UNIVERSITI PUTRA MALAYSIA

REGULATION OF IGE-MEDIATED MAST CELL DEGRANULATION IN ALLERGY BY GERANYL ACETOPHENONE

TAN JI WEI

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By

TAN JI WEI

Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in Fulfillment of the Requirement for the Degree of Doctor of Philosophy

November 2017

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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfillment of the requirement for the degree of Doctor of Philosophy

REGULATION OF IgE-MEDIATED MAST CELL DEGRANULATION IN ALLERGY BY GERANYL ACETOPHENONE

By

TAN JI WEI

November 2017

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Faculty: Medicine and Health Sciences

The worldwide prevalence of IgE-antigen-mediated allergic diseases, such as asthma and anaphylaxis, has increased dramatically over the past decades. Mast cells, which play a critical role in IgE-mediated allergy by contributing through its ability to release various proinflammatory mediators during degranulation. 2, 4, 6-trihydroxy-3-geranylacetophenone (tHGA), is an active compound originated from a local shrub known as *Melicope ptelefolia*. Previous studies demonstrated the potential therapeutic effects of tHGA in murine model of allergic airway inflammation. However, the underlying mechanism of the inhibitory effects of tHGA on mast cell degranulation remains unknown. Therefore, the current study aim is to investigate the *in vitro* and *in vivo* inhibitory effects of tHGA on IgE-mediated mast cell degranulation beside exploring its underlying mechanism. Three non-toxic concentrations of tHGA were used to investigate the cell morphology changes and selected key mediators release by DNP-IgE-sensitized RBL-2H3 cells during degranulation process. Apart from that, real time qPCR was used to study the effect of tHGA on gene expression of the mediators' release. Calcium assay kit was used to determine whether tHGA affects the mast cell activation whereas Western blotting was employed to explore the inhibitory role of tHGA by studying the signaling molecules located along LAT and LAT2 axis signaling pathways. In order to justify the speculated molecular target of tHGA, siRNA was utilized to silence the gene of protein of interest. Finally, *in vivo* study was carried out to determine tHGA's pharmaceutical effects in an animal model of anaphylaxis. For the results, pre-treatment of tHGA was able to preserve the cell morphology and actin microfilaments rearrangement challenged by DNP-BSA. This has led to a significant decrease in the release of both selected preformed and *de novo* synthesis mediators as well as the gene expression of mediators' release. Within the signalling pathway during IgE-mediated mast cell activation, tHGA was shown to play a major inhibitory role in LAT-PLCγ-MAPK pathway,

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which involves mainly the role of calcium ions. However, tHGA only plays a partial inhibition in the downstream of LAT2-PI3K pathway. Interestingly, the inhibition tHGA does not affect the activity of Syk tyrosine kinase molecule, which is responsible for the activation of both LAT and LAT2 pathways. This shows that the possible molecular target of tHGA in IgE-mediated mast cell degranulation might be the adapter transmembrane protein LAT. Apparently, LAT-deficient RBL-2H3 cells demonstrated that tHGA was unable to inhibit the IgE-mediated mast cell degranulation process, thus further justified the speculation that LAT is the possible molecular target of tHGA. Finally, tHGA is shown to be effective in the *in vivo* study where pre-treatment of tHGA was able to significantly decrease the serum mediators level while preserving the morphology of isolated peritoneal mast cells in DNP-BSA challenged Sprague Dawley rats by demonstrating less release of granules into the surrounding environment. As a conclusion, tHGA was shown to play a significant inhibitory effect in IgE-mediated mast cell degranulation both in *in vitro* and *in vivo* model. Specifically, tHGA targets the transmembrane LAT during the IgE-mediated mast cell activation.

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PENGAWALAN IgE-DIGRANULASI SEL MAST DALAM PENYAKIT ALERGI OLEH GERANYL ACETOPHENONE

Oleh

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Sejak dekad yang lalu, kelaziman penyakit alahan di seluruh dunia yang disebabkan oleh IgE / Antigen seperti asma dan anafilaksis telah meningkat secara mendadak. Sel mast memainkan peranan yang kritikal dalam penyakit alahan IgE melalui keupayaannya untuk menghasilkan pelbagai induksi keradangan ketika pengaktifannya. 2, 4, 6-trihydroxy-3-geranylacetophenone, atau dikenali sebagai tHGA, merupakan kompaun aktif yang berasal dari pokok renek tempatan iaitu tenggek burung. Kajian terdahulu menunjukkan bahawa tHGA mempunyai kesan terapeutik yang berpotensi dalam keradangan saluran udara yang berasaskan model haiwan. Tetapi, mekanisme asas yang menyebabkan kesan rencatan oleh tHGA dalam pengaktifan sel mast masih belum diterokai. Oleh itu, tujuan kajian semasa ini adalah untuk menyiasat kesan-kesan rencatan bagi tHGA dalam IgE pengaktifan sel mast dengan menggunakan model sel dan haiwan disamping menentukan peranannya dalam isyarat laluan. Tiga kepekatan tHGA yang tidak bertoksik telah digunakan untuk menyelidik perubahan morfologi sel serta penghasilan induksi keradangan yang terpilih oleh IgE-sensitasi sel-sel RBL-2H3 yang aktif. Disamping itu, teknik real time qPCR telah dugunakan untuk menggaji kerencatan tHGA terhadap gen induksi keradangan. Ujian kalsium juga telah dilaksanakan untuk menentukan sama ada tHGA menjejaskan penggerudian kalsium dalam sel semasa sel mast diaktifkan. Dari segi penentuan peranan penghalangan tHGA dalam lata isyarat sel, teknik Western blot telah digunakan untuk mengkaji ungkapan protein molekul isyarat yang terletak di sepanjang laluan isyarat LAT dan LAT2. Untuk menjustifikasikan hasil spekulasi molekul sasaran tHGA, Short interference RNA (siRNA) telah digunakan untuk tujuan menurunkan ungkapan gen protein yang dispekulasi. Akhirnya, bagi memastikan bahawa kesan perencatan tHGA dapat berfungsi dalam model haiwan, tHGA telah digunakan dalam model anafilaksis haiwan supaya kesan farmaseutikalnya dapat ditentukan. Dari segi keputusan eksperimen, perawatan awal tHGA dapat mengekalkan morfologi sel serta penyusunan

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microfilamen-filamen actin yang diaktifkan oleh DNP-BSA. Ini menyebabkan pengurangan yang ketara dalam kesemua penghasilan induksi keradangan yang terpilih dan juga gen-gen yang berkaitan denganya. Dari segi isyarat laluan dalam proses pengaktifan sel mast, tHGA memainkan peranan kerencatan yang utama dalam isyarat LAT-PLCγ-MAPK yang terutamanya melibatkan ion-ion calcium. Akan tetapi, tHGA hanya memainkan peranan separa dalam kerencatan isyarat LAT2-PI3K yang merupakan laluan alternatif isyarat pengaktifan. Apa yang menarik adalah tHGA tidak memainkan peranannya dalam perencatan aktiviti molekul Syk tyrosine kinase yang bertanggungjawab mengaktifkan laluan isyarat LAT dan LAT2. Ini membuktikan bahawa kemungkinan besar sasaran molekul bagi tHGA dalam isyarat lata huluan IgE-pengaktifan sel mast merupakan transmembran LAT. Justifikasi telah dibuktikan di mana IgE-sensitasi sel-sel RBL-2H3 yang telah didedahkan dengan siRNA LAT masih menghasilkan induksi-induksi keradangan yang ketara walaupun telah dirawat awal oleh tHGA. Dari segi kajian haiwan, hasil-hasil keputusan daripada rawatan awal tHGA terhadap tikus-tikus Sprague Dawley yang didedahkan dengan DNP-BSA menujukkan tahap mediator dalam serum darah telah menurun secara ketara. Bukan sahaja itu, hasil pemeriksaan mikroskop bagi sel-sel mast peritoneal yang terpencil menunjukan bahawa tHGA dapat mengekalkan morfologi sel walaupun telah diaktifkan disamping mengurangkan pelepasan granul-granul dalam sel ke sekeliling luar. Kesimpulannya, tHGA dibukti dapat memainkan peranan yang ketara dalam perencatan IgE-pengaktifan sel mast scara *in vitro* dan *in vivo*. Secara khususnya, sasaran perencatan tHGA dalam proses pengaktifan sel mast adalah transmembran LAT.

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Tan Ji Wei

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I certify that a Thesis Examination Committee has met on 21st of November 2017 to conduct the final examination of Tan Ji Wei on his thesis entitled "Regulation of IgE-mediated Mast Cell Degranulation in Allergy by Geranyl Acetophenone" in accordance with the Universities and University Colleges Act 1971 and the Constitution of the Universiti Putra Malaysia [P.U.(A) 106] 15 March 1998. The Committee recommends that the student be awarded the Doctor of Philosophy.

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LIST OF ABBREVIATIONS / NOTATIONS

AE	Atopic Eczema
AEBSF	4-(2-Aminoethyl) benzenesulfonyl fluoride hydrochloride
AChe	Acetylcholinesterase
AKT	Protein kinase B
ANOVA	One-way analysis of variance
ATCC	American Type Culture Collection
ATP	Adenosine triphosphate
BALF	Bronchoalveolar lavage fluid
BCA	Bicinchoninic acid
BMMC	Bone marrow mast cell
DNP-BSA	Dinitrophenyl bovine serum albumin
BTK	Bruton's tyrosine kinase
Ca^{2+}	Calcium ion
$CaCl_2$	Calcium chloride
CCD	Charge-coupled device
CH_2Cl_2	Dichloromethane
COX	Cyclooxygenase
CO_2	Carbon Dioxide
$cPLA_2$	Cytosolic phospholipases A2
CTMC	Connective tissue mast cells
cysLT	cysteinyl leukotrienes
DAG	Diacylglycerol
dd.H_2O	Deionized water
DMSO	Dimethylsulfoxide
ECL	Enhanced chemiluminescence
EDTA	Ethylenediaminetetraacetate
EIA	Enzyme-linked immunoassay
ELISA	Enzyme-linked immunosorbent assay
EMEM	Eagle's minimum essential medium
ERK	Extracellular signal–regulated kinases
FBS	Fetal bovine serum
FcεRI	High-affinity IgE receptor
Fig.	Figure
FITC	Fluorescein isothiocyanate
FYN	Proto-oncogene tyrosine-protein kinase
GAB2	GRB2-associated-binding protein 2
GADS	GRB2-related adaptor protein
GAPDH	Glyceraldehyde 3-phosphate dehydrogenase
gDNA	Genomic DNA
GRB2	Growth-factor-receptor-bound protein 2
HCl	Hydrochloric acid
HMC	Human mast cell
H_2O_2	Hydrogen peroxide
HRP	Horseradish peroxidase
IL	Interleukin
IC_{50}	Half maximal inhibitory concentration
DNP-IgE	Dinitrophenyl Immunoglobulin E

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IKK	IκB kinase
IP_2	Phosphatidylinositol-4,5-bisphosphate
IP_3	Inositol-1,4,5-trisphosphate
JAK	Janus kinase 3
JNK	c-Jun N-terminal kinases
KCl	Potassium chloride
KF	Ketotifen Fumarate
LAT	Linker for Activation of T cells
LAT2	Non–T Cell Activation Linker
LOX	lipoxygenase
LTC	Leukotriene
LYN	Tyrosine-protein kinase
MAPK	Mitogen activated protein kinase
$MgCl_2$	Magnesium chloride
MOA	Mechanism of action
MCs	Mast cells
mRNA	Messenger RNA
MTT	Diphenyltetrazolium Bromide
MMC	Mucosal mast cells
MW	Molecular weight
NFκB	Nuclear factor-κB
NaCl	Sodium chloride
Na_2HPO_4	Sodium hydrogen phosphate
NADH	Nicotinamide adenine dinucleotide (reduced form)
NSF	N-ethylmaleimide-sensitive factor
OD	Optical density
OVA	Ovalbumin
p65	Transcription factor p65
PAGE	Polyacrylamide gel electrophoresis
PBS	Phosphate buffered saline
PCA	Passive cutaneous anaphylaxis
PCR	Polymerase chain reaction
PDK1	3-phosphoinositide dependent protein kinase 1
PET	Positron emission tomography
PI3K	Phosphoinositide 3-kinase
PG	Prostaglandin
PLCγ	Phosphoinositide phospholipase C gamma
PMSF	Phenylmethanesulfonylfluoride
PMC	Peritoneal mast cell
PNAG	4-Nitrophenyl N-acetyl-β-D-glucosaminide
PSA	Passive systemic anaphylaxis
PTL	Parthenolide
PVDF	Polyvinylidene fluoride
qPCR	Quantitative polymerase chain reaction
R^2	Coefficient of determination
RAF	Rapidly Accelerated Fibrosarcoma
RBL	Rat basophilic leukaemia
RIPA	Radio immuno precipitation assay
RNA	Ribonucleic acid
RNAi	RNA interference
RPM	Revolutions per minute

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RT	Room Temperature
rRNA	Ribosomal RNA
SDS	Sodium dodecyl sulphate
S.E.M	Standard error of mean
SCF	Stem cell factor
SG	Secretory granule
SHC	SH2-domain-containing transforming protein C
shRNA	short hairpin RNA
SIN	Sinomenine
siRNA	small interfering RNA
SK	Sphingosine kinase
SLP76	SH2-domain-containing leukocyte protein of 65 kDa
SOS	Son of sevenless homologue
SPSS	Statistical Package for the social Science
SRC	proto-oncogene c-Src
SYK	Spleen tyrosine kinase
TAE	Tris-acetate-EDTA
TB	Toluidine blue
TMB	3,3',5,5'-Tetramethylbenzidine
TBS	Tris Buffered Saline
TBST	Tris Buffered Saline-Tween 20
TEC	Tyrosine-protein kinase Tec
TEMED	Tetramethylethylenediamine
TFIIB	Transcription factor II B
tHGA	2, 4, 6-trihydroxy-3-geranylacetophenone
Th2	T helper cell type 2
TNF-α	Tumor necrosis factor alpha
TUNEL	TdT-mediated dUTP-nickend labeling
WAO	World Allergy Organisation
WHO	World Health Organisation
WST	Water soluble tetrazolium

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Notations

α	Alpha
β	Beta
γ	Gamma
U	Units
°C	Degree Celsius
M	Molar
mM	Millimolar
µM	Micromolar
nmol	nanomol
g	Gram
H	Hour
Hrs	Hours
mg	Milligram
µg	Microgram
L	Litre
mL	Millilitre
µL	Microlitre
cm^2	Centimeter square
nm	Nano meter
V	Volt
A	Ampere
kDa	Kilodalton
Min	Minutes
%	Percent
±	Plus and/or minus
P	Probability
<	Lesser than
*	Asterisk
#	Number sign

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CHAPTER 1

INTRODUCTION

Allergies are a number of reactions caused by hypersensitivity of the immune system to innocuous antigens in the environment that are normally harmless and cause minute problems in most people. The exposure of atopic individuals to potential allergens such as dust, mites, and pollen might lead to tissue damage and undesirable allergic reactions including allergic asthma, food allergies, atopic dermatitis, and anaphylaxis (Bieber et al., 2013; Fiocchi et al., 2013; Richard et al., 2013; Stephen et al., 2013). The prevalence of allergic diseases worldwide is rising dramatically in both developed and developing countries (Pawankar et al, 2013). This increase is especially problematic in children, who are bearing the greatest burden of this rising trend over the last two decades (Pawankar et al, 2013). The pathogenesis behind allergic diseases is complex and until now, is still not fully understood. However, there are increasing evidences that mast cells (MCs) play a crucial role in Type I allergy, as well as in innate and adaptive immune responses (Harvima et al., 2014).

Mast cells are known as critical participants in various biologic allergic disease processes (Bruhns et al., 2005; Plaut et al., 1989). They originate in the bone marrow from a lineage-specific multipotent hematopoietic progenitor after which they migrate to tissues and mature into effector cells in the proximity of organs and blood vessels (Gri et al., 2012; Migalovich-Sheikhet et al., 2012). These cells express receptors on their surface membranes that have high affinity and specificity for IgE (Wang et al., 2012). Interactions between multivalent antigens and surface-bound IgE will cause FcεRI aggregation, resulting in mast cell activation and release of various proinflammatory mediators (Wang et al., 2012). These released mediators will result in vasodilatation, bronchoconstriction, and other inflammatory responses (Sakai et al, 2010). Therefore, mast cell activation is widely recognised as a critical event in many IgE-mediated immune responses including allergic asthma, allergic rhinitis, atopic dermatitis, and anaphylaxis (Galli and Tsai, 2012; Sheinkopf et al., 2008; Holgate et al., 2005).

As mast cell degranulation is an important pathophysiological feature of allergic diseases in the initiation of immediate responses following exposure to allergens (Locksley, 2010), the activation of mast cells by antigens has been reported as strictly dependent on the influx of extracellular calcium, in which a complex interaction between signaling molecules and various pathways located

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within the cells is involved (Sanchez-Miranda et al., 2010; Shumilina et al., 2008). Mast cell activation in allergic reactions is regulated by the aggregation of FcεRI, which immediately releases chemical mediators such as histamine and proteases in the early phase, as well as newly synthesised inflammatory mediators including prostaglandins, leukotrienes, and other proinflammatory cytokines in the late phase of allergic reactions (Yamaguchi et al., 1999). Although during mast cell activation, the immediate receptor-proximal signalling events seem common in the release of all categories of mediators, the receptor-distal signalling events must diverge to regulate the different mechanisms by which these mediators are released (Gilfillan and Tkaczyk, 2006).

Many prescription medicines commonly used for the treatment of allergies such as glucocorticoids, ketotifen, and cromolyn have been shown to have inhibitory effects on mast cell degranulation and mediator release. However, in patients with allergy, as in those with many other diseases, several different cell types are involved in causing symptoms, and therefore a number of potential targets exist (Harvima et al., 2014). The problem of selectivity and attempting to hit multiple targets simultaneously increases potential side effects and adverse drug reactions (Harvima et al., 2014). Therefore, considerable progress has been made in the last decade to explore new possibilities for treating allergic diseases, specifically by targeting MCs.

Currently, anti-histamine drugs and steroids remain the mainstay of therapies to combat allergic diseases (Kaliner, 2009). Apart from that, small-molecule inhibitors that target leukotrienes (zileuton and pranlukast) or histamine receptors (clemastine and loratadine) are also being used to treat allergic diseases, although adverse effects such as cardiac toxicity and angioedema have occasionally been reported (Van Hoecke et al., 2007). There are also drugs that prevent mediator release from activated mast cells, also known as mast cell stabilisers, such as cromolyn sodium and ketotifen fumarate (Finn and Walsh, 2013). However, these treatments cause unwanted side effects such as drowsiness, upset stomach, chest congestion, and dry mouth (Oppenheimer and Casale, 2002). As such, many researchers are now shifting their attention towards natural compounds or other traditional medicinal herbs to control immune responses and combat allergic reactions with less side effects (Tang et al, 2015; Chung et al, 2013). A diverse range of mast cell stabilising compounds have been identified from natural sources in the last decade, and are currently under clinical trial (Nugroho et al., 2009; Mazuc et al. 2008; D'Cruz and Uckun, 2007; Altounyan, 1967). Although in many cases, the precise mode of action of these molecules is unclear, all of these substances have demonstrated mast cell stabilisation activity and therefore may have potential therapeutic use in the treatment of allergic and related diseases where mast cells are intrinsically involved (Finn and Walsh, 2013).

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2, 4, 6-trihydroxy-3-geranylacetophenone (tHGA) is an active compound that can be found in a local shrub namely *Melicope ptelefolia* (Abas et al., 2006). Due to its exceptional antipyretic, analgesic, and anti-inflammatory activities, *M. ptelefolia* has been traditionally used for treating a wide range of diseases including eczema and dermatitis (Shaari et al., 2006; Van et al., 1998). tHGA is also shown to exhibit anti-inflammatory activity through LOX/COX inhibition (Shaari et al., 2011). Recently, it has been reported that this compound can be synthesised in the laboratory for which its anti-inflammatory effects in a murine model of allergic asthma was demonstrated (Ismail et al., 2012). In addition, previous findings confirm the effects of tHGA in reducing airway hyperresponsiveness, eosinophilia, goblet cell hyperplasia, inflammatory cell infiltration, cysteinyl leukotriene, and Th2-associated cytokines (IL-4, IL-5 and IL-13) synthesis (Lee et al., 2017; Ismail et al., 2012). However, there have been been no attempts as yet to relate the anti-inflammatory and anti-asthmatic effects of tHGA with mast cell degranulation.

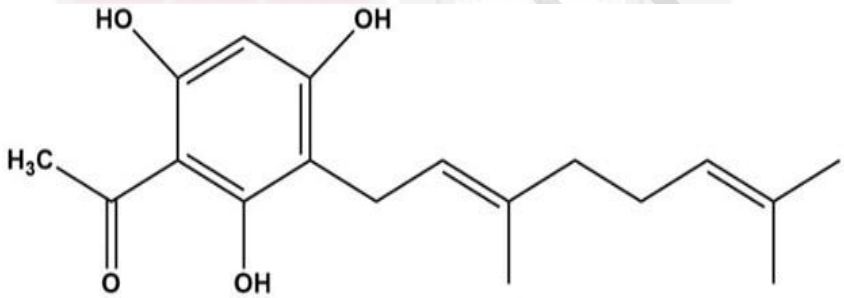

Figure 1: Molecular structure of tHGA

Since previous studies have shown that tHGA is able to suppress the production of many of these cytokines and mediators in murine models of allergic asthma, it is hypothesised that tHGA may exert its anti-inflammatory and anti-allergic properties via inhibition of IgE-mediated mast cell degranulation. Therefore, this study is designed to explore the potential inhibitory effects of tHGA in *in vitro* and *in vivo* models of IgE-mediated mast cell degranulation. Apart from that, the mechanism of action underlying its inhibitory action within the signaling cascade during mast cell activation is also studied in order to identify the possible molecular target of tHGA.

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1.1. General objective

To determine the *in vitro* and *in vivo* inhibitory effects of tHGA on IgE-mediated mast cell degranulation.

1.2. Specific objectives

i. To determine the effects of tHGA on the morphology of mast cells and the production of mast cell related proinflammatory mediators using *in vitro* model of IgE-mediated mast cell degranulation.

ii. To dissect the mechanism of action underlying the mast cell stabilizing effects of tHGA using *in vitro* model of IgE-mediated mast cell degranulation.

iii. To identify the potential molecular target(s) associated with the mast cell stabilizing effects of tHGA using *in vitro* model of IgE-mediated mast cell degranulation.

iv. To determine the effects of tHGA on the morphology of mast cells and the production of mast cell related mediators using *in vivo* model of IgE-mediated mast cell degranulation.

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1.3. Hypotheses

i. tHGA is able to inhibit IgE-mediated mast cell degranulation in an *in vitro* model by stabilizing the mast cell morphology and attenuates the release of mast cell related proinflammatory mediators.

ii. tHGA is able to play a vital part in the inhibition of protein molecules within the signalling pathways, including the LAT-PLCγ-MAPK and LAT2-PI3K axis pathways, during IgE-mediated mast cell activation.

iii. The possible molecular target of inhibition for tHGA in IgE-mediated mast cell degranulation is located on the upstream signalling cascade.

iv. tHGA is able to inhibit IgE-mediated mast cell degranulation in an *in vivo* model of allergic diseases by preserving the peritoneal mast cell structure and attenuates the release of proinflammatory mediators in the blood sera.

1.4. Limitation of study

i. The mast cell stabilizer included in this study, ketotifen fumarate, was not intentionally used to do comparison with the effects of tHGA, but rather as an experimental control group to ensure that there is an intended inhibitory effect on all the experimental designs throughout this study.

ii. The ketotifen fumarate's concentration (300 µM) and dose (1 mg/kg) selection for the respective *in vitro* and *in vivo* study were based on previous reported studies (Wang et al., 2012; Singh et al., 2012).

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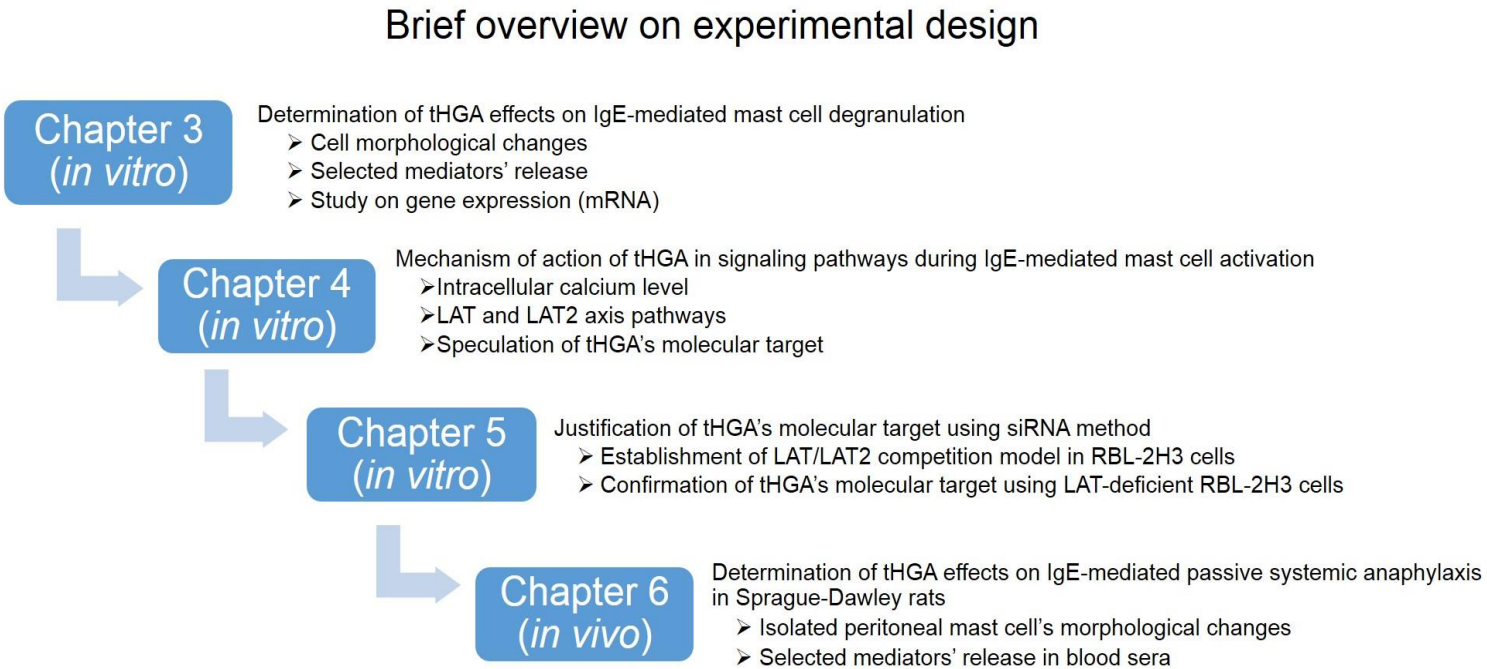

Figure 2: Overview of experimental works on chapter 3 to chapter 6

REFERENCES

Abas, F., Shaari, K., Israf, D.A., Syafri, S., Zainal, Z. and Lajis, N.H. 2010. LC-DAD-ESI-MS analysis of nitric oxide inhibitory fractions of tenggek burung (*Melicope ptelefolia* Champ Ex Benth.). *Journal of Food Composition and Analysis* 23: 107–112.

Abas, F., Lajis, N.H., Israf, D.A., Shaari, K. and Umi Kalsomb, Y. 2006. Antioxidant and nitric oxide inhibition activities of selected Malay traditional vegetables. *Food Chemistry* 95: 566–573.

Ab Karim, M.S., Nasouddin, S.S., Hussin, S.R. Othman, M., Khozirah, S., and Mohd Adzahan, N. 2011. Consumers' knowledge and perception towards *Melicope ptelefolia* (Daun Tenggek Burung): A preliminary qualitative study. *International Food Research Journal* 18: 1481-1488.

Ali, K. Bilancio, A., Thomas, M., Pearce, W., Gilfillan, A.M., Tkaczyk, C., Kuehn, N., Gray, A., Giddings, J., Peskett, E., Fox, R., Bruce, I., Walker, C., Sawyer, C., Okkenhaug, K., Finan, P. and Vanhaesebroeck, B. 2004. Essential role for the p110δ phosphoinositide 3-kinase in the allergic response. *Nature* 431: 1007–1011.

Altounyan, R.E.C., 1967. Inhibition of experimental asthma by a new compound, cromolyn sodium "Intal". *Acta Allergologica* 22: 487–495.

Amin, K. 2012. The role of mast cells in allergic inflammation. *Respiratory Medicine* 106: 9-14.

Aranda, P.S., LaJoie, D.M. and Jorcyk, C.L. 2012. Bleach gel: a simple agarose gel for analyzing RNA quality. *Electrophoresis* 33: 366-369.

Arruda, L.K. 2005. Cockroach allergens. *Current Allergy and Asthma Reports* 5: 411-416.

Asher, M.I., Anderson, H.R., Stewart, A.W. and Crane, J. 1998. Worldwide variations in the prevalence of asthma symptoms: The International Study of Asthma and Allergies in Childhood (ISAAC). *The European Respiratory Journal* 12: 315-335.

Aung, G., Niyonsaba, F., Ushio, H., Kajiwara, N., Saito, H., Ikeda, S., Ogawa, H. and Okumura, K. 2011. Catestatin, a neuroendocrine antimicrobial peptide, induces human mast cell migration, degranulation and production of cytokines and chemokines. *Immunology* 132: 527–539.

Baba, A., Tachi, M., Ejima, Y., Endo, Y., Toyama, H., Matsubara, M., Saito, K., Yamauchi, M., Miura, C. and Kazama, I. 2016. Anti-allergic drugs tranilast and ketotifen dose-dependently exert mast cell-stabilizing properties. *Cellular Physiology and Biochemistry* 38: 15-27.

© COPYRIGHT UPM

Baba, Y., Nishida, K., Fujii, Y., Hirano, T., Hikida, M. and Kurosaki, T. 2008. Essential function for the calcium sensor STIM1 in mast cell activation and anaphylactic responses. *Nature Immunology* 9: 81–88.

Balagopalan, L., Kortum, R.L., Coussens, N.P., Barr, V.A. and Samelson, L.E. 2015. The linker for activation of T cells (LAT) signalling hub: from signalling complexes to microclusters. *Journal of Biological Chemistry* 290: 26422-26429.

Barker, S.A., Caldwell, K.K., Pfeiffer, J.R. and Wilson, B.S. 1998. Wortmannin-sensitive phosphorylation, translocation, and activation of PLC-γ1, but not PLC-γ2, in antigen-stimulated RBL-2H3 mast cells. *Molecular Biology of the Cell* 6: 483–496.

Barnes, P.J. 2008. The cytokine network in asthma and chronic obstructive pulmonary disease. *Journal of Clinical Investigation* 118: 3546-3556.

Barnes, P.J., 2011. Glucocorticosteroids: current and future directions. *British Journal of Pharmacology* 163, 29–43.

Barnetson, R.S. and Rogers, M. 2002. Childhood atopic eczema. *British Medical Journal* 324: 1376-1379.

Barsumian, E.L., Isersky, M.G., Petrino, M.G. and Siraganian, R.P. 1981. IgE-induced histamine release from rat basophilic leukemia cell lines: isolation of releasing and non-releasing clones. *European Journal of Immunology* 11: 317-322.

Beaven, M.A. and Baumgartner, R.A. 1996. Downstream signals initiated in mast cells by FcεRI and other receptors. *Current Opinion in Allergy and Clinical Immunology* 8: 766–772.

Beaven, M.A. and Metzger, H. 1993. Signal transduction by Fc receptors: the FcεRI case. *Immunology Today* 14: 222–226.

Beaven, M.A., Rogers, J., Moore, J.P., Hesketh, T.R., Smith, G.A. and Metcalfe, J.C. 1984. The mechanism of the calcium signal and correlation with histamine release in 2H3 cells. *Journal of Biological Chemistry* 259: 7129-7136.

Belliveau, P.P. 2005. Omalizumab: a monoclonal anti-IgE antibody. *Medscape General Medicine* 7: 27.

Beyer, K. and Teuber, S. 2005. Food allergy diagnostics: Scientific and unproven procedures. *Current Opinion in Allergy and Clinical Immunology* 5: 261-266.

Bieber, T. 2008. AD. *New England Journal of Medicine* 358: 1483-1494.

Bieber, T., Leung, D., Gamal, Y.E. and Ivancevich, J.C. 2013. Atopic Eczema. In *WAO White Book on Allergy update 2013*, eds. R. Pawankar, G.W. Canonica, S.T. Holgate, R.F. Lockey, M.S. Blaiss, pp. 44-48. USA.

© COPYRIGHT UPM

Blank, U., Madera-Salcedo, I.K., Danelli, L., Claver, J., Tiwari, N., Sánchez-Miranda, E., Vázquez-Victorio, G., Ramírez-Valadez, K.A., Macias-Silva, M. and González-Espinosa, C. 2014. Vesicular trafficking and signaling for cytokine and chemokine secretion in mast cells. *Frontiers in Immunology* 5: 453.

Blechman, J.M., Lev, S., Brizzi, M.F., Leitner, O., Pegoraro, L., Givol, D. and Yarden, Y. 1993. Soluble c-kit proteins and antireceptor monoclonal antibodies confine the binding site of the stem cell factor. *Journal of Biological Chemistry* 268: 4399-4406.

Bohl B. 2013. Food allergy. In Clinical Immunology 4th edition, ed R.R. Rich, T.A. Fleisher, W.T. Shearer, H. Schroeder, A.J. Frew and C.M. Weyand, pp 543-549. Philadelphia: Elsevier.

Borish, L. and Joseph, B.Z. 1992. Inflammation and the allergic response. *Medical Clinics of North America* 76: 765-787.

Bousquet, J., Bieber, T., Fokkens, W., Humbert, M., Kowalski, M.L., Niggemann, B., Simon, H.U. and Schünemann, H. 2008. Consensus statements, evidence-based medicine and guidelines in allergic diseases. *Allergy* 63: 1-4.

Boyce, J.A., Assa'ad, A., Burks, A.W., Jones, S.M., Sampson, H.A., Wood, R.A., Plaut, M., Cooper, S.F., Fenton, M.J., Arshad, S.H., Bahna, S.L., Beck, L.A., Byrd-Bredbenner, C., Camargo, C.A.Jr., Eichenfield, L., Furuta, G.T., Hanifin, J.M., Jones, C., Kraft, M., Levy, B.D., Lieberman, P., Luccioli, S., McCall, K.M., Schneider, L.C., Simon, R.A., Simons, F.E., Teach, S.J., Yawn, B.P. and Schwaninger, J.M. 2011. Guidelines for the diagnosis and management of food allergy in the United States: summary of the NIAID-sponsored expert panel report. *Nutrition research* 31: 61-75.

Bradbury, C.M., Markovina, S., Wei, S.J., Rene, L.M., Zoberi, I., Horikoshi, N. and Gius, D. 2001. Indomethacin-induced radiosensitization and inhibition of ionizing radiation-induced NF-kappaB activation in HeLa cells occur via a mechanism involving p38 MAP kinase. *Cancer Research* 61: 7689–7696.

Bradding, P., Walls, A.F. and Holgate, S.T. 2006. The role of the mast cell in the pathophysiology of asthma. *Journal of Allergy and Clinical Immunology* 117: 1277-1284.

Braselmann, S., Taylor, V., Zhao, H., Wang, S., Sylvain, C., Baluom, M., Qu, K., Herlaar, E., Lau, A., Young, C., Wong, B.R., Lovell, S., Sun, T., Park, G., Argade, A., Jurcevic, S., Pine, P., Singh, R., Grossbard, E.B., Payan, D.G. and Masuda, E.S. 2006. R406, an orally available spleen tyrosine kinase inhibitor blocks fc receptor signalling and reduces immune complex-mediated inflammation. *Journal of Pharmacology and Experimental Therapeutics* 319: 998-1008.

© COPYRIGHT UPM

Bric, A., Miething, C., Bialucha, C.U., Scuoppo, C., Zender, L., Krasnitz, A., Xuan, Z., Zuber, J., Wigler, M., Hicks, J., McCombie, R.W., Hemann, M.T., Hannon, G.J., Powers, S. and Lowe, S.W. 2009. Functional identification of tumor-suppressor genes through an in vivo RNA interference screen in a mouse lymphoma model. *Cancer Cell* 16: 324-335.

Brown, J.M., Wilson, T.M. and Metcalfe, D.D. 2008. The mast cell and allergic diseases: role in pathogenesis and implications for therapy. *Clinical & Experimental Allergy* 38: 4-18.

Bruhns, P., Fremont, S. and Daeron, M. 2005. Regulation of allergy by Fc receptors. *Current Opinion in Immunology* 17: 662–669.

Buell, D.N., Fowlkes, B.J., Metzher, H. and Isersky, C. 1976. Cell cycle and morphological changes during growth and differentiation of a rat basophilic leukemia cell line. *Cancer Research* 36: 3131–3137.

Butler, M.S. 2004. The role of natural product in chemistry in drug discovery. *Journal of Natural Products* 67: 2141–2153.

Calogero, C., Kusel, M.M., Van Bever, H.P. and Sly, P.D. 2009. Management of childhood asthma in Western Australia. *Journal of Paediatric and Child Health* 45: 139-148.

Carlos E.B.C. and Héctor, B. 2013. Pharmacotherapy of Allergic Diseases. In *WAO White Book on Allergy update 2013*, eds. R. Pawankar, G.W. Canonica, S.T. Holgate, R.F. Lockey, M.S. Blaiss, pp. 114-118. USA.

Carter, A. 2008. Curry compound fights cancer in the clinic. *Journal of the National Cancer Institute* 100: 616-617.

Castanedo-Tardan, M.P., Matiz, C. and Jacob, S.E. 2011. Contact dermatitis in children: A review of current opinions. *Actas Dermo-sifiliográficas* 102: 8-18.

Chew, F.T., Lim, S.H., Shang, H.S., Dahlia, M.D., Goh, D.Y., Lee, B.W., Tan, H.T. and Tan, T.K. 2000. Evaluation of the allergenicity of tropical pollen and airborne spores in Singapore. *Allergy* 55: 340-347.

Chinn, D.J. and Sheikh, A. 2009. Epidemiology of anaphylaxis. In *Allergy frontiers: Epigenetics, allergens and risk factors*, eds. R. Pawankar, S.T. Holgate, L.J. Rosenwasser, pp 123-144. Tokyo: Springer.

Choi, O.H., Adelstein, R.S. and Beaven, M.A. 1994. Secretion from rat basophilic RBL-2H3 cells is associated with diphosphorylation of myosin light chains by myosin light chain kinase as well as phosphorylation by protein kinase C. *Journal of Biological Chemistry* 269, 536-541.

Choi, O.H., Lee, J.H., Kassessinoff, T., Cunha-Melo, J.R., Jones, S.V. and Beaven, M.A. 1993. Antigen and carbachol mobilize calcium by similar mechanisms in a transfected mast cell line (RBL-2H3 cells) that expresses m1 muscarinic receptors. *Journal of Immunology* 151: 5586–5595.

© COPYRIGHT UPM

Chong, Y.J., Firdaus, M.N., Chean, Hui. A.N., Shaari, K., Israf, D.A. and Tham, C.L. 2016. Barrier Protective Effects of 2,4,6-Trihydroxy-3-Geranyl Acetophenone on Lipopolysaccharides-Stimulated Inflammatory Responses in Human Umbilical Vein Endothelial Cells. *Journal of Ethnopharmacology* 192: 248-255.

Chung, M.J., Sohng, J.K., Choi, D.J. and Park, Y.I. 2013. Inhibitory effect of phloretin and biochanin A on IgE-mediated allergic responses in rat basophilic leukemia RBL-2H3 cells. *Life Sciences* 93: 401-408.

Church, M.K. and Gradidge, C.F. 1980. The activity of sodium cromoglycate analogues in human lung *in vitro*: a comparison with rat passive cutaneous anaphylaxis and clinical efficacy. *British Journal of Pharmacology* 70: 307-311.

Church, M.K. and Levi-Schaffer, F. 1997. The human mast cell. *Journal of Allergy and Clinical Immunology* 99: 155-160.

Cook, D.N., Pisetsky, D.S. and Schwartz, D.A. 2004. Toll-like receptors in the pathogenesis of human disease. *Nature Immunology* 5: 975-979.

Cox, L., Larenas-Linnemann, D., Lockey, R.F. and Passalacqua, G. 2010. Speaking the same language: The World Allergy Organization Subcutaneous Immunotherapy Systemic Reaction Grading System *Journal of Allergy and Clinical Immunology* 125: 569-574.

Cragg, G.M. and Newman, D.J. 2005. Biodiversity: A continuing source of novel drug leads. *Pure and Applied Chemistry* 77: 7–24.

Daikonya, A., Katsuki, S., Wu, J.B. and Kitanaka, S. 2002. Anti-allergic agents from natural sources (4): anti-allergic activity of new phloroglucinol derivatives from *Mallotus philippensis (Euphorbiaceae)*. *Chemical & pharmaceutical bulletin* 50: 1566-1569.

Darsow, U., Wollenberg, A., Simon, D., Taïeb, A., Werfel, T., Oranje, A., Gelmetti, C., Svensson, A., Deleuran, M., Calza, A.M., Giusti, F., Lübbe, J., Seidenari, S., Ring, J. and European Task Force on Atopic Dermatitis/EADV Eczema Task Force. 2010. ETFAD/EADV Eczema Task Force 2009 position paper on diagnosis and treatment of atopic dermatitis. *Journal of the European Academy of Dermatology and Venereology* 24: 317-328.

da Silva, E.Z., Jamur, M.C. and Oliver, C. 2014. Mast cell function: a new vision of an old cell. *Journal of Histochemistry & Cytochemistry* 62: 698–738.

D'Cruz, O.J. and Uckun, F.M. 2007. Targeting mast cells in endometriosis with janus kinase 3 inhibitor, JANEX-1. *American Journal of Reproductive Immunology* 58: 75–97.

© COPYRIGHT UPM

Decker, W.W. Campbell, R.L., Manivannan, V., Luke, A., St Sauver, J.L., Weaver, A., Bellolio, M.F., Bergstralh, E.J., Stead, L.G. and Li, J.T. 2008. The ethology and incidence of anaphylaxis in Rochester, Minnesota: A report from the Rochester Epidemiology Project. *Journal of Allergy and Clinical Immunology* 122: 1161-1165.

Denyer, J. and Patel, V., 2009. Syk kinase inhibitors in allergic diseases. *Drug News & Perspectives* 22: 146-150.

de Souza Santos, M., Freire de Morais Del Lama, M.P., Deliberto, L.A., da Silva Emery, F., Tallarico Pupo, M. and Zumstein Georgetto Naal, R.M. 2013. In situ screening of 3-arylcoumarin derivatives reveals new inhibitors of mast cell degranulation. *Archives of Pharmacal Research* 36: 731–738.

de Vries, V.C., Pino-Lagos, K., Elgueta, R. and Noelle, R.J. 2009. The enigmatic role of mast cells in dominant tolerance. *Current Opinion in Organ Transplantation* 14: 332-337.

Dias, D.A., Urban, S. and Roessner, U. 2012. A Historical Overview of Natural Products in Drug Discovery. *Metabolites* 2: 303-336.

Doffek, K., Chen, X., Sugg, S.L. and Shilyansky, J. 2011. Phosphatidylserine inhibits NFκB and p38 MAPK activation in human monocyte derived dendritic cells. *Molecular Immunology* 48: 1771-1777.

Dombrowicz, D., Flamand, V., Brigman, K.K., Koller, B.H. and Kinet, J.P. 1993. Abolition of anaphylaxis by targeted disruption of the high affinity immunoglobulin E receptor α chain gene. *Cell* 75: 969–976.

Draber, P., Halova, I., Levi-Schaffer, F. and Draberova, L. 2012a. Transmembrane adaptor proteins in the high-affinity IgE receptor signalling. *Frontiers in Immunology* 2: 95.

Draber, P., Sulimenko, V. and Dráberová, E. 2012b. Cytoskeleton in mast cell signalling. *Frontiers in Immunology* 3: 130.

Duke, J.A., Duke, P.A.K. and du Cellier, J.L. 2008. Camel's Thorn (Alhagi maurorum Medik.) ++ Fabaceae. In *Duke's Handbook of Medicinal Plants of the Bible*, ed. J.A. Duke, pp. 13-15. USA: CRC Press Taylor and Francis Group.

Dvorak, A.M., Massey, W., Warner, J., Kissell, S., Kagey-Sobotka, A. and Lichtenstein, L.M. 1991. IgE-mediated anaphylactic degranulation of isolated human skin mast cells. *Blood* 77: 569–578.

Eccleston, E., Leonard, B.J., Lowe, J.S. and Welford, H.J. 1973. Basophilic leukaemia in the albino rat and a demonstration of the basopoietin. *Nature: New Biology* 244: 73-76.

Edwards, A.M. and Norris, A.A. 1994. Cromoglycate and asthma. *Lancet* 343: 426.

© COPYRIGHT UPM

El-Agamy, D.S. 2012. Anti-allergic effects of nilotinib on mast cell-mediated anaphylaxis like reactions. *European Journal of Pharmacology* 680: 115-121.

Elbashir, S.M., Harborth, J., Lendeckel, W., Yalcin, A., Weber, K. and Tuschl, T. 2001. Duplexes of 21-nucleotide RNAs mediate RNA interference in cultured mammalian cells. *Nature* 411: 494-498.

Elias, P.M., Hatano, Y. and Williams, M.L. 2008. Basis for the barrier abnormality in atopic dermatitis: Outside-inside-outside pathogenic mechanisms. *Journal of Allergy and Clinical Immunology* 121: 1337-1343.

El-Sibai, M. and Backer, J.M. 2007. Phospholipase C gamma negatively regulates Rac/Cdc42 activation in antigen-stimulated mast cells. *European Journal of Immunology* 37: 261–270.

Enerback, L. 1966. Mast cells in rat gastrointestinal mucosa. 2. Dye binding and metachromatic properties. *Acta Pathologica et Microbiologica Scandinavica* 66: 303-312.

Engström, L., Ruud, J., Eskilsson, A., Larsson, A., Mackerlova, L., Kugelberg, U., Qian, H., Vasilache, A.M., Larsson, P., Engblom, D., Sigvardsson, M., Jönsson, J.I. and Blomqvist, A. 2012. Lipopolysaccharide-induced fever depends on prostaglandin E2 production specifically in brain endothelial cells. Endocrinology 153: 4849–4861.

Epstein, J.B., Scully, C. and Spinelli J. 1992. Toluidine blue and Lugol's iodine application in the assessment of oral malignant disease and lesions at risk of malignancy. *Journal of Oral Pathology & Medicine* 21: 160–163.

Erdei, A., Toth, G.K., Andrasfalvy, M., Matko, J., Bene, L., Bajtay, Z., Ischenko, A., Rong, X. and Pecht, I. 1999. Inhibition of IgE-mediated triggering of mast cells by complement-derived peptides interacting with the Fc epsilon RI. *Immunology Letters* 68: 79–82.

Fadyia Y. Al-Hamdani. 2010. Comparative clinical evaluation of ketotifen and montelukast sodium in asthmatic Iraqi patients. *Saudi Pharmaceutical Journal* 18: 245–249.

Fahrner, M., Derler, I., Jardin, I. and Romanin, C. 2013. The STIM1/Orai1 - signalling machinery. *Channels* 7: 330-343.

Finn, D.F. and Walsh, J.J. 2013. Twenty-first century mast cell stabilizers. *British Journal of Pharmacology* 170: 23-37.

Fiocchi, A., Sampson, H.A., Bahna, S.L. and Lack, G. 2013. Food Allergy. In *WAO White Book on Allergy update 2013*, eds. R. Pawankar, G.W. Canonica, S.T. Holgate, R.F. Lockey, M.S. Blaiss, pp. 54-59. USA.

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Flohr, C., Weiland, S.K., Weinmayr, G., Björkstén, B., Bråbäck, L., Brunekreef, B., Büchele, G., Clausen, M., Cookson, W.O., von Mutius, E., Strachan, D.P., Williams, H.C. and ISAAC Phase Two Study Group. 2008. The role of atopic sensitization in flexural eczema: findings from the International Study of Asthma and Allergies in Childhood Phase Two. *Journal of Allergy and Clinical Immunology* 121: 141-147.

Fukuishi, N., Murakami, S., Ohno, A., Yamanaka, N., Matsui, N., Fukutsuji, K., Yamada, S., Itoh, K. and Akagi, M. 2014. Does β-hexosaminidase function only as a degranulation indicator in mast cells? The primary role of β-hexosaminidase in mast cell granules. *Journal of Immunology* 193: 1886-1894.

Fuller. D.M., Zhu, M., Ou-Yang, C., Sullivan, S.A. and Zhang, W. 2011. A Tale of Two TRAPs: LAT and LAB in the Regulation of Lymphocyte Development, Activation, and Autoimmunity. Immunologic research. 49: 97-108.

Funaba, M., Ikeda, T. and Abe M. 2003. Degranulation in RBL-2H3 cells: regulation by calmodulin pathway. *Cell Biology International* 27: 879–885.

Furomoto, Y., Nunomura, S., Terada, T., Rivera, J. and Ra, C. 2004. The FcεRIβ immunoreceptor tyrosine-based activation motif exerts inhibitory control on MAPK and IκB kinase phosphorylation and mast cell cytokine production. *Journal of Biological Chemistry* 279: 49177–49187.

Galli, S.J., Nakae, S. and Tsai, M. 2005. Mast cells in the development of adaptive immune responses. *Nature Immunology* 6: 135–142.

Galli, S.J., Starkl, P., Marichal, T. and Tsai, M. 2016. Mast cells and IgE in defense against venoms: Possible "good side" of allergy? *Allergology International* 65:3-15.

Galli, S.J. and Tsai, M. 2012. IgE and mast cells in allergic disease. *Nature Medicine* 18: 693-704.

Galli, S.J., Tsai, M. and Piliponsky, A.M. 2008. The development of allergic inflammation. Nature 454: 445–454.

Gandolfo, S., Pentenero, M., Broccoletti, R., Pagano, M., Carrozzo, M. and Scully, C. 2006. Toluidine blue uptake in potentially malignant lesions *in vivo*: Clinical and histological assessment. *Oral Oncology* 42: 89–95.

Gentle, A., Anastasopoulos, F. and McBrien, N.A. 2001. High resolution semi-quantitative real-time PCR without the use of a standard curve. *BioTechniques* 31: 502-508.

Ghosh, M., Tucker, D.E., Burchett, S.A. and Leslie, C.C. 2006. Properties of the Group IV phospholipase A2 family. *Progress in Lipid Research* 45: 487–510.

© COPYRIGHT UPM

Giangrieco, I., Rafaiani, C., Liso, M., Palazzo, P., Pomponi, D., Tuppo, L., Crescenzo, R., Tamburrini, M., Mari, A. and Ciardiello, M.A. 2012. Allergens in allergy diagnosis: a glimpse at emerging new concepts and methodologies. *Translational Medicine @ UniSa* 4: 27-33.

Gilfillan, A.M. and Beaven, M.A., 2011. Regulation of mast cell responses in health and disease. *Critical Reviews™ in Immunology* 31: 475-529.

Gilfillan, A.M. and Tkaczyk, C. 2006. Integrated signalling pathways for mast-cell activation. *Nature Reviews Immunology* 6: 218-230.

Giudice, E.D., Rinaldi, L., Passarotto, M., Facchinetti, F., D'Arrigo, A., Guiotto, A., Carbonare, M.D., Battistin, L. and Leon, A. 2007. Cannabidiol, unlike synthetic cannabinoids, triggers activation of RBL-2H3 mast cells. *Journal of Leukocyte Biology* 81: 1512-1522.

Gordon, J.R. and Galli, S.J. 1991. Release of both preformed and newly synthesized tumor necrosis factor alpha (TNF-alpha)/cachectin by mouse mast cells stimulated via the Fc epsilon RI. A mechanism for the sustained action of mast cell-derived TNF-alpha during IgE-dependent biological responses. *Journal of Experimental Medicine* 174: 103-107.

Gri, G., Frossi, B., D'Inca, F., Danelli, L., Betto, E., Mion, F., Sibilano, R. and Pucillo, C. 2012. Mast cell: an emerging partner in immune interaction. *Frontiers in Immunology* 3: 120.

Gu, H., Saito, K., Klaman, L.D., Shen, J., Fleming, T., Wang, Y., Pratt, J.C., Lin, G., Lim, B., Kinet, J.P. and Neel, B.G. 2001. Essential role for Gab2 in the allergic response. *Nature* 412: 186–190.

Gurish, M.F. and Austen, K.F. 2012. Developmental origin and functional specialization of mast cell subsets. *Immunity* 37: 25–33.

Haefner, B. 2003. Drugs from the deep: Marine natural products as drug candidates. *Drug Discovery Today* 8: 536–544.

Hahtela, T., Tuomisto, L.E., Pietinalho, A., Klaukka, T., Erhola, M., Kaila, M., Nieminen, M., Kontula, E. and Laitinen, L.A. 2006. A 10-year asthma programme in Finland: major change for the better. *Thorax* 61: 663-670.

Halonen, M., Stern, D.A., Wright, A.L., Taussig, L.M. and Martinez, F.D. 1997. Alternaria as a major allergen for asthma in children raised in a desert environment. *American Journal of Respiratory and Critical Care Medicine* 155: 1356-1361.

Han, E.H., Hwang, Y.P., Kim, H.G., Choi, J.H., Im, J.H., Yang, J.H., Lee, H.U., Chun, S.S., Chung, Y.C. and Jeong, H.G. 2011. Inhibitory effect of *Pleurotus eryngii* extracts on the activities of allergic mediators in antigen-stimulated mast cells. *Food and Chemical Toxicology* 49: 1416-1425.

© COPYRIGHT UPM

Han, S.Y., Bae, J.Y., Park, S.H., Kim, Y.H., Park, J.H. and Kang, Y.H. 2013. Resveratrol inhibits IgE-mediated basophilic mast cell degranulation and passive cutaneous anaphylaxis in mice. *Journal of Nutrition* 143: 632-639.

Harvima, I.T., Levi-Schaffer, F., Draber, P., Friedman, S., Polakovicova, I., Gibbs, B.F., Blank, U., Nilsson, G. and Maurer, M. 2014. Molecular targets on mast cells and basophils for novel therapies. *Journal of Allergy and Clinical Immunology* 134: 530-544.

Hefner, Y., Borsch-Haubold, A.G., Murakami, M., Wilde, J.I., Pasquet, S., Schieltz, D., Ghomashchi, F., Yates, J.R.3rd, Armstrong, C.G., Paterson, A., Cohen, P., Fukunaga, R., Hunter, T., Kudo, I., Watson, S.P. and Gelb, M.H. 2000. Serine 727 phosphorylation and activation of cytosolic phospholipase A2 by MNK1-related protein kinases. *Journal of Biological Chemistry* 275: 37542–37551.

Hirano, T., Higa, S., Arimitsu, J., Naka, T., Ogata, A., Shima, Y., Fujimoto, M., Yamadori, T., Ohkawara, T., Kuwabara, Y., Kawai, M., Matsuda, H., Yoshikawa, M., Maezaki, N., Tanaka, T., Kawase, I. and Tanaka, T. 2006. Luteolin, a flavonoid, inhibits AP-1 activation by basophils. *Biochemical and Biophysical Research Communications* 340: 1–7.

Hirasawa, N., Santini, F. and Beaven, M. A. 1995. Activation of the mitogen activated protein kinase/cytosolic phospholipase A2 pathway in a rat mast cell line. Indications of different pathways for release of arachidonic acid and secretory granules. *Journal of Immunology* 154: 5391–5402.

Hisamichi, H., Naito, R., Toyoshima, A., Kawano, N., Ichikawa, A., Orita, A., Orita, M., Hamada, N., Takeuchi, M., Ohta, M. and Tsukamoto, S. 2005. Synthetic studies on novel Syk inhibitors. Part 1: Synthesis and structure-activity relationships of pyrimidine-5-carboxamide derivatives. *Bioorganic & Medicinal Chemistry Letters* 13: 4936-4951.

Hoesel, B. and Schmid, J.A. 2013. The complexity of NF-κB signalling in inflammation and cancer. *Molecular Cancer* 12: 86.

Holgate, S.T. 2011. Pathophysiology of asthma: what has our current understanding taught us about new therapeutic approaches? Journal of Allergy and Clinical Immunology 128: 495–505.

Holgate, S.T., Djukanovic, R., Casale, T. and Bousquet, J. 2005. Anti-immunoglobulin E treatment with omalizumab in allergic diseases: an update on anti-inflammatory activity and clinical efficacy. *Clinical & Experimental Allergy* 35: 408–416.

Horejsi, V. 2004. Transmembrane adaptor proteins in membrane microdomains: important regulators of immunoreceptor signalling. *Immunology Letters* 92: 43–49.

Hoth, M. and Penner, R. 1992. Depletion of intracellular calcium stores activates a calcium current in mast cells. *Nature* 355: 353–356.

© COPYRIGHT UPM

Huang, F., Tong, X., Deng, H., Fu, L. and Zhang, R. 2008a. Inhibition of the antigen-induced activation of RBL-2H3 cells by Gab2 siRNA. *Cellular & Molecular Immunology* 5: 433-438.

Huang, F., Yamaki, K., Tong, X., Fu, L., Zhang, R., Cai, Y., Yanagisawa, R., Inoue, K.i., Takano, H. and Yoshino, S. 2008b. Inhibition of the antigen-induced activation of RBL-2H3 cells by sinomenine. *International Immunopharmacology* 8: 502–507.

Huang, L., Li, T., Zhou, H., Qiu, P., Wu, J. and Liu, L. 2015. Sinomenine potentiates degranulation of RBL-2H3 basophils via up-regulation of phospholipase A2 phosphorylation by Annexin A1 cleavage and ERK phosphorylation without influencing on calcium mobilization. *International Immunopharmacology* 28: 945-951.

Hundley, T.R., Gilfillan, A.M., Tkaczyk, C., Andrade, M.V., Metcalfe, D.D. and Beaven, M.A. 2004. Kit and FcepsilonRI mediate unique and convergent signals for release of inflammatory mediators from human mast cells. *Blood* 104: 2410-2417.

Ikawati, Z., Wahyuono, S. and Maeyama, K. 2001. Screening of several Indonesian medicinal plants for their inhibitory effect on histamine release from RBL-2H3 cells. *Journal of Ethnopharmacology* 75: 249–56.

Inoue, T., Suzuki, Y. and Ra, C. 2010. Epigallocatechin-3-gallate inhibits mast cell degranulation, leukotriene C4 secretion, and calcium influx via mitochondrial calcium dysfunction. *Free Radical Biology and Medicine* 49: 632–640.

Interchim. 2013. Cell viability by Interchim: Water Soluble Tetrazolium Salts (WSTs). Retrieved 11 July 2016 from http://www.interchim.fr/ft/F/F98881.pdf.

Ishijima, Y., Ohmori, S., Uenishi, A. and Ohneda, K. 2012. GATA transcription factors are involved in IgE-dependent mast cell degranulation by enhancing the expression of phospholipase C-γ1. *Genes to Cells* 17: 285-301.

Ismail, N., Jambari, N.N., Zareen, S., Akhtar, M.N., Shaari, K., Zamri-Saad, M., Tham, C.L., Sulaiman, M.R., Lajis, N.H. and Israf, D.A. 2012. A geranyl acetophenone targeting cysteinyl leukotriene synthesis prevents allergic airway inflammation in ovalbumin-sensitized mice. *Toxicology and Applied Pharmacology* 259: 257-262.

Iwaki, S., Jensen, B.M. and Gilfillan, A.M. 2007. Ntal/Lab/Lat2. *International Journal of Biochemistry & Cell Biology* 39: 868–873.

Iwaki, S., Tkaczyk, C., Metcalfe, D.D. and Gilfillan, A.M. 2005. Roles of adaptor molecules in mast cell activation. *Chemical Immunology and Allergy* 87: 43–58.

© COPYRIGHT UPM

Jiang, Y., Kanaoka, Y., Feng, C., Nocka, K., Rao, S. and Boyce, J.A. 2006. Cutting edge: Interleukin 4-dependent mast cell proliferation requires autocrine/intracrine cysteinyl leukotriene-induced signalling. *Journal of Immunology* 177: 2755-2759.

Johansson, S.G.O., Bieber, T., Dahl, R., Friedmann, P.S., Lanier, B.Q., Lockey, R.F., Motala, C., Ortega, M.J.A., Platts-Mills, T.A., Ring, J., Thien, F., Van Cauwenberge, P. and Williams, H.C. 2004. A revised nomenclature for allergy for global use: Report of the Nomenclature Review Committee of World Allergy Organization. *Journal of Allergy and Clinical Immunology* 113: 832-836.

Johnzon, C.F., Rönnberg, E. and Pejler, G. 2016. The Role of Mast Cells in Bacterial Infection. *American Journal of Pathology* 186: 4-14.

Jones, D.T., Soepadmo, E. and Wong, K.M. 1995. Rutaceae: The flora of Sabah and Sarawak. Kuala Lumpur: Ampang Press Sdn Bhd.

Jutel, M. and Akdis, C.A. 2011. Immunological mechanisms of allergen-specific immunotherapy. *Allergy* 66: 725–732.

Jyonouchi, H. 2010. Autism spectrum disorders and allergy: observation from a paediatric allergy/immunology clinic. *Expert Review of Clinical Immunology* 6: 397-411.

Kalesnikoff, J. and Galli, S. J. 2008. New developments in mast cell biology. *Nature Immunology* 9: 1215–1223.

Kaliner, M.A. 2009. Azelastine and olopatadine in the treatment of allergic rhinitis. *Annals of Allergy, Asthma & Immunology* 103: 373–380.

Kaplan, D.H., Igýartó, B.Z. and Gaspari, A.A. 2012. Early immune events in the induction of allergic contact dermatitis. *Nature Reviews Immunology* 12: 114–124.

Karin, M. 2005. Inflammation-activated protein kinases as target for drug development. *Proceedings of the American Thoracic Society* 2: 386-390.

Kawakami, T. and Galli, S.J. 2002. Regulation of mast-cell and basophil function and survival by IgE. *Nature Reviews Immunology* 2: 773–786.

Kawakami, T., Inagaki, N., Takei, M., Fukamachi, H., Coggeshall, K.M., Ishizaka, K. and Ishizaka, T. 1992. Tyrosine phosphorylation is required for mast cell activation by Fc epsilon RI cross-linking. *Journal of Immunology* 148: 3513–3519.

Kempuraj, D., Madhappan, B., Christodoulou, S., Boucher, W., Cao, J., Papadopoulou, N., Cetrulo, C.L. and Theoharides, T.C. 2005. Flavonols inhibit proinflammatory mediator release, intracellular calcium ion levels and protein kinase C theta phosphorylation in human mast cells. *British Journal of Pharmacology* 145: 934–944.

© COPYRIGHT UPM

Kang, O.H., Lee, J.H and Kwon, D.Y. 2011. Apigenin inhibits release of inflammatory mediators by blocking the NF-κB activation pathways in the HMC-1 cells. *Immunopharmacology and Immunotoxicology* 33: 473–479.

Kato, A., Chustz, R.T., Ogasawara, T., Kulka, M., Saito, H., Schleimer, R.P. and Matsumoto, K. 2009. Dexamethasone and FK506 inhibit expression of distinct subsets of chemokines in human mast cells. *Journal of Immunology* 182: 7233–7243.

Kidd, M., McKenzie, S.H., Steven, I., Cooper, C., Lanz, R., and the Australian Ketotifen Study Group. 2003. Efficacy and safety of ketotifen eye drops in the treatment of seasonal allergic conjunctivitis. *British Journal of Ophthalmology* 87: 1206–1211.

Kimata, M., Inagaki, N. and Nagai, H. 2000a. Effects of luteolin and other flavonoids on IgE-mediated allergic reactions. *Planta Medica* 66: 25–29.

Kimata, M., Shichijo, M., Miura, T., Serizawa, I., Inagaki, N. and Nagai, H. 2000b. Effects of luteolin, quercetin and baicalein on immunoglobulin E-mediated mediator release from human cultured mast cells. *Clinical & Experimental Allergy* 30: 501–508.

Kim, D.K., Lee, J.H., Kim, J.W., Kim, H.S., Kim, A.R., Kim, B.K., Yi, K.Y., Park, H.J., Park, D.K. and Choi, W.S. 2011. A novel imidazo[1,5-b]isoquinolinone derivative, U63A05, inhibits syk activation in mast cells to suppress IgE-mediated anaphylaxis in mice. *Journal of Pharmaceutical Sciences* 115: 500–508.

Kim, K.M., Lee, S.B., Lee, S.H., Lee, Y.K. and Kim, K.N. 2005. Comparison of Validity between WST-1 and MTT Test in Bioceramic Materials. *Key Engineering Materials* 284-286: 585-588.

Kim, M.S., Rådinger, M. and Gilfillan, A.M. 2008. The multiple roles of phosphoinositide 3-kinase in mast cell biology. *Trends in Immunology* 29: 493-501.

Kim, N.H., Jeong, H.J. and Kim, H.M. 2012. Theanine is a candidate amino acid for pharmacological stabilization of mast cells. *Amino Acids* 42: 1609–1618.

Kitaura, J., Asai, K., Maeda-Yamamoto, M., Kawakami, Y., Kikkawa, U. and Kawakami, T. 2000. Akt-dependent cytokine production in mast cells. *Journal of Experimental Medicine* 192: 729–740.

Knani, J., Campbell, A., Enander, I., Peterson, C.G., Michel, F.B. and Bousquet, J. 1992. Indirect evidence of nasal inflammation assessed by titration of inflammatory mediators and enumeration of cells in nasal secretions of patients with chronic rhinitis. *Journal of Allergy and Clinical Immunology* 90: 880-809.

© COPYRIGHT UPM

Komiyama, H., Miyake, K., Asai, K., Mizuno, K. and Shimada. T. 2014. Cyclical mechanical stretch enhances degranulation and IL-4 secretion in RBL-2H3 mast cells. *Cell biochemistry and function* 32: 70-76.

Kovarova, M. 2013. Isolation and Characterization of Mast Cells in Mouse Models of Allergic Diseases. *Methods in Molecular Biology* 1032: 109-119.

Krause, K., Spohr, A., Zuberbier, T., Church, M.K. and Maurer, M. 2013. Up-dosing with bilastine results in improved effectiveness in cold contact urticaria. *Allergy* 68: 921-928.

Krystel-Whittemore, M., Dileepan, K.N. and Wood, J.G. 2016. Mast Cell: A Multi-Functional Master Cell. *Frontiers in Immunology* 6: 620.

Kurisaki, K., Kurisaki, A., Valcourt, U., Terentiev, A.A., Pardali, K., Ten Dijke, P., Heldin, C.H., Ericsson, J. and Moustakas, A. 2003. Nuclear Factor YY1 Inhibits Transforming Growth Factor {beta}- and Bone Morphogenetic Protein-Induced Cell Differentiation. *Molecular and Cellular Biology* 23: 4494-4510.

Kustermans, G., Piette, J. and Legrand-Poels, S. 2008. Actin-targeting natural compounds as tools to study the role of actin cytoskeleton in signal transduction. *Biochemical Pharmacology* 76: 1310-1322.

Kwon, J.H., Lee, J.H., Kim, K.S., Chung, Y.W. and Kim, I.Y. 2012. Regulation of cytosolic phospholipase A2 phosphorylation by proteolytic cleavage of annexin A1 in activated mast cells. *Journal of Immunology* 188: 5665-5673.

Lee, E.J., Ji, G.E. and Sung, M.K. 2010. Quercetin and kaempferol suppress immunoglobulin E-mediated allergic inflammation in RBL-2H3 and Caco-2 cells. *Inflammation Research* 59: 847–854.

Lee, M., Lee, N.Y., Chung, K.S., Cheon, S.Y., Lee, K.T. and An, H.J., 2017. Roxatidine attenuates mast cell-mediated allergic inflammation via inhibition of NF-κB and p38 MAPK activation. *Scientific Reports* 7: 41721.

Lee, Y.Z., Shaari, K., Cheema, M.S., Tham, C.L., Sulaiman, M.R. and Israf, D.A. 2017. An orally active geranyl acetophenone attenuates airway remodeling in a murine model of chronic asthma. *European Journal of Pharmacology* 797: 53-64.

Leung, K.B.P., Flint, K.C., Brostoff, J., Hudspith, B.N., Johnson, N., Lau, H.Y.A., Liu, W.L. and Pearce, F.L. 1988. Effects of sodium cromoglycate and nedocromil sodium on histamine secretion from human lung mast cells. *Thorax* 43: 756-761.

Levi-Schaffer, F., Slovik, D., Armetti, L., Pickholtz, D. and Touitou, E. 2000. Activation and inhibition of mast cells degranulation affect their morphometric parameters. *Life Sciences* 66: PL283-290.

© COPYRIGHT UPM

Lian, Q., Cheng, Y., Zhong, C. and Wang, F. 2015. Inhibition of the IgE-mediated activation of RBL-2H3 cells by TIPP, a novel thymic immunosuppressive pentapeptide. *International Journal of Molecular Sciences* 16: 2252-2268.

Lindquist, J.A. Simeoni, L. and Schraven, B. 2003. Transmembrane adapters: attractants for cytoplasmic effectors. *Immunological Reviews* 191: 165–182.

Lin, L.L., Wartmann, M., Lin, A.Y., Knopf, J.L., Seth, A. and Davis, R.J. 1993. $cPLA_2$ is phosphorylated and activated by MAP kinase. *Cell* 72: 269-278.

Li, Y., Lee, S.H., Le, Q.T., Kim, M.M. and Kim, S.K. 2008. Anti-allergic effects of phlorotannins on histamine release via binding inhibition between IgE and Fc epsilonRI. J. Agric*. Food Chemistry* 56: 12073-12080.

Locksley, R.M. 2010. Asthma and allergic inflammation. *Cell* 140: 777-783.

Lorentz, A., Klopp, I., Gebhardt, T., Manns, M.P. and Bischoff, S.C. 2003. Role of activator protein 1, nuclear factor-kappaB, and nuclear factor of activated T cells in IgE receptor-mediated cytokine expression in mature human mast cells. *Journal of Allergy and Clinical Immunology* 111: 1062-1068.

Lusková, P. and Dráber, P. 2004. Modulation of the Fcε receptor I signaling by tyrosine kinase inhibitors: search for therapeutic targets of inflammatory and allergy diseases. *Current Pharmaceutical Design* 10: 1727-1737.

Lu, Y., Li, Y., Jin, M., Yang, J.H., Li, X., Chao, G.H., Park, H.H., Park, Y.N., Son, J.K., Lee, E. and Chang, H.W., 2012a. *Inula japonica* extract inhibits mast cell-mediated allergic reaction and mast cell activation. *Journal of Ethnopharmacology* 143, 151-157.

Lu, Y., Li, Y., Seo, C.S., Murakami, M., Son, J.K. and Chang, H.W. 2012b. Saucerneol D inhibits eicosanoid generation and degranulation through suppression of Syk kinase in mast cells. *Food and Chemical Toxicology* 50: 4382-4388.

Lu, Y., Yang, J.H., Li, X., Hwangbo, K., Hwang, S.L., Taketomi, Y., Murakami, M., Chang, Y.C., Kim, C.H., Son, J.K. and Chang, H.W. 2011. Emodin, a naturally occurring anthraquinone derivative, suppresses IgE-mediated anaphylactic reaction and mast cell activation. *Biochemical Pharmacology* 82: 1700-1708.

Mahmood, T. and Yang, P.C. 2012. Western Blot: Technique, Theory, and Trouble Shooting. *North American Journal of Medical Sciences* 4: 429-434.

Ma, H.T. and Beaven, M.A. 2011. Regulators of Ca^{2+} signalling in mast cells: potential targets for treatment of mast cell-related diseases? *Advances in Experimental Medicine and Biology* 716: 62-90.

Malacombe, M., Bader, M.F. and Gasman, S. 2006. Exocytosis in neuroendocrine cells: new tasks for actin. *Biochimica et Biophysica Acta* 1763: 1175-1183.

© COPYRIGHT UPM

Malaviya, R. and Abraham, S.N. 2000. Role of mast cell leukotrienes in neutrophil recruitment and bacterial clearance in infectious peritonitis. *Journal of Leukocyte Biology* 67: 841–846.

Malaviya, R. and Jakschik, B.A. 1993. Reversible translocation of 5-lipoxygenase in mast cells upon IgE/antigen stimulation. *Journal of Biological Chemistry* 268: 4939-4944.

Mario, S.B., Juan, C.I., Noel, R.P. and Ignacio, A. 2013. Evidence based approaches to diagnosis and management. In *WAO White Book on Allergy update 2013*, eds. R. Pawankar, G.W. Canonica, S.T. Holgate, R.F. Lockey, M.S. Blaiss, pp. 109-113. USA.

Marquardt, D.L. and Walker, L.L. 2000. Dependence of mast cell IgE-mediated cytokine production on nuclear factor-kappaB activity. *Journal of Allergy and Clinical Immunology* 105: 500-505.

Marshall, J.S. 2004. Mast-cell responses to pathogens. *Nature Reviews. Immunology* 4: 787–799.

Martin, V.V., Beierlein, M., Morgan, J.L., Rothe, A. and Gee, K.R. 2004. Novel Fluo-4 analogues for fluorescent calcium measurements. *Cell Calcium* 36: 509-514.

Masoli, M., Fabian, D., Holt, S. and Beasley, R. 2004. The global burden of asthma: executive summary of the GINA Dissemination Committee Report. *Allergy* 59: 469-478.

Mastuda, H., Morikawa, T., Ueda, K., Managi, H. and Yoshikawa, M. 2002. Structural requirements of flavonoids for inhibition of antigen-Induced degranulation, TNF-alpha and IL-4 production from RBL-2H3 cells. *Bioorganic & Medicinal Chemistry* 10: 3123–3128.

Matsuda, K, Piliponsky, A.M., Iikura, M., Nakae, S., Wang, E.W., Dutta, S.M., Kawakami, T., Tsai, M. and Galli, S.J., 2005. Monomeric IgE enhances human mast cell chemokine production: IL-4 augments and dexamethasone suppresses the response. *Journal of Allergy and Clinical Immunology* 116, 1357–1363.

Matsui, T., Ito, C., Furukawa, H., Okada, T. and Itoigawa, M. 2013. Lansiumamide B and SB-204900 isolated from Clausena lansium inhibit histamine and TNF-a release from RBL-2H3 cells. *Inflammation Research* 62, 333-341.

Mazuc, E., Villoutreix, B.O., Malbec, O., Roumier, T., Fleury, S., Leonetti, J.P. Dombrowicz, D., Daëron, M., Martineau, P. and Dariavach, P. 2008. A novel druglike spleen tyrosine kinase binder prevents anaphylactic shock when administered orally. *Journal of Allergy and Clinical Immunology* 122: 188–194.

Mestas, J. and Hughes, C.C. 2004. Of mice and not men: differences between mouse and human immunology. *Journal of immunology* 172: 2731-2738.

© COPYRIGHT UPM

Metcalfe, D.D., Baram, D. and Mekori, Y.A. 1997. Mast cells. *Physiological Reviews* 77: 1033–1079.

Metcalfe, D.D., Kaliner, M. and Donlon, M.A., 1981. The mast cell. *Critical Reviews™ in Immunology* 3: 23-74.

Metcalfe, D.D., Peavy, R.D. and Gilfillan, A.M. 2009. Mechanisms of mast cell signalling in anaphylaxis. *Journal of Allergy and Clinical Immunology* 124: 639-646.

Meyer, U. 2002. From khellin to sodium cromoglycate--a tribute to the work of Dr. R. E. C. Altounyan (1922-1987). *Die Pharmazie* 57: 62-69.

Migalovich-Sheikhet, H., Friedman, S., Mankuta, D. and Levi-Schaffer, F. 2012. Novel identified receptors on mast cells. *Frontiers in Immunology* 3: 238.

Mishra, B.B. and Tiwari, V.K. 2011. Natural products: An evolving role in future drug discovery. *European Journal of Medicinal Chemistry* 46: 4769–4807.

Miyata, N., Gon, Y., Nunomura, S., Endo, D., Yamashita, K., Matsumoto, K., Hashimoto, S. and Ra, C. 2008. Inhibitory effects of parthenolide on antigen-induced microtubule formation and degranulation in mast cells. *International Immunopharmacology* 8: 874–880.

Moon, T.C., Murakami, M., Ashraf, M.D., Kudo, I. and Chang, H.W. 1998. Regulation of cyclooxygenase-2 and endogenous cytokine expression by bacterial lipopolysaccharide that acts in synergy with c-kit ligand and Fc epsilon receptor I crosslinking in cultured mast cells. *Cellular Immunology* 185: 146–152.

Moran, L.A., Horton, R.A., Scrimgeour, G. and Perry, M. 2012. Principles of Biochemistry. 5th ed. New York: Pearson Education.

Moriya, K., Rivera, J., Odom, S., Sakuma, Y., Muramato, K., Yoshiuchi, T., Miyamoto, M. and Yamada, K., 1997. ER-27319, an acridine-related compound, inhibits release of antigen-induced allergic mediators from mast cells by selective inhibition of Fcε receptor I-mediated activation of Syk. *Proceedings of the National Academy of Sciences of USA* 94: 12539-12544.

Mousli, M., Bronner, C., Bueb, J.L., Tschirhart, E., Gies, J.P. and Landry, Y. 1989. Activation of rat peritoneal mast cells by substance P and mastoparan. *Journal of Pharmacology and Experimental Therapeutics* 250: 329–335.

Muraro, A., Roberts, G., Clark, A., Eigenmann, P.A., Halken, S., Lack, G., Moneret-Vautrin, A., Niggemann, B., Rancé, F. and EAACI Task Force on Anaphylaxis in Children. 2007. The management of anaphylaxis in childhood: position paper of the European Academy of Allergology and Clinical Immunology. *Allergy* 62: 857-871.

© COPYRIGHT UPM

Muraro, A., Roberts, G., Worm, M., Bilò, M.B., Brockow, K., Fernández, R.M., Santos, A.F., Zolkipli, Z.Q., Bellou, A., Beyer, K., Bindslev-Jensen, C., Cardona, V., Clark, A.T., Demoly, P., Dubois, A.E., DunnGalvin, A., Eigenmann, P., Halken, S., Harada, L., Lack, G., Jutel, M., Niggemann, B., Ruëff, F., Timmermans, F., Vlieg-Boerstra, B.J., Werfel, T., Dhami, S., Panesar, S., Akdis, C.A. and Sheikh, A. 2014. EAACI Food Allergy and Anaphylaxis Guidelines Group. *Allergy* 69: 1026-1045.

Murray, J.J., Tonnel, A.B., Brash, A.R., Roberts, L.J.2nd., Gosset, P., Workman, R., Capron, A. and Oates, J.A. 1986. Release of prostaglandin D2 into human airways during acute antigen challenge. *The New England Journal of Medicine* 315: 800-804.

National Institute of Allergy and Infectious Diseases. Something in the Air: Airborne Allergens. NIH Publication No. 03-7045: USA, 2003.

Ng, C.H., Rullah, K., Aluwi, M.F., Abas, F., Lam, K.W., Ismail, I.S., Narayanaswamy, R., Jamaludin, F. and Shaari, K. 2014. Synthesis and Docking Studies of 2,4,6-Trihydroxy-3Geranylacetophenone Analogs as Potential Lipoxygenase Inhibitor. *Molecules* 19: 11645-11659.

Ng, N.M., Jiang, S.P. and Zhang, W. 2012. 2-Aminoethoxydiphenyl borate reduces degranulation and release of cytokines in a rat mast cell line. *European Review for Medical and Pharmacological Sciences* 16: 1017-1021.

Nishida, K., Yamasaki, S., Ito, Y., Kabu, K., Hattori, K., Tezuka, T., Nishizumi, H., Kitamura, D., Goitsuka, R., Geha, R.S., Yamamoto, T., Yagi, T. and Hirano, T. 2005. FcεRI-mediated mast cell degranulation requires calcium-independent microtubule-dependent translocation of granules to the plasma membrane. *Journal of Cell Biology* 170: 115-126.

Norman, P. 2011. Selective PI3Kd inhibitors, a review of the patent literature. *Expert Opinion on Therapeutic Patents* 21: 1773-90.

Nugroho, A.E., Ikawati, Z., Sardjiman and Maeyama, K. 2009. Effects of benzylidenecyclopentanone analogues of curcumin on histamine release from mast cells. *Biological and Pharmaceutical Bulletin* 32: 842–849.

Odom, S., Gomez, G., Kovarova, M., Furumoto, Y., Ryan, J.J., Wright, H.V., Gonzalez-Espinosa, C., Hibbs, M.L., Harder, K.W. and Rivera, J. 2004. Negative regulation of immunoglobulin E-dependent allergic responses by Lyn kinase. *Journal of Experimental Medicine* 199: 1491–1502.

Office of Animal Resources. 2014. IACUC Guidelines: Anesthesia by University of Lowa. Retrieved 20 June 2016 from http://animal.research.uiowa.edu/iacuc-guidelines-anesthesia

O'Hollaren, M.T., Yunginger, J.W., Offord, K.P., Somers, M.J., O'Connell, E.J., Ballard, D.J. and Sachs, M.I. 1991. Exposure to an aeroallergen as a possible precipitating factor in respiratory arrest in young patients with asthma. *The New England Journal of Medicine* 324: 359-363.

© COPYRIGHT UPM

Ok, H.C. and Chang H.S. 2015. Role of mast cell in the late phase of contact hypersensitivity induced by *trimellitic anhydride*. *Anatomy & Cell Biology* 48: 225–234.

Oka, T., Sato, K., Hori, M., Ozaki, H. and Karaki, H. 2002. Xestospongin C, a novel blocker of IP3 receptor, attenuates the increase in cytosolic calcium level and degranulation that is induced by antigen in RBL-2H3 mast cells. *British Journal of Pharmacology* 135: 1959–1966.

Okkenhaug, K. 2013. Two birds with one stone: dual p110δ and p110γ inhibition. *Chemistry & Biology* 20: 1309–1310.

Onose, J., Yoshioka, Y., Ye, Y.Q., Sugaya, K., Yajima, A., Taniguchi, K., Okada, K., Yajima, S., Takahashi, S., Koshino, H. and Abe, N. 2012. Inhibitory effects of vialinin A and its analog on tumor necrosis factor-α release and production from RBL-2H3 cells. *Cellular immunology* 279: 140-144.

Oppenheimer, J.J. and Casale, T.B. 2002. Next generation antihistamines: therapeutic rationale, accomplishments and advances. *Expert Opinion on Investigational Drugs* 11: 807–817.

Ortega, E., Schweitzer-Stenner, R. and Pecht, I. 1988. Possible orientational constraints determine secretory signals induced by aggregation of IgE receptors on mast cells. *EMBO Journal* 7: 4101–4109.

Orr, S.J. and McVicar, D.W. 2011. LAB/NTAL/Lat2: a force to be reckoned with in all leukocytes? *Journal of Leukocyte Biology* 89: 11-19.

Orr, T.S.C., Hall, D.E., Gwilliam, J.M. and Cox, J.S.G. 1971. The effect of cromolyn sodium on the release of histamine and degranulation of rat mast cells induced by Compound 48/80. *Life Sciences* 10: 805–812.

Oskeritzian, C.A., Zhao, W., Min, H.K., Xia, H.Z., Pozez, A., Kiev, J. and Schwartz, L.B. 2005. Surface cd88 functionally distinguishes the MCTC from the mct type of human lung mast cell. *Journal of Allergy and Clinical Immunology* 115: 1162-1168.

Overed-Sayer, C., Rapley, L., Mustelin, T. and Clarke, D.L. 2014. Are mast cells instrumental for fibrotic diseases? *Frontiers in Pharmacology* 4: 174.

Park, H.H., Lee, S., Son, H.Y., Park, S.B., Kim, M.S., Choi, E.J. Singh, T.S., Ha, J.H., Lee, M.G., Kim, J.E., Hyun, M.C., Kwon, T.K., Kim, Y.H. and Kim, S.H. 2008. Flavonoids inhibit histamine release and expression of proinflammatory cytokines in mast cells. *Archives of Pharmacal Research* 31: 1303–1311.

Parravicini, V., Gadina. M., Kovarova, M., Odom, S., Gonzalez-Espinosa, C., Furumoto, Y., Saitoh, S., Samelson, L.E., O'Shea, J.J. and Rivera, J. 2002. Fyn kinase initiates complementary signals required for IgE-dependent mast cell degranulation. *Nature Immunology* 3: 741–748.

© COPYRIGHT UPM

Passante, E., Ehrhardt, C., Sheridan, H. and Frankish, N. 2009. RBL-2H3 cells are an imprecise model for mast cell mediator release. *Inflammation Research* 58: 611-618.

Passante, E. and Frankish, N. 2009. The RBL-2H3 cell line: its provenance and suitability as a model for the mast cell. *Inflammatory Research* 58: 737-745.

Pawankar, R., Canonica, G.W., Holgate, S.T., Lockey, R.F. and Blaiss, M.S. 2013. Allergic Diseases as a Global Public Health Issue. In *WAO White Book on Allergy update 2013*, eds. R. Pawankar, G.W. Canonica, S.T. Holgate, R.F. Lockey, M.S. Blaiss, pp. 86-90. USA.

Pearce, N., Ait-Khaled, N., Beasley, R., Mallol, J., Koll, U., Mitchel, E. and Robertson, C. 2007. Worldwide trends in the prevalence of asthma symptoms: Phase III of the International Study of Asthma and Allergies in Childhood (ISAAC). *Thorax* 62: 758-766.

Peavy, R.D. and Metcalfe, D.D. 2009. Understanding the mechanisms of anaphylaxis. *Current Opinion in Allergy and Clinical Immunology* 8: 310–315.

Pendleton, A. and Koffer, A. 2001. Effects of latrunculin reveal requirements for the actin cytoskeleton during secretion from mast cells. *Cell Motility and the Cytoskeleton* 48: 37-51.

Penissi, A.B., Vera, M.E., Mariani, M.L., Rudolph, M.I., Cenal, J.P., de Rosas, J.C., Fogal, T.H., Tonn, C.E., Favier, L.S., Giordano, O.S. and Piezzi, R.S. 2009. Novel anti-ulcer alpha,beta-unsaturated lactones inhibit compound 48/80-induced mast cell degranulation. *European Journal of Pharmacology* 612: 122–130.

Pfeiffer, J.R., Seagrave, J.C., Davis, B.H., Deanin, G.G. and Oliver, J.M. 1985. Membrane and cytoskeletal changes associated with IgE-mediated serotonin release from rat basophilic leukemia cells. *Journal of Cell Biology* 101: 2145-2155.

Pickett, J.A. and Edwardson, J.M. 2006. Compound exocytosis: mechanisms and functional significance. *Traffic* 7: 109–116.

Plaut, M., Pierce, J.H., Watson, C.J., Hanley-Hyde, J., Nordan, R.P. and Paul, W.E. 1989. Mast cell lines produce lymphokines in response to cross-linkage of FcεRI or to calcium ionophores. *Nature* 339: 64–67.

Qiao, H., Andrade, M.V., Lisboa, F.A., Morgan, K. and Beaven, M.A. 2006. FcepsilonR1 and toll-like receptors mediate synergistic signals to markedly augment production of inflammatory cytokines in murine mast cells. *Blood* 107: 610-618.

Quader, A., Armstrong, J.A., Gray, A.I., Hartley, T.G. and Waterman, P.G. 1991. Chemosystematics of *Acradenia* and general significance of acetophenones in the Rutaceae. *Biochemical Systematics and Ecology* 19: 171-176.

© COPYRIGHT UPM

Ra, C., Jouvin, M.H. and Kinet, J.P. 1989. Complete structure of the mouse mast cell receptor for IgE (FcεRI) and surface expression of chimeric receptors (rat–mouse–human) on transfected cells. *Journal of Biological Chemistry* 264: 15323–15327.

Rao, D.D., Vorhies, J.S., Senzer, N. and Nemunaitis, J. 2009. siRNA vs. shRNA: similarities and differences. *Advanced Drug Delivery Reviews* 61: 746-759.

Rao, K. and Brown, M. 2008. Mast cells: multifaceted immune cells with diverse roles in health and disease. *Annals of the New York Academy of Sciences* 1143: 83-104.

Repetto, B., Bandara, G., Kado-Fong, H., Larigan, J.D. and Kochan, J.P. 1996. Functional contribution of the FcεRIα and FcεRIγ subunit domains in FcεRI-mediated signalling in mast cells. *Journal of Immunology* 156: 4876–4883.

Rey-Ladino, J., Ross, A.G., Cripps, A.W., McManus, D.P. and Quinn, R. 2011. Natural products and the search for novel vaccine adjuvants. *Vaccine* 29: 6464–6471.

Reynolds, A., Leake, D., Boese, Q., Scaringe, S., Marshall, W.S. and Khvorova, A. 2004. Rational siRNA design for RNA interference. *Nature Biotechnology* 22: 326-330.

Riccaboni, M., Bianchi, I. and Petrillo, P. 2010. Spleen tyrosine kinases: biology, therapeutic targets and drugs. *Drug Discovery Today* 15: 517-530.

Ricciotti, E. and FitzGerald, G.A. 2011. Prostaglandins and inflammation. *Arteriosclerosis, Thrombosis, and Vascular Biology* 31: 986-1000.

Richard, F.L., Stephen, F.K., Philip, L.L. and Aziz, S. 2013. Anaphylaxis. In WAO White Book on Allergy update 2013, eds. R. Pawankar, G.W. Canonica, S.T. Holgate, R.F. Lockey, M.S. Blaiss, pp. 48-53. USA.

Rivera, J. and Gilfillan, A.M. 2006. Molecular regulation of mast cell activation. *Journal of Allergy and Clinical Immunology* 117: 1214–1225.

Rivera, J. 2005. NTAL/LAB and LAT: a balancing act in mast-cell activation and function. *Trends in Immunology* 26: 119-122.

Rivera, J. 2002. Molecular adapters in FcεRI signalling and the allergic response. *Current Opinion in Allergy and Clinical Immunology* 14: 688–693.

Röhlich, P., Anderson, P. and Uvnäs, B. 1971. Electron microscope observation on compound 48/80-induced degranulation in mast cells. *Journal of Cell Biology* 51: 465–483.

Rossi, A.B., Herlaar, E., Braselmann, S., Huynh, S., Taylor, V., Frances, R. Issakani, S.D., Argade, A., Singh, R., Payan, D.G. and Masuda, E.S. 2006. Identification of the Syk kinase inhibitor R112 by a human mast cell screen. *Journal of Allergy and Clinical Immunology* 118: 749–755.

© COPYRIGHT UPM

Ruster, G.U., Hoffmann, B. and Hamburger, M. 2004. Inhibitory activity of indolin-2-one derivatives on compound 48/80-induced histamine release from mast cells. *Die Pharmazie* 59: 236–237.

Saitoh, S., Arudchandran, R., Manetz, T.S., Zhang, W., Sommers, C.L., Love, P.E., Rivera, J. and Samelson, L.E. 2000. LAT is essential for FcεRI-mediated mast cell activation. *Immunity* 12: 525–535.

Saitoh, S., Odom, S., Gomez, G., Sommers, C.L., Young, H.A., Rivera, J. and Samelson, L.E. 2003. The four distal tyrosines are required for LAT-dependent signaling in Fc3RI-mediated mast cell activation. *Journal of Experimental Medicine* 198: 831–843.

Sakai, S., Sugawara, T., Kishi, T., Yanagimoto, K. and Hirata, T. 2010. Effect of glucosamine and related compounds on the degranulation of mast cells and ear swelling induced by dinitrofluorobenzene in mice. *Life Sciences* 86: 337-343.

Samelson, L.E. 2002. Signal transduction mediated by the T cell antigen receptor: The role of adapter proteins. *Annual Review of Immunology* 20: 371–394.

Sampson, H.A., Munoz-Furlong, A., Bock, S.A., Schmitt, C., Bass, R., Chowdhury, B.A., Decker, W.W., Furlong, T.J., Galli, S.J., Golden, D.B., Gruchalla, R.S., Harlor, A.D.Jr., Hepner, D.L., Howarth, M., Kaplan, A.P., Levy, J.H., Lewis, L.M., Lieberman, P.L., Metcalfe, D.D., Murphy, R., Pollart, S.M., Pumphrey, R.S., Rosenwasser, L.J., Simons, F.E., Wood, J.P. and Camargo, C.A.Jr. 2005. Symposium on the definition and management of anaphylaxis: summary report. *Journal of Allergy and Clinical Immunology* 115: 584–591.

Sampson, H.A., Munoz-Furlong, A., Campbell, R.L., Adkinson, N.F., Bock, S.A., Branum, A.B., Brown, S.G., Camargo, C.A.Jr., Cydulka, R., Galli, S.J., Gidudu, J., Gruchalla, R.S., Harlor, A.D.Jr, Hepner, D.L., Lewis, L.M., Lieberman, P.L., Metcalfe, D.D., O'Connor, R., Muraro, A., Rudman, A., Schmitt, C., Scherrer, D., Simons, F.E., Thomas, S., Wood, J.P. and Decker, W.W. 2006. Second symposium on the definition and management of anaphylaxis: summary report Second National Institute of Allergy and Infectious Disease/Food Allergy and Anaphylaxis Network symposium. *Journal of Allergy and Clinical Immunology* 117: 391–397.

Samuelsson, B. 1987. An elucidation of the arachidonic acid cascade. Discovery of prostaglandins, thromboxane and leukotrienes. *Drugs* 33: 2-9.

Sanchez-Miranda, E., Ibarra-Sanchez, A. and Gonzalez-Espinosa, C. 2010. Fyn kinase controls FcεRI receptor-operated calcium entry necessary for full degranulation in mast cells. *Biochemical and Biophysical Research Communications* 391: 1714-1720.

Savage, J. and Johns, C.B. 2015. Food allergy: epidemiology and natural history. *Immunology and Allergy Clinics of North America* 35: 45–59.

© COPYRIGHT UPM

Schroeder, J.T. 2011. Basophils: emerging roles in the pathogenesis of allergic disease. *Immunological Reviews* 242: 144–160.

Schwartz, L.B., Irani, A.M., Roller, K., Castells, M.C. and Schechter, N.M. 1987. Quantitation of histamine, tryptase, and chymase in dispersed human T and TC mast cells. *Journal of Immunology* 138: 2611-2615.

Schwarzer, G., Bassler, D., Mitra, A., Ducharme, F.M. and Forster, J. 2004. Ketotifen alone or as additional medication for long-term control of asthma and wheeze in children. Cochrane Database Systematic Reviews. 2004: CD001384.

Sears, M.R., Herbison, G.P., Holdaway, M.D., Hewitt, C.J., Flannery, E.M. and Silva, P.A. 1989. The relative risks of sensitivity to grass pollen, house dust mite and cat dander in the development of childhood asthma. *Clinical and Experimental Allergy* 19: 419-424.

Shaari, K., Safri, S., Abas, F., Lajis, N.H. and Israf, D.A. 2006. A geranylacetophenone from the leaves of *Melicope ptelefolia*. *Natural Product Research* 20: 415-419.

Shaari, K., Suppaiah, V., Wai, L.K., Stanslas, J., Tejo, B.A., Israf, D.A., Abas, F., Ismail, I.S., Shuaib, N.H., Zareen, S. and Lajis, N.H. 2011. Bioassay-guided identification of an anti-inflammatory prenylated acylphloroglucinol from *Melicope ptelefolia* and molecular insights into its interaction with 5-lipoxygenase. *Bioorganic & Medicinal Chemistry* 9: 6340-6347.

Sheinkopf, L.E., Rafi, A.W., Do, L.T., Katz, R.M. and Klaustermeyer, W.B. 2008. Efficacy of omalizumab in the treatment of atopic dermatitis: a pilot study. *Allergy & Asthma Proceedings* 29: 530–537.

Sheridan, H., Walsh, J.J., Jordan, M., Cogan, C. and Frankish, N. 2009. A series of 1, 2-coupled indane dimers with mast cell stabilisation and smooth muscle relaxation properties. *European Journal of Medicinal Chemistry* 44: 5018–5022.

Shibata, H. and Yagi, T. 1993. Rate assay of N-acetyl-beta-D-hexosaminidase with 4-nitrophenyl N-acetyl-beta-D-glucosaminide as an artificial substrate. *Clinica Chimica Acta* 251: 53-64.

Shoji, N., Umeyama, A., luchi, A., Saito, N. and Ariahara, S. 1989. Two novel alkaloids from *Eudia rutaceacarpa*. *Journal of Natural Products* 52: 1160-1162.

Shuiba, N.H., Shaari, K. Khatiba, A., Maulidiania, Kneer, R., Zareena, S., Raofa, S.M., Lajisa, N.H. and Neto, V. 2011. Discrimination of young and mature leaves of Melicope ptelefolia using 1H NMR and multivariate data analysis. *Food Chemistry* 126: 640-645.

© COPYRIGHT UPM

Shumilina, E., Lam, R.S., Wölbing, F., Matzner, N., Zemtsova, I.M., Sobiesiak, M., Mahmud, H., Sausbier, U., Biedermann, T., Ruth, P., Sausbier, M. and Lang, F. 2008. Blunted IgE-mediated activation of mast cells in mice lacking the Ca^{2+}-activated K^{+} channel KCa3.1. *Journal of Immunology* 180: 8040-8047.

Sibilano, R., Frossi, B. and Pucillo, C.E. 2014. Mast cell activation: a complex interplay of positive and negative signalling pathways. *European Journal of Immunology* 44: 2558-2566.

Sicherer, S.H. and Sampson, H.A. 2006. Food allergy. *Journal of Allergy and Clinical Immunology* 117: 470-475.

Sicherer, S. and Leung D. 2010. Advances in allergic skin disease, anaphylaxis, and hypersensitivity reactions to foods, drugs, and insects in 2009. *Journal of Allergy and Clinical Immunology* 125: 85-97.

Sicherer, S.H. and Sampson, H.A. 2010. Food allergy. *Journal of Allergy and Clinical Immunology* 125: 116-125.

Silver, R. and Curley, J.P. 2013. Mast cells on the mind: new insights and opportunities. *Trends in Neurosciences* 36: 513-521.

Simmons, D.L. 2013. Targeting kinases: a new approach to treating inflammatory rheumatic diseases. *Current Opinion in Pharmacology* 13: 426-434.

Simons, F.E.R., Ardusso, L.R.F., Bilò, M.B., l-Gamal, Y.M., Ledford, D.K., Ring, J., Sanchez-Borges, M., Senna, G.E., Sheikh, A., Thong, B.Y. and World Allergy Organization. 2011. World Allergy Organization anaphylaxis guidelines: Summary. *Journal of Allergy and Clinical Immunology* 127: 587-593.

Simons, F.E. 2008. 9. Anaphylaxis. *Journal of Allergy and Clinical Immunology* 121: S402–S407.

Singh, B, Nadkarni, J.R., Vishwakarma, R.A., Bharate, S.B., Nivsarkar, M. and Anandjiwala, S. 2012. The hydroalcoholic extract of *Cassia alata* (Linn.) leaves and its major compound rhein exhibits antiallergic activity via mast cell stabilization and lipoxygenase inhibition. *Journal of Ethnopharmacology* 141: 469-473.

Singh, R. and Masuda, E.S., 2007. Chapter 24 Spleen Tyrosine Kinase (Syk) Biology, Inhibitors and Therapeutic Applications. *Annual Reports in Medicinal Chemistry* 42: 379-391.

Sinniah, A., Yazid, S., Perretti, M., Solito, E. and Flower, R.J., 2016. The role of the Annexin-A1/FPR2 system in the regulation of mast cell degranulation provoked by compound 48/80 and in the inhibitory action of nedocromil. *International Immunopharmacology* 32: 87-95.

© COPYRIGHT UPM

Siraganian, R.P., de Castro, R.O., Barbu, E.A. and Zhang, J., 2010. Mast cell signaling: the role of protein tyrosine kinase Syk, its activation and screening methods for new pathway participants. *FEBS Letters* 584: 4933-4940.

Siraganian, R.P., McGivney, A., Barsumian, E.L., Crews, F.T., Hirata, F. and Axelrod, J. 1982. Variants of the rat basophilic leukaemia cell line for the study of histamine release. *Federation proceedings* 41: 30–34.

Soderstrom, L., Kober, A. and Ahlstedt, S. 2003. A further evaluation of the clinical use of specific IgE antibody testing in allergic diseases. *Allergy* 58: 921-928.

Son, J.K., Son, M.J., Lee, E., Moon, T.C., Son, K.H., Kim, C.H., Kim, H.P., Kang, S.S. and Chang, H.W. 2005. Ginkgetin, a Biflavone from Ginko biloba leaves, inhibits cyclooxygenases-2 and 5-lipoxygenase in mouse bone marrow-derived mast cells. *Biological and Pharmaceutical Bulletin* 28: 2181-2184.

Spinas, E., Kritas, S.K., Saggini, A., Mobili, A., Caraffa, A., Antinolfi, P., Pantalone, A., Tei, M., Speziali, A., Saggini, R. and Conti, P. 2014. Role of mast cells in atherosclerosis: a classical inflammatory disease. *International Journal of Immunopathology and Pharmacology* 27: 517-521.

Sridharan, G. and Shankar, A.A. 2012. Toluidine blue: A review of its chemistry and clinical utility. *Journal of Oral and Maxillofacial Pathology* 16: 251-255.

Stephens, L., Anderson, K., Stokoe, D., Erdjument-Bromage, H., Painter, G.F., Holmes, A.B., Gaffney, P.R., Reese, C.B., McCormick, F., Tempst, P., Coadwell, J. and Hawkins, P.T. 1998. Protein kinase B kinases that mediate phosphatidylinositol 3,4,5-trisphosphate dependent activation of protein kinase B. *Science* 279: 710–714.

Stephen, T.H., Giorgio W.C., Carlos, E.B.C., Thomas, B.C., Myron, Z., Harold, N. and Pakit, V. 2013. Asthma. In *WAO White Book on Allergy update 2013*, eds. R. Pawankar, G.W. Canonica, S.T. Holgate, R.F. Lockey, M.S. Blaiss, pp. 34-39. USA.

St John, A.L. and Abraham, S.N. 2013. Innate immunity and its regulation by mast cells. *Journal of Immunology* 190: 4458–4463.

Stone, K.D., Prussin, C. and Metcalfe, D.D. 2010. IgE, mast cells, basophils, and eosinophils. *Journal of Allergy and Clinical Immunology* 125: S73-80.

Struwe, W.B. and Warren C.E. 2010. High-throughput RNAi screening for N-glycosylation dependent loci in Caenorhabditis elegans. *Methods in Enzymology* 480: 477-493.

Suryati, S. 2005. Phytochemicals and Anti-Inflammatory Activity of *Melicope ptelefolia* Champ Ex Benth. Master's Thesis, Universiti Putra Malaysia, Malaysia.

© COPYRIGHT UPM

Suzuki, H., Takei, M., Yanagida, M., Nakahata, T., Kawakami, T. and Fukamachi, H. 1997. Early and late events in Fc epsilon RI signal transduction in human cultured mast cells. *Journal of Immunology* 159: 5881–5888.

Tan, C.H., Rasool, S. and Johnston, G.A. 2014. Contact dermatitis: allergic and irritant. *Clinics in Dermatology* 32: 116-124.

Tang, J.M., Liu, J. and Wu, W. 2012. Studies on the Degranulation of RBL-2H3 Cells Induced by Traditional Chinese Medicine Injections. *Chinese Medicine* 3: 200-208.

Tang, F., Chen, F., Ling, X., Huang, Y., Zheng, X., Tang, Q. and Tan, X. 2015. Inhibitory effect of methyleugenol on IgE-mediated allergic inflammation in RBL-2H3 cells. *Mediators of Inflammation* 463530. doi: 10.1155/2015/463530.

Theoharides, T.C. and Kalogeromitros, D. 2006. The critical role of mast cells in allergy and inflammation. *Annals of the New York Academy of Sciences* 1088: 78-99.

Thomas, A.E., Platts-Mills., Lee, B.W., Arruda, L.K., Chew, F.T. 2013. Allergens as Risk Factors for Allergic Disease. In *WAO White Book on Allergy update 2013*, eds. R. Pawankar, G.W. Canonica, S.T. Holgate, R.F. Lockey, M.S. Blaiss, pp. 86-90. USA.

Thompson, S.L., Dhar, V., Bond, M.W., Mosmann, T.R., Moore, K.W. and Rennick, D.M. 1991. Interleukin 10: a novel stimulatory factor for mast cells and their progenitors. *Journal of Experimental Medicine* 173: 507-510.

Tkaczyk, C., Beaven, M. A., Brachman, S. M., Metcalfe, D. D. & Gilfillan, A. M. 2003. The phospholipase Cγ-dependent pathway of FcεRI-mediated mast cell activation is regulated independently of phosphatidylinositol 3-kinase. *Journal of Biological Chemistry* 278: 48474–48484.

Tkaczyk, C., Horejsi, V., Iwaki, S., Draber, P., Samelson, L.E., Satterthwaite, A.B., Nahm, D.H., Metcalfe, D.D. and Gilfillan, A.M., 2004. NTAL phosphorylation is a pivotal link between the signaling cascades leading to human mast cell degranulation following Kit activation and FcεRI aggregation. *Blood* 104, 207–214.

Tkaczyk, C., Iwaki, S., Metcalfe, D.D. and Gilfillan, A.M. 2005. Roles of adaptor molecules in mast cell activation. *Chemical Immunology and Allergy* 87: 43–58.

Turnbull, J.L., Adams, H.N. and Gorard, D.A. 2015. Review article: the diagnosis and management of food allergy and food intolerances. *Alimentary Pharmacology & therapeutics* 41: 3-25.

© COPYRIGHT UPM

Vanhaesebroeck, B., Leevers. S.J., Ahmadi, K., Timms, J., Katso, R., Driscoll, P.C., Woscholski, R., Parker, P.J. and Waterfield, M.D. 2001. Synthesis and function of 3-phosphorylated inositol lipids. *Annual Review of Biochemistry* 70: 535–602.

Van Hoecke, H., Vandenbulcke, L. and Van Cauwenberge, P. 2007. Histamine and leukotriene receptor antagonism in the treatment of allergic rhinitis: an update. *Drugs* 67: 2717-2726.

Van, N. H., Kamperdick, Sung, T.V. and Adam, G. 1998. Benzopyan diminers from *Melicope ptelefolia*, *Phytochemistry* 48: 1055-1057.

Velema, W.A., van der Toorn, M., Szymanski, W. and Feringa, B.L., 2013. Design, synthesis, and inhibitory activity of potent, photoswitchable mast cell activation inhibitors. *Journal of Medicinal Chemistry* 56: 4456–4464.

Volna, P., Lebduska, P., Dráberová, L., Símová, S., Heneberg, P., Boubelík, M., Bugajev, V., Malissen, B., Wilson, B.S., Horejsí, V., Malissen, M. and Dráber, P. 2004. Negative regulation of mast cell signalling and function by the adaptor LAB/NTAL. *Journal of Experimental Medicine* 200: 1001–1013.

Von Pirquet, C. 1906. Allergie. *MMW Munch Med Wochenschr* 53: 388–390.

Walker, P.G., Woollen, J.W. and Heyworth, R. 1961. Studies on glucosaminidase. 5. Kidney N-acetyl-β-glucosaminidase and N-acetyl-bgalactosaminidase. *Biochemical Journal* 79: 288–294.

Wang, T. and Brown, M.T. 1999. mRNA quantification by real time TaqMan polymerase chain reaction: validation and comparison with RNase protection. *Analytical Biochemistry* 269: 198-201.

Wang, W., Zhou, Q., Liu, L. and Zou, K. 2012. Anti-allergic activity of emodin on IgE-mediated activation in RBL-2H3 cells. *Pharmacological Reports* 64: 1216-1222.

Wambre, E., James, E.A. and Kwok, W.W. 2012. Characterization of CD4+ T cell subsets in allergy. *Current opinion in immunology* 24: 700-706.

Waserman, S. and Watson W. 2011. Food allergy. *Allergy, Asthma & Clinical Immunology* 7(Suppl 1): S7.

Weinblatt, M.E., Kavanaugh, A., Genovese, M.C., Jones, D.A., Musser, T.K., Grossbard, E.B. and Magilavy, D.B. 2013. Effects of fostamatinib (R788), an oral spleen tyrosine kinase inhibitor, on health-related quality of life in patients with active rheumatoid arthritis: analyses of patient-reported outcomes from a randomized, double-blind, placebo controlled trial. *Journal of Rheumatology* 40: 369-378.

Wendeler, M. and Sandhoff, K. 2009. Hexosaminidase assays. *Glycoconjugate Journal* 26: 945-952.

© COPYRIGHT UPM

Wernersson, S. and Pejler, G. 2014. Mast cell secretory granules: armed for battle. *Nature reviews. Immunology* 14: 478-494.

Wijesinghe, M., Weatherall, M., Perrin, K., Crane, J. and Beasley, R. 2009. International trends in asthma mortality rates in the 5 to 34 years' age group: A call for closer surveillance. *Chest* 135: 1045-1049.

Wilde, J.I. and Watson, S.P. 2001. Regulation of phospholipase C γ isoforms in hematopoietic cells. Why one, not the other? *Cellular Signalling* 13: 691–701.

Williams, H.C. 2005. Clinical practice. Atopic dermatitis. *New England Journal of Medicine* 352: 2314-2324.

Williams, H., Stewart, A., von Mutius, E. Cookson, W., Anderson, H.R. and International Study of Asthma and Allergies in Childhood (ISAAC) Phase One and Three Study Groups. 2008. Is eczema really on the increase worldwide? *Journal of Allergy and Clinical Immunology* 121: 947-954.

Wong, M.L. and Medrano, J.F. 2005. Real-time PCR for mRNA quantitation. *BioTechniques* 39: 75-85.

Xu, H., Gu, L.N. Yang, Q.Y., Zhao, D.Y. and Liu, F. 2016. MiR-221 promotes IgE-mediated activation of mast cells degranulation by PI3K/Akt/PLCγ/Ca(2+) pathway. *Journal of Bioenergetics and Biomembranes* 48: 293-299.

Xu, X., Zhang, D., Zhang, H., Wolters, P.J., Killeen, N.P., Sullivan, B.M., Locksley, R.M., Lowell, C.A. and Caughey, G.H. 2006. Neutrophil histamine contributes to inflammation in mycoplasma pneumonia. *Journal of Experimental Medicine* 203: 2907-2917.

Yamaguchi, M., Sayama, K., Yano, K., Lantz, C.S., Noben-Trauth, N., Ra, C., Costa, J.J. and Galli, S.J. 1999. IgE enhances Fc epsilon receptor I expression and IgE dependent release of histamine and lipid mediators from human umbilical cord blood-derived mast cells: synergistic effect of IL-4 and IgE on human mast cell Fc epsilon receptor I expression and mediator release. *Journal of Immunology* 162: 5455–5465.

Yang, Y.H., Lu, J.Y.L., Wu, X.S., Summer, S., Whoriskey, J., Saris, C. and Reagan, J.D. 2010. G-protein-coupled receptor 35 is a target of the asthma drugs cromolyn disodium and nedocromil sodium. *Pharmacology* 86: 1–5.

Yagi, N., Taniuchi, Y., Hamada, K., Sudo, J. and Sekikawa, H. 2002. Pharmacokinetics of ketotifen fumarate after intravenous, intranasal, oral and rectal administration in rabbits. *Biological & pharmaceutical bulletin* 25: 1614-1618.

Yazid, S., Sinniah, A., Solito, E., Calder, V. and Flower, R.J., 2013. Anti-allergic cromones inhibit histamine and eicosanoid release from activated human and murine mast cells by releasing Annexin A1. *PLoS One* 8: e58963.

© COPYRIGHT UPM

Yokoi, H., Choi, O.H., Hubbard, W., Lee, H.S., Canning, B.J., Lee, H.H., Ryu, S.D., von Gunten, S., Bickel, C.A., Hudson, S.A., Macglashan, D.W.Jr. and Bochner, B.S. 2008. Inhibition of FcepsilonRI-dependent mediator release and calcium flux from human mast cells by sialic acid-binding immunoglobulin-like lectin 8 engagement. *Journal of Allergy and Clinical Immunology* 121: 499–505.

Yoo, J.M., Sok, D.E. and Kim, M.R. 2013. Effect of endocannabinoids on IgE-mediated allergic response in RBL-2H3 cells. *International Immunopharmacology* 17: 123-131.

Yuk, C.S., Lee, S.J., Yu, Kim, T.H., Hahn, Y.K., Lee, S.Y. Men, Y.H., Hahn, M.W. and Lee, K.S. 1981. Korean Herbal Medicine, pp 267. Seoul: Cyechuk Publishing Company.

Zhang, A., Chi, X., Luo, G., Hei, Z., Xia, H., Luo, C., Wang, Y., Mao, X. and Xia, Z. 2013a. Mast cell stabilization alleviates acute lung injury after orthotopic autologous liver transplantation in rats by downregulating inflammation. *PLoS One* 8: e75262.

Zhang, N.N., Park, D.K. and Park, H.J. 2013b. The inhibitory activity of atractylenolide V, a sesquiterpenoid, on IgE-mediated mast cell activation and passive cutaneous anaphylaxis (PCA). *Journal of Ethnopharmacology* 145: 278-285.

Zhang, T., Finn, D.F., Barlow, J.W. and Walsh, J.J. 2016. Mast cell stabilisers. *European Journal of Pharmacology* 778:158-168.

Zhu, M., Liu, Y., Koonpaew, S., Granillo, O. and Zhang, W. 2004. Positive and negative regulation of FcεRImediated signalling by the adaptor protein LAB/NTAL. *Journal of Experimental Medicine* 200: 991–1000.

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LIST OF PUBLICATIONS

Publications

Tan, J.W., Israf, D.A., Nur Fariesha, M.H., Cheah, Y.K., Hanis, H.H., Shaari, K. and Tham., C.L. 2017. LAT is Essential for the Mast Cell Stabilising Effect of tHGA in IgE-Mediated Mast Cell Activation. *Biochemical Pharmacology* 144:132-148.

Tan, J.W., Israf, D.A., Hanis, H.H., Nur Fariesha, M.H., Ng, C.H., Shaari, K. and Tham., C.L. 2017. Anti-allergic Activity of 2,4,6-Trihydroxy-3-Geranylacetophenone (tHGA) via Attenuation of IgE-Mediated Mast Cell Activation and Inhibition of Passive Systemic Anaphylaxis. *Toxicology and Applied Pharmacology* 319: 47-58.

Tan, J.W. and Kim, M.K. 2016. Neuroprotective Effects of Biochanin A against β-Amyloid-Induced Neurotoxicity in PC12 Cells via a Mitochondrial-Dependent Apoptosis Pathway. *Molecules* 21: pii: E548.

Tan, J.W., Tham, C.L., Israf, D.A., Lee, S.H. and Kim, M.K. 2013. Neuroprotective Effects of Biochanin A Against Glutamate-Induced cytotoxicity in PC12 Cells Via Apoptosis Inhibition. *Neurochemical Research* 38: 512-518.

Audrey, K.S.F., Fong, L.Y., Khoo, L.W., **Tan, J.W.**, Shaari, K., Lee, M.T., Israf, D.A., Abas, F. and Tham., C.L. 2017. *Clinacanthus nutans* Aqueous Extract But Not Ethanolic Extract Inhibits the Release of Preformed Mediators in *In Vitro* Model of IgE-Mediated Mast Cell Degranulation. *Progress in Bioscience and Bioengineering* 1: 57-68.

Patents

PI 2016702942. 2,4,6-Trihydroxy-3-Geranylacetophenone for use as a mast cell stabilizer. Inventors: Tham Chau Ling, Khozirah Shaari, **Tan Ji Wei** and Ahmad Daud Israf Ali. 12th August 2016.

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Proceedings

Ji Wei Tan, Daud Ahmad Israf Ali, Nur Fariesha Md Hashim, Khozirah Shaari and Chau Ling Tham. Molecular Insight of A Geranyl Acetophenone in IgE-mediated Mast Cell Activation of Allergy. International Anatomical and Biomedical Scientific Conference (IABS) 2017 "Research Advances in Health Sciences" 1st-2nd August 2017, Faculty of Medicine and Health Sciences, University Putra Malaysia.

Ji Wei Tan, Daud Ahmad Israf Ali, Khozirah Shaari and Chau Ling Tham. Natural Geranylacetophenone compound from Melicope ptelefolia, a possible mast cell stabilizer against allergy in the future. APAAACI and APAPARI 2016. 17th October – 20th October 2016. Shangri-La Hotel KL, Kuala Lumpur.

Ji Wei Tan, Daud Ahmad Israf Ali, Khozirah Shaari and Chau Ling Tham. *In vitro* Inhibitory Effects of 2, 4, 6,-Trihydroxy-3-Geranylacetophenone (tHGA) on IgE-Mediated Mast Cell Degranulation. INTRACOM 2014. 30 October – 2 November 2014. Sunway Convention Centre, Kuala Lumpur.

Ji Wei Tan, Daud Ahmad Israf Ali, Khozirah Shaari and Chau Ling Tham. 2, 4, 6,-Trihydroxy-3-Geranylacetophenone (tHGA) Inhibits IgE-Mediated Degranulation by Suppressing The Production of Key Mediators in Rat Basophilic Leukemia RBL-2H3 Cells. Faculty Excellence Month 2015, Faculty of Medicine and Health Sciences, Universiti Putra Malaysia.

Ji Wei Tan, Daud Ahmad Israf Ali, Khozirah Shaari and Chau Ling Tham. 2, 4, 6,-Trihydroxy-3-Geranylacetophenone (tHGA) Inhibits Newly Synthesized Mediators Release in IgE-Mediated Mast Cell Degranulation. International Anatomical and Biomedical Scientific Conference (IABS) 2015 "From Cell Towards Translational Medicine" 18-20th August 2015, Faculty of Medicine and Health Sciences, University Putra Malaysia.

Ji Wei Tan and Min Kyu Kim. Protective effect of biochanin A on in-vitro model of alzheimer's disease: a possible future frontier against ageing. IAGG 2013. 23rd June – 27th June 2013. Seoul, South Korea.

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Awards

In vitro Inhibitory Effects of 2, 4, 6,-Trihydroxy-3-Geranylacetophenone (tHGA) on IgE-Mediated Mast Cell Degranulation. INTRACOM 2014. Sunway Convention Centre, Kuala Lumpur. **Best poster award**.

2, 4, 6,-Trihydroxy-3-Geranylacetophenone (tHGA) Inhibits IgE-Mediated Degranulation by Suppressing The Production of Key Mediators in Rat Basophilic Leukemia RBL-2H3 Cells. Faculty Excellence Month 2015, Faculty of Medicine and Health Sciences, Universiti Putra Malaysia. **Best poster award**.

2, 4, 6,-Trihydroxy-3-Geranylacetophenone (tHGA) Inhibits Newly Synthesized Mediators Release in IgE-Mediated Mast Cell Degranulation. International Anatomical and Biomedical Scientific Conference (IABS) 2015 “From Cell Towards Translational Medicine” Faculty of Medicine and Health Sciences, University Putra Malaysia. **Best poster award**.

Zerumbone protects against house dust mite-induced airway epithelial barrier disruption by preserving junctional permeability and localization. International Anatomical and Biomedical Scientific Conference (IABS) 2017 “Research Advances in Health Sciences” 1st-2nd August 2017, Faculty of Medicine and Health Sciences, University Putra Malaysia. **Best oral presentation**.

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