispettando un **rapporto di 1:100**

Stesse dinamiche secretorie di insulina

doi:10.1016/j.tem.2010.06.003 Trends in Endocrinology and Metabolism 21 (2010) 599–609

https://doi.org/10.4239/wjd.v7. i9.189.
https://doi.org/10.1002/jcb.240550004.
https://doi.org/10.1016/ 0167-4889(89)90220-6.

AMILINA breve storia

1901

….Hyaline degeneration ff the islands of Langerhans….

[Opie E.L. (1901) J. Of Experimental Medicine]

1943-1961

….Extracellular material called AMYLOID….

[Ahronheim 1943 American Journal of Pathology]
[Ehrlich and Ratner 1961 American Journal of Pathology]

1986-1987

[Westermark et al. 1986 BBRC]
[Clark et al. 1987 Lancet]
[Cooper et al. 1987 PNAS]

….sequence of peptide Amylin….

1989

….identified amylin gene ….

1990

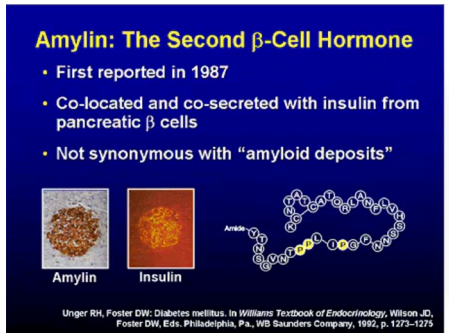
…. ***amylin in beta cell*** ….

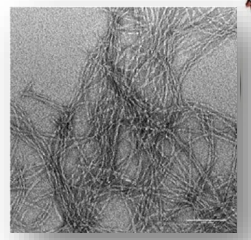
1995-1996

….first full-length amylin analogue….

PRAMLINTIDE

1992





??omeostasi glicemica??

Azioni ed effetti contrastanti ne hanno frenato il successo

…. Amylin's effect….

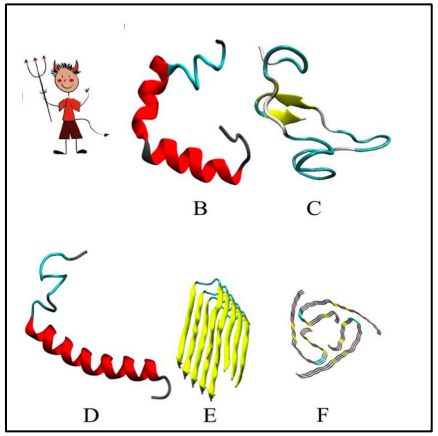
…. Metabolic effects….
…. Adiposity signals ….
…. Brain and islet actions ….

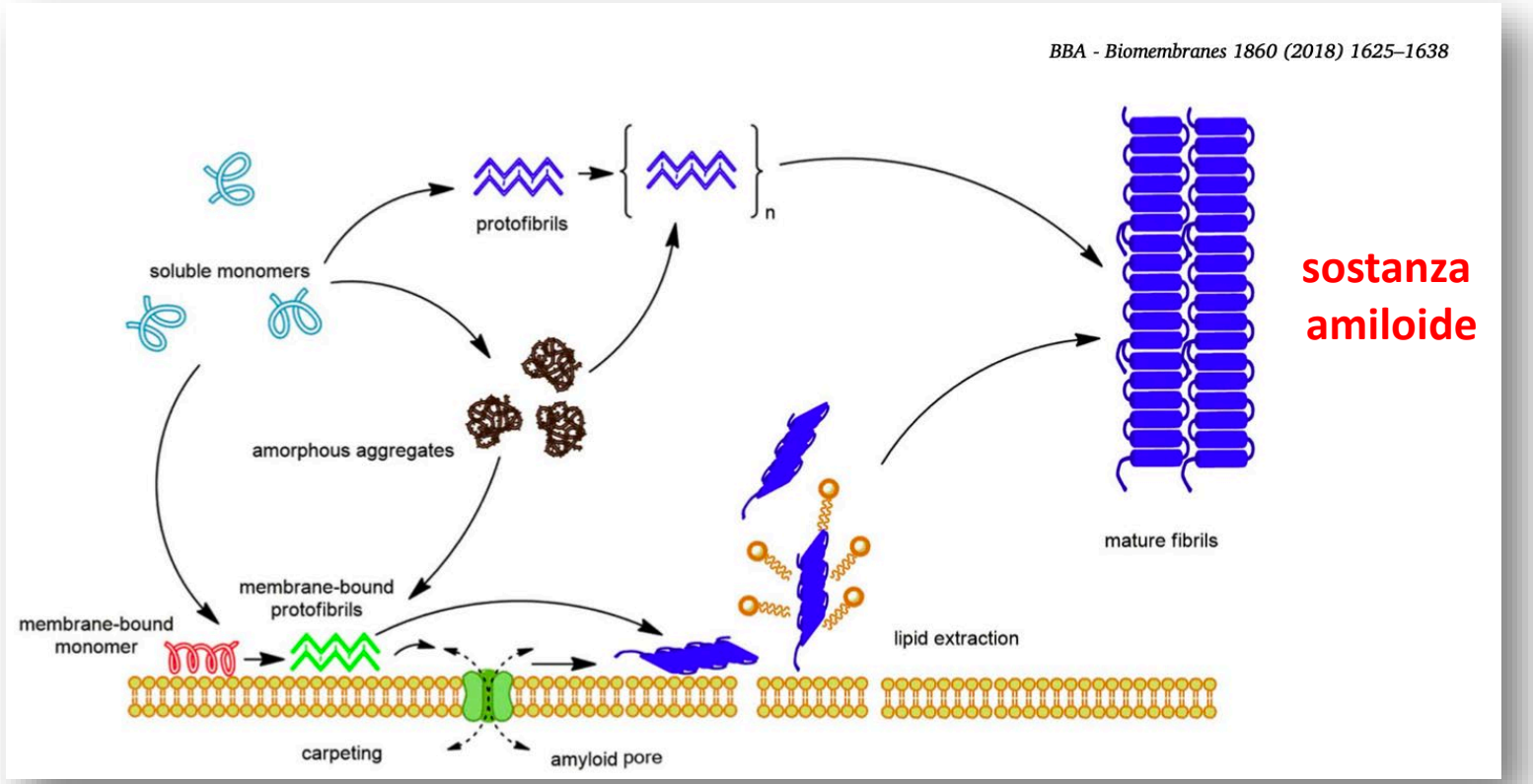
https://doi.org/10.1016/j.appet.2022.105965

TRA le CONTROVERSIE…….

Amilina (forma solubile)

Amilina (possibili errori di conformazione)





sostanza amiloide

Formazione di sostanza amiloide, dannosa per le cellule beta

Amilina umana forma aggregati

Amilina di roditori NON forma aggregati

… sequenza??

https://doi.org/10.1016/j.bbamem.2018.02.022

Condizioni pro-diabetogene

Base fisiopatologica

2002

Metabolism, Vol 51, No 10 (October), 2002: pp 1340-1347

Chronic Exposure to Free Fatty Acids or High Glucose Induces Apoptosis in Rat Pancreatic Islets: Possible Role of Oxidative Stress

Salvatore Piro, Marcello Anello, Cinzia Di Pietro, Maria Natalia Lizzio, Giovanni Patanè, Agata Maria Rabuazzo, Riccardo Vigneri, Michele Purrello, and Francesco Purrello

Condizioni pro-diabetogene

→iperglicemia cronica

→iper FFA cronica

2005

DIABETES, VOL. 54, JULY 2005

Aberrant Processing of Human Proislet Amyloid Polypeptide Results in Increased Amyloid Formation

Johan F. Paulsson[1,2] and Gunilla T. Westermark[1,2]

→**attivazione** promotore del **gene di IAPP**

→**aumento espressione gene** IAPP

→**aumento rilascio** di IAPP

→**aumento enzimi** che **producono IAPP**

→riduzione della secrezione insulinica

→stress reticolo endoplasmatico

→aumento **apoptosi beta cellule**

2008

Carboxypeptidase E mediates palmitate-induced β-cell ER stress and apoptosis

Kristin D. Jeffrey*, Emilyn U. Alejandro*, Dan S. Luciani*, Tatyana B. Kalynyak*, Xiaoke Hu*, Hong Li*, Yalin Lin*, R. Reid Townsend†, Kenneth S. Polonsky†, and James D. Johnson*‡

*Diabetes Research Group, Laboratory of Molecular Signalling in Diabetes, Department of Cellular and Physiological Sciences, University of British Columbia, Vancouver, BC, Canada V6T 1Z3; and †Departments of Internal Medicine and Cell Biology and Physiology, Washington University School of Medicine, St. Louis, MO 63110

Edited by Melanie H. Cobb, University of Texas Southwestern Medical Center, Dallas, TX, and approved April 2, 2008 (received for review December 14, 2007)

PNAS

8452–8457 | PNAS | June 17, 2008 | vol. 105 | no. 24

→aumento **formazione sostanza amiloide**

2010

The American Journal of Pathology, Vol. 176, No. 2, February 2010
Copyright © American Society for Investigative Pathology
DOI: 10.2353/ajpath.2010.090532

Metabolic, Endocrine and Genitourinary Pathobiology

Evidence for Proteotoxicity in β Cells in Type 2 Diabetes

Toxic Islet Amyloid Polypeptide Oligomers Form Intracellularly in the Secretory Pathway

2010

The American Journal of Pathology, Vol. 176, No. 2, February 2010

DOI: 10.2353/ajpath.2010.090532

Metabolic, Endocrine and Genitourinary Pathobiology

Evidence for Proteotoxicity in β Cells in Type 2 Diabetes

Toxic Islet Amyloid Polypeptide Oligomers Form Intracellularly in the Secretory Pathway

human insulinoma and **pancreas from humans** with and without T2DM,

➡ Oligomeri di amiloide

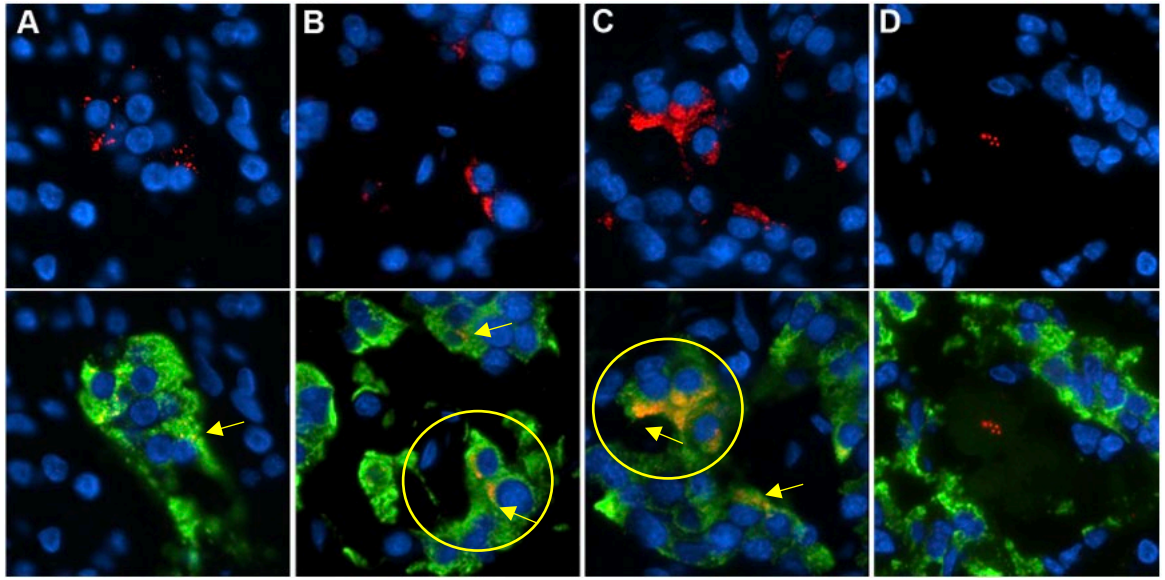
⇨ Autofagosomi che inglobano

patient 1 # patient 2 # patient 3 # patient 4

Anti Amilina

Anti Amilina

Anti Insulina



Co-localization insulin+amylin

human beta cells from T2DM

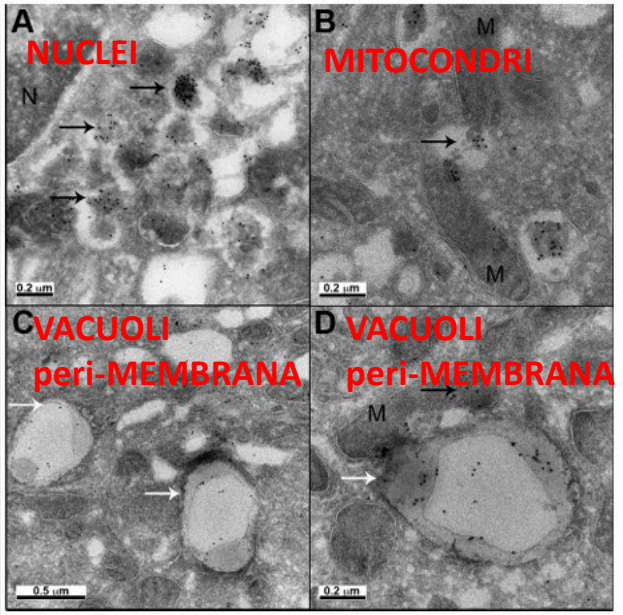


Figure 4. Electron micrographs of human insulinoma cells expressing IAPP. **A:** Oligomer immunoreactivity (10 nm gold) was detected in perinuclear area in vesicle-like structures and aggregates (N, nucleus). **B:** Some oligomer aggregates appear to compromise mitochondria (M) integrity. **C, D:** Oligomer labeling was detected in vacuoles located in membrane rich areas, which appear to be autophagosomes. **Black arrows** point to oligomers (**A, B, D**); **white arrows** point to autophagosomes (**C, D**). Original magnification: ×60,000 (**C**), ×80,000 (**A**), ×100,000 (**B, D**).

human insulinoma

Sequenza aminoacidica di AMILINA e dei suoi analoghi

Name	N-term mod.	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	C-term mod.	Receptor targets
αCGRP		A	C	D	T	A	T	C	V	T	H	R	L	A	G	L	L	S	R	S	G	G	V	V	K	N	N	F	V	P	T	N	V	G	S	K	A	F	$-NH_2$	CGRP-R
Human amylin		K	C	N	T	A	T	C	A	T	Q	R	L	A	N	F	L	V	H	S	S	N	N	F	G	A	I	L	S	S	T	N	V	G	S	N	T	Y	$-NH_2$	AMY-R
Rat amylin		K	C	N	T	A	T	C	A	T	Q	R	L	A	N	F	L	V	H	S	S	N	N	F	G	P	V	L	P	P	T	N	V	G	S	N	T	Y	$-NH_2$	AMY-R
Pramlintide		K	C	N	T	A	T	C	A	T	Q	R	L	A	N	F	L	V	H	S	S	N	N	F	G	P	I	L	P	P	T	N	V	G	S	N	T	Y	$-NH_2$	AMY-R
KBP-088	AC-	C	S	N	L	S	T	C	M	L	G	R	L	S	Q	E	L	H	R	L	Q	T	F	P	K	T	D	V	G	A	N	A	P					$-NH_2$	CTR AMY-R
KBP-089	AC-	C	S	N	L	S	T	C	M	L	G	R	L	S	Q	D	L	H	R	L	Q	T	Y	P	K	T	D	V	G	A	N	A	P					$-NH_2$	CTR AMY-R

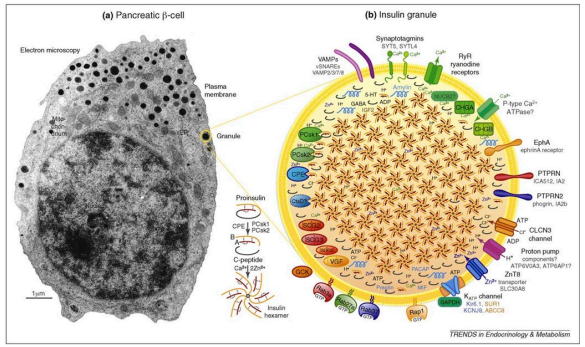
….Extracellular material called AMYLOID….

[Ahronheim **1943** American Journal of Pathology]
[Ehrlich and Ratner **1961** American Journal of Pathology]

https://doi.org/10.1016/j.molmet.2020.101109

20! Prima possibile interazione

Dato che l'AMILINA è contenuta all'interno del granulo secretorio



A

Questa è **Amilina**

Come interagisce con l'insulina?

PROVE DI CONVIVENZA

doi:10.1016/j.tem.2010.06.003 Trends in Endocrinology and Metabolism 21 (2010) 599–609

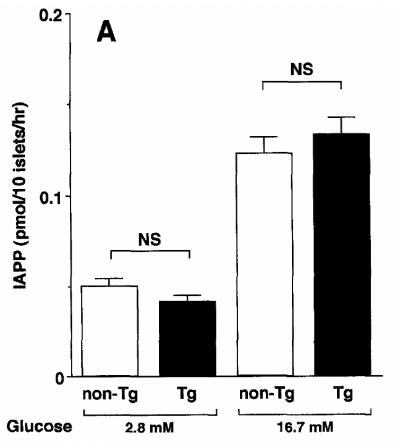
Come interagiscono AMILINA ed INSULINA?

Expression of Human Islet Amyloid Polypeptide/Amylin Impairs Insulin Secretion in Mouse Pancreatic β Cells

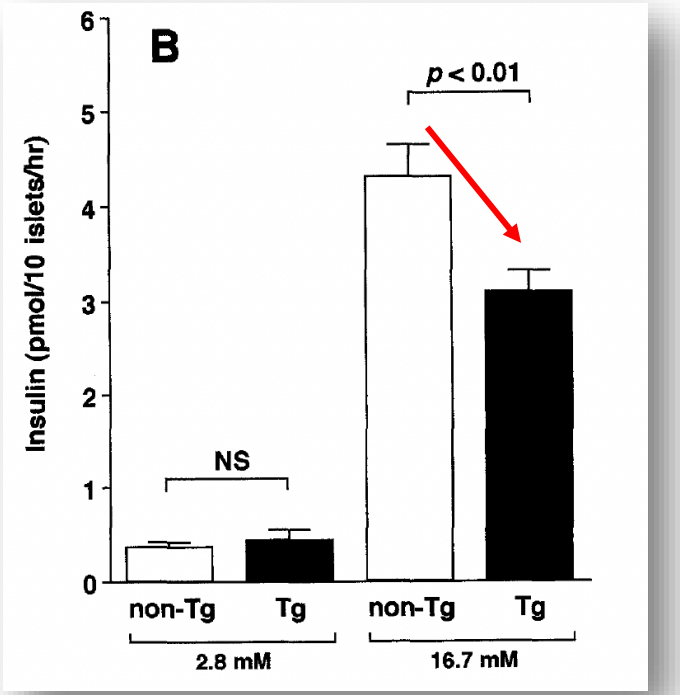
Tatsuhiko Tokuyama, Kazuo Yagui, Takahide Yamaguchi, Choung I. Huang, Nobuhiko Kuramoto, Fumio Shimada, Jun-ichi Miyazaki, Hiroshi Horie, Yasushi Saito, Hideichi Makino, and Azuma Kanatsuka

Non–insulin-dependent diabetes mellitus (NIDDM) is associated histopathologically with islet amyloid deposits of which a major component is islet amyloid polypeptide (IAPP)/amylin. We examined whether endogenous IAPP controls insulin secretion via a local effect within pancreatic islets and whether overexpression of this peptide contributes to pancreatic β-cell dysfunction in this disease. Transgenic mice expressing human IAPP in pancreatic β cells were used in this study. Human IAPP expression did not influence the mouse proinsulin mRNA level and insulin content. Glucose-induced insulin secretion was decreased in the isolated pancreatic islets of transgenic mice. MIN6, a glucose-responsive pancreatic β-cell line, was transfected with human IAPP cDNA by a lipofectin method. Human IAPP expression was confirmed by RNA blot and immunohistochemical analysis. In two transfectants expressing the largest amount of human IAPP, insulin secretion was increased in response to glucose stimulation; however, the magnitude of the insulin response in cells transfected with human IAPP was smaller than in control clones. Insulin content was not influenced by the expression. We conclude that endogenous IAPP inhibits insulin secretion via an autocrine effect within pancreatic islets, and that the impaired insulin secretion in this disease may be partly caused by overexpression of IAPP.

Studio del 1997



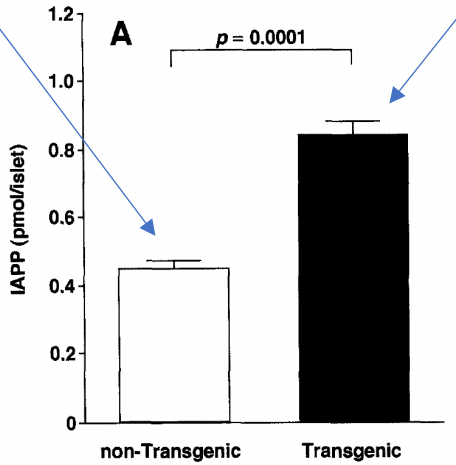
Secrezione di Amilina nelle isole



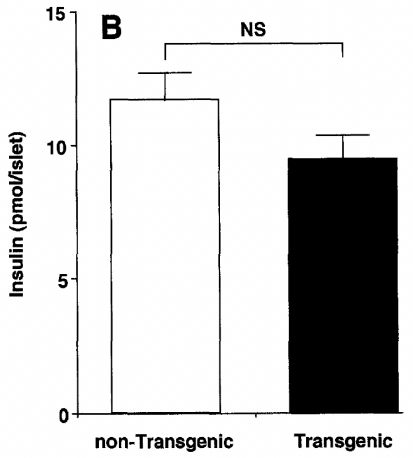
Secrezione di INSULINA nelle isole

controllo

Iper-espressione nelle isole di IAPP umana

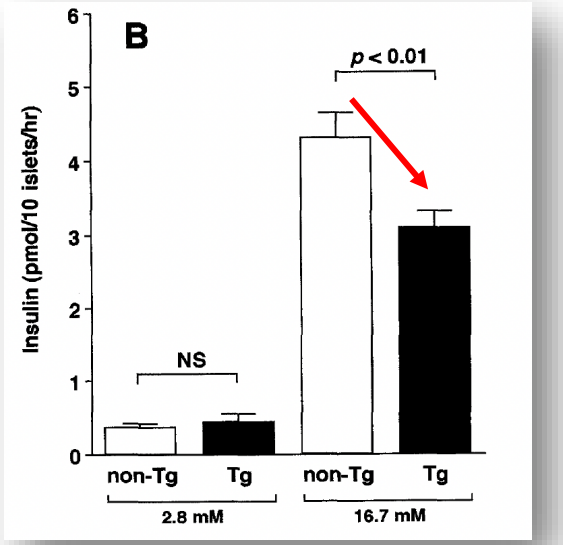


Contenuto di Amilina nelle isole



Contenuto di INSULINA nelle isole

CONVIVENZA TRAVAGLIATA



Secrezione di INSULINA nelle isole

Sembra esistere una competizione tra i due ormoni

La competizione sembra 'apparentemente' funzionale, in termini di rilascio da parte del granulo…

PROVE DI MATRIMONIO

Per analizzare le azioni….

modello 2

KO per IAAP

BIOCHEMICAL AND BIOPHYSICAL RESEARCH COMMUNICATIONS 250, 271–277 (1998)
ARTICLE NO. RC989308

1998

Increased Insulin Secretion and Glucose Tolerance in Mice Lacking Islet Amyloid Polypeptide (Amylin)

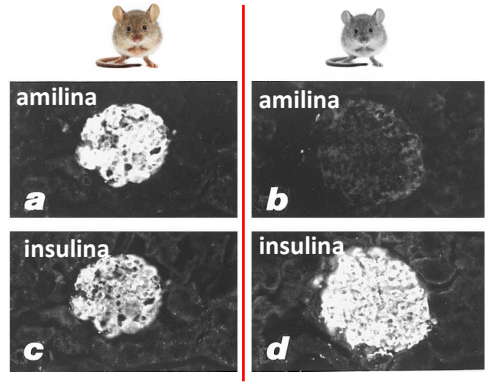
Samuel Gebre-Medhin,*,[1] Hindrik Mulder,†‡ Milos Pekny,* Gunilla Westermark,§ Jan Törnell,¶ Per Westermark,§ Frank Sundler,† Bo Ahrén,‖ and Christer Betsholtz*

*Department of Medical Biochemistry, Göteborg University, Göteborg, Sweden; †Department of Physiology and Neuroscience, Section for Neuroendocrine Cell Biology, ‡Department of Cell and Molecular Biology, Lund University, Lund, Sweden; §Department of Pathology I, Linköping University, Linköping, Sweden; ¶Department of Physiology, Section for Endocrinology, Göteborg University, Göteborg, Sweden; and ‖Department of Medicine, Wallenberg Laboratoriet, Lund University, Malmö, Sweden

Received August 14, 1998

Topi di controllo con gene amilina **[*IAPP* +/+]**

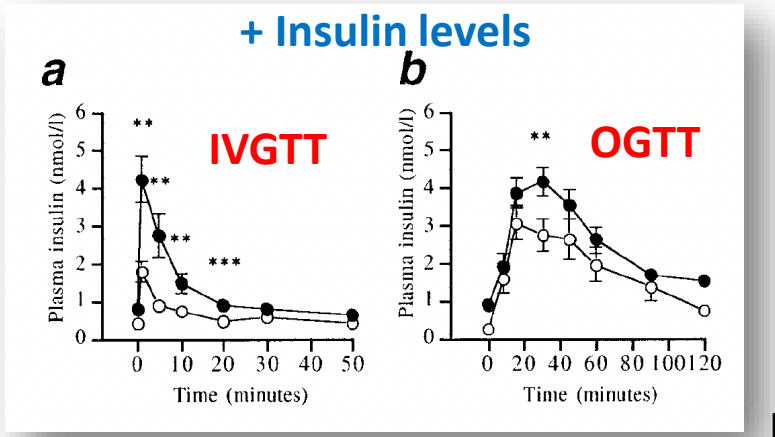
Knockout mice per gene amilina [*IAPP* -/-]



Isole pancreatiche

transgenic rescue experiment

Aggiungendo nuovamente AMILINA

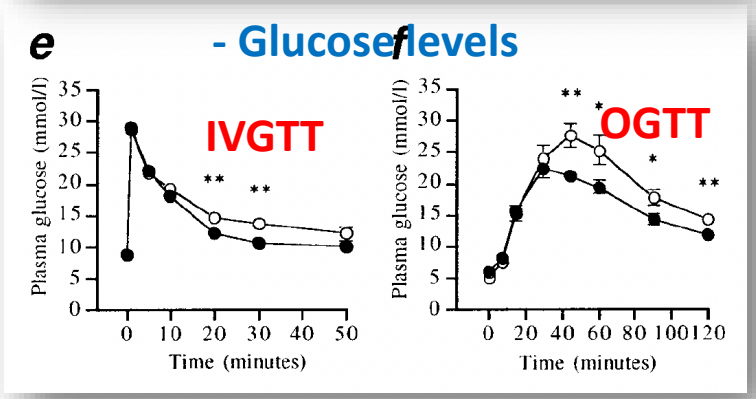


Topi *KO* **senza amilina**

Topi di controllo

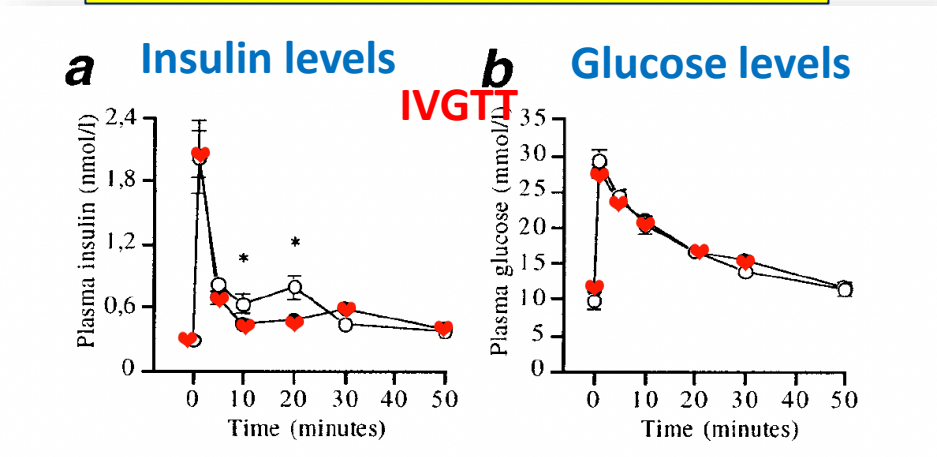
In assenza di AMILINA

Maggiore secrezione di insulina e minore escursione di glucosio



Topi *KO* **senza amilina**

Topi di controllo



Si ripristina la condizione iniziale di controllo
(si perde il beneficio secretorio??)

Topi *KO* **senza amilina + amilina**

Topi di controllo

ELSEVIER

Physiology & Behavior 91 (2007) 566–572

PHYSIOLOGY & BEHAVIOR

Modulation of the satiating effect of amylin by central ghrelin, leptin and insulin

M. Osto, P.Y. Wielinga, B. Alder, N. Walser, T.A. Lutz *

Institute of Veterinary Physiology, and Zurich Center for Integrative Human Physiology, Vetsuisse Faculty University of Zurich, Winterthurerstrasse 260, 8057 Zurich, Switzerland

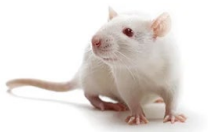
Received 12 October 2006; received in revised form 8 March 2007; accepted 21 March 2007

AMILINA *sinergizza* e potenzia l'azione dell'insulina
Effetti additivi di potenziamento

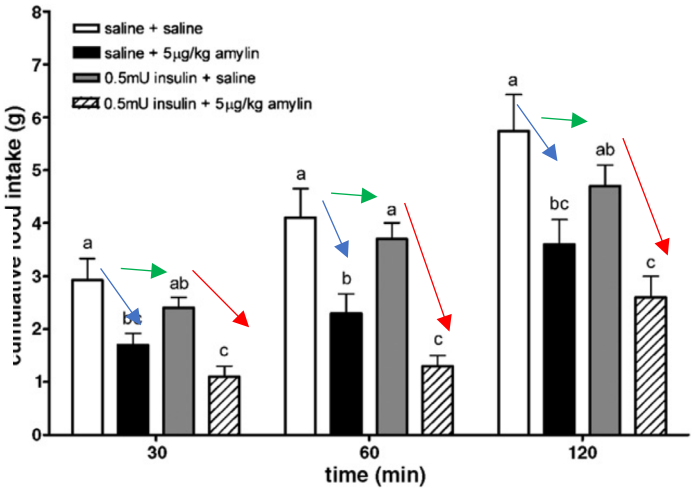
Insulina →normalmente iperfagia

Esperimenti in vivo di infusione

Fenotipo normale



Ratti Wistar



AMILINA riduce fame (consumo cibo)

AMILINA+Insulina riducono maggiormente fame (consumo cibo)

Effetto additivo e sinergizzante

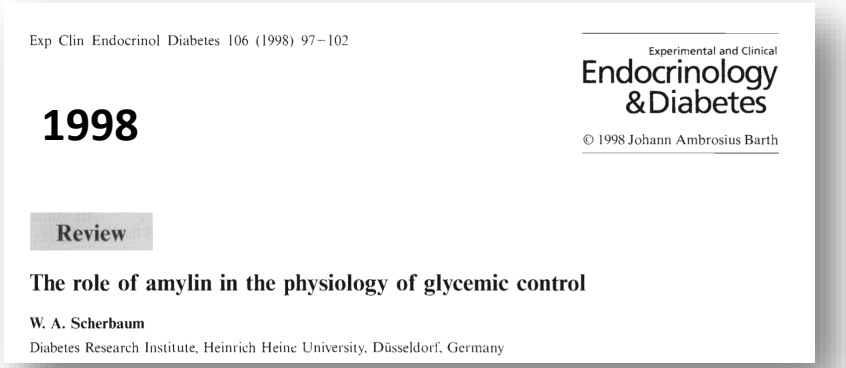
AMILINA ⟷ INSULINA

AMILINA ⟷ INSULINA

SONO NECESSARIE ENTRAMBE

Cosa succede nell'uomo?

In fisiologia


Exp Clin Endocrinol Diabetes 106 (1998) 97–102

Experimental and Clinical Endocrinology & Diabetes

1998

© 1998 Johann Ambrosius Barth

Review

The role of amylin in the physiology of glycemic control

W. A. Scherbaum
Diabetes Research Institute, Heinrich Heine University, Düsseldorf, Germany

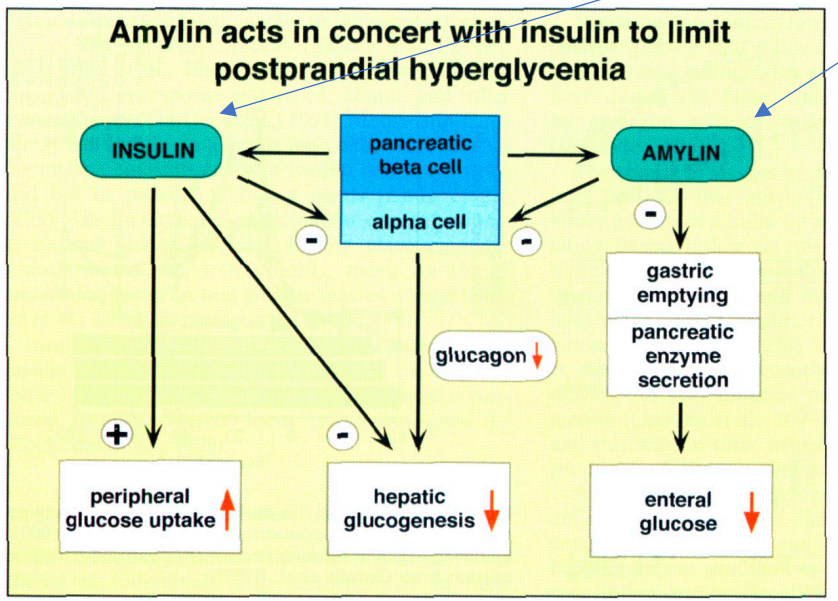
E se dovessimo integrare anche l'amilina???

L'AMILINA ha azioni simili all'insulina

sia insulina che Amilina

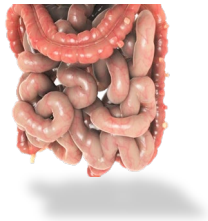
controllano la secrezione di glucagone

Necessità di comprendere il significato



In fisiopatologia

Quale ruolo nell'integrazione con altro ormoni GI??



Lavoro di squadra

AMILINA e controllo glicemico

- Controllo della glicemia **(interazione con insulina)**

Risultati deludenti e **non conclusivi**

Anni ‘90

Azioni buone ma …minimali

Effetto prorompente dei GLP-1RA

Interessi ridotti

V SESSIONE

OK per GLP-1 RA. Ma insieme a chi?

Moderatori: Katherine Esposito, Gian Paolo Fadini

09.30	GIP (e antiGIP)	*Francesco Giorgino*
09.50	Discussione	
10.00	Glucagone	*Riccardo C. Bonadonna*
10.20	Discussione	
10.30	Amilina	*Salvatore Piro*
10.50	Discussione	

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SCIENTIFIC REPORTS

OPEN

Combined Amylin/GLP-1 pharmacotherapy to promote and sustain long-lasting weight loss

Received: 15 February 2019
Accepted: 20 May 2019
Published online: 11 June 2019

Claudia G. Liberini, Kieran Koch-Laskowski, Evan Shaulson, Lauren E. McGrath, Rachele K. Lipsky, Rinzin Lhamo, Misgana Ghidewon, Tyler Ling, Lauren M. Stein & Matthew R. Hayes

A growing appreciation of the overlapping neuroendocrine mechanisms controlling energy balance has highlighted combination therapies as a promising strategy to enhance sustained weight loss. Here, we investigated whether amylin- and glucagon-like-peptide-1 (GLP-1)-based combination therapies produce greater food intake- and body weight-suppressive effects compared to monotherapies in both lean and diet-induced obese (DIO) rats. In chow-maintained rats, systemic amylin and GLP-1 combine to reduce meal size. Furthermore, the amylin and GLP-1 analogs salmon calcitonin (sCT) and liraglutide produce synergistic-like reductions in 24 hours energy intake and body weight. The administration of sCT with liraglutide also led to a significant enhancement in cFos-activation in the dorsal-vagal-complex (DVC) compared to mono-therapy, suggesting an activation of distinct, yet overlapping neural substrates in this critical energy balance hub. In DIO animals, long-term daily administration of this combination therapy, specifically in a stepwise manner, results in reduced energy intake and greater body weight loss over time when compared to chronic mono- and combined-treated groups, without affecting GLP-1 receptor, preproglucagon or amylin-receptor gene expression in the DVC.

Obesity affects more than one-third of individuals in the world, creating an enormous health and economic burden, yet effective non-invasive treatments are limited[1,2]. The failure of behavioral interventions to produce sustained weight loss in an overwhelming majority of cases underscores the importance of identifying novel therapeutic strategies to promote and sustain reductions in food intake and body weight. As energy homeostasis involves innumerous interactions among redundant neural mechanisms[3–5], it is reasonable to consider that treating a complex disease such as obesity may require combinatorial therapies that target multiple energy balance-regulating pathways. However, identifying the right combinatorial therapeutic strategies has proven challenging. Ideally, an effective combination drug therapy for treating obesity would enhance the magnitude and temporal duration of weight loss but would also engage a set of partially-overlapping, complementary neural circuits to mechanistically suppress feeding. To this end, the amylin and glucagon-like peptide-1 (GLP-1) hormonal systems have been independently identified as promising targets for pharmacotherapy intervention[6–11].

While the appetite suppressive effects of amylin and GLP-1 receptor agonists are known to involve direct actions in the brain[6,7,9,10,12–14], uncovering the neuroanatomical site(s) of action and the physiological mechanisms that mediate the anorectic response remains an area of intense research focus. In the hindbrain, the dorsal-vagal-complex (DVC), collectively comprised of the nucleus tractus solitarius (NTS), area postrema (AP) and dorsal motor nucleus of the vagus (DMV), mediates the anorectic effects of both amylin and GLP-1 receptor agonists[15–19]. As amylin and GLP-1 target different populations of neurons in the DVC[20], this suggests that combination drug therapies affecting both systems may provide a greater magnitude and sustainable degree of weight loss compared to monotherapies due to a reduced likelihood of tachyphylaxis[21–23]. Indeed, a critical barrier that the obesity research field must overcome is to find pharmacotherapies that can break-through the plateau of weight loss that occurs with every existing FDA-approved treatment for obesity[1,24–28].

Due in part to the short half-life of endogenous amylin and GLP-1[29,30], long acting agonists for amylin (*e.g.* pramlintide) and GLP-1 receptors (GLP-1R; *e.g.* liraglutide) have been developed[21–23,31–35]. Pramlintide and liraglutide are already FDA-approved monotherapies for the treatment of type II diabetes[36]. In addition, peripheral

Department of Psychiatry, Perelman School of Medicine, University of Pennsylvania, 19104, Philadelphia, PA, USA. Claudia G. Liberini and Kieran Koch-Laskowski contributed equally. Correspondence and requests for materials should be addressed to M.R.H. (email: hayesmr@pennmedicine.upenn.edu)

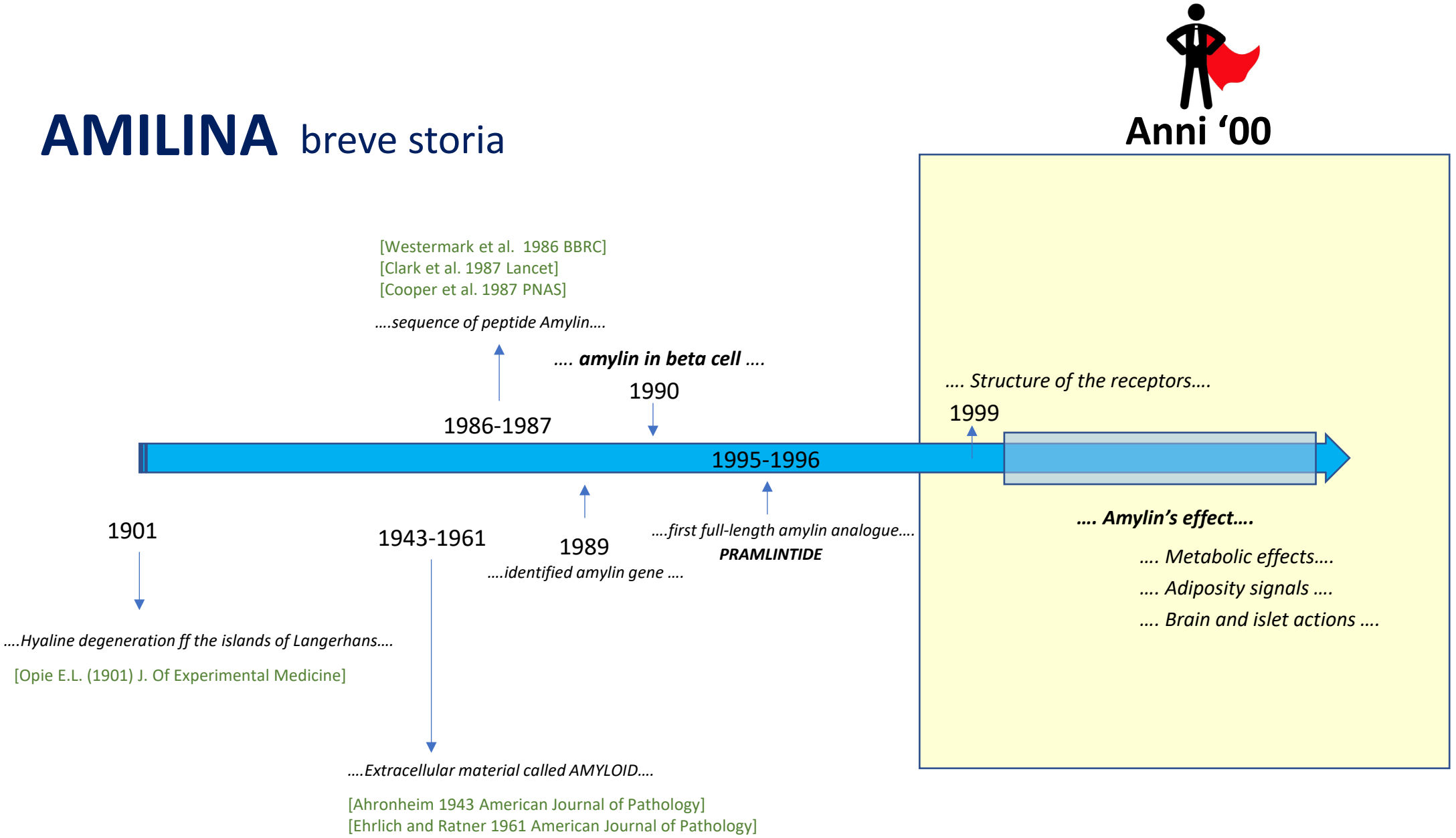
Indipendentemente da insulina e da glicemia, **l'AMILINA ha molteplici potenziali da scoprire, meglio se in compagnia**

AMILINA + GLP-1 RA

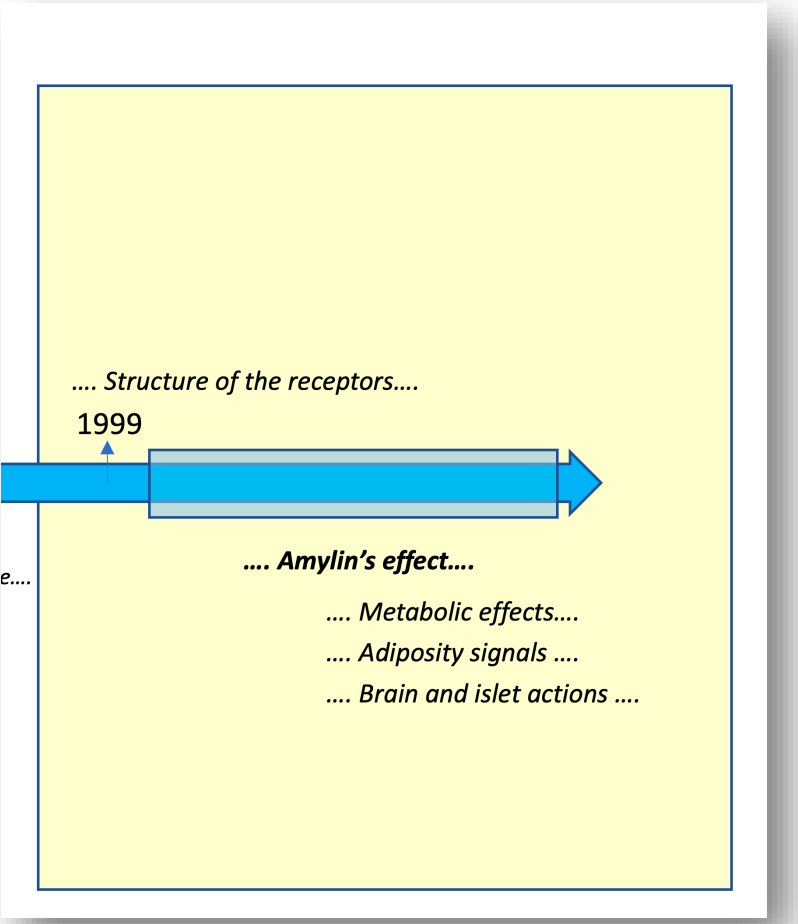
- **Controllo della secrezione di glucagone (modula la secrezione)**
- **Controllo del peso corporeo**
- **Controllo dello svuotamento gastrico**
- **Controllo della fame e della sazietà**
- **Controllo sul cervello**

Anni '00

AMILINA breve storia

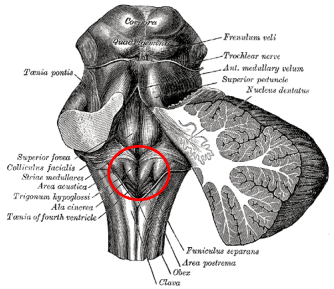


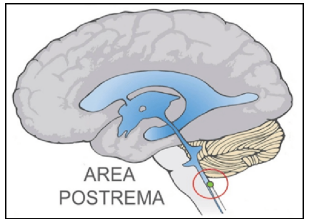
https://doi.org/10.1016/j.appet.2022.105965



Amilina è l'ormone della fame e della sazietà.
Questa azione sembra essere ''diretta'' tramite bersagli specifici cerebrali

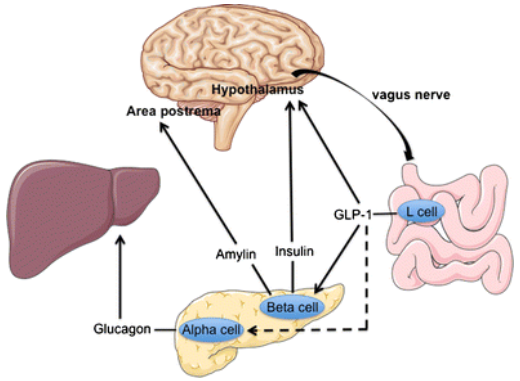
[Lutz, Althaus, Rossi and Scharrer 1998 *Petide*]





Presenza di recettori
nell'**AREA POSTREMA** (AP)

Indipendenza da barriera emato-encefalica

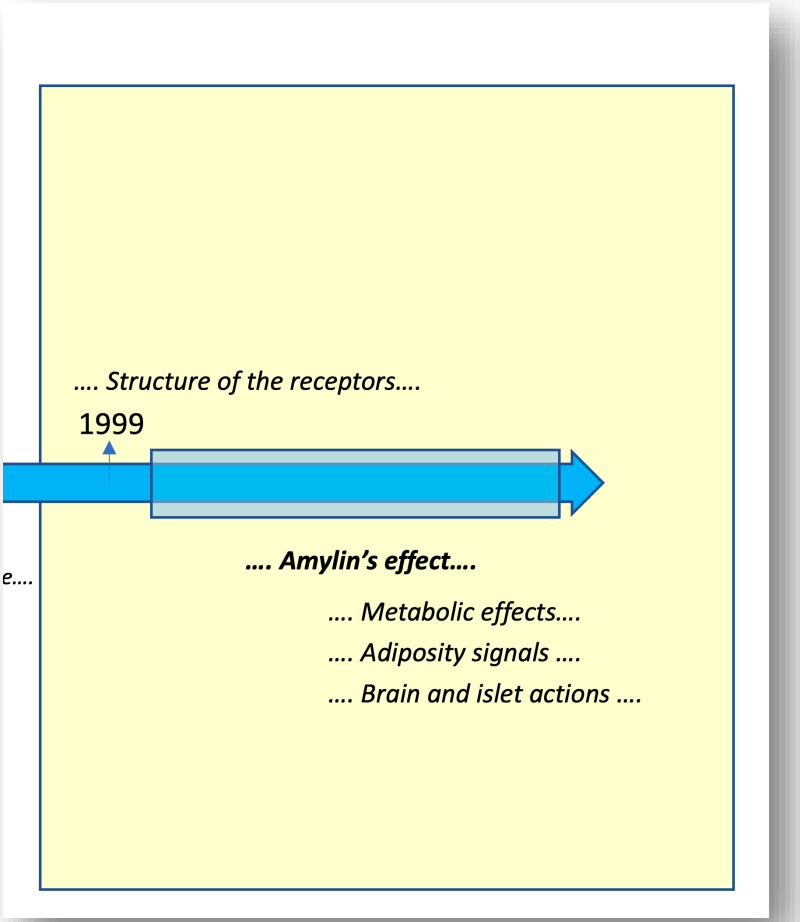


Ipotizzato un *loop* diretto
Pancreas-cervello

e possibili
connessioni nervose
Cervello-periferia

[Le Foll and Lutz, 2020 *Comprehensive Physiology*]

Che tipo di recettori e quale risposta?

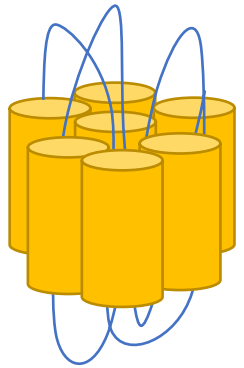


*Amilina si lega a **recettori** della famiglia della **calcitonina** **(CTR)***

Sono recettori di membrana a 7 domini trans-membrana

Spesso mediano l'effetto tramite la proteina G e cAMP

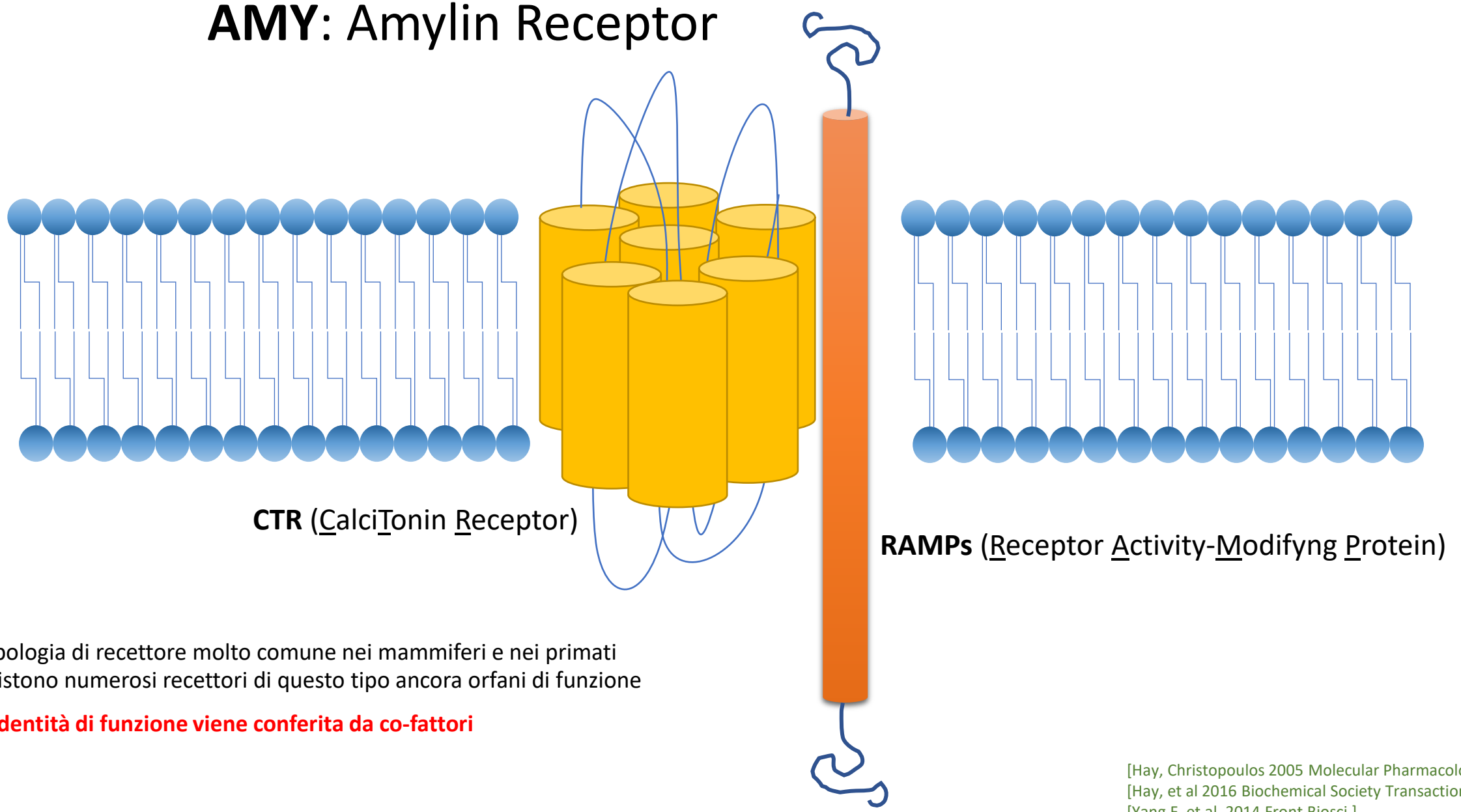
Modulano azioni pleiotropiche molteplici in funzione di cofattori



Esistono un numero elevato di questi recettori ancora orfani di funzioni e ligandi

[Hay, Christopoulos 2005 Molecular Pharmacology]
[Hay, et al 2016 Biochemical Society Transactions]
[Yang F. et al. 2014 Front Biosci.]

AMY: Amylin Receptor



CTR (CalciTonin Receptor)

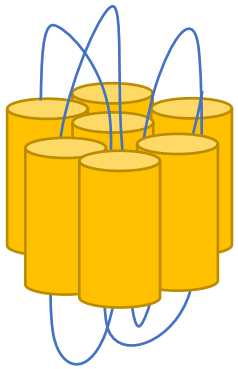
RAMPs (Receptor Activity-Modifyng Protein)

Tipologia di recettore molto comune nei mammiferi e nei primati
Esistono numerosi recettori di questo tipo ancora orfani di funzione

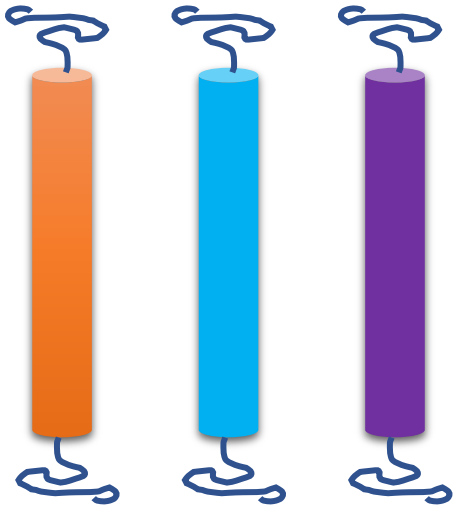
L'Identità di funzione viene conferita da co-fattori

[Hay, Christopoulos 2005 Molecular Pharmacology]
[Hay, et al 2016 Biochemical Society Transactions]
[Yang F. et al. 2014 Front Biosci.]

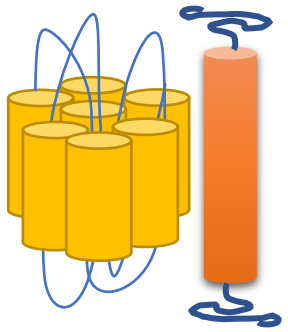
CTR



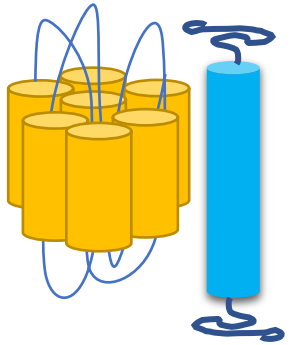
Almeno 3 forme di RAMP



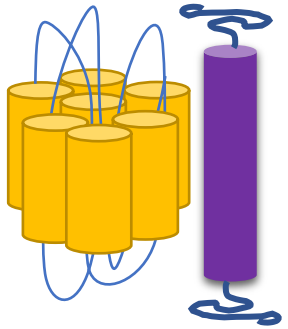
RAMP1 RAMP2 RAMP3



AMY 1

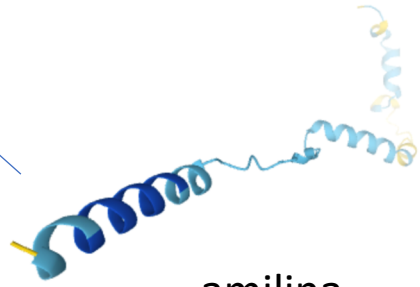


AMY 2



AMY 3

Controllo adiposità, fame sazietà

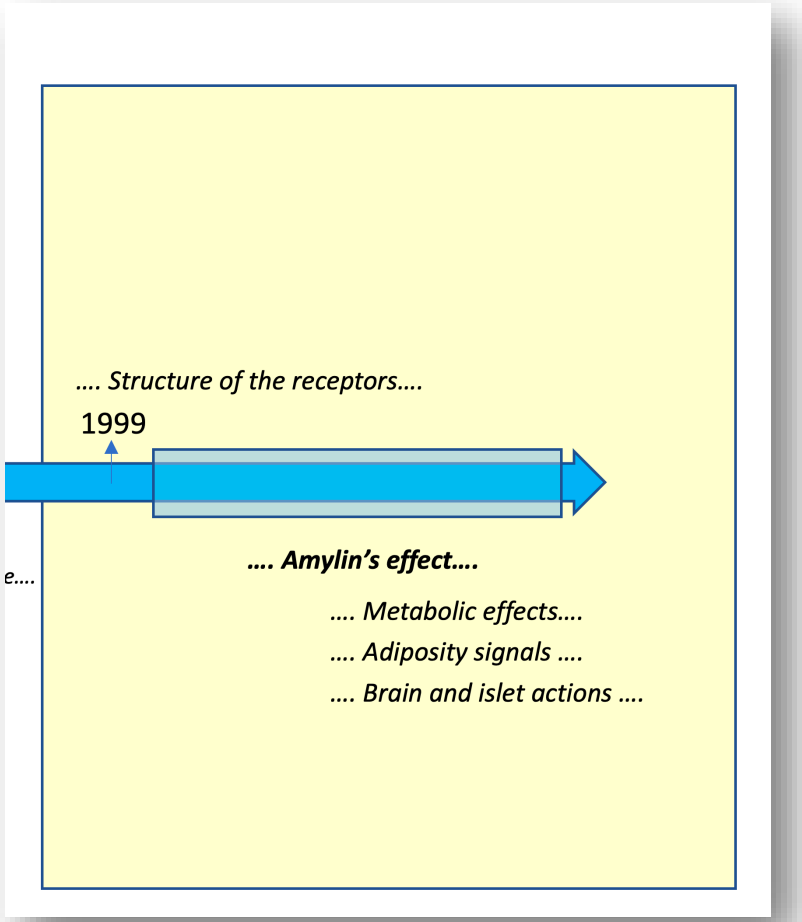


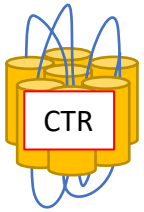
amilina

Controllo metabolismo del glucosio

[Coster, Pence et al. 2020 Neuroscience]

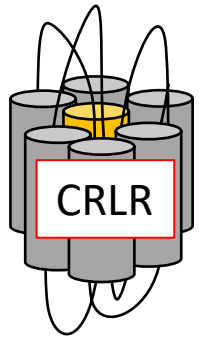
AMILINA e recettori (oltre CTR anche CRLR)





Data l'origine di Amilina, oltre a CTR, puo' legare anche:*

→ *i recettori per* ***(CRLRs)*** *Calcitonin Receptor-****Like*** *Receptors*

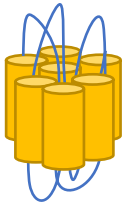


Questi recettori legano i peptidi derivanti da Calcitonin-Gene-Related Peptide (CGRP):

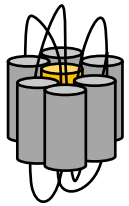
- *Amilina*
- *Calcitonina*
- *Intermedina/adrenomedullina*

gene **calcitonina, localizzato braccio p cromosoma 12, contiene sequenze di altri ormoni.*

[Hay, Christopoulos 2005 Molecular Pharmacology]
[Hay, et al 2016 Biochemical Society Transactions]
[Yang F. et al. 2014 Front Biosci.]

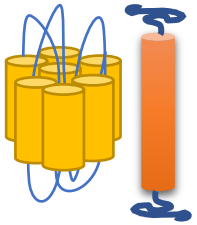


CTR
(recettore calcitonina)

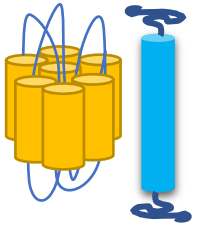


CRLR
(calcitonin-receptor like receptor)

Considerato Recettore ORFANO



AMY 1



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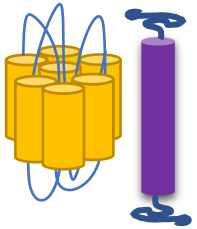

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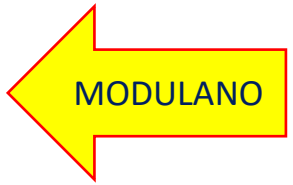
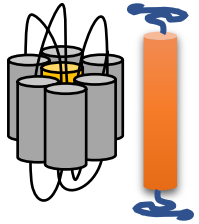
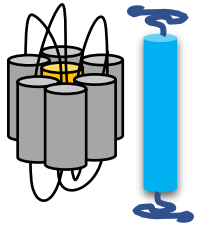
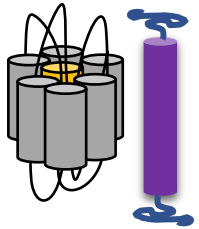




CGRP-R



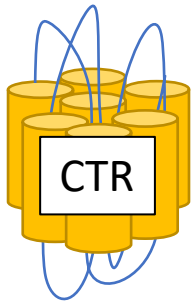
AM1-R

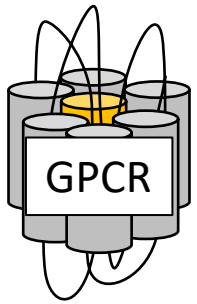


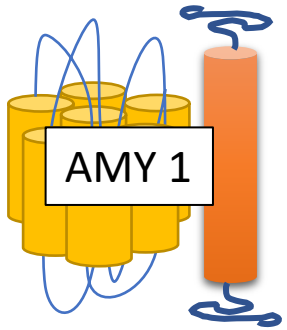
AM2-R

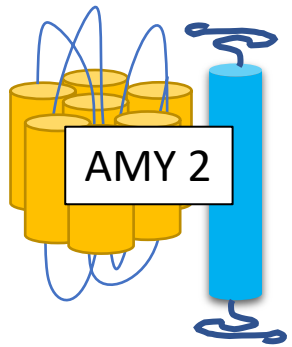
Quando interagisce con i RAMPs
-legano Adrenomedullina 1 e 2
-legano peptidi dericati da calcitonina
-MODULANO azione di AMYs

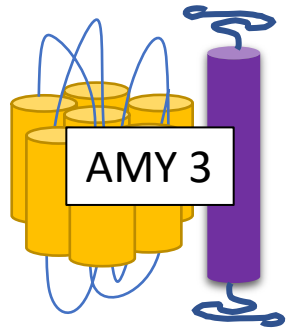
https://doi.org/10.1016/j.molmet.2020.101109



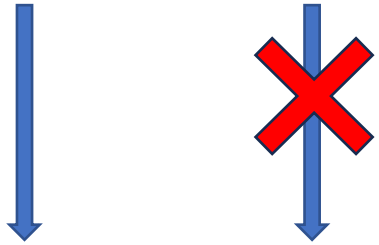


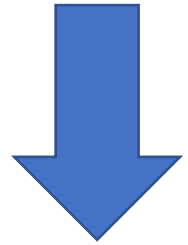






Potenza metabolica rilevata





Manca il reale **numero** di RAMPs

Manca il reale numero **polimorfismi** di RAMPs

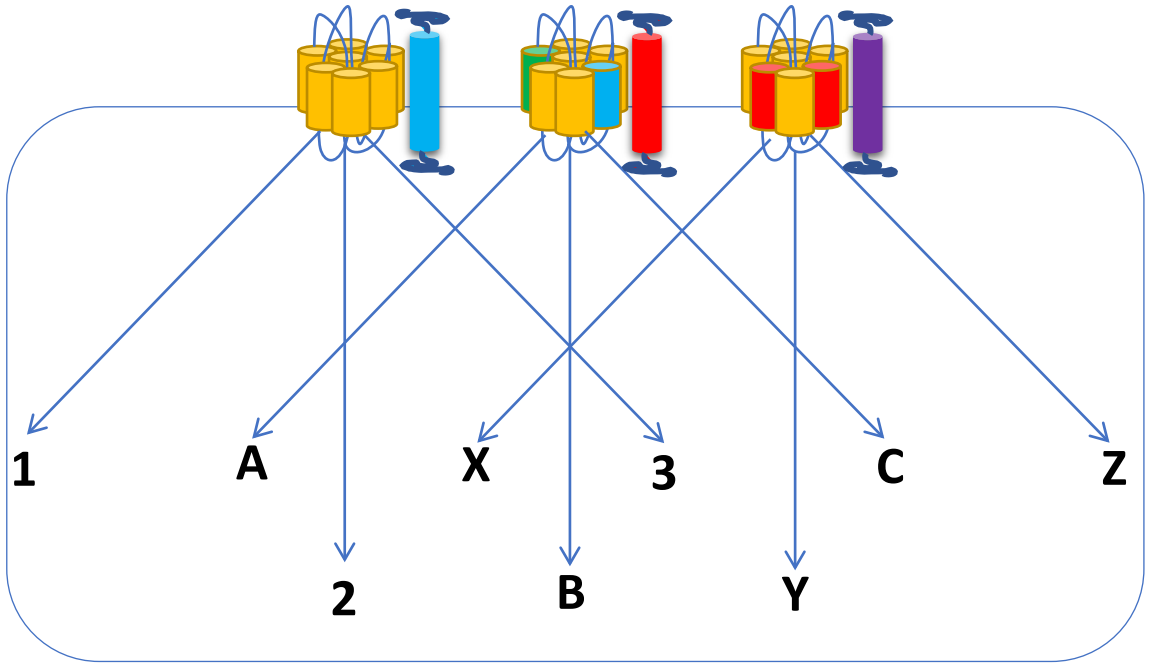
Manca il reale numero **polimorfismi** di CTR

Manca la **regola di distribuzione** di AMYs

[Coster, Pence et al. 2020 Neuroscience]
https://doi.org/10.1016/j.molmet.2020.101109

Effetto camaleontico dipendente dal recettore legato

Recettori differenti



Lo stesso tessuto può mostrare pattern di espressione recettoriale diverso

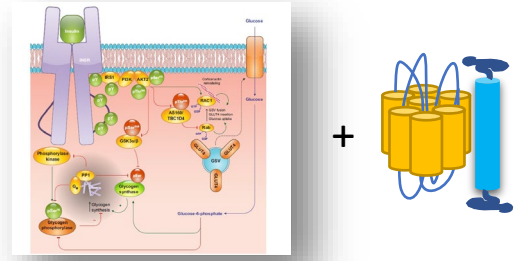
Effetti tessuto-specifici
Effetti dose-dipendente
Eterogeneità **recettori**ale

Effetto intracellulare dipendente da:

- Tipo di recettore legato
- Affinità recettoriale
- Secondo messaggero intracellulare
- Interazione con altre vie di segnale

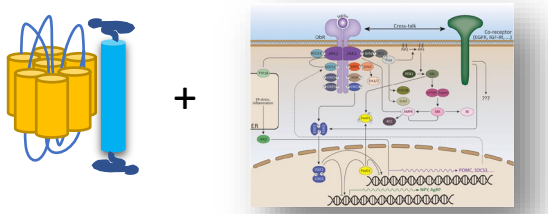
Interazione con altri recettori

Recettore per **insulina**



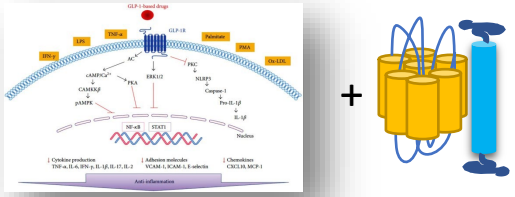
https://www.google.com/search?q=recettore+insulinico

Recettore per **leptina**



https://www.google.com/search?q=recettore+leptina

Recettore **GLP-1**



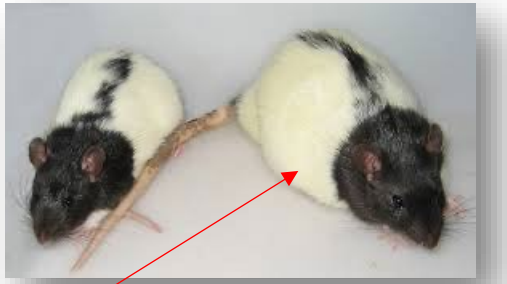
https://www.google.com/search?q=recettore+GLP1

interazione con altri recettori potrebbe influenzare ulteriormente l'effetto
→ **vedi leptina**

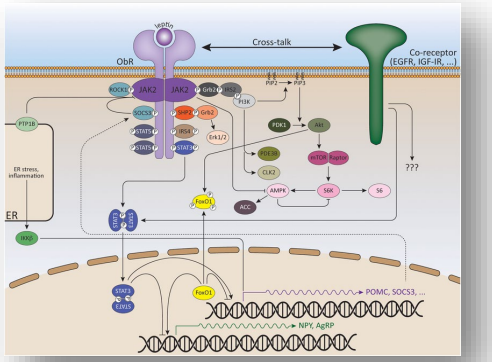
Amilina → RECETTORI → Leptina

Un altro enigma da riscoprire

Ratti ZDF



Recettore per leptina

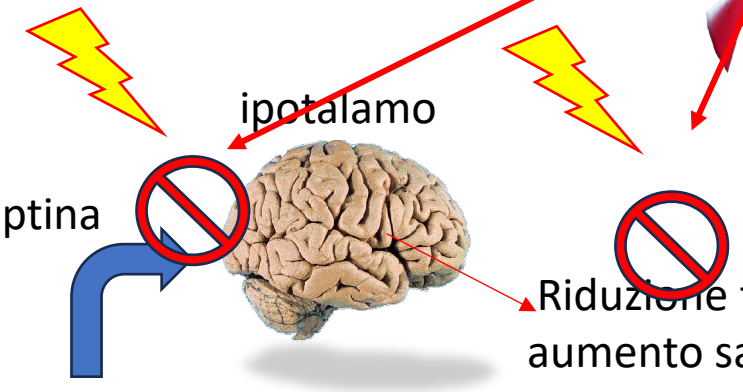


Assenza Recettore per leptina

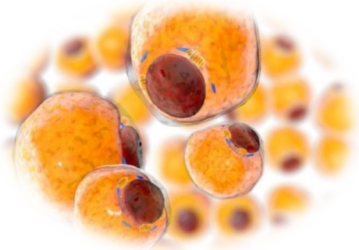
estrema obesità e diabete

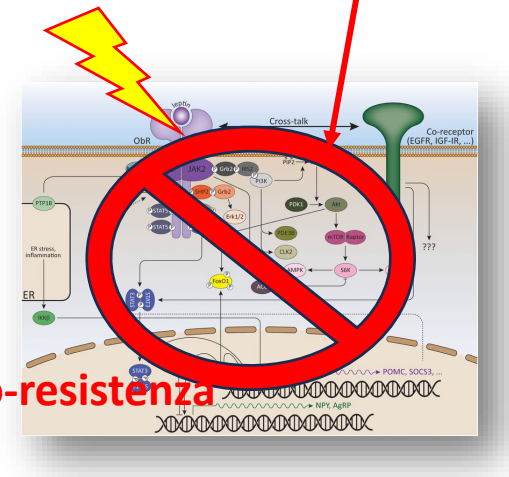
Descritta leptino-resistenza

Studiato ruolo di leptina in obesità



Tessuto adiposo





leptino-resistenza

Prodotto e Modificato da: https://doi.org/10.1016/j.appet.2022.105965

Obesity Science & Practice doi: 10.1002/osp4.62

doi: 10.1002/osp4.62

ORIGINAL ARTICLE

Amylin/leptin synergy is absent in extreme obesity and not restored by calorie restriction-induced weight loss in rats

J. L. Trevaskis†, C. Wittmer, J. Athanacio, P. S. Griffin, D. G. Parkes and J. D. Roth‡

doi:10.1038/ijo.2009.238

International Journal of Obesity (2010) 34, 385–395

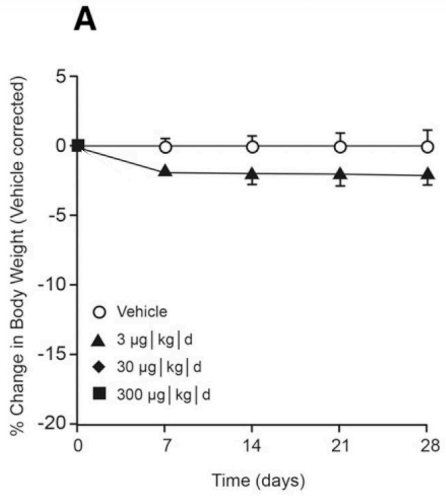
0307-0565/10 $32.00
www.nature.com/ijo

ORIGINAL ARTICLE

Davalintide (AC2307), a novel amylin-mimetic peptide: enhanced pharmacological properties over native amylin to reduce food intake and body weight

CM Mack, CJ Soares, JK Wilson, JR Athanacio, VF Turek, JL Trevaskis, JD Roth, PA Smith, B Gedulin, CM Jodka, BL Roland, SH Adams, A Lwin, J Herich, KD Laugero, C Vu, R Pittner, JR Paterniti Jr, M Hanley, S Ghosh and DG Parkes

DIO RATS



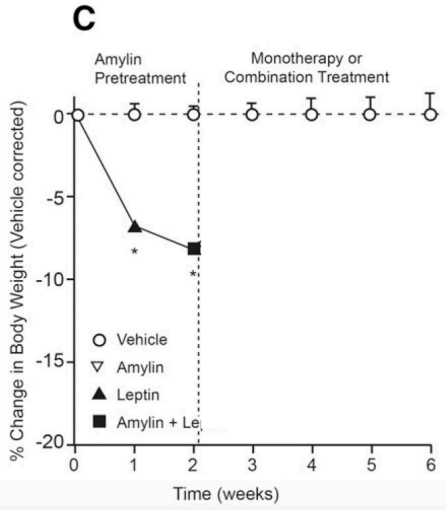
controllo

Amilina impiantata sc infusione continua 3 microgr/die

Amilina 30 microgr/die

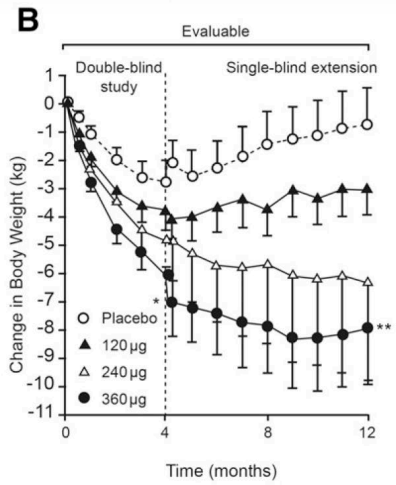
Amilina 300 microgr/die

EFFETTO POSITIVO dose dipendente



EFFETTO ADDITIVO
In co-presenza con Leptina

OBESE HUMANS



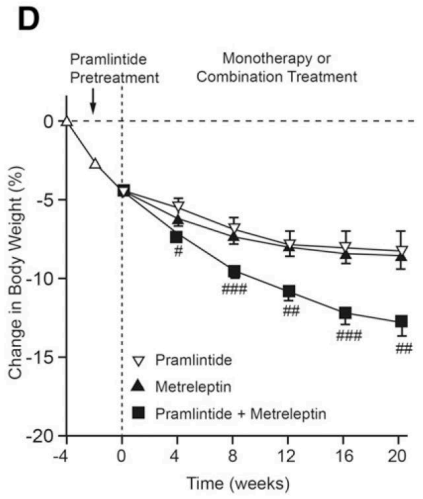
controllo

Amilina (analogo) impiantata sc infusione continua 120 microgr/die

Amilina (analogo) 240 microgr/die

Amilina (analogo) 360 microgr/die

EFFETTO POSITIVO dose dipendente

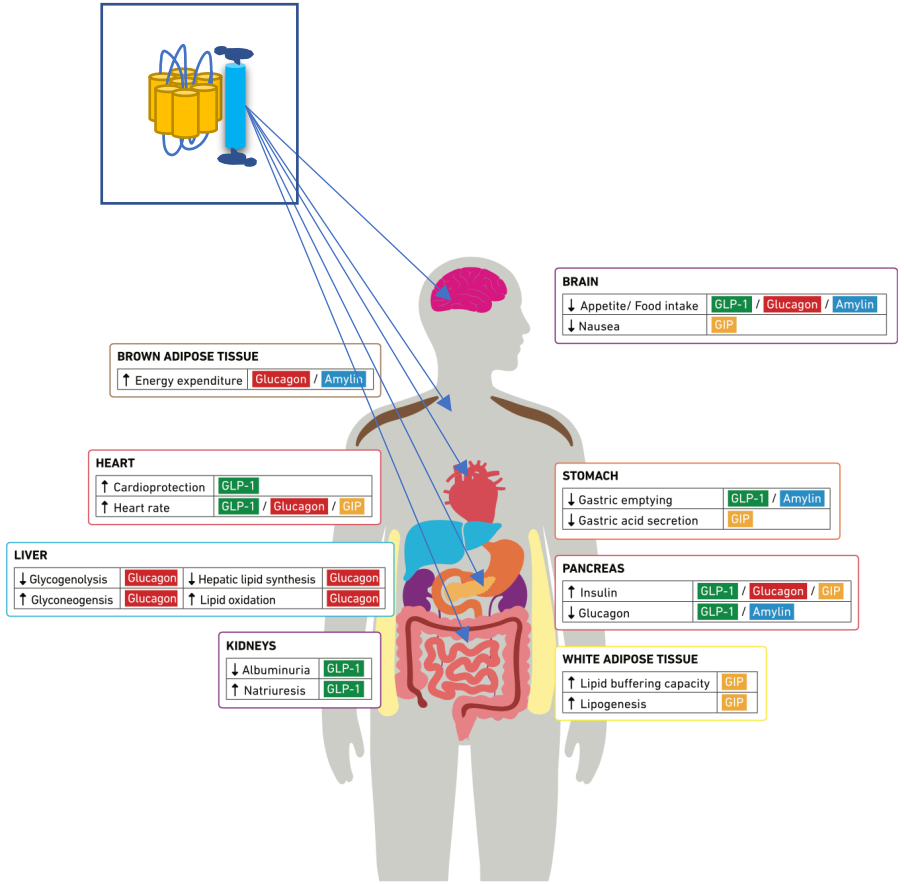


EFFETTO ADDITIVO
In co-presenza con Leptina

http://dx.doi.org/10.1124/pr.115.01062
Pharmacol Rev 67:564–600, July 2015

Il campo di studio dei recettori per AMILINA sarà molto attivo nei prossimi anni

Per aggiungere ulteriori elementi
ai benefici già noti



Modificato da:

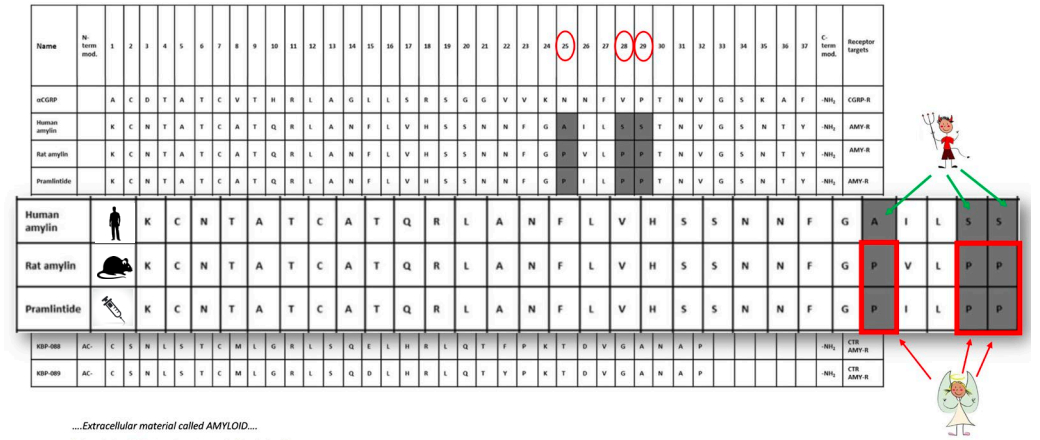
EXPERT OPINION ON PHARMACOTHERAPY
2024, VOL. 25, NO. 7, 801–818
https://doi.org/10.1080/14656566.2024.2356254

Che altro sappiamo di….

Effetto di AMILINA in vivo nell'uomo

Studi con *Pramlintide*

Sequenza aminoacidica di AMILINA e dei suoi analoghi



PRAMLINTIDE (approvato in USA per il trattamento di T1 e T2DM)

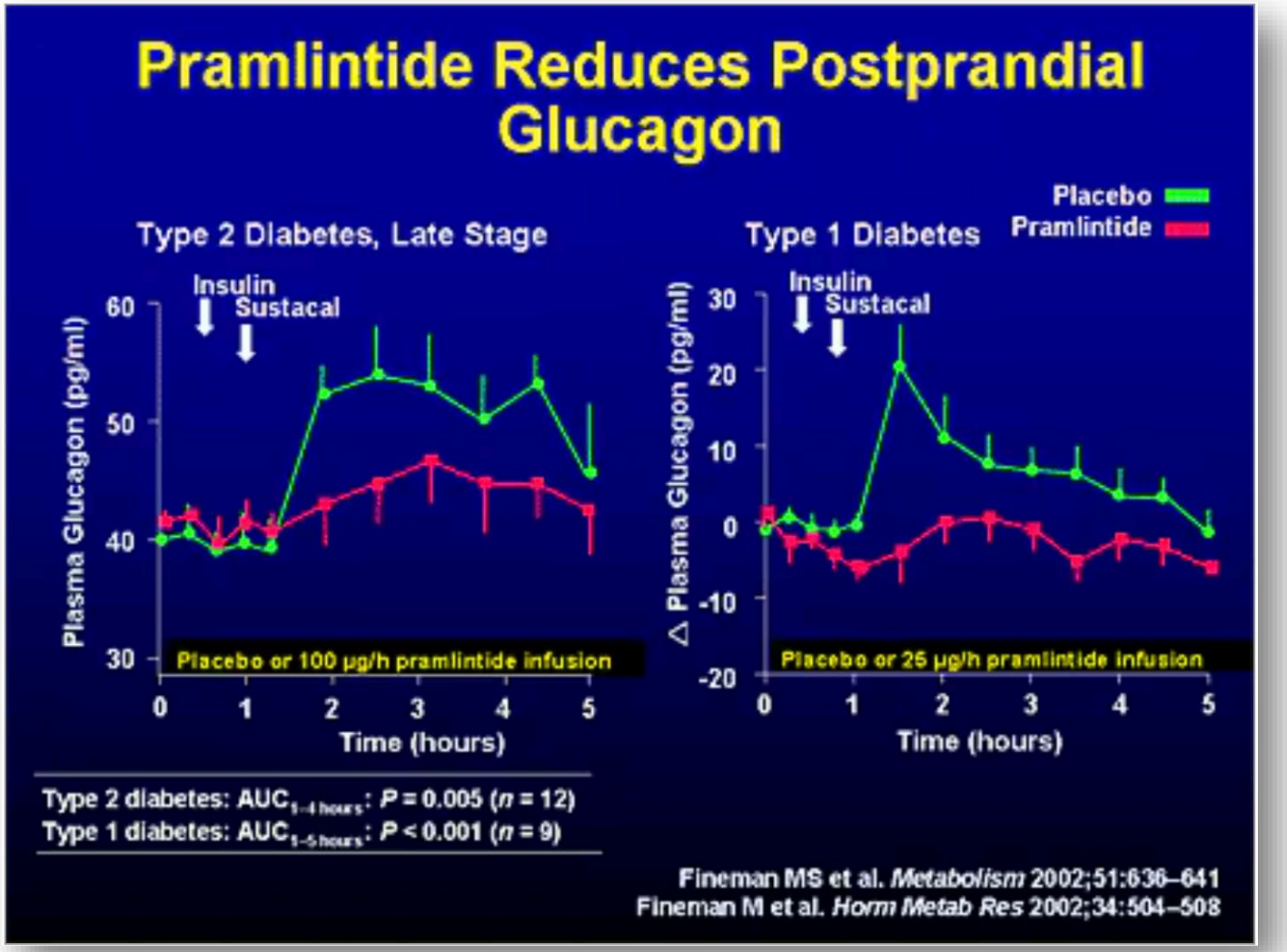
Pramlintide	K	C	N	T	A	T	C	A	T	Q	R	L	A	N	F	L	V	H	S	S	N	N	F	G	P	I	L	P	P

Perché?

- L'emivita plasmatico di **Amilina** è breve (minuti)
- La sequenza umana crea aggregati
- La differenza tra uomo e roditore rende difficile lo studio della fisiologia

Gli studi con PRAMLINTIDE hanno definito il reale significato clinico

Nel DMT2



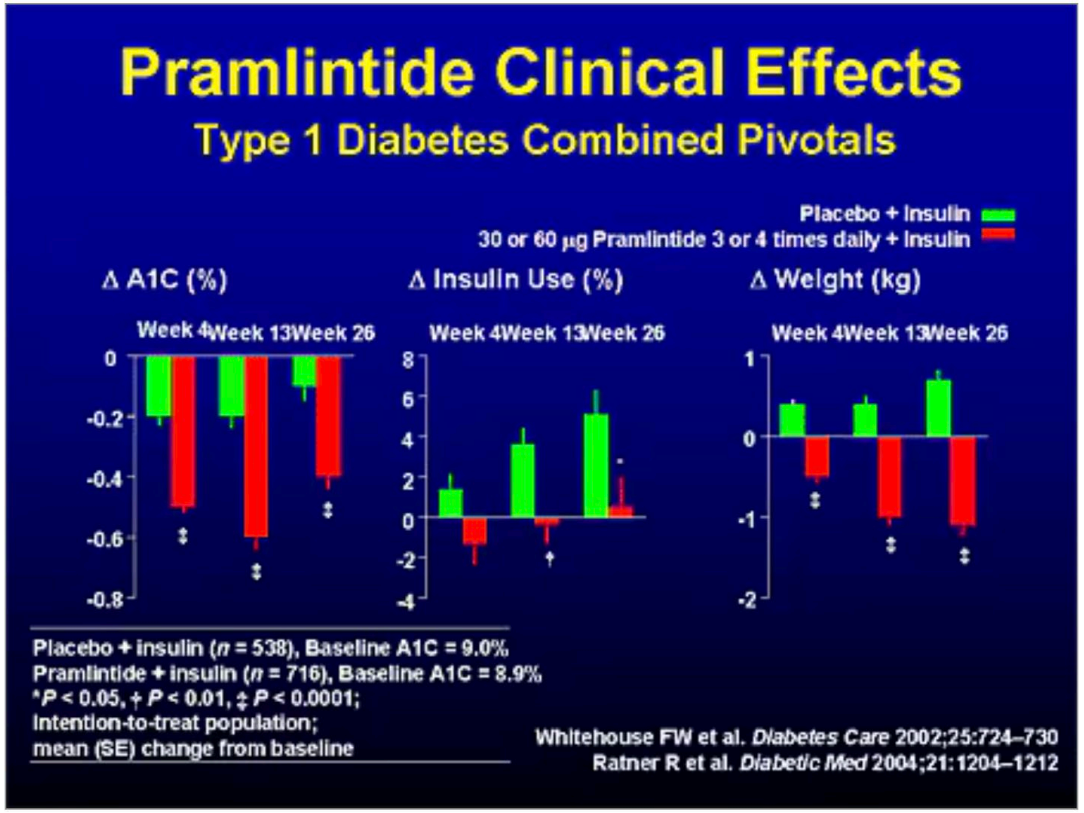
Nel DMT1

Dimostra l'importanza della **perdita** delle **cellule beta** e dei **granuli di insulina……**

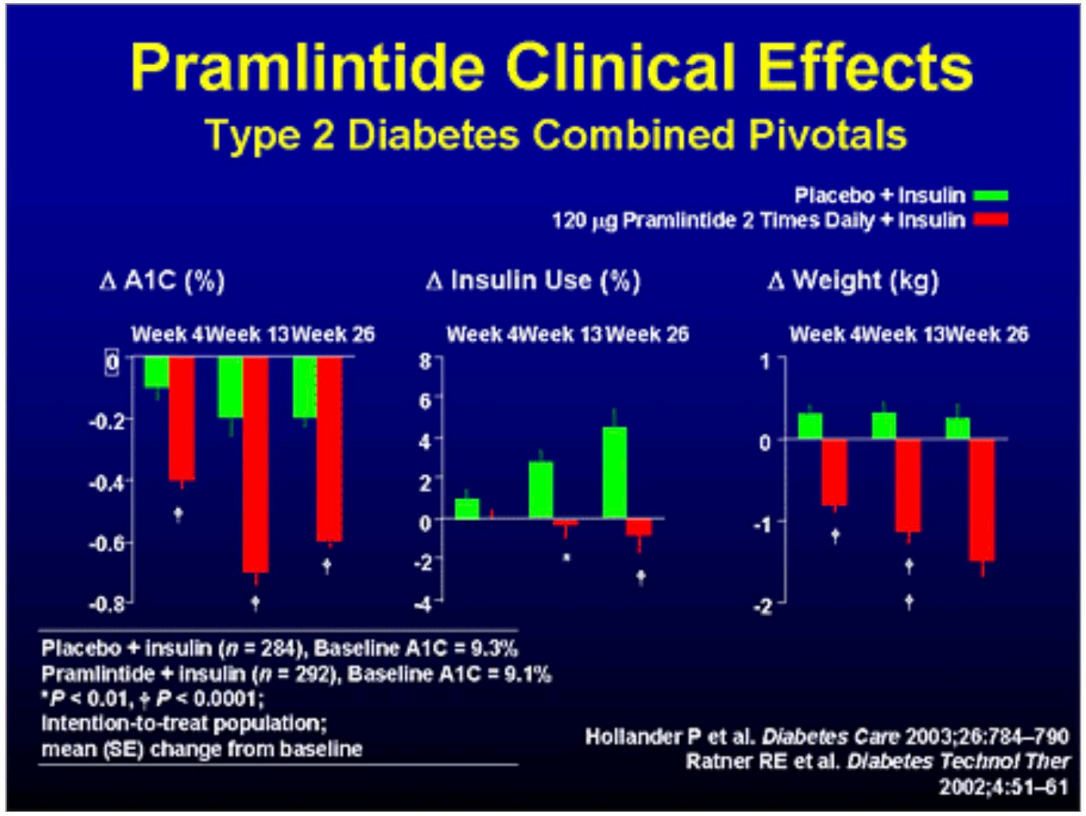
PRAMLINTIDE modula la secrezione di glucagone (anche in assenza di insulina)

Fonte: https://www.medscape.org/viewarticle/541782_5

Nel DMT1



Nel DMT2



PRAMLINTIDE modula compenso glicemico e peso corporeo

Somministrazione s.c. prima dei pasti principali (in aggiunta ad insulina basale)

Fonte: https://www.medscape.org/viewarticle/541782_9

Trials clinici con PRAMLINTIDE sia in T1DM che in T2DM

Authors	Research Design (Primary Goal, Duration)	Number of Patients	Results	Main Conclusions
Whitehouse et al., 2002 [70]	A double-blinded clinical trial with parallel assignment. Effects of pramlintide on HBA1c and weight. Duration: 52 weeks.	480 patients with **T1DM**	HBA1c was lower in patients with pramlintide (−0.39%) in comparison with placebo (−0.12%). The patients with pramlinitide had a weight loss (−0.5%) in contrast to placebo patients with weight gain (+1.0%).	Pramlintide has a positive effect on HBA1c and weight loss compared to placebo.
Ratner et al., 2000 [62]	The study was double-blinded with parallel assignment. Effects of pramlinitide on weight and HBA1c. Duration: 52 weeks.	538 pat… insulin-tre…		
Hollander et al., 2003 [63]	The study was double-blinded with parallel assignment. Pramlintide effects on HBA1c and weight. Duration: 52 weeks.	498 patient…		
Gottlieb et al., 1999 [71]	The study was double-blinded with parallel assignment. Effects of pramlintide on HBA1c and weight. Duration: 26 weeks.	499 patients with **T2DM**	dosage); on the contrary, placebo patients had a weight gain (+0.1%). HBA1c was lower in patients with pramlinitide (−0.3% to −0.4% depending on the dosage) in comparison with placebo (−0.1%).	Pramlintide has a positive effect on HBA1c and weight loss compared to the placebo.

Clin. Pharmacol. Adv. Appl. 2018, 10, 23–29.

Clinical Pharmacology: Advances and Applications

Dovepress

open access to scientific and medical research

Open Access Full Text Article

ORIGINAL RESEARCH

Pramlintide, an antidiabetic, is antineoplastic in colorectal cancer and synergizes with conventional chemotherapy

This article was published in the following Dove Press journal:
Clinical Pharmacology: Advances and Applications

Authors	Research Design (Primary Goal, Duration)	Number of Patients	Results	Main Conclusions
Karl D et al., 2007 [66]	Open-label study. Safety and efficacy of pramlintide therapy.	166 insulin-treated patients with **T2DM**	The change in HBA1c from baseline was −0.56%. Pramlintide therapy notably reduced weight (−2.8 kg) and postprandial glucose excursions.	Pramlintide initiation and mealtime insulin reduction led to weight loss. It also improved postprandial glucose excursions and HBA1c.
		…nts with **T1DM** …**T2DM** with …ate glycaemic …control	After 3 months, the incidence of patient-ascertained severe hypoglycemia (PASH) was 2.8% in patients with T2DM and 4.8% in patients with T1DM.	The possibility of insulin-induced severe hypoglycemia after pramlintide therapy initiation is low in patients with T2DM or T1DM.
		…nts with **T2DM** …ulin glargine in …pramlintide or …lacebo	Reductions in HBA1c (−0.70% against −0.36%) and postprandial glucose increase were more significant in pramlintide-treated patients. Pramlinitide-treated patients experienced weight loss (−1.6 kg), while placebo gained weight (+0.7 kg).	Pramlintide improved HBA1c and postprandial glucose with weight reduction in T2DM patients.
Peyrot M et al., 2010 [69]	effectiveness of basal insulin regimens with rapid-acting insulin or pramlintide. Duration: 9 months.	…nts with **T2DM** and basal insulin therapy in addition to pramlintide or rapid-acting insulin	Total diabetes distress in pramlintide patients improved significantly. On the other hand, patients with rapid-acting insulin did not. The perception of hypoglycemia was improved only in pramlintide patients.	Adding pramlintide to basal insulin treatment improved life quality and satisfaction compared with rapid-acting insulin analogs.

Riduzione peso (circa 1 %)

Migliore compenso glicemico (riduzione HbA1c circa 0,5%)

Effetti collaterali lievi (nausea, vomito, riduzione appetito)

Effetti benefici inattesi:

Potenziamento effetto citotossico in corso di chemioterapia in pazienti con K colon (evidenze da confermare)

Int. J. Mol. Sci. 2024, 25, 1517. https://doi.org/10.3390/ijms25031517

Trials clinici con analoghi in corso.....

Table 2. Summary of the studies in patients with diabesity treated with cagrilintide. T1DM—type 1 diabetes mellitus; T2DM—type 2 diabetes mellitus; HbA1C—hemoglobin A1C.

Authors	Research Design (Duration, Comparator)	Number of Patients	Results	Main Findings
Frias J P et al., 2023 [83]	Multicenter, double-blinded randomized study. The safety and effect of cagrilintide and semaglutide combination in **T2DM** patients. Duration: one year.	Patients were divided into three groups: 31 with CagriSema, 31 with semaglutide, and 30 with cagrilintide	CagriSema had a significant HbA1c reduction compared with cagrilintide (−1.3%) but not with semaglutide (−0.4%). CagriSema had better results regarding the change in body weight (−15.6%) in comparison with cagrilintide –(8.1%) and semaglutide (−5.1%).	CagriSema therapy led to better glycaemic control in T2DM patients. CagriSema had better results in HBA1c reduction than cagrilinitide but not semaglutide. CagriSema had the best results regarding weight loss.
NCT06065540 Study start: 2023 [84]	The study is randomized with parallel assignment. The effect of CagriSema, cagrilintide, semaglutide, and placebo on blood glucose and body weight) in **T2DM** patients treated with metformin.	Estimated enrollment of 2700 participants	Not available	Ongoing study
NCT05394519 Study start: 2023 [85]	The study is randomized with parallel assignment. The effect of CagriSema on weight loss in patients with excess body weight and **T2DM**.	Estimated enrollment of 1200 participants	Not available	Ongoing study
NCT06131372 Study start: 2024 [86]	Randomized with parallel assignment. Comparison between CagriSema, cagrilintide, semaglutide, and placebo regarding the renal damage in people with chronic kidney disease, **T2DM**, and **excess body weight**.	Estimated enrollment of 618 participants	Not available	Ongoing study
NCT05567796 Study start: 2022 [87]	The study is randomized with parallel assignment. Safety and effectiveness of CagriSema in patients with **excess body weight**.	Estimated enrollment of 3400 participants	Not available	Ongoing study

Cagrilintide
(analogo settimanale)

Int. J. Mol. Sci. 2024, 25, 1517. https://doi.org/10.3390/ijms25031517

Articles

Efficacy and safety of co-administered once-weekly cagrilintide 2·4 mg with once-weekly semaglutide 2·4 mg in type 2 diabetes: a multicentre, randomised, double-blind, active-controlled, phase 2 trial

Juan P Frias, Srikanth Deenadayalan, Lars Erichsen, Filip K Knop, Ildiko Lingvay, Stanislava Macura, Chantal Mathieu, Sue D Pedersen, Melanie Davies

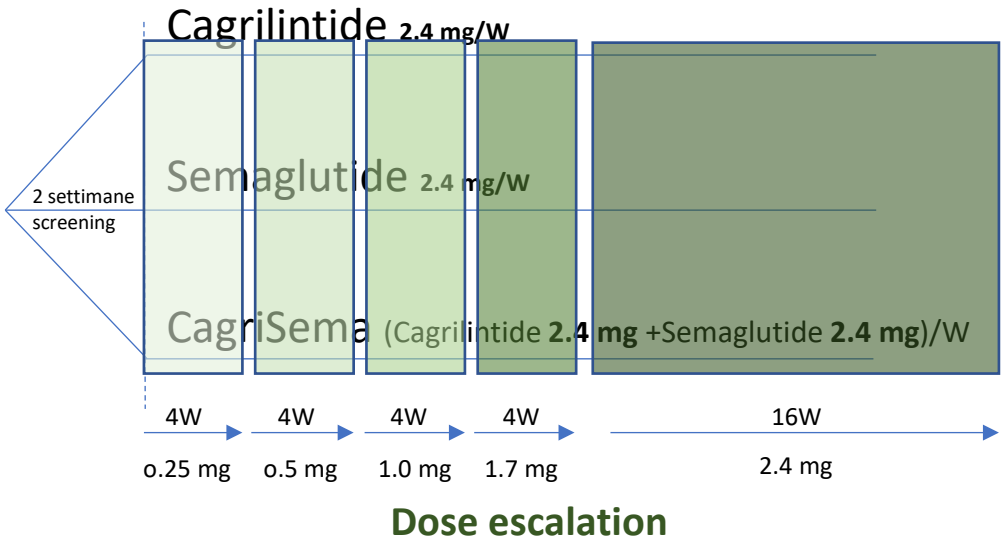
Lancet 2023; 402: 720–30
Published Online
June 23, 2023
https://doi.org/10.1016/
S0140-6736(23)01163-7

Studio di 32 settimane, doppio cieco, **fase 2**, multicentrico

T2DM

BMI >27 kg/m2

In terapia con metformina e/o SGLT2i

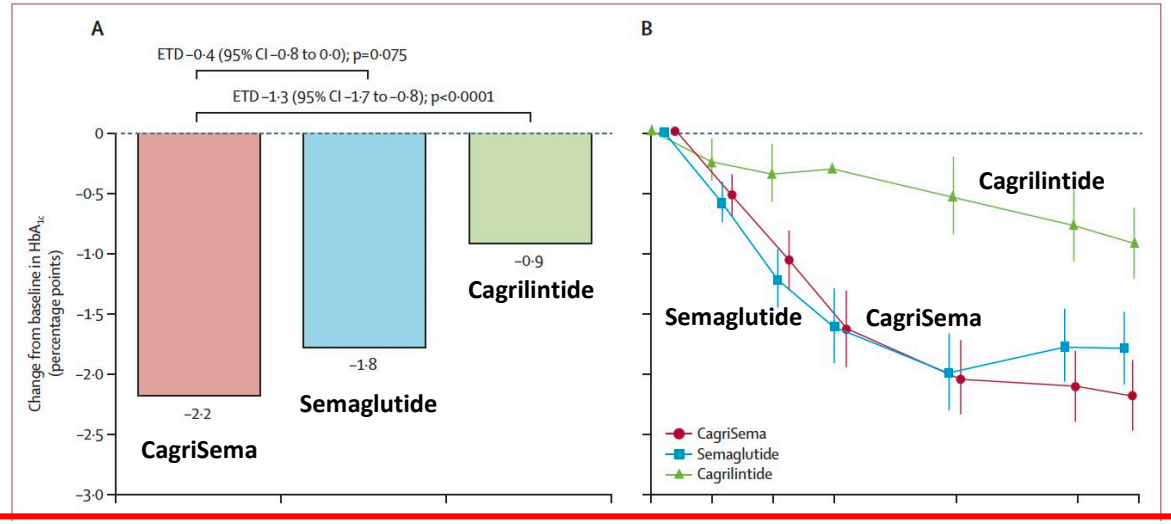


End-point primario:
Riduzione HbA1c

End-point secondario:
Riduzione peso
Miglioramento glicemia
Migliore profili glicemici

End-point primario:
Riduzione HbA1c

Non inferiore a Semaglutide



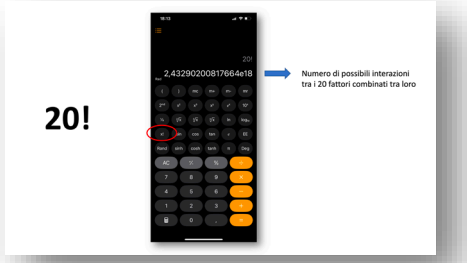
Lancet 2023; 402: 720–30
Published Online
June 23, 2023
https://doi.org/10.1016/
S0140-6736(23)01163-7

In sintesi

Ci sarebbero tante cose da raccontare ancora su AMILINA

AMILINA fa innamorare

AMILINA conferma la necessità
di comprendere il gioco di squadra necessario per gestire l'omeostasi



AMILINA ci ricorda che le interazioni sono molteplici e da scoprire

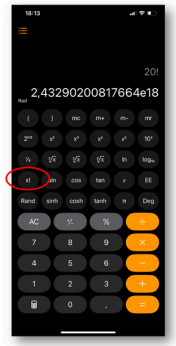
Almeno **20!**

In sintesi

“take-home messages”

1

20!



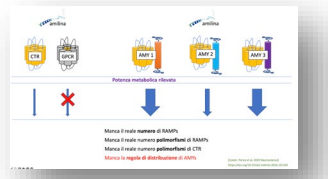
Numero di possibili interazioni tra i 20 fattori combinati tra loro

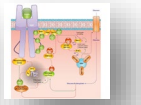
Anche se siamo lontani dalla verità

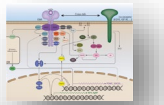
2

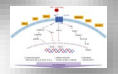
3

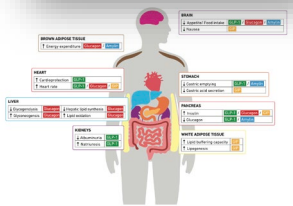
Studiare interazioni ormone-recettore-recettori per comprenderne il VERO significato









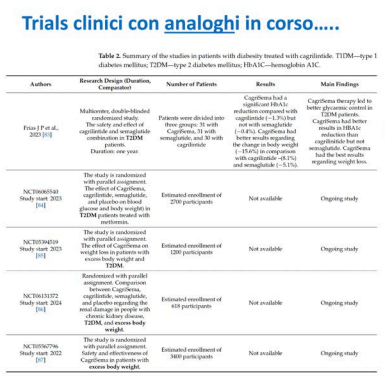


Il campo di studio dei recettori per AMILINA sarà molto attivo nei prossimi anni

4

L’interpretazione dei dati necessità di una visione complessiva integrata

Trials clinici con analoghi in corso.....



grazie

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