4年ぶりの同窓会


清水研
合同教室忘年会
2019

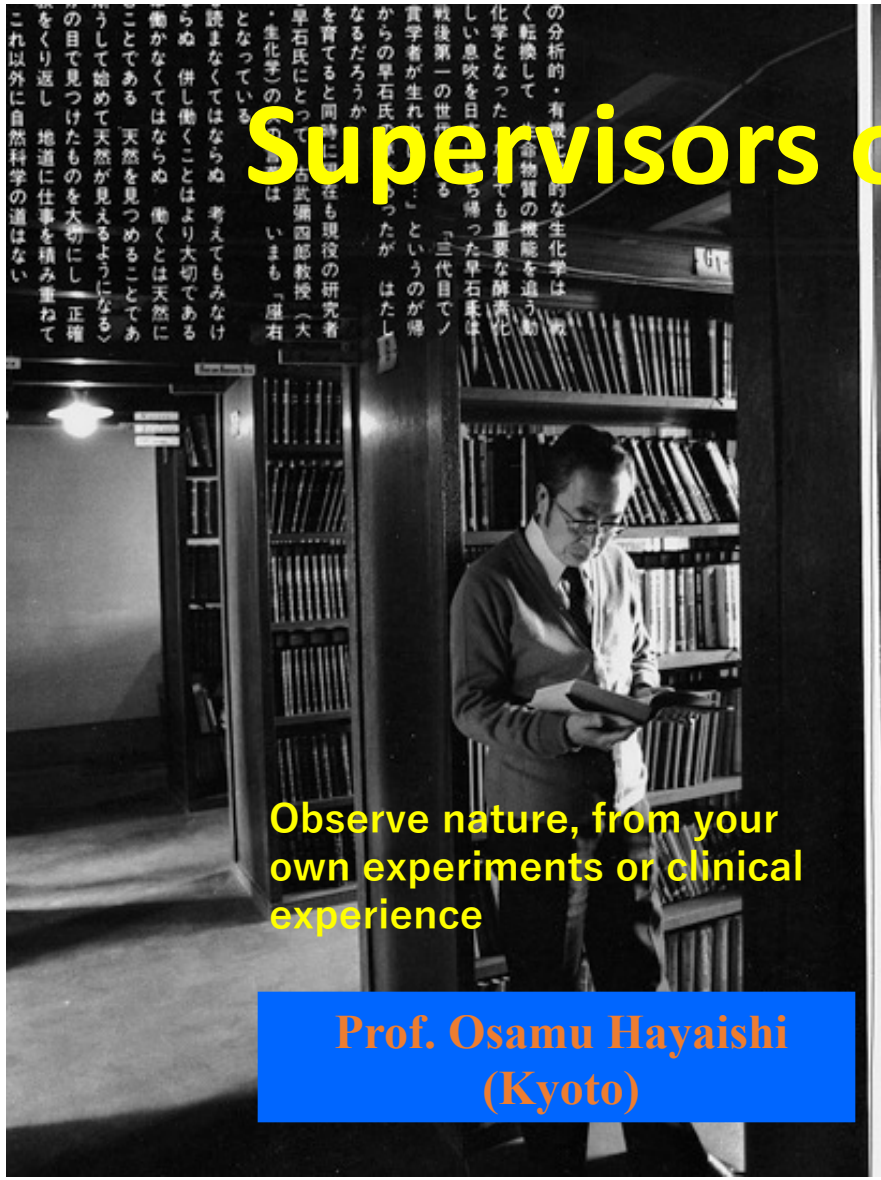

ARTham
Therapeutics
"Make Drug A Drug"


探創
新薬探索の基盤技術を創り、創薬を探求する

私の履歴・遍歴

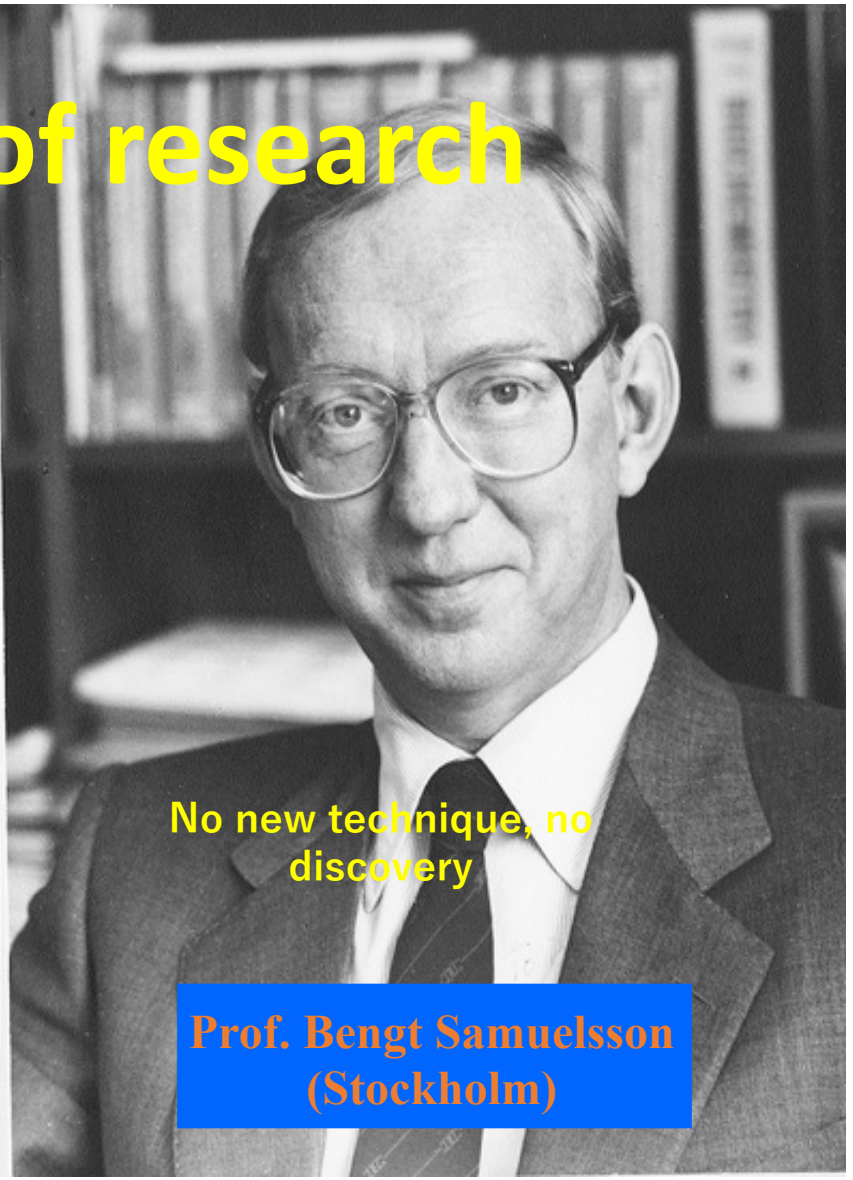
- 1973～1975　　東大病院、清瀬結核療養所　内科臨床医
- 1975～1982　　京大医化学ータンパク精製、有機化学
  トリプトファン代謝、プロスタグランディン代謝、有機合成
- 1982～1984（86）　カロリンスカ研究所
  5–リポキシゲナーゼ、ロイコトリエンA4水解酵素、有機合成
- 1984～1991　　東大栄養学助教授
  ロイコトリエン酵素cDNAクローニングから、受容体研究
- 1991～2012　　東大生化・分子生物（細胞情報）教授
  PAF受容体、ロイコトリエン受容体、LPA受容体、cPLA2研究、アシル転位酵素、リピドミクス
- 2012～2022　　国立国際医療研究センター研究所
  アシル転位酵素、マウス、ヒトサンプル
- 2022～？　　微生物化学研究所

Supervisors of research



Observe nature, from your own experiments or clinical experience

Prof. Osamu Hayaishi (Kyoto)



No new technique, no discovery

Prof. Bengt Samuelsson (Stockholm)

THE JOURNAL OF BIOLOGICAL CHEMISTRY
Vol. 253, No. 13, Issue of July 10, pp. 4700–4706, 1978
Printed in U.S.A.

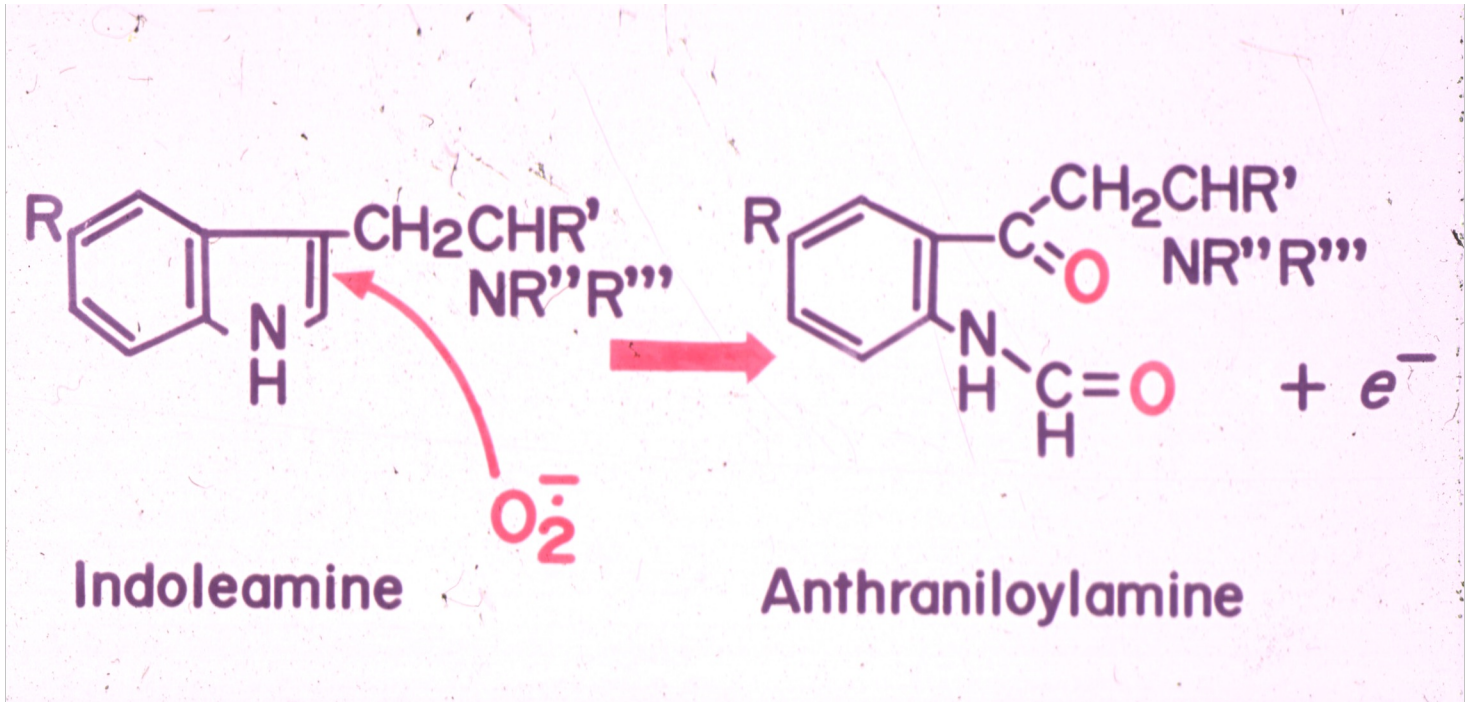
Indoleamine 2,3-Dioxygenase

PURIFICATION AND SOME PROPERTIES*

(Received for publication, January 24, 1978)

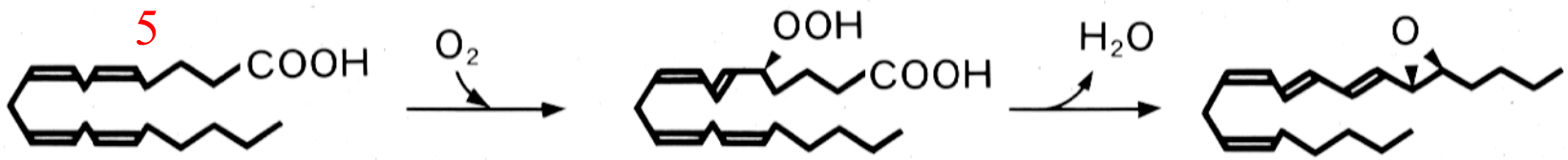
Takao Shimizu, Sumitsugu Nomiyama,‡ Fusao Hirata, and Osamu Hayaishi

From the Department of Medical Chemistry, Kyoto University Faculty of Medicine, Kyoto 606, Japan



アラキドン酸５−リポキシゲナーゼ

Dual activities, Ca requirement, membrane translocation (800 x g ppt)



Arachidonic acid **5-HPETE** **LTA4**

Proc. Natl. Acad. Sci. USA
Vol. 81, pp. 689–693, February 1984
Biochemistry

Enzyme with dual lipoxygenase activities catalyzes leukotriene A_4 synthesis from arachidonic acid

(potato lipoxygenase/bishomo-γ-linolenic acid/8-lipoxygenase/*D*-hydrogen/5-hydroperoxyicosatetraenoic acid)

TAKAO SHIMIZU*, OLOF RÅDMARK, AND BENGT SAMUELSSON†

Department of Physiological Chemistry, Karolinska Institutet, S-104 01 Stockholm, Sweden

Purification of a mammalian 5-lipoxygenase from rat basophilic leukemia cells

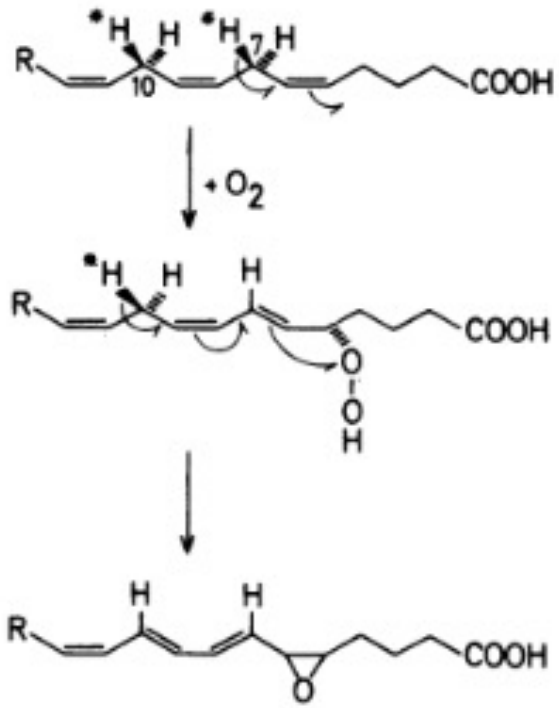
Andrew M. Goetze, Liz Fayer, Jennifer Bouska, DirkBornemeier and George W. Carter
Immunosciences Research, D-47K, Abbott Laboratories, North Chicago, IL 60064, U.S.A.

Abstract

5-Lipoxygenase (5-lipox) has been purified to homogeneity from the 20,000 xg supernatant of sonicated rat basophilic leukemia (RBL-1) cells using a 4-step procedure. Purification was achieved primarily through the use of anion-exchange HPLC on two different media. Using the supernatant from 1 × 10 9 cells, approximately 33 μg of the enzyme can be routinely isolated with an estimated net yield of 5–10%. Purified 5-lipox consists of a single Mr 73,000 band on SDS gels (reduced or unreduced). When the purified enzyme was incubated with radiolabeled arachidonic acid and products analyzed by both straight phase and reversed phase HPLC, **5-hydroperoxyeicosate-traenoic acid (5-HPETE) was the only enzymatic product detected**.

Prostaglandins 29 689–701. 1985

A proposed mechanism for LTA4 formation by 5-lipoxygenase



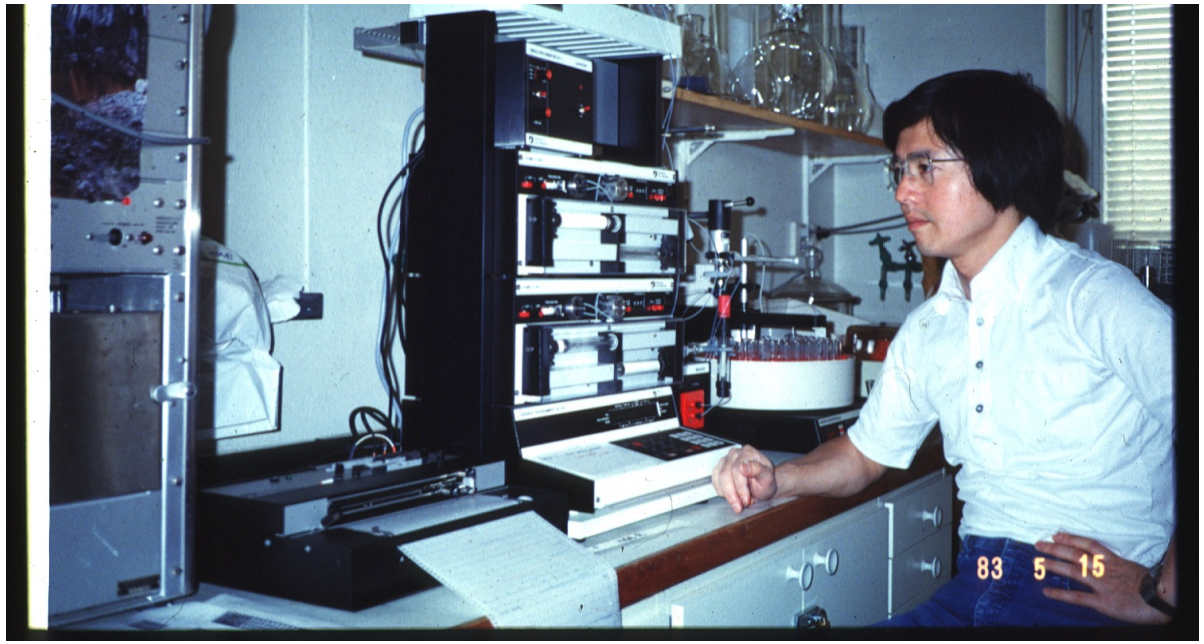
Proc. Natl. Acad. Sci. USA
Vol. 83, pp. 4175–4179, June 1986
Biochemistry

Characterization of leukotriene A_4 synthase from murine mast cells: Evidence for its identity to arachidonate 5-lipoxygenase

TAKAO SHIMIZU*, TAKASHI IZUMI*†, YOUSUKE SEYAMA*, KENJI TADOKORO‡, OLOF RÅDMARK§, AND BENGT SAMUELSSON§

*Department of Physiological Chemistry and Nutrition and ‡Department of Medicine and Physical Therapy, Faculty of Medicine, University of Tokyo, Bunkyo-ku, Hongo, Tokyo 113, Japan; §Department of Physiological Chemistry, Karolinska Institutet, S-104 01 Stockholm, Sweden; and †Third Department of Internal Medicine, Faculty of Medicine, University of Tokyo, Bunkyo-ku, Hongo, Tokyo 113, Japan

FIG. 5. Proposed reaction mechanism for the formation of LTA_4 from arachidonic acid by successive elimination of *D*-hydrogens at C-7 and C-10. R is $—CH_2—CH=CH—(CH_2)_4—CH_3$.


83 5 15

生化学講座９０年代

Receptors

PAF receptor, *Nature* 1991, *Neuron*, 1992

LTB4 receptors (BLT1, 2), *Nature* 1997,

J. Exp. Med., 2000, *J. Exp. Med*, 2005;

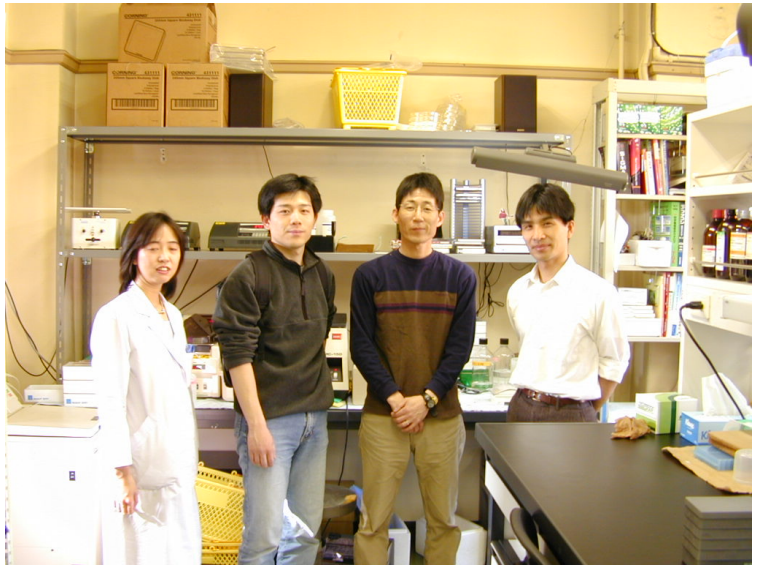
Nature CB, 2018

Non-edg LPA4 and LPA6, *JBC*,2003, *JBC*,2009

JCI Insight, 2018; *Human Mol. Genetics*, 2022

Z. Honda, I. Miki
(Firenze, 1991)

S. Ishii, K. Yanagida, K. Noguchi
Snowmass, 2005



T. Yokomizo group

Expression cloning of PAF receptor
the 1st example of lipid GPCRs cloning

Cloning by functional expression of platelet-activating factor receptor from guinea-pig lung

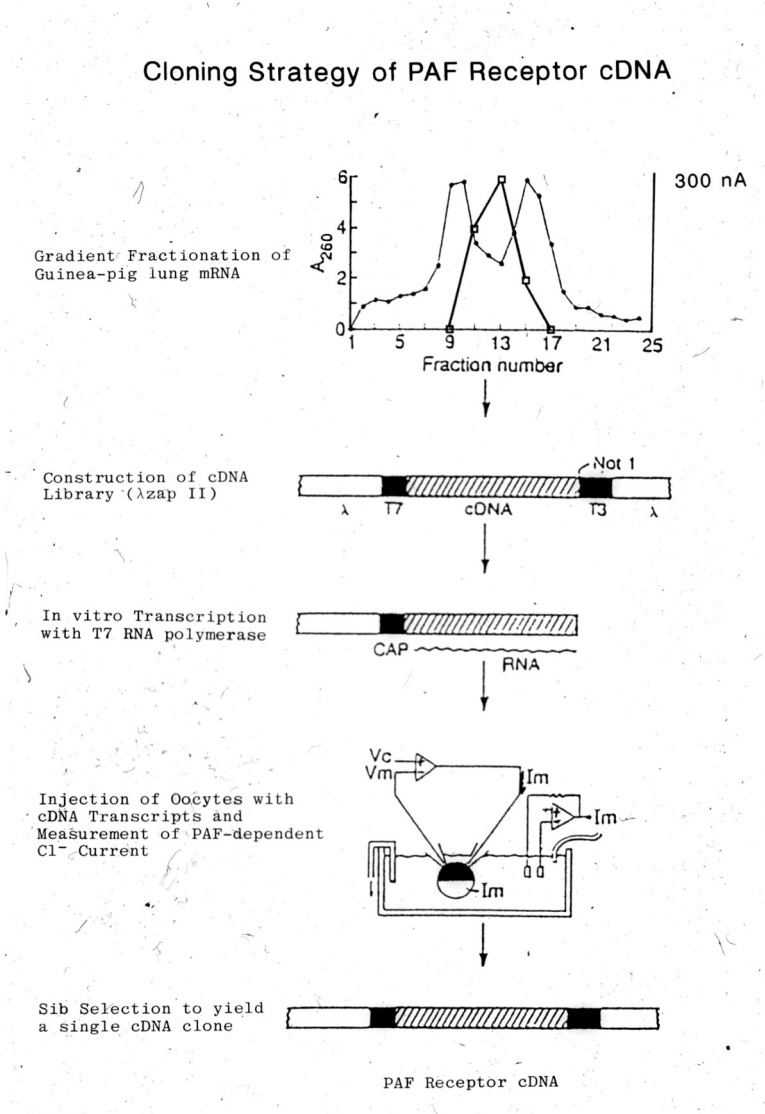
Zen-ichiro Honda*†, Motonao Nakamura*, Ichiro Miki*, Michiko Minami*, Tsuyoshi Watanabe*, Yousuke Seyama*, Haruo Okado‡, Hiroyuki Toh§, Kohji Ito‖, Terumasa Miyamoto‖ & Takao Shimizu*¶

* Department of Physiological Chemistry and Nutrition,
‡ Department of Neurobiology, Institute of Brain Research, and
‖ Department of Internal Medicine and Physical Therapy,
Faculty of Medicine, University of Tokyo, 7-3-1 Hongo, Bunkyo-ku, Tokyo 113, Japan
§ Protein Engineering Research Institute, 6-2-3 Furuedai, Suita, Osaka 565, Japan

PLATELET-activating factor (PAF), a unique phospholipid mediator, possesses potent proinflammatory, smooth-muscle contractile and hypotensive activities, and appears to be crucial in the pathogenesis of bronchial asthma and in the lethality of endotoxin and anaphylactic shock[1–3]. Despite this, little is known of the molecular properties of the PAF receptor and related signal transduction systems. Although several lines of evidence suggest that activation of the PAF receptor stimulates phospholipase C and subsequent inositol trisphosphate formation through G protein(s)[4,5], the PAF receptor and calcium channel are reported to show a close relation[2,6]. As a first approach to cloning lipid autacoid

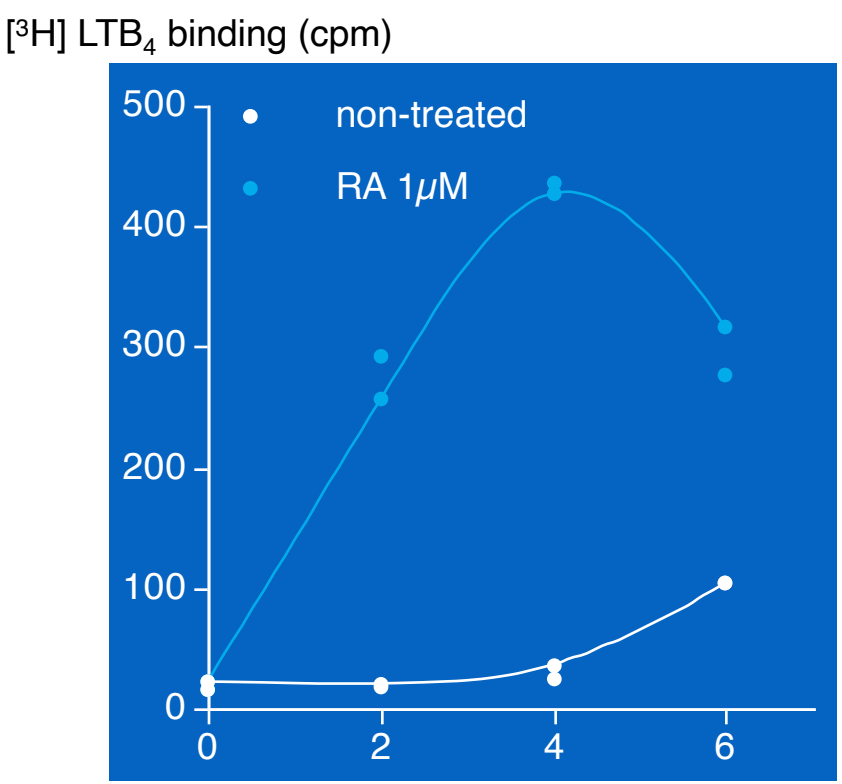
† On leave from Department of Internal Medicine and Physical Therapy, Faculty of Medicine, University of Tokyo, Japan.
¶ To whom correspondence should be addressed.

NATURE · VOL 349 · 24 JANUARY 1991

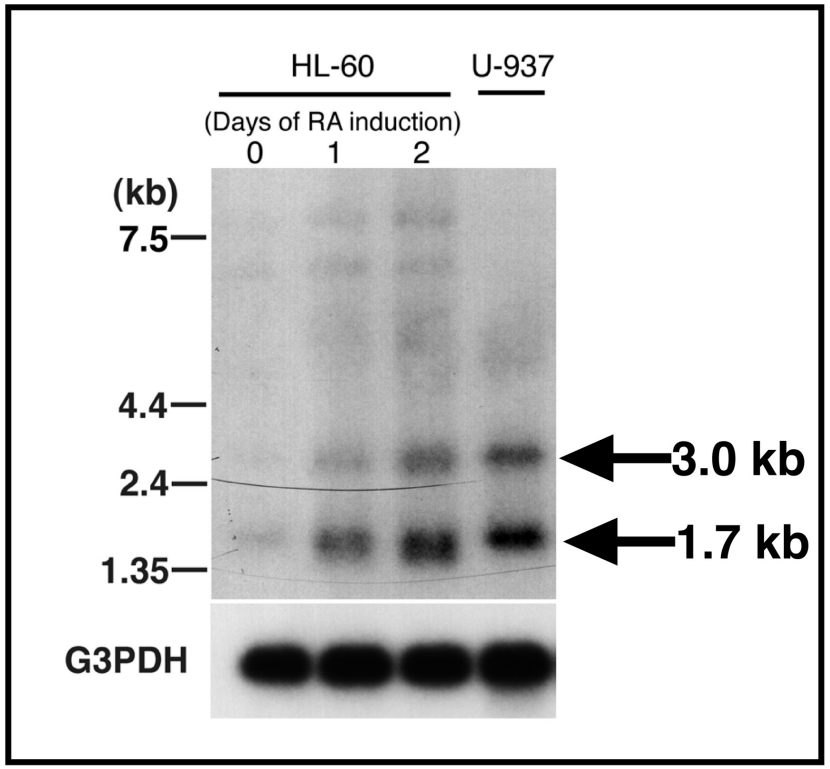


Induction of BLT in HL-60 cells —Subtractive PCR by T. Yokomizo

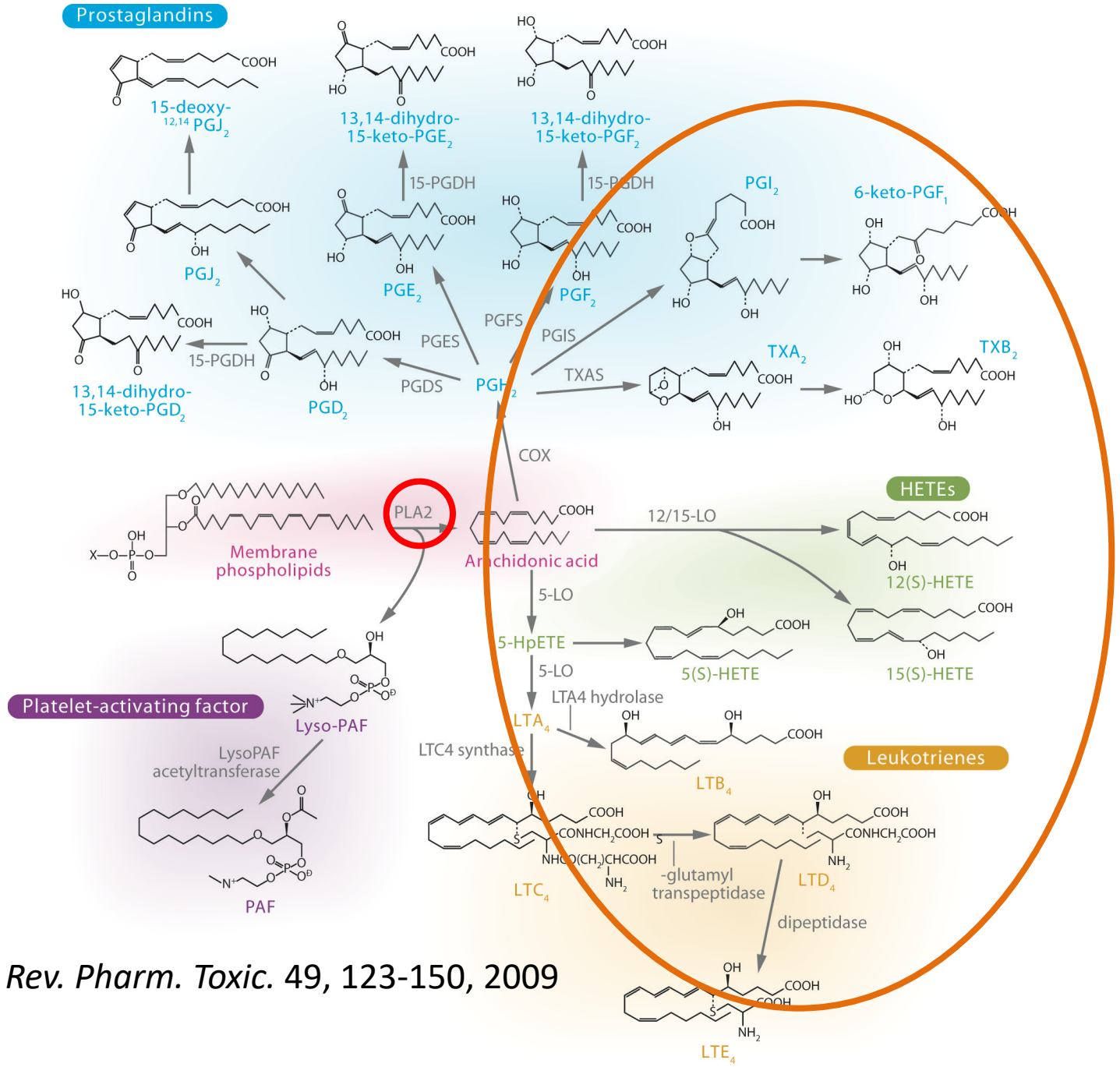
[^{3}H] LTB_4 binding



Northern Blot



Nature, 1997; J. Exp. Med. 2000



Shimizu, T. *Annu. Rev. Pharm. Toxic.* 49, 123-150, 2009

Phenotypes of cPLA$_2\alpha$ (-/-) mice

1. Reduced symptom of bronchial asthma (*Nature*,1997).
2. Decreased mortality & symptoms of ARDS (*Nature Immunol*, 2001).
3. Milder symptoms in bleomycin-induced fibrosis (*Nature Med*, 2002)
4. Reduced mortality due to thromboembolism (*J. Exp. Med*, 2002, *Blood* 2009)
5. Marked reduction of collagen-induced arthritis (*J. Exp. Med,* 2003).
6. Milder symptoms in inflammatory bone resorption (*J. Exp. Med,* 2003)
7. Milder symptoms in allergic encephalomyelitis (*J. Exp. Med* 2005, *PNAS,* 2010)
8. Prevention from atherosclerosis (*Amer. J. Physiol.* 2012)
9. *Impairment of synaptic plasticity* (*PNAS*, 2010)

Most of phenotypes are explained by the deficiency of downstream lipid mediators

Kume, Uozumi, Kita

- Phospholipase A2 meeting


Medical Tribune
MMJ

Failure of cPLA2α inhibitor for clinical use

- Collaboration with A-company for 12 years
- Screening out a potent and selective inhibitor (20 mg per day, po, good PK and PD)
- Target, rheumatoid arthritis, bronchial asthma, osteoporosis (all from animal data)
- Stop development in 2014, because of adverse effects at high doses during phase III clinical trial in US

Turning point in 2003

a year of whole human genome was sequenced,
and 10 years before my formal retirement

- **How arachidonate is located at *sn*-2 position, and how membrane diversity is made?**

- **Need development of comprehensive lipidomics techniques**

(Ono Pharmaceutical and Shimadzu supported establishment of a metabolome laboratory at U-Tokyo.)

ノーベル
た田中耕
かす寄付
、東京大
細胞内の
う物質を
技術を駆
命活動の
気の原因
。
品工業が
億円を出
系研究科

田中氏の研究成果生かせ

島津、東大に寄付講座

に設置する。名古屋市立大の田口良・助教授が寄付講座を率いる客員教授に就任する。

研究の対象は細胞内にあるアミノ酸や脂質、糖質など代謝産物と呼ばれる物質。これらは生命活動に必要な物質やエネルギーを作るとともに、病気の発症にも関係しているとみられ、代謝産物を網羅的に解析するメタボ

2002.10.7　日経新聞

小野薬品と島津製作所に篤く御礼

2005~2010

清水孝雄先生

How does the intellectual address authority:
as a professional supplicant or as its unrewarded
amateurish conscience?
—Ftr. Said
二〇〇八・九・二八 大江健三郎

「伝える言葉」プラス

大江健三郎

朝日新聞社

「伝える言葉」プラス

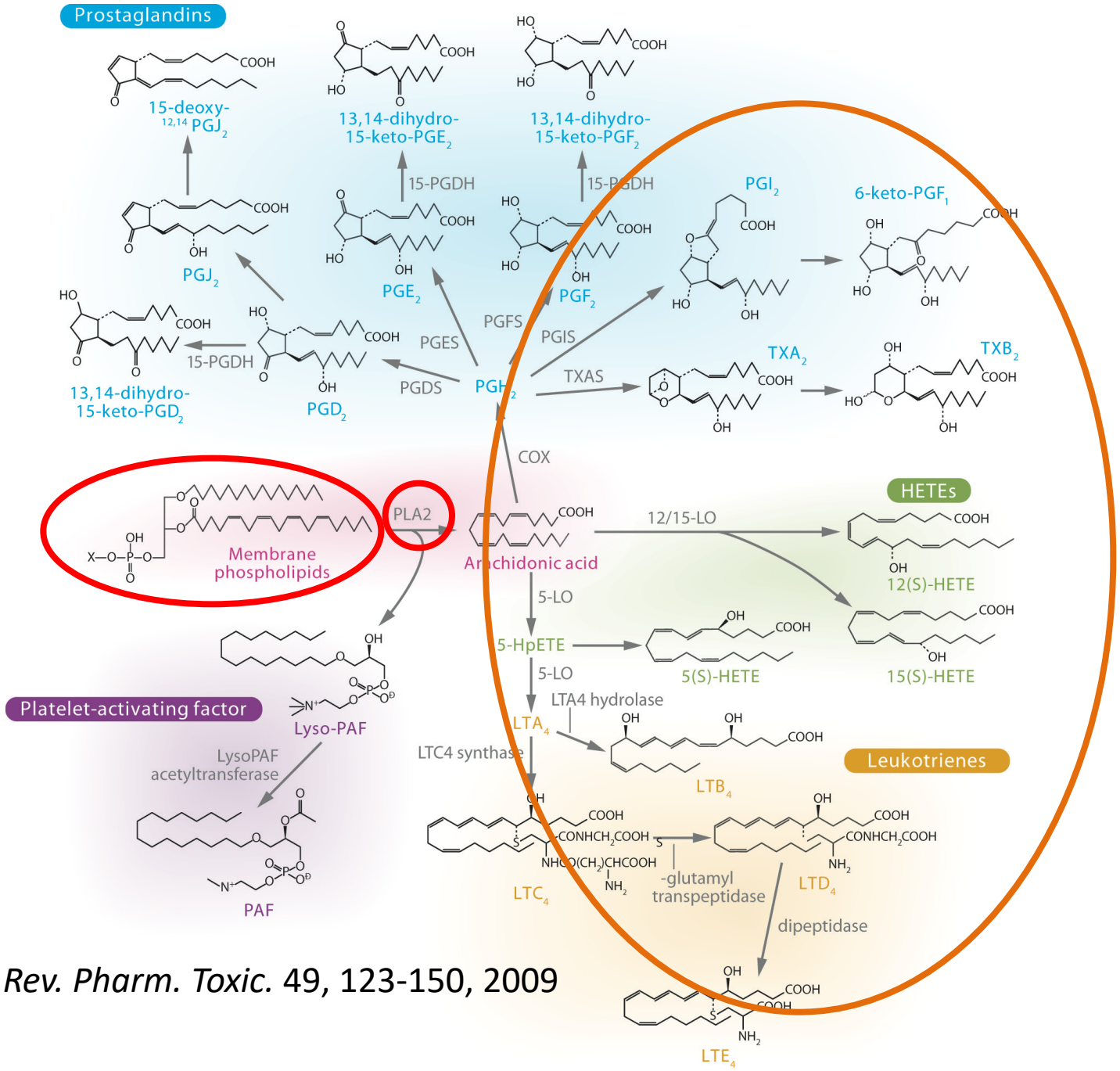
大江健三郎

人生の困難な折々に、本当に出会った27の奥深い言葉

著者自身の家族の歴史「苦しみと良いこと」が
繰り返される不思議とともに語る、
感銘と励ましに満ちたエッセイ集

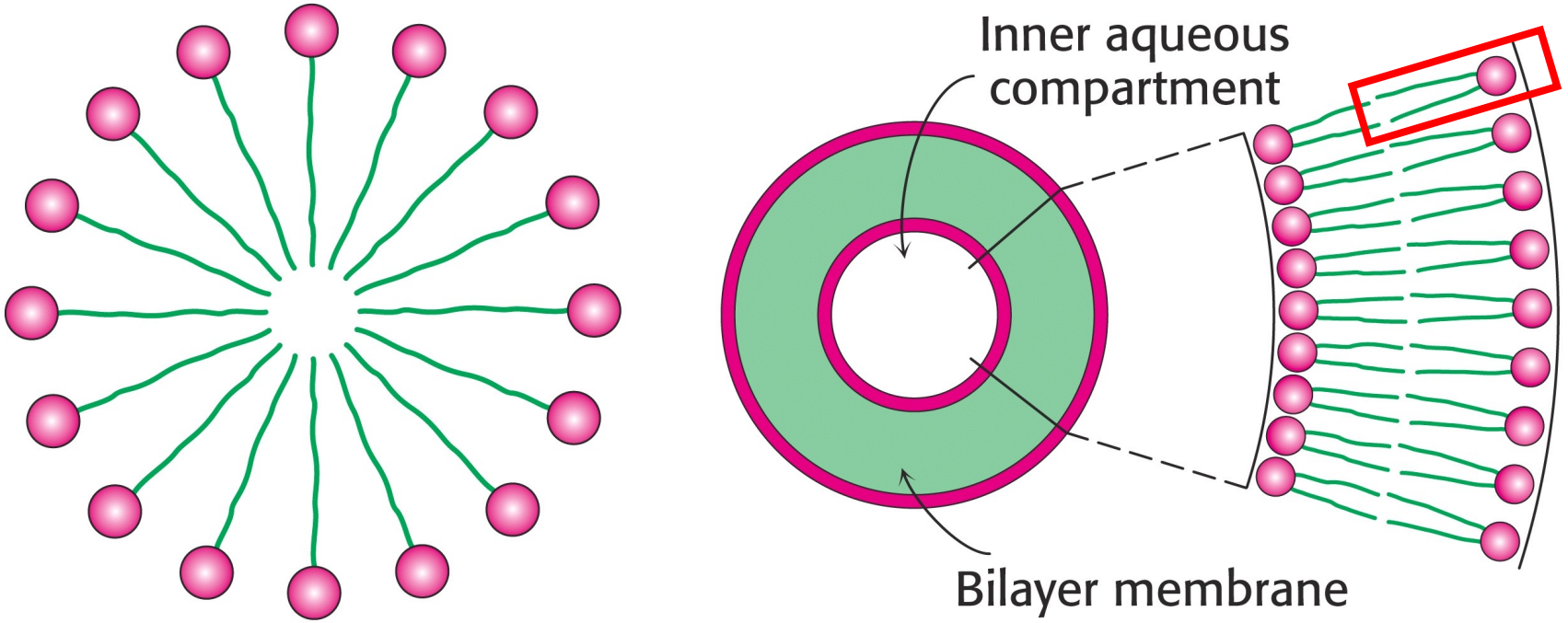
朝日新聞社

Reunion after March 11 Great East Japan Earthquake (2011.4.7)



Shimizu, T. *Annu. Rev. Pharm. Toxic.* 49, 123-150, 2009

Amphipathic properties of glycerophospholipids useful for lipid bilayer



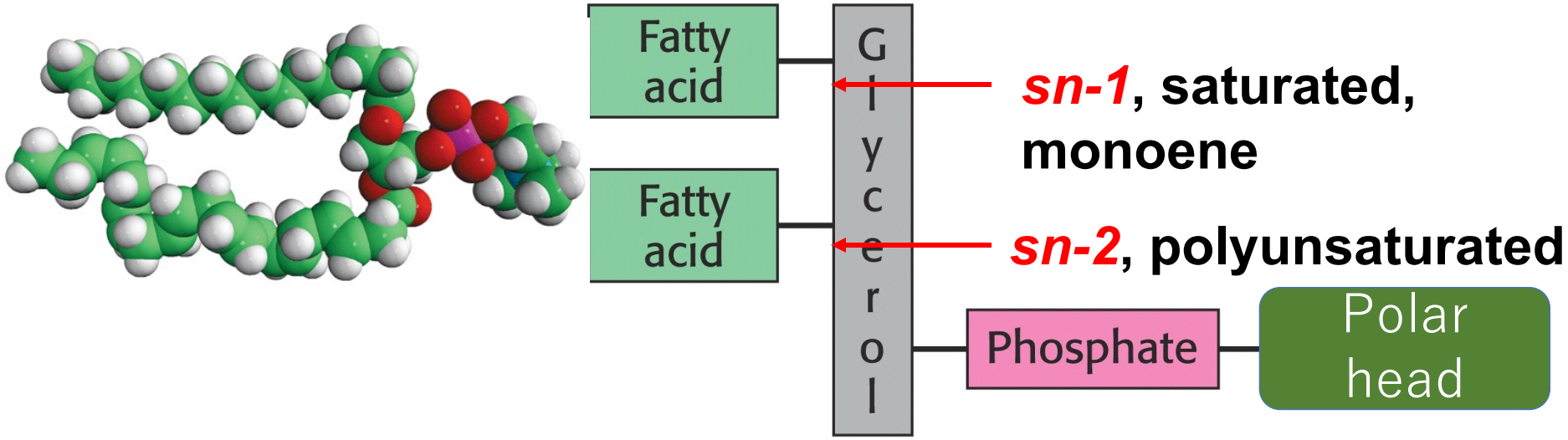
Micelle

Liposome

Stryer, 8th ed

Essential for cell structure in aqueous environments

Fatty acyl diversity and asymmetry of glycerophospholipids



- *How? Biochemical mechanisms*
- *So what? Biological consequence*

Acyltransferase Family

(Diacylglycerol acyltransferase 2)

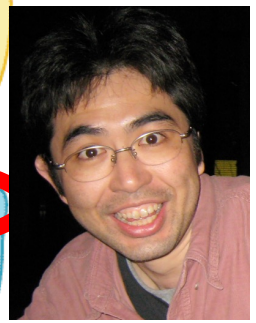
DGAT2 family

DGAT2
DGAT2 Like3
DGAT2 Like4
DGAT2 Like6
MGAT1
MGAT2

MBOAT family

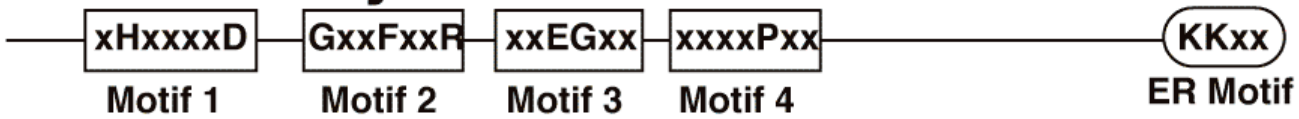
(membrane bound O-acyltransferase)

Porcupine-a
GOAT
HHAT
LPCAT3
LPCAT4
LPEAT1
ACAT2
ACAT1
DGAT1

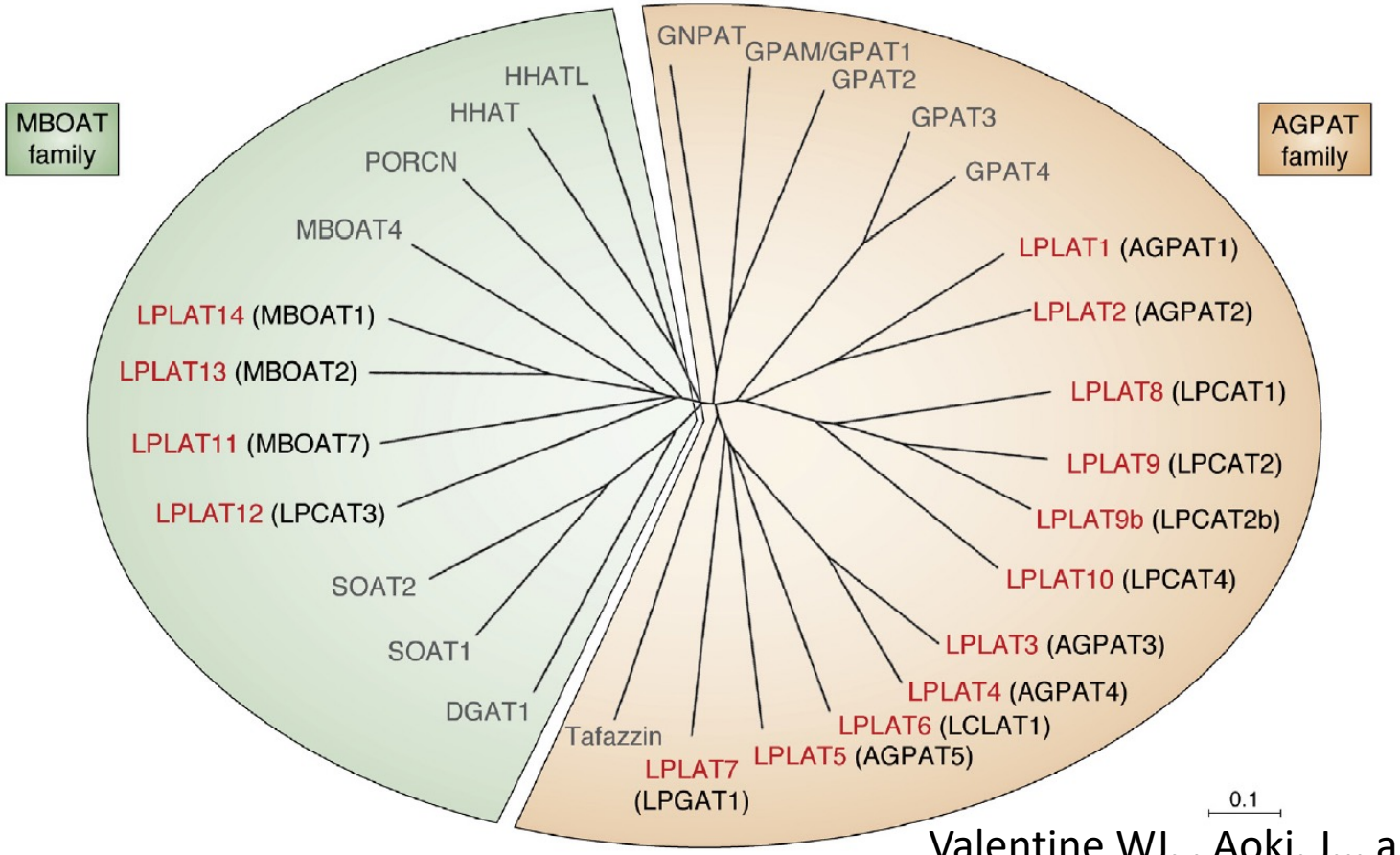


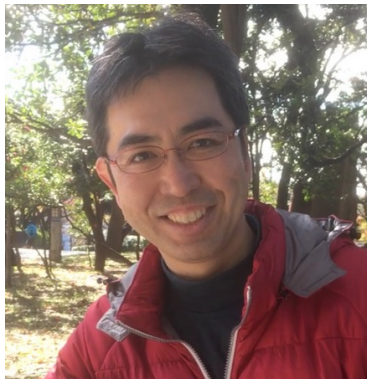
LPAAT1
LPAAT2
LPGAT1
AGPAT5
GPAT4
GPAT3
LCLAT1
AGPAT4
LPCAT2
LPAAT3
LPCAT1
GPAT2
LPEAT2

AGPAT family



Update of nomenclature proposal for LPL acyltransferases





Valentine WJ, , Aoki, J.,, and Shindou, H *JBC*, 2021

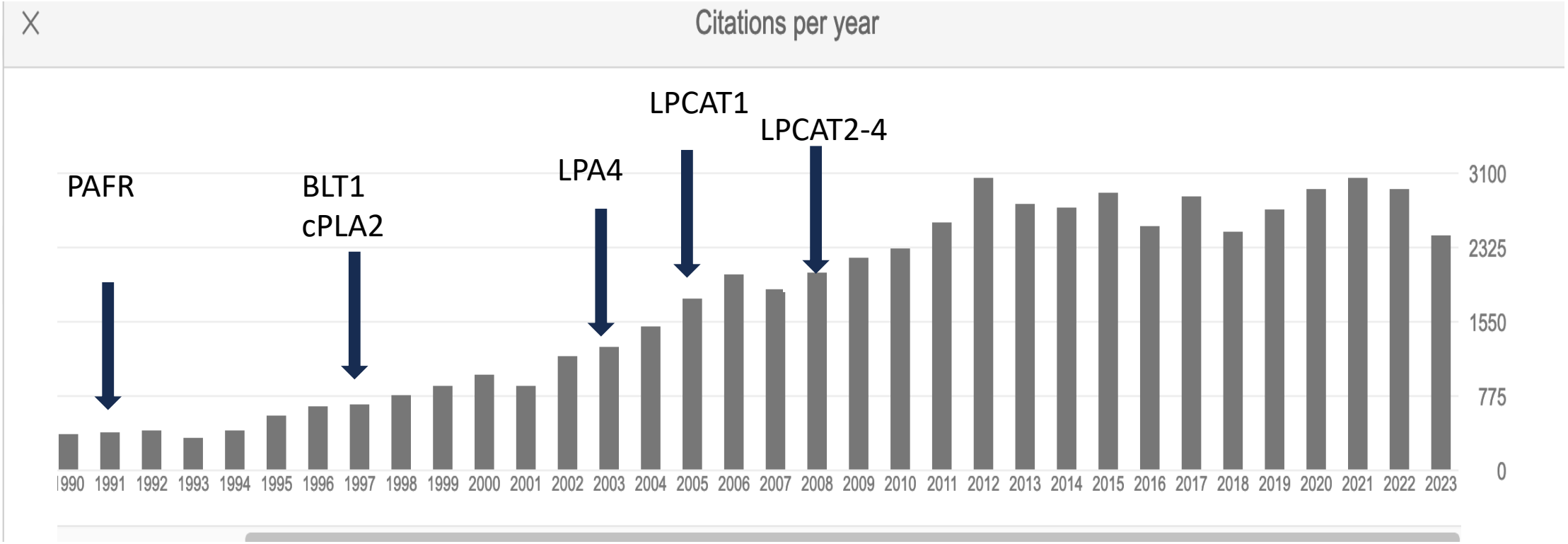
Conversations with Lipid Leaders: Dr. Bruno Antonny

Posted on September 02, 2021

What do you consider the greatest breakthrough in lipid research in recent years?

The discovery of lipid remodeling enzymes, notably by the Shimizu lab in Japan because it opens an avenue for understanding how and why cells in real tissues control so well the acyl chain profiles their organelles. Classical cell lines used by cell biologists are very rudimentary in this respect.

Scientific activity



h-index, 119. from Google Scholar

国立国際医療研究センター (2012~)

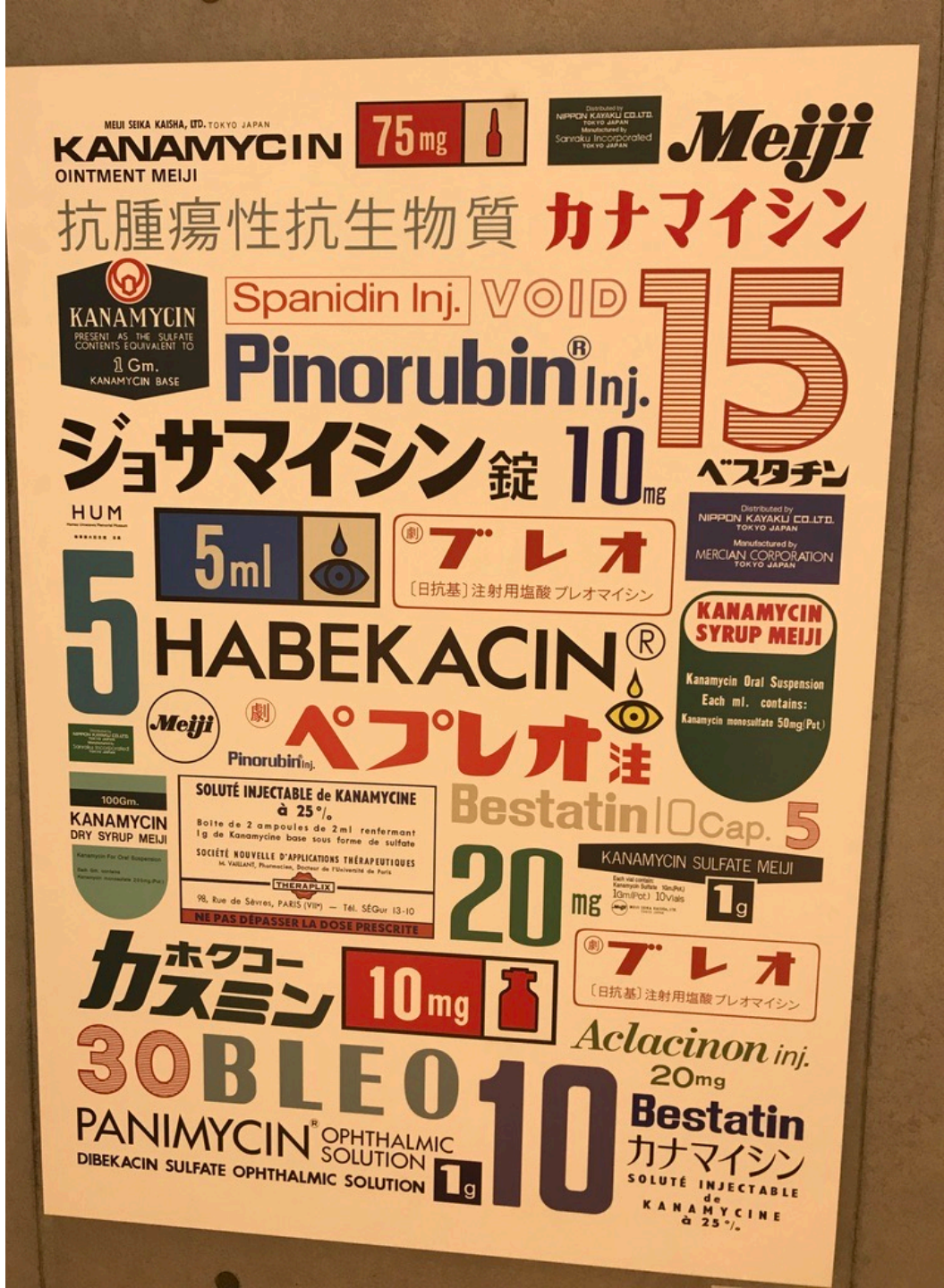
- 任期付き年俸制
- 優れた動物施設、共通機器、
- ナショナルバイオバンク、ヒト検体へのアクセス

2022-　微生物化学研究所（五反田、沼津）

微生物化学研究所の特長と未来

- 豊富な天然物資源、ライブラリー、中分子化合物
- 微生物培養、精製技術、人材豊富
- 構造解析（NMR、X線、電顕、各種のMS)が充実
- 創薬に繋がる有機合成、また、沼津での動物実験
- 抗菌剤P1、抗NTM薬、神経筋接合部強化剤などの進展

- 良いアッセイ系を導入、ライブラリーの公開
- 抗菌剤開発に国や民間の協力必要
- 神経因性疼痛、抗がん剤などNCGMと共同研究



最後に、多くの方に感謝

- 小野薬品工業（1977年～）、
- 島津製作所（2003年～）

- Funding Agency,
  文科省、学術振興会、JSPS、JST、AMED
  各種民間財団

- 雇用先

京都大学→カロリンスカ研究所→東京大学→国立国際医療研究センター→微生物化学研究所

- 共同して研究を進めてくれた人々

海外在住14日人、国内教員５６人、企業勤務２８人、ベンチャー起業3人

- 現在、未来の共同研究者、家族