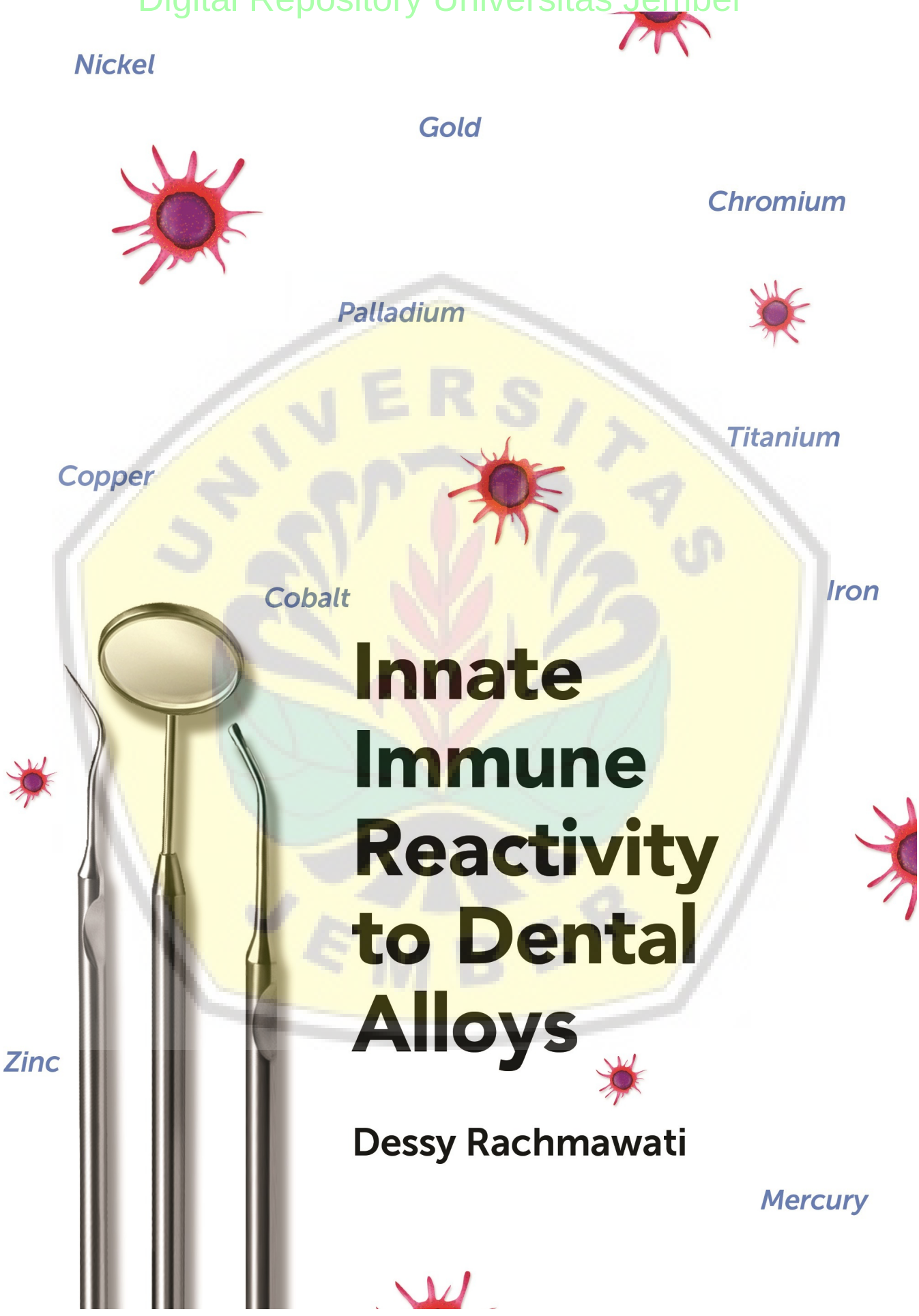

Innate Immune Reactivity to Dental Alloys

Dessy Rachmawati

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Innate immune reactivity to dental alloys

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dr. I.M.W. van Hoogstraten

Dedicated to

"My mom and my daughter Icha"

"And, when you want something,
all the universe conspires in helping you
to achieve it."

— Paulo Coelho, *The Alchemist*

“The journey of a thousand miles begins with one step.”
– Lao Tzu

CONTENTS

List of abbreviations

ANA	*Anti-nuclear antibodies*
AI	*Autoimmune/autoimmunity*
AID	*Autoimmune disease*
ACD	*Allergic contact dermatitis*
ALS	*Amyotrophic Lateral Sclerosis*
APC	*Antigen-presenting cell(s)*
CCP	*Cyclic citrullinated peptides*
CD	*Cluster of differentiation*
CFS	*Chronic Fatigue Syndrome*
CLR	*C-type lectin receptors*
CNS	*Central nervous system*
CpG	*Cytosine-phosphodiester-Guanine*
CSF	*Cerebrospinal fluid*
DC	*Dendritic cell(s)*
ELISA	*Enzyme-linked immunosorbent assay*
FACS	*Fluorescence-activated cell sorter*
FDP	*fixed dental prosthese(s)*
GM-CSF	*Granulocyte–macrophage colony stimulating factor*
HEK	*Human embryonic kidney*
HLA	*Human leukocyte antigen*
iDC	*Immature dendritic cell(s)*
IFNα	*Interferon α*
IFNβ	*Interferon β*
IL	*Interleukin*
KC	*Keratinocytes*
LC	*Langerhans cells*
LN	*Lymph node(s)*
LPS	*Lipopolysaccharide*
MAPK	*Mitogen-activated protein kinase*
MCP-1	*Monocyte chemotactic protein-1*
MHC	*Major histocompatibility complex*
MIP-1α	*Macrophage inflammatory protein-1alpha*
MS	*Multiple sclerosis*
MoDC	*Monocyte-derived dendritic cells*
MW	*Molecular weight*
NPG	*Non-plaque related gingivitis*
NLR	*NOD-like receptors*
OLP/ OLL	*Oral Lichen Planus/ Oral Lichenoid Lessions*

PAMP	*Pathogen-associated molecular pattern*
PBMC	*Peripheral blood mononuclear cell*
PGCG	*Peripheral giant cell*
PE	*Phycoerythrin*
PFM	*Porcelain fused to metal*
PRR	*Pattern Recognition Receptor*
Ps	*Psoriasis*
RA	*Rheumatoid arthritis*
RF	*Rheumatoid factors*
SD	*Standard deviation*
TA	*Thyroid antibodies*
TD	*Thyroid dysfunction*
TNF	*Tumour necrosis factor*
TLR	*Toll like receptor*
TPO	*Thyroid peroxidase*

CONTENTS

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Summary

Innate immune reactivity to dental alloys

Oral exposure to metals and metal alloys is frequently associated with local and systemic adverse reactions. The studies presented in this thesis shed a new light on the mechanisms by which dentally applied metals can cause irritation, inflammation or allergy. In addition the question is addressed why some metals, such as nickel, are known as strong sensitizers, while others seldom cause allergy. Overall, this thesis contributes to the understanding why metal-sensitivity occurs so frequently and it helps the researcher, dentist and patient to become more aware of the (potential) health effects of dental alloys. This data might also be useful for dentists to select alloys which have minimal immune stimulatory capacity. In addition, the results can contribute to a rational basis for future guidelines on the use of metal alloys in the oral cavity. Below, a summary of the chapters is given and the most relevant findings are highlighted.

In **chapter 1** six major questions on the relation between metal exposure and inflammation or allergy are selected and introduced by evaluating the present knowledge on the immune events that can lead to allergy. It is explained how the activation of innate immune cells by metals, *in casu* dendritic cells (DC), can induce inflammation and eventually adaptive immune responses to these metals. To become activated, DC receive 'danger signals', that are normally given by micro-organisms but can be mimicked by metals, such as nickel. Important receptors for such 'danger signals' are the so called toll like receptors (TLR), that are expressed on DC and other innate immune cells..

The results of **chapter 2** show that, like nickel, also cobalt and palladium strongly activate DC *in vitro*, as shown by the production of IL-8, an important inflammatory mediator. Copper and zinc, but not iron and chromium, induced low but significant innate activation. As shown in experiments with different TLR transfected cell lines next to nickel, cobalt and palladium could trigger the cells via TLR4 ligation, the receptor for bacterial endotoxin.

In **chapter 3** also the high molecular weight transition metals gold and mercury were tested for their innate stimulatory capacity. Gold and, to a lesser extent, mercury caused distinct DC activation and maturation. Both metal salts induced IL-8 production by DC as well as by the cell line THP-1, although to a lower extent than we had found for nickel, cobalt and palladium. Importantly, when studying gold induced responsiveness in the TLR transfected cell lines, TLR3 rather than TLR4 ligation was shown to be involved. TLR3 is the (intracellular) receptor for dsRNA associated with viral infection. The nature of the low-level innate response to mercury remains to be clarified.

Chapter 4 describes the direct stimulatory effects of gold, mercury, copper and nickel salts on keratinocytes (KC). First we observed that human KC as well as skin or gingiva derived KC cell lines express functional TLR3, but not TLR4, 5, 7/8 or 9. Indeed, gold induced robust IL-8 production by KC. Of note, also mercury, copper and nickel did activate KC. Whereas

these findings confirm our hypothesis that gold triggers TLR3, the mechanism(s) by which mercury, copper and nickel trigger KC still remain to be elucidated.

In **chapter 5** we tested dental cast alloys as solid specimens, reflecting the actual situation in the oral cavity, in DC and THP-1 cell cultures. The results fit with our earlier findings that most metals used for dental alloys show innate stimulatory activity. Importantly, the latter finding indicates that such stimulatory activity can be observed at low metal concentrations such as released from alloys in regular media. Of note, strongest and consistent IL-8 release was found with gold and palladium-copper containing alloys. In the presence of bacterial endotoxin, the exposure to these alloys, as well as to the 24 hrs supernatants of them, resulted in even stronger innate stimulation, suggesting a synergy between the metal exposure and endotoxin. The use of actual dental cast alloys in *in vitro* studies, instead of metal salt solutions, provides an effective strategy to study potential immune stimulatory effects of orally applied metals.

In **chapter 6** we investigated whether metal exposure can activate brain microglia, thereby providing a potential clue to the development of neuro-degenerative disease. The results confirmed that microglia can be triggered by metal salts, although at concentrations which may not readily be seen *in situ*. Nevertheless, also microglial cells reveal a strong synergy between exposure to metals, in particular to copper and zinc, and microbial endotoxin, indicating that such unfortunate coinciding activities may contribute to or augment chronic inflammation and neurotoxicity in humans.

Chapter 7 focused on clinical and serological parameters for autoimmune disease (AID) in relation to oral metal exposure in non-allergic and metal allergic individuals, as well as in patients with oral lesions attributed to dental restorations. Metal exposure, anamnesis and immune responsiveness to metals were evaluated extensively. The results of this study support the view that oral exposure to palladium, gold and mercury does not facilitate the development of AID. Surprisingly, in this limited group, we found a correlation between oral exposure to nickel and the presence of clinical autoimmune disease. Therefore, further investigations into a possible role of transition metals, in particular nickel, in the development of autoimmune disease are warranted.

In **chapter 8** the six major questions on the relation between oral metal exposure and inflammation, allergy, neurotoxicity and autoimmune disease have been answered and discussed in the light of the results of this thesis.

In short, the use of metal alloys in dentistry as well as in numerous other applications will never be avoided. Metal alloys cannot always be replaced by other materials such as methacrylates since their characteristics are essential for distinct clinical requirements.

Results from this study indicate that dental metal alloys may initiate local and systemic immune reactivities. Still, we should emphasize that careful production of dental metal alloys and appropriate clinical monitoring all contribute to their safe use in oral applications.

Nederlandse samenvatting

Immuunreactiviteit op tandheelkundige legeringen

Blootstelling aan metalen in de mondholte kan gepaard gaan met allergische klachten, met plaatselijk ontstoken slijmvlies, of met vage klachten over of door het gehele menselijke lichaam. Dit proefschrift laat zien waarom allergie voor metalen zo vaak voorkomt en werpt een nieuw licht op de mechanismen waarmee tandheelkundige legeringen irritatie, ontsteking en allergie kunnen veroorzaken. Daarbij staat de vraag centraal waarom tandheelkundige toegepaste metalen, zoals bij voorbeeld nikkel, palladium en goud, het innate immuunsysteem kunnen activeren. Dit zou een rol kunnen spelen bij acute ontstekingsreacties en ook het adaptieve immuunsysteem kunnen stimuleren.

De resultaten dragen bij aan de bewustwording t.a.v. de gezondheidsrisico's die blootstelling aan metalen bij onderzoekers, tandartsen, en patiënten kunnen geven. Ook zijn de resultaten van belang voor de tandarts die adequate legeringen voor zijn of haar patiënten moet selecteren: legeringen met minimaal ongewenste bijwerkingen op het immuunsysteem.

Tot slot kunnen de resultaten van waarde zijn voor toekomstige richtlijnen ten behoeve van het vervaardigen van nieuwe tandheelkundige legeringen.

Hieronder volgt een samenvatting van de hoofdstukken en de meest belangwekkende bevindingen.

In **hoofdstuk 1** zijn zes belangrijke vragen over de gezondheidsrisico's van orale metaal-blootstelling toegelicht. Dit is gedaan aan de hand van wat we momenteel weten over het ontstaan van de immuunreacties die uiteindelijk tot allergie kunnen leiden. De activatie van zogenaamde dendritische cellen (DC), welke deel uit maken van het 'innate' immuunsysteem, is hier essentieel. Bacteriën en virussen kunnen `danger`-signalen geven die de DC activeren en doen uitrijpen, zodat zij een specifieke, adaptieve immuunrespons kunnen initiëren. De DC vangen dergelijke signalen op met een reeks receptoren, waarvan de zogenaamde Toll like receptors (TLR) een belangrijke groep vormen. Opmerkelijk is dat sommige metalen, zoals nikkel, zo´n `danger`-signaal kunnen imiteren.

De resultaten van **hoofdstuk 2** laten zien dat naast nikkel ook kobalt en palladium de DC *in vitro* kunnen aanzetten tot de productie van IL-8, een belangrijke ontstekingsmediator. Koper en zink stimuleerden minder sterk, terwijl chroom en ijzer de DC helemaal niet activeerden. In experimenten met verschillende TLR-getransfecteerde cellijnen werd aangetoond dat palladium en kobalt, evenals nikkel, de cellen kunnen activeren via binding aan TLR4, de receptor voor bacterieel endotoxine.

In **hoofdstuk 3** werden ook zouten van de zware metalen goud en kwik getest op hun vermogen om 'innate' cellen te activeren. Goud, en in mindere mate kwik, induceerden de uitrijping van de DC en de productie van IL-8. Dit was ook aantoonbaar in THP-1 cellen, een

cellijn die als model voor DC gebruikt werd. De activatie was over het geheel minder sterk dan die voor nikkel, kobalt en palladium. Belangwekkend waren de experimenten waarbij goud en kwik getest werden op hun vermogen verschillende TLR-cellijnen te activeren. Terwijl de, op zich zwakke 'innate' respons, op kwik niet toegeschreven kon worden aan een bepaalde TLR binding, bleek goud met name TLR3 te triggeren, de receptor voor viraal dsRNA.

Hoofdstuk 4 beschrijft in welke mate goud-, kwik-, koper- en nikkelzouten in staat zijn om ook keratinocyten (KC) te stimuleren. Vers opgewerkte KC en KC-cellijnen, gemaakt vanuit huid of mucosa, bleken alle functioneel TLR3 tot expressie te brengen, maar geen TLR4, 5, 7, 8, of 9. Inderdaad induceerde goud een robuuste IL-8 productie door KC. Echter, ook kwik, koper en nikkel activeerden de KC. Terwijl dit resultaat onze hypothese bevestigt, dat goud via binding aan TLR3 stimuleert, moet het mechanisme waarmee kwik, koper en nikkel KC activeren nog opgehelderd worden.

In **hoofdstuk 5** hebben we tandheelkundige legeringen als solide stukjes metaal *in vitro* getest, omdat een dergelijke test de situatie in de mondholte het beste weerspiegelt. De resultaten bevestigen onze eerdere bevindingen, dat veel van de metalen in tandheelkundige legeringen het `innate` immuunsysteem kunnen activeren, ook in de betrekkelijk lage concentraties die vrijkomen uit de legeringen. De sterkste activatie van DC en de THP-1 cellijn vond plaats in aanwezigheid van goud en palladium-koper bevattende legeringen of met de metaal kweek-supernatanten. Deze activatie werd significant versterkt in aanwezigheid van geringe hoeveelheden endotoxine. Het gebruik van reële stukjes tandheelkundig reconstructiemateriaal *in vitro* verschaft een interessante strategie voor het uittesten van nieuwe tandheelkundige metaal legeringen.

In **hoofdstuk 6** hebben we onderzocht in hoeverre metaalzouten in staat zijn om ook uit hersenweefsel geïsoleerde `innate` cellen (microglia) te activeren. Dit om een eventueel verband te kunnen onderbouwen tussen orale metaalblootstelling en het ontstaan van neuro-degeneratieve ziekten. De resultaten bevestigen dat ook microglia wordt aangezet tot IL-8 productie in de aanwezigheid van metaalzouten, zij het in concentraties die gewoonlijk niet in de hersenen voorkomen. In fysiologische concentraties gaven alleen zink en koper een geringe stimulatie van microglia en THP-1 cellen. Deze respons bleek aanzienlijk versterkt te worden door de aanwezigheid van lage spiegels endotoxine. Dit suggereert dat het onfortuinlijk samengaan van deze inflammatoire stimuli (metaalblootstelling en infectie) mogelijk kan bijdragen tot een staat van chronische ontsteking, tenslotte uitmondend in een neuro-degeneratief ziektebeeld.

In **hoofdstuk 7** zijn klinische en serologische parameters voor auto-immuunziekten (AIZ) geëvalueerd in relatie tot orale metaal blootstelling, in metaal allergische en niet-allergische

individuen, alsmede in patiënten met orale laesies die toegeschreven konden worden aan tandheelkundige restauraties. Metaalblootstelling en metaal-specifieke immuun reactiviteit werden nauwkeurig in kaart gebracht. De resultaten van deze studie onderbouwen het idee dat orale blootstelling aan palladium, goud en kwik de ontwikkeling van auto-immuniteit niet faciliteert. Tot onze verrassing bleek, althans in deze beperkte groep (n=78), dat oraleblootstelling aan nikkel geassocieerd was met het hebben van een AIZ. Derhalve is nader onderzoek geboden naar een eventuele rol van metalen, in het bijzonder nikkel, in de ontwikkeling van AIZ.

In **hoofdstuk 8** zijn de zes cruciale vragen uit hoofdstuk 1, met betrekking tot de relatie tussen metaalblootstelling en ontsteking, allergie, neurotoxiciteit en AIZ beantwoord en bediscussieerd, in het licht van de resultaten van dit proefschrift.

Benadrukt wordt dat het gebruik van metaal legeringen voor tandheelkundige en andere toepassingen nooit vermeden zal kunnen worden omdat alternatieven, zoals gebaseerd op methacrylaten en andere kunststoffen, niet voldoen aan alle gebruikseisen. Hoewel de meeste tandheelkundig toegepaste metalen locale en systemische immuunreacties kunnen opwekken, kunnen zorgvuldige productie en testen van metaallegeringen en gepaste klinische monitoring bijdragen tot hun verantwoord veilig gebruik bij orale toepassingen.

Ikhtisar Bahasa Indonesia

Reaktivitas imun bawaan logam paduan kedokteran gigi

Paparan logam dan logam paduan (alloy) di rongga mulut sering kali dikaitkan dengan reaksi yang merugikan kesehatan baik lokal maupun sistemik. Hasil penelitian yang diuraikan dalam tesis ini memberikan pemahaman baru tentang mekanisme logam-logam yang dipakai di kedokteran gigi dalam menyebabkan iritasi, inflamasi atau alergi. Beberapa pertanyaan yang diajukan dalam thesis ini diantaranya adalah mengapa beberapa logam, salah satu contohnya nikel, adalah logam yang telah dikenal luas mempunyai daya sensitisasi yang kuat; sedangkan beberapa logam yang lain, didapati lebih jarang menyebabkan alergi. Secara keseluruhan, tesis ini diharapkan dapat memberikan kontribusi dalam memberikan pemahaman mengapa sensitivitas terhadap logam sangat sering terjadi dan juga diharapkan dapat membantu peneliti, dokter gigi dan pasien untuk lebih berhati-hati terhadap efek yang ditimbulkan oleh logam-logam paduan kedokteran gigi terhadap kesehatan. Data yang dihasilkan dari penelitian ini juga dapat digunakan sebagai panduan bagi dokter gigi dalam memilih paduan logam yang memiliki kapasitas stimulasi terhadap imunitas bawaan paling minimal. Selain itu, hasil penelitian ini dapat dijadikan sebagai panduan yang rasional tentang penggunaan logam paduan di rongga mulut. Dalam bab ini, hasil-hasil penelitian tersebut dirangkum dan hasil yang paling relevan diuraikan lebih lanjut.

Dalam **bab 1** enam pertanyaan pokok tentang hubungan antara paparan logam dan inflamasi atau alergi telah disajikan melalui evaluasi dengan pendekatan imunologis tentang mekanisme penyebab alergi. Dalam ikhtisar ini akan dibahas pula tentang bagaimana mekanisme aktivasi sel-sel imun bawaan oleh karena paparan logam, salah satu diantaranya adalah kemampuan logam dalam memicu terjadinya inflamasi dan respon imun adaptif melalui aktifasi sel-sel dendritik (DC). Untuk dapat menjadi aktif, DC menerima 'sinyal bahaya' yang biasanya diberikan oleh mikro-organisme tetapi dalam hal ini mampu ditirukan oleh logam, sebagai salah satu contoh yang konkrit adalah nikel. Reseptor yang berperan penting sebagai 'sinyal bahaya' dikenal dan disebut sebagai *toll like receptor (TLR)*, sinyal tersebut diekspresikan oleh DC dan sel-sel imun bawaan yang lain.

Hasil bab 2 menunjukkan bahwa, seperti halnya nikel, kobalt dan paladium sangat kuat dalam mengaktifkan DC secara *in vitro*, hal ini ditunjukkan melalui parameter produksi IL-8, yang merupakan salah satu mediator inflamasi yang cukup penting. Tembaga (Cuprum) dan seng (Zn), mempunyai daya stimulasi yang tidak begitu kuat tetapi cukup signifikan dalam memicu aktivasi imunitas bawaan, sedangkan besi dan kromium tidak dapat mengaktivasi imunitas bawaan. Hal ini ditunjukkan melalui percobaan dengan menggunakan *cell line* yang diinduksi (*transfected*) dengan TLR, hasilnya menunjukkan bahwa hanya nikel, kobalt dan paladium yang mampu memicu aktivasi sel melalui TLR4, reseptor yang dikenal sebagai reseptor bagi bakteri endotoksin.

Dalam **bab 3** ini logam-logam transisi dengan massa molekul yang sangat tinggi seperti emas dan merkuri juga diuji untuk mengetahui kapasitasnya dalam menstimulasi imunitas

bawaan. Emas dan, dengan kapasitas yang lebih rendah, merkuri menunjukkan kemampuan aktivasi dan maturasi DC yang berbeda. Kedua garam logam dapat menginduksi IL-8 pada DC dan cell line THP-1, meskipun kemampuan aktivasinya lebih rendah dibandingkan dengan nikel, kobalt dan paladium. Yang perlu diperhatikan, ketika meneliti kemampuan emas dalam menginduksi *cell line* yang diinduksi TLR, TLR 3 merupakan reseptor yang lebih berperan penting untuk emas dibandingkan dengan TLR4. Sedangkan mekanisme respon imun bawaan terhadap merkuri masih harus diklarifikasi melalui penelitian lebih lanjut .

Bab 4 menjelaskan tentang efek langsung dari stimulasi garam-garam logam emas, merkuri, tembaga dan nikel pada sel-sel keratinosit (KC). Sel keratinosit pada tubuh manusia dapat dijumpai di kulit ataupun gingiva. KC kulit dan cell line KC dari gingiva telah diketahui dapat mengekspresikan TLR3, tetapi tidak dijumpai ekspresi TLR4, 5, 7/8 atau 9. Hasil yang didapat pada bab ini menunjukkan bahwa emas cukup kuat dalam menginduksi produksi IL-8 pada KC. Selain itu, merkuri, tembaga dan nikel juga mempunyai kapasitas serupa dalam mengaktifkan KC. Hasil dari penelitian ini juga mengkonfirmasi hipotesis kami bahwa emas memicu aktivasi KC melalui TLR3, sedangkan mekanisme aktivasi KC oleh karena paparan merkuri, tembaga dan nikel masih harus dibuktikan melalui penelitian lebih lanjut.

Dalam **bab 5** ini kami menguji logam tuang (cast) paduan kedokteran gigi dalam bentuk spesimen yang solid, penggunaan bentuk spesimen ini diharapkan mampu mencerminkan kondisi yang sebenarnya di dalam rongga mulut. Hasil yang didapatkan pada bab ini sejalan dengan hasil pada penelitian sebelumnya yang menguraikan bahwa sebagian besar dari logam yang digunakan sebagai logam paduan kedokteran gigi menunjukkan kapasitas stimulasi terhadap imunitas bawaan. Selain itu, hasil ini menunjukkan bahwa kemampuan stimulasi tersebut bahkan didapati pada konsentrasi logam yang cukup rendah yang biasanya dilepaskan oleh logam di dalam media cair. Selain itu, produksi IL-8 yang paling tinggi dan konsisten ditemukan melalaui paparan terhadap logam paduan yang mengandung emas dan campuran logam paladium–tembaga (Pd-Cu). Dengan adanya penambahan komponen bakteri endotoksin, paparan terhadap logam paduan dan juga supernatan yang didapatkan setelah perendaman spesimen logam selama 24 jam, menghasilkan stimulasi imun bawaan yang lebih kuat dibandingkan dengan paparan tanpa bakteri endotoksin, hal ini menunjukkan adanya sinergi antara paparan logam dan endotoksin. Selain penggunaan larutan garam logam yang selama ini telah dikenal, penggunaan paduan logam tuang kedokteran gigi dalam bentuk solid spesimen pada studi *in vitro* ini juga dapat memberikan penanda sebagai metode alternatif yang cukup efektif dalam meneliti kapasitas potensial yang dimiliki oleh logam yang diaplikasikan di dalam rongga mulut dalam menstimulasi imunitas bawaan.

Dalam **Bab 6** ini kami meneliti apakah paparan logam dapat mengaktifkan sel mikroglia otak, sehingga hasil penelitian ini diharapkan dapat memberikan petunjuk yang cukup potensial dalam menjelaskan terjadinya penyakit *neuro-degeneratif*. Hasil penelitian ini

mengkonfirmasi bahwa aktivasi mikroglia dapat dipicu oleh paparan garam-garam logam, sekalipun pada konsentrasi yang rendah yang tidak dapat dideteksi dengan mudah secara *in situ* di dalam tubuh manusia. Selain itu, didapatkan pula adanya sinergi yang kuat antara paparan logam, khususnya tembaga dan seng, dengan bakteri endotoksin, sehingga studi ini dapat memberikan petunjuk tentang mekanisme imun dalam meningkatkan kemungkinan terjadinya peradangan kronis dan neurotoksisitas pada manusia.

Bab 7 difokuskan pada parameter klinis dan serologis penyakit autoimun (AID) dalam kaitannya dengan paparan oral logam pada individu yang non-alergi dan individu yang alergi terhadap logam, serta pada pasien dengan lesi oral yang berkaitan erat dengan restorasi gigi. Paparan logam, anamnesis dan respon imun terhadap logam dievaluasi secara menyeluruh. Hasil penelitian ini mendukung pandangan bahwa paparan oral terhadap paladium, emas dan merkuri tidak berkaitan dengan terjadinya AID. Secara mengejutkan, dalam kelompok yang terbatas ini, kami menemukan korelasi antara paparan oral terhadap nikel dan adanya penyakit autoimun secara klinis. Oleh karena itu, penelitian lebih lanjut tentang kemungkinan peran dari logam transisi, khususnya nikel, dalam perkembangan penyakit autoimun sangat dibutuhkan.

Dalam **bab 8** enam pertanyaan pokok tentang hubungan antara paparan oral logam dan inflamasi, alergi, neurotoksisitas dan penyakit autoimun telah dijawab dan dibahas dengan jelas melalui hasil tesis ini.

Singkatnya, penggunaan paduan logam dalam kedokteran gigi serta dalam berbagai aplikasi lainnya tidak akan dapat dihindari. Paduan logam tidak selalu bisa diganti dengan bahan lain seperti metakrilat mengingat bahwa karakteristik logam sangat cocok untuk beberapa kebutuhan klinis yang berbeda. Hasil dari penelitian ini menunjukkan bahwa paduan logam gigi dapat menimbulkan terjadinya reaktivitas imun lokal dan sistemik. Namun, harus ditekankan pula bahwa produksi yang cermat dan hati-hati dari paduan logam gigi serta pemantauan klinis yang tepat sangat berkontribusi dalam menghasilkan penggunaan yang aman dalam aplikasi oral.

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Biography

ABOUT THE AUTHOR

Dessy Rachmawati was born on the 23rd of December 1976, in Jember, East Java, Indonesia. In 1995 she entered the Faculty of Dentistry at the University of Jember, Indonesia, and obtained her Bachelor of Dental Science (BDS) in 2000. She graduated as a Doctor of Dental surgery (**DDs**) in 2002.

Thereafter she worked at the Department of Biomedical Science and the Department of Prosthodontics at the Faculty of Dentistry, University of Jember, where she was appointed as a lecturer in 2002, and worked there until 2009. Since 2004 she studied for her master degree (MDSc) in Biomaterial Science in Dentistry, at the Graduate School Gadjah Mada University, Yogyakarta, Indonesia, funded by the Directorate General Higher Education, Ministry of Education, Republic of Indonesia. She graduated cum laude for her master degree in 2006.

She was involved in both the clinical and academic teaching for dental students of all years. Initially, she taught in the discipline of Prosthetics Dentistry and Biomedical Science, and worked as a dentist at the Dental Hospital, Faculty of Dentistry, University of Jember.

Since January 2010 she is following a PhD program in the section of Medical Immunology, Department of Pathology, VU University Medical Centre, Amsterdam. This program was supported by the Directorate General Higher Education (DIKTI), Ministry of Research, Technology and Higher Education of the Republic of Indonesia, Scholarship for Overseas (2010 - 2013) until March 2013 and subsequently, until March 2015, by the Department of Pathology of the VUmc. E-mail address: d.rachmawati@unej.ac.id

PHD PORTFOLIO

Summary of PhD Training
PhD student: Dessy Rachmawati
Period: January 2010-April 2016
PhD supervisors: Prof. dr. R.J. Scheper
dr. B.M.E. von Blomberg-van der Flier
dr. I.M.W. van Hoogstraten

PHD TRAINING

Courses	Year	ECTS
Basic Immunology	2010	2
Effective Time Management	2010	0.3
Skin immunology and cancer vaccines: from allergy to resistance	2010	0.5
Working With Radioactivity	2010	2
Radiation Oncology	2010	1.5
Writing A Scientific Article	2010	3
The Course on Molecular Diagnostics VII	2012	1.5
Advance Immunology	2013	3
Presenting Scientific Research	2013	0.3
ESCD: Immunology of allergic contact dermatitis	2013	1
Seminars, Meetings and Lecturers		
Participation in VUmc CCA evening lecturers	2010-2015	0.5
Participation in weekly CCA meetings	2010-2016	6
Participation in weekly Dental Immunology meetings	2010-2016	6
National and international conferences		
European Research Group on Experimental Contact Dermatitis (ERGECD) in Trier, Germany (Oral presentation)	2012	2
Dental Scientific Meeting (DSM) in Jember, Indonesia (Oral presentation)	2013	2
Dutch Society for Immunology Meeting 49 th (NVVI) in noordwijkerhout, The Netherlands (Oral poster presentation)	2013	2
Experimental Contact Dermatitis (ERGECD) in Paris,		

France(Oral presentation)	2014	2
Dutch Society for Immunology Meeting 50 th (NVVI) in Kaatsheuvel, The Netherlands (Oral poster presentation)	2014	2

Activities

Supervision master students internship		
- Lizalo Galama	2011	5
- Magda Baroud	2012	5
- Heleen Tromp	2013	5
- Inás Alsalem	2014	5
Writing Scientific articles under supervision of Professor	2010-2016	9
Total ECTS		**66.33**

COLLABORATIONS

Skin and oral mucosal equivalent dental metal exposures
Ilona Kosten – Department of Dermatology, VU University Medical Centre, Amsterdam, The Netherlands

Gingival cells exposure to TLR ligands
Jeroen K. Buskermolen - Department of Oral Cell Biology, Academic Centre for Dentistry Amsterdam, University of Amsterdam and VU University Amsterdam, The Netherlands

Innate stimulatory capacity of high molecular weight transition metals Au (gold) and Hg (mercury), exposure of HEK-TLR transfectant cells to the metals.
Marleen I. Verstege - Department of Molecular Cell Biology (MCBI), VU University Amsterdam, the Netherlands

Clinical oral examination of allergic patients to dental alloys
Joris Muris - Department of Dental Materials Science, Academic Centre for Dentistry Amsterdam (ACTA), VU University Amsterdam and University of Amsterdam, The Netherlands

Prof. dr. Cees Kleverlaan - Department of Dental Materials Science, Academic Centre for Dentistry Amsterdam (ACTA), VU University Amsterdam and University of Amsterdam, The Netherlands
Innate immune reactivity to dental alloy specimens

Capacity of metal-ions activate human brain-microglia
Laura A.N. Peferoen – Neuropathology, Dept. of Pathology, VU University Medical Centre, Amsterdam

Metal ions potentiate microglia responsiveness to endotoxin
Daphne Y.S. Vogel - Neuroimmunology Unit Department of Molecular Cell Biology Immunology, VU University, Amsterdam

LIST OF PUBLICATIONS

PEER REVIEWED PUBLICATIONS

Rachmawati D, Bontkes HJ, Verstege MI, Muris J, von Blomberg BM, Scheper RJ, van Hoogstraten IM. Transition metal sensing by Toll-like receptor-4: next to nickel, cobalt and palladium are potent human dendritic cell stimulators. *Contact Dermatitis. 2013 Jun;68(6):331-8.*

Rachmawati D, Bontkes HJ, Verstege MI, von Blomberg BM, Scheper RJ, van Hoogstraten IM.: "Innate stimulatory capacity of high molecular weight transition metals: Au (gold) and Hg (mercury)". *Toxicology in Vitro Journal.2015 29, 363-369.*

Rachmawati D, Joris Muris J, Scheper RJ, Rustemeyer T, Kleverlaan CJ, Feilzer AJ, von Blomberg BME, van Hoogstraten, IMW. Continuing the quest for autoimmunity due to oral metal exposure. *Autoimmunity 48 (7), 494-501, 2015*

Rachmawati D, Jeroen K. Buskermolen, Rik J. Scheper, Susan Gibbs, B.M.E. von Blomberg, Ingrid M.W. van Hoogstraten: 'Dental metal-induced innate immune responses in keratinocytes, *Toxicology in vitro, 2015, Vol.30, pp.325-30*

Rachmawati D, Laura A.N. Peferoen, Daphne Y.S. Vogel, Inás W Alsalem, Hetty J Bontkes, Sandra Amor, B Mary E von Blomberg, Rik J Scheper, Ingrid MW van Hoogstraten. Metals released from dental materials are unlikely to cause innate immune reactivity of microglia, *Journal of Neuroimmunology, 2016, Vol 291, Pp.89–95*

Rachmawati D, B. Mary E. von Blomberg, Cornelis J. Kleverlaan, Rik J. Scheper , Ingrid M.W. van Hoogstraten: 'Immunostimulatory capacity of dental casting alloys on endotoxin responsiveness', (submitted)

Rachmawati D, 'Immune reactivity to dental alloys: innate responses may facilitate allergies but not autoimmunity (Review)' (in progress)

CONFERENCE AND NON-PEER REVIEWED PUBLICATIONS

Rachmawati D, Laura A.N. Peferoen, Daphne Y.S. Vogel, Inás W Alsalem, Hetty J Bontkes, Sandra Amor, B Mary E von Blomberg, Rik J Scheper, Ingrid MW van Hoogstraten. Capacity of metal ions to activate human brain microglia (oral poster presentation). Dutch Society for *Immunology Meeting, 17-19 Dec 2014, Efteling, The Netherlands*

Rachmawati D, B Mary E von Blomberg, Rik J Scheper, Ingrid MW van Hoogstraten. Innate stimulatory capacity of high molecular weight transition metals: Au (gold) and Hg (mercury)

(oral presentation). Group on Experimental Contact Dermatitis (ERGECD) Meeting 201413-14 June 2014, Paris Sud University, France

Rachmawati D, B Mary E von Blomberg, Rik J Scheper, Ingrid MW van Hoogstraten. Differential triggering of the innate immune system by metals (oral poster presentation). Dutch Society for Immunology Meeting, 18-19 Dec 2013, Noordwijkehout, The Netherlands

Rachmawati D, B Mary E von Blomberg, Rik J Scheper, Ingrid MW van Hoogstraten. Regulation of the immune responses of metal allergy (oral presentation). Dies Natalis and Indonesian dental scientific Meeting, 10-12 Oct 2013 Faculty of Dentistry Jember University

Rachmawati D, B Mary E von Blomberg, Rik J Scheper, Ingrid MW van Hoogstraten. Transition metal-sensing of human TLR4: Cobalt and Palladium are, next to Nickel, potent human Dendritic Cell stimulators (oral presentation). European Research Group on Experimental Contact Dermatitis (ERGECD) Meeting 2012 10-12 Oct 2012, Universitat Trier, Germany