



UNIVERSITI PUTRA MALAYSIA

***OIL NANO-EMULSION FORMULATIONS OF AZADIRACHTIN
FOR CONTROL OF Bemisia tabaci GENNADIUS***

NOORHAZWANI BINTI KAMARUDIN

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NOORHAZWANI BINTI KAMARUDIN

**MASTER OF SCIENCE
UNIVERSITI PUTRA MALAYSIA
2013**



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CONTROL OF *Bemisia tabaci* GENNADIUS**

By

NOORHAZWANI BINTI KAMARUDIN

**Thesis Submitted to the School of Graduate Studies,
Universiti Putra Malaysia, in Fulfilment of the
Requirements for the Degree of Master of Science**

July 2013

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DEDICATION

Dedicated to:

My mother (Bahiah Bt Abd Aziz) and My Father (Kamarudin Harun)

For their true love, support and inspiration

Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirement for the degree of Master of Science

**OIL NANO-EMULSION FORMULATIONS OF AZADIRACHTIN FOR
CONTROL OF *Bemisia tabaci* GENNADIUS**

By

NOORHAZWANI BT KAMARUDIN

July 2013

Chairman : Dzolkhifli Omar, PhD.

Faculty : Agriculture

Current water emulsion insecticides only provide limited control of *Bemisia tabaci*. Oil droplets were found to be more effective as they spread much better on leaf surfaces compared to either water alone or water that contained adjuvant. Thus oil nano-emulsion formulation derived from azadirachtin was developed as an effort to control the population of whiteflies, *B. tabaci*. Oil nano-emulsion system was developed for insecticide formulations by constructing ternary phase diagrams with 70% (w/w) emulsion system constituted of non-ionic surfactant(s), carrier, water, and 30% (w/w) neem oil as an active ingredient. The non-ionic surfactant was alkylpolyglucosides while carrier or oil phase was dimethylamide. Ternary phase diagrams of the mixed surfactant systems MBL510H: MBL530B at mixed surfactant

ratios (MSRs) of 5:5, 6:4, 7:3, 8:2, 9:1 exhibited larger isotropic (I) phase than the single surfactants of either MBL510H or MBL530B.

The points were selected from the 'I' phase and homogenous region for pre-formulation. Most of the points selected were from regions with high proportion of oil, low proportion of water and adequate proportion of surfactant to mix with active ingredient and to form water-in-oil (W/O) emulsion. Sixteen formulations miscible with neem oil were selected. In the stability study, all the selected formulations were stable under centrifugation and storage at room temperature (25°C). However, at 54°C after 14 days storage, F3, F7, F9, F10, and F12 showed phase separation, transformed to two opaque phases. The mean particle size of nano-emulsions ranged between 150.00 and 450.00nm except for F9 with mean particle size of 640.44nm. All sixteen formulations showed surface tension lower than water (72.00mN/m). The formulation F14 (29.90mN/m), F15 (29.93mN/m) and F16 (29.86mN/m) showed lower surface tension compared to other formulations. The zeta potential values of F14 (39.60mV), F15 (39.20mV) and F16 (38.80mV) were higher compared to the other formulations. The value is related to the stability of colloidal dispersions and high zeta potential value will confer stability.

In the biological activity study, the adult *B. tabaci* were used to test the toxicity of the oil nano-emulsion formulation. The result showed the mortality of the adults was higher with the increase of time exposure. The mortality rate of *B. tabaci* showed that the oil nano-emulsion formulations gave excellent efficacy with LC₅₀ value of 3.70ppm at 96 h after treatment. In the measurement of spread area study, three

different levels of formulation toxicities were used to determine the spreading coefficient and evaluate the mode of action of the formulation on the early nymphal instar's *B. tabaci*. The studies have proved the interaction between spread area and mortality rate. The larger the spread area of the droplet result in increased of mortality. In this study, F15 formulation with low mean lethal concentration gave the larger spread area on the leaves surfaces. As a result, the formulation also gave highest mortality rate on early nymphal instar of whiteflies due to the spreading ability of this formulation. This finding has proved the mode of action of oil nano-emulsion formulation in killing the early nymphal instars of *B.tabaci* by giving wider coverage of active material on leaves surface and brings larger areas of cuticle into contact with the insecticides, resulting in better retention and enhanced the biological effect.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia
Sebagai memenuhi keperluan untuk Ijazah Master Sains

NANO-EMULSI MINYAK DARI AZADIRACHTIN UNTUK PENGAWALAN
***Bemisia tabaci* GENNADIUS**

Oleh

NOORHAZWANI BT KAMARUDIN

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Racun serangga emulsi air yang sedia ada hanya memberikan kawalan terhadap kepada *Bemisia tabaci*. Titisan minyak didapati lebih berkesan kerana ia merebak lebih baik pada permukaan daun berbanding air sama ada bersendirian atau air yang mengandungi adjuvan. Oleh itu, formulasi minyak nano-emulsi yang bersumberkan dari azadirachtin telah dihasilkan sebagai satu usaha untuk mengawal populasi lalat putih, *B. tabaci*. Sistem minyak nano-emulsi telah dihasilkan untuk formulasi racun serangga dengan membina diagram fasa 'terner' pada sistem emulsi 70% (b/b) yang mengandungi surfaktan nonionik, pembawa, air, dan minyak mambu 30% (b/b) sebagai bahan aktif. Surfaktan bukan ionik yang digunakan adalah akilpoliglukosida manakala pembawa atau minyak adalah dimetiamid. Diagram fasa terner bagi sistem

surfaktan campuran MBL510H: MBL530B pada nisbah campuran (MSR_S) 5:5, 6:4, 7:3, 8:2, 9:1 mempamerkan fasa isotropic (I) yang lebih besar berbanding surfaktan tunggal MBL510H atau MBL530B.

Kawasan fasa I dan fasa homogenus adalah kawasan di mana pra-formulasi dipilih. Kebanyakan titik yang dipilih adalah dari kawasan yang mempunyai kadar minyak yang tinggi, kadar air yang rendah dan kadar surfaktan yang mencukupi untuk bercampur dengan bahan aktif serta untuk membentuk emulsi air dalam minyak (W/O). Enam belas formulasi terlarut campur dengan minyak mambu telah dipilih. Dalam ujian kestabilan, semua formulasi yang dipilih stabil pada proses emparan dan simpanan pada suhu bilik (25°C). Walau bagaimanapun, pada 54°C selepas 14 hari penyimpanan, F3, F7, F9, F10 dan F12 menunjukkan pemisahan fasa, berubah kepada dua fasa legap. Min saiz zarah bagi nano emulsi ialah di antara 150.00 dan 450.00nm kecuali untuk formulasi F9 dengan min saiz zarahnya 640.44nm. Keseluruhan 16 formulasi menunjukkan ketegangan permukaan lebih rendah daripada air (72.00mN/m). Formulasi F14 (29.90mN/m), F15 (29.93mN/m) dan F16 (29.86mN/m) menunjukkan ketegangan permukaan yang lebih rendah berbanding dengan formulasi yang lain. Nilai potensi zeta bagi formulasi F14 (29.90mN / m), F15 (29.93mN / m) dan F16 (29.86mN / m) adalah lebih tinggi berbanding dengan formulasi lain. Nilai yang diperolehi mempunyai kaitan dengan kestabilan penyebaran koloid dan nilai potensi zeta yang tinggi akan memberikan kestabilan.

Dalam kajian aktiviti biologi, *B. tabaci* dewasa telah digunakan untuk menguji ketoksikan formulasi minyak nano emulsi. Kematian lalat putih dewasa meningkat

seiring dengan peningkatan masa pendedahan. Kadar kematian *B. tabaci* menunjukkan bahawa formulasi minyak nano-emulsi memberi keberkesanan yang sangat baik dengan nilai LC_{50} sebanyak 3.70ppm pada 96 jam selepas rawatan. Dalam kajian penentuan kawasan penyebaran, tiga formulasi dengan aras toksik yang berbeza telah digunakan untuk menentukan pekali penyebaran dan menilai ketoksikan formulasi pada pada nimfa lalat putih peringkat awal. Kajian telah membuktikan terdapat interaksi antara luas kawasan penyebaran dan kadar kematian. Semakin besar kawasan penyebaran titisan, semakin meningkat kadar kematian. Dalam kajian ini, formulasi F15 yang mempunyai kepekatan LC_{50} paling rendah telah memberikan penyebaran kawasan yang lebih besar pada permukaan daun. Hasilnya, formulasi juga turut memberikan kadar kematian tertinggi kepada peringkat awal nimfa lalat putih disebabkan keupayaan penyebaran formulasi ini. Hasil penemuan ini telah membuktikan kesan tindakan formulasi minyak nano-emulsi dalam membunuh nimfa lalat putih peringkat awal iaitu dengan memberi liputan bahan aktif yang lebih meluas di atas permukaan daun dan memberi kawasan yang lebih besar bagi kutikel bersentuhan dengan racun serangga, lantas menyebabkan pengekalannya yang lebih baik dan meningkatkan kesan biologi.

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APPROVAL

I certify that a Thesis Examination Committee has met on 29 July 2013 to conduct the final examination of Noorhazwani binti Kamarudin on her thesis entitled “Oil Nano-Emulsion Formulations Of Azadirachtin for Control of *Bemisia tabaci* Gennadius” 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 Master of Science.

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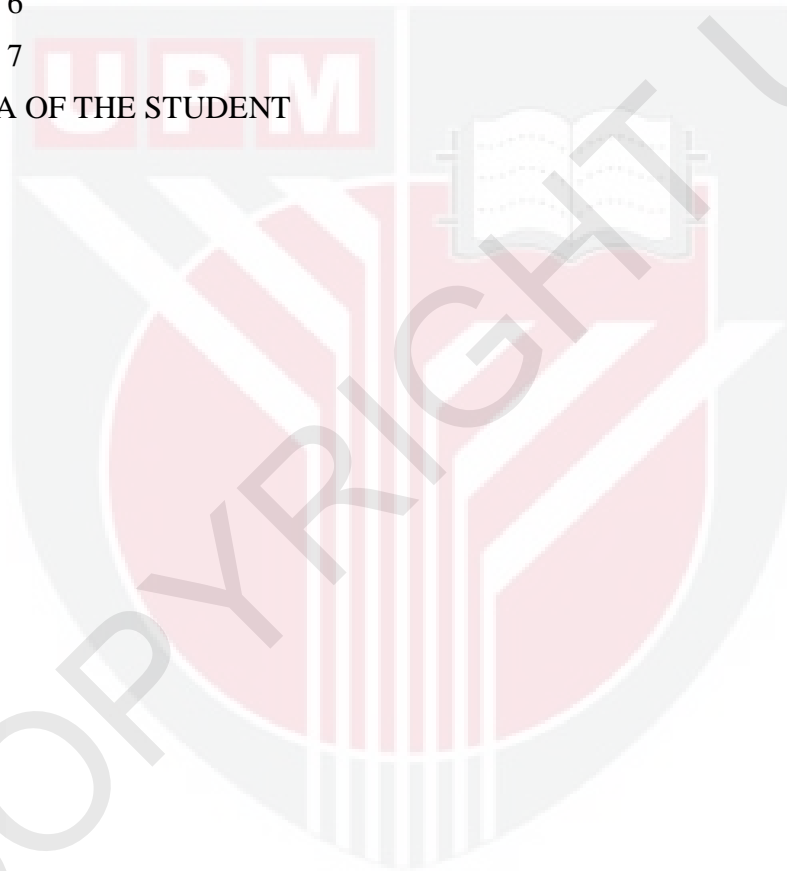
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LIST OF ABBREVIATIONS

g	gram (s)
kg	kilogram(s)
mL	mililitre(s)
cm	centimetre(s)
mm	milimetre(s)
d	day(s)
h	hour(s)
%	percent
°	degree
°C	degree(s) Celsius
APG	Alkylpolyglucosides
CRD	Complete Randomized Design
DAT	Day after treatment
PM	<i>post meridem</i> , after noon
RM	Ringgit Malaysia
S.E	Standard Error
UPM	Universiti Putra Malaysia
a.i	active ingredient
w/w	weight over weight
w/v	weight over volume
ppm	parts per million
no.	number
&	and
<i>viz.</i>	<i>videlicet</i> , that is, namely

i.e. *exempli gratia*, for example

et al. *et alii*, and others



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CHAPTER 1

INTRODUCTION

Whiteflies are among the major key pest of many fruits, vegetables and ornamental crops. They are highly polyphagous, and damage a broad range of food and non-food crops by direct feeding, impairing product quality through the excretion of honeydew, and transmission of over 100 plant viruses (Jones, 2003). Some of these viruses such as tomato yellow leaf curl virus (TYLCV) are of high economic importance and cause high economic losses on tomato in the Mediterranean basin (Morione & Luis-Arteaga, 1999). Although there are approximately 1,200 species of whiteflies worldwide, only a few of their species cause the highest damage on agricultural crops. Among the species, *Bemisia tabaci* is the most important species in agriculture.

Bemisia tabaci is often difficult to control using insecticides as all stages are normally located on the underside of the leaf (S. Chu et al., 1998). Furthermore, *B. tabaci* has developed high levels of resistance against several chemical classes of insecticides. Pesticide resistance usually arises from the overuse and misuse of pesticides, which is often due to lack of available alternatives (Denholm, 1988). The use of insecticides also has a negative impact on the environment, non-target organisms and human health. These have encouraged the development of alternative methods of control. Thus, biopesticides are being developed to control *B. tabaci* around the world.

Biopesticides are pesticides in which the active ingredient (a.i) is derived from virus, fungus, bacteria or natural product from plant sources. The use of biopesticide in crop protection is a practical and sustainable alternative to the synthetic organic-based insecticides. They could maintain biological diversity of predators (Grange & Ahmed, 1988), reduce environmental contamination and human health hazards. Plant sources commonly used as biopesticide include *Azadirachta indica*, *Derris sp.*, and *Cymbopogon nardus*. Azadirachtin extracted from *Azadirachta indica* has a broad mode of action. Thus, it is difficult for the insects to build resistance to this compound. Besides, the use of agro-based carrier materials in the pesticide formulation has become more important as they are relatively biodegradable, low in toxicity and from renewable resources than those from mineral oil derived commodities (Chow et al., 1992).

Water-based formulation cannot fully control the whiteflies due to morphological and ecological characteristics of the leaf such as a waxy cuticle, and the whiteflies tendency to colonize the underside of leaves making it difficult for active ingredient (a.i) to reach the target (Osborne & Landa, 1992). Oil-based formulations droplets were found to spread much better on leaf surfaces than either water alone or water that contained adjuvant (McWhorter & Barrentine, 1988). The wider spread enables the active ingredient (a.i) to reach the target pest especially sessile insects such as whiteflies.

Aside from having good spreading ability, the formulations should also have good penetration of the active ingredient (a.i) towards the target pest. This can be achieved

by having a nano droplet size formulation. Nano-emulsion is a non-equilibrium colloidal system comprising of oil phase, surfactants and water, offers better absorption having extremely a small size droplets (100-600nm) (Shafiq et al., 2007; Solans et al., 2003) and thus could be uniformly distributed (Gutierrez et al., 2008). Oil-phase in nano-emulsion increase bioavailability of active ingredient (a.i) which allows better penetration into the waxy layers and cuticle of the leaf. However, there is limited information on the development of nano-emulsion system for oil-based biopesticide.

Thus, the objectives of this study were to:

1. Prepare oil nano-emulsion formulation of azadirachtin and determine the physicochemical properties of the formulations;
2. Evaluate the toxicity of oil nano-emulsion formulations against *Bemisia tabaci* and,
3. Verify the mode of action on oil nano-emulsion formulation in killing *Bemisia tabaci*.

BIBLIOGRAPHY

- Abbott, H. A., Van Dyk, L. P., & Grobbelaar, N. (1990). Spreading of spray mixtures on leaf surfaces. *Pesticide Science*, (28), 419–429.
- Abbott, W. S. (1925). A method of computing the effectiveness of an insecticides. *Journal of Economic Entomology*, 18, 265–267.
- Anjali, C. H., Sharma, Y., Mukherjee, A., & Chandrasekaran, N. (2012). Neem oil (*Azadirachta indica*) nanoemulsion--a potent larvicidal agent against *Culex quinquefasciatus*. *Pest management science*, 68(2), 158–63.
doi:10.1002/ps.2233
- Anonymous. (1994). *PPCS, Annual Report GAU* (pp. 15–18).
- Anton, N., Benoit, J. ., & Saulnier, P. (2008). Design and production of nanoparticles formulated from nano-emulsion templates-a review. *Journal of Controlled Release*, 128, 185–199.
- Anwar, T., Tahir, S., & Jabbar, A. (1993). Effect of neem oil on longevity and fecundity of chickpea pod borer. *Pakistan Journal of Agricultural Research*, 14(4), 340–343.
- Attwood, D., Mallon, C., Ktistis, G., & Taylor, C. J. (1992). A study on factors influencing the droplet size in nonionic oil-in-water microemulsions. *International Journal of Pharmaceutics*, 88, 417–422.
- Authority, V. (2010). A Publication of the Agri-Food & Veterinary Authority of Singapore Decemember 2010, 0–1.
- Avidov, Z. (1956). Bionomics of the tobacco whitefly (*Bemisia tabaci* Gennad.) in Israel. *Katvim*, 7, 25–41.
- Azab, A. K., Megahed, M. M., & El-Mirsawi, D. H. (1971). On the Biology of *Bemisia tabaci* (Genn.). *Societe Entomologie D'Egypte Bulletin*, 55, 305–315.
- Azeem, A., Rizwan, M., Ahmad, F. J., Iqbal, Z., Khar, R. K., Aqil, M., & Talegaonkar, S. (2009). Nanoemulsion components screening and selection: a technical note. *AAPS PharmSciTech*, 10, 69–76.
- Baseeth, S. ., & Sebree, B. R. (2010). Renewable surfactants in spray adjuvants. *Lipid Technology*, 22, 79–82.
- Bellows TS Jr, M, P. T., RJ, G., & DH, H. (1994). Description of a species of *Bemisia tabaci* (Homoptera: Aleyrodidae). *Annals of the Entomological Society of America*, (87), 195–206.

- Bethke, J., Canas, L., Chamberlin, J., Cloyd, R., Counter, H., Dennehy, T., ... Sanderson, J. (2012). Whiteflies. Retrieved January 02, 2011, from <http://mrec.ifas.ufl.edu/lso/bemisia/bemisia.htm>
- Butler, G. D., Jr., Henneberry, T. J., & Clayton, T. E. (1983). *Bemisia tabaci* (Homoptera: Aleyrodidae): Development, Oviposition, and Longevity in Relation to Temperature. *Annals of the Entomological Society of America*, 76(2), 310–313.
- Byrne, D. N., Bellow, T. S., & Parella, M. P. (1990). Whiteflies in agricultural systems. In *Whiteflies: their bionomics, pest status and management*. (D. Gerling., pp. 227–261). Andover, United Kingdom.: Intercept.
- Caboni, P., Sarais, G., Angioni, A., Garcia, A. J., Lai, F., Dedola, F., & Cabras, P. (2008). Residues and persistence of neem formulations on strawberry after field treatment. *Journal Agricultural Food Chemistry*, 54, 10026–10032.
- Carpinella, M. C., Ferrayoli, C. G., & Palacios, S. M. (2005). Antifungal synergistic effect of scopoletin, a hydroxycoumarin isolated from *Melia azedarach* L. fruits. *Journal Agricultural Food Chemistry*, 53, 2922–2927.
- Chen, F., Wang, Y., Zheng, F., Wu, Y., & Liang, W. (2000). Studies on cloud point of agrochemical microemulsions. *Colloids and Surfaces: Physicochemical Engineering Aspects*, 175, 257–262.
- Cheng, G., & Tao, D. (2005). An experimental study of stability of oil-water emulsion. *Fuel Processing Technology*, 86, 499–508.
- Chow PNP, Grant CA, & Foy CL. (1992). Research and development of agro-adjuvants: a bibliographic survey. In *Second International Symposium on Adjuvants for Agrichemicals: Adjuvants for agrichemicals* (pp. 3–15). Blacksburg, USA.: Second International Symposium on Adjuvants for Agrichemicals.
- Chu, D., Jiang, T., Liu, G. X., Jiang, D. F., Tao, Y. L., Fan, Z. X., Bi., Y. P. (2007). Biotype status and distribution of *Bemisia tabaci* (Hemiptera: Aleyrodidae) in Shandong Province of China based on mitochondrial DNA markers. *Environmental Entomology*, 36, 1290–1295.
- Chu, D., Zhang, Y. J., Brown, J. K., Cong, B., Xu, B. Y., Wu, Q. J., & Zhu, G. R. (2006). The introduction of the exotic Q biotype of *Bemisia tabaci* from the Mediterranean region into China on ornamental crops. *Florida Entomologists*, 89, 168–174.
- Chu, S., DeRisi, J., Eisen, M., Mulholland, J., Botstein, D., Brown, P. O., & Herskowitz, I. (1998). The transcriptional program of sporulation in budding yeast. *Science* 282, 1421, 699–705.

- Cohen, A. C., Chu, C. C., Henneberry, T. J., Freeman, T., Nelson, D., Buckner, J., Aung, L. H. (1998). Feeding biology of the silverleaf whitefly (Homoptera: aleyrodidae). *Chinese Journal of Entomology*, 18, 65–82.
- DeBarro, J. P. (1995). *Bemisia tabaci* biotype B: a review of its biology, distribution and control. (Division of Entomology, Ed.) (2nd ed., p. 36). Canberra, Australia: CSIRO.
- Denholm, I. (1988). Insecticide resistance: an avoidable pest management problem. *Aspects of Applied Biology*, 17, 239–246.
- Devendra, R., Umamahesh, V., Prasad, T. V. R., Prasad, T. G., Ashok, & Asha, S. T. (2004). Influence of surfactants on efficacy of different herbicides in control of *Cyperus rotundus* and *Oxalis latifolia*. *Current Science*, 86(8), 1148–1151.
- Dodia, D. A., Patel, I. S., & Patel, G. M. (2008). Trees of botanical pesticides importance. In D. A. Dodia, I. S. Patel, & G. M. Patel (Eds.), *Botanical pesticides for pest management* (p. 53). Jodhpur, India: SCIENTIFIC PUBLISHERS (INDIA).
- Dreyer, M. (1987). Field and laboratory trials with simple neem products as protectants against pests of vegetables and field crops in Togo. In H. Schmutterer & K. R. S. Ascher (Eds.), *Natural Pesticides from the Neem Tree (Azadirachta Indica A. Juss.) and other Tropical Plants* (pp. 431–447). Nairobi, GTZ, Eschborn: Proc. 3rd International Neem Conference,.
- Dukhin, A. S., & Goetz, P. J. (2002). Ultrasound for characterizing colloids. *Journal of Dairy Sciences*, 88, 1320–1334.
- Eichelkraut, K., & Cardona, C. (1989). Biología, cría massal y aspectos ecológicos de la mosca blanca *Bemisia tabaci* (Gennadius) (Homoptera: Aleyrodidae), como plaga del frijol común. *Turrialba, San José*, 39(1), 51–55.
- El-Aasser, M. S., & Miller, C. M. (1997). In *Polymeric Dispersions: Principles and Applications*. (J. M. Asua, Ed.) (pp. 109–126). Dordrecht, The Netherlands: Kluwer Academic Publishers.
- El-Helaly, M. S., El-Shazil, A. Y., & El-Gayar, F. H. (1971). Biological studies on *Bemisia tabaci* Genn. (Homoptera: Aleyrodidae) attacking cotton in the coastal plain of Israel. *Bulletin of entomological research*, 70, 213–219.
- El-Shafei, G. M. S., El-Said, M. M., Attia, H. A. E., & Mohammed, T. G. M. (2010). Environmentally friendly pesticides: Essential oil-based w/o/w multiple emulsions for anti-fungal formulations. *Industrial Crops and Products*, 31, 99–106.
- Flanagan, J., Kortegaard, K., Pinder, D. N., Rades, T., & Singh, H. (2006). Solubilisation of soybean oil in microemulsions using various surfactants. *Food Hydrocolloids*, 20, 253–260.

- Forgiarini, A., Esquena, J., Gonzalez, C., & Solans, C. (2001). Formation of nano-emulsions by low-energy emulsification methods at constant temperature. *Langmuir*, *17*, 2076–2083.
- Fukuda, K., Olsson, U., & Ueno, M. (2001). Microemulsion formed by alkyl polyglucosides and alkyl glycerol ether with weakly charged films. *Colloid Surfaces B-Biointerfaces*, *20*, 129–135.
- Gahukar, R. T., & Balpande, P. B. (1997). Field evaluation of a new neem based formulation against major insect pests of brinjal. *Pestology*, *21*(11), 14–18.
- Gi, H. J., Chen, S. N., Hwang, J. S., Tien, C., & Kuo, M. T. (1992). Studies of Formation and Interface of Oil-Water Microemulsion. *Chinese Journal of Physics*, *30*, 665–678.
- Gill, J. S., & Lewis, T. (1971). Systemic action of an insect feeding deterrent. *Nature*, *232*, 402–403.
- Gonzalez-Zamora, J. E., Leira, D., Bellido, M. J., & Avilla, C. (2004). Evaluation of the effect of different insecticides on the survival and capacity of *Eretmocerus mundus* Mercet to control *Bemisia tabaci* (Gennadius) populations. *Crop Protection*, *23*(7), 611–618. doi:10.1016/j.cropro.2003.11.011
- Grange, N., & Ahmed, S. (1988). *Handbook of plants with pest control properties*. (John Wiles & Sons, Eds.). New York.
- Green, J. M. (2001). Herbicide adjuvants. *UC Davis WRIC Weed Science School*.
- Green, J. M., & Beestman, G. B. (2007). Recently patented and commercialized formulation and adjuvant technology. *Crop Protection*, *26*, 320–327.
- Gutierrez, J. M., Gonzalez, C., Maestro, A., Sole, I., Pey, C. M., & Nolla, J. (2008). Nano-emulsions: New applications and optimization of their preparation. *Current Opinion in Colloid and Interface Science*, *13*, 245–251.
- Hammad A.F.M, Zournajian H., & Talhouk S. (2001). Efficacy of extracts of *Melia azedarach* L. callus, leaves and fruits against adults of the sweet potato whitefly *Bemisia tabaci* (Homoptera: Aleyrodidae). *Journal Application Entomology*, *125*, 483–488.
- He, Y., Yang, B., Cheng, G., & Pan, H. (2004). Influence of the thermodynamic stability of microemulsion on the size of nanoparticles prepared by a coupling route of microemulsion with homogenous precipitation. *Materials Letters*, *58*, 2019–2022.
- Hess, F. D. (1985). Herbicide absorption and translocation and their relationship to plant tolerances and susceptibility. In S. O. Duke (Ed.), *Weed Physiology. Volume II. Herbicide Physiology* (pp. 191–124). Boca Raton, FL: CRC Press.

- Hoffmann, B., & Platz, G. (2001). Phase and aggregation behaviour of alkylglucosides. *Current Opinion in Colloid and Interface Science*, 6, 171–177.
- Howatt, K. (1994). *Azadirachta indica*: One tree's Arsenal Against Pests. Retrieved from www.colostate.edu/Depts/Entomology/courses/en570/papers/howatt.html.
- Iglauer, S., Wu, Y., Shuler, P., Tang, Y., & Goddard, W. A. (2010). New surfactant classes for enhanced oil recovery and their tertiary oil recovery potential. *Journal of Petroleum Science and Engineering*, 71, 23–29.
- IRAC. (2009). Susceptibility Test Methods Series. *Method No: 015, Insecticide Resistance Action Committee*. Retrieved from www.iraconline.org
- Isman, M. B. (2006). Botanical insecticides, deterrents, and repellents in modern agriculture and increasingly regulated world. *Annual Review of Entomology*, 51, 45–66.
- Jabatan Pertanian Serdang (2011). *Serangan lalat putih, Bemisia tabaci (Hemiptera: Aleyrodidae) ke atas tanaman sayuran (Solanacea sp)*. Serdang, Selangor, Malaysia.
- Jafari, S. M., Assadpoor, E., He, Y., & Bhandari, B. (2008). Re-coalescence of emulsion droplets during high energy emulsification. *Food Hydrocolloids*, 22, 1191–1202.
- James, R. R. (2003). Combining azadirachtin and *Paecilomyces fumosoroseus* (Deuteromycotina: Hyphomycetes) to control *Bemisia argentifolii* (Homoptera: aleyrodidae). *Journal of Economic Entomology*, 96, 25–30.
- Jones, D. R. (2003). Plant viruses transmitted by whiteflies. *European Journal Plant Pathology*, 109(3), 195–219.
- Jurado, E., Bravo, V., Vicaria, J. M., Fernandez-Arteaga, A., & Garcia-Lopez, A.I. (2008). Triolein solubilization using highly biodegradable non-ionic surfactants. *Colloids and Surfaces: Physicochemical Engineering Aspects*, 326, 162–168.
- Kartsev, V. N., Shtykov, S. N., Bogomolova, I. V., & Ryzhov, I. P. (2009). Thermodynamic stability of microemulsion based on sodium dodecyl sulfate. *Journal of Molecular Liquid*, 145, 173–176.
- Knowles, A. (2008). Recent developments of safer formulations of agrochemicals. *Environmentalists*, 28, 35–44.
- Koul, O., & Wahab, S. (2004). Neem research in Asian continents: presents status and future outlook. In G. . Dhaliwal, R. Arora, & Opendar Koul (Eds.), *Neem. Today and In the New Millenium*, (p. 65:74). Kluwer Academic Publishers.
- Kumar, J., & Parmar, S. B. (1998). Neem oil - Chemistry and Bioactivity. *Pesticide Research Journal*, 10(1), 41–43.

- Larew, H. G., & Locke, J. C. (1990). Repellency and toxicity of a horticultural oil against whiteflies on *Chrysanthemum*. *HortScience*, 25, 1406–1407.
- Lee, G. W. J., & Tadros, T. F. (1982). Formation and stability of emulsions produced by dilution of emulsifiable concentrates. Part I. An investigation of the dispersion on dilution of emulsifiable concentrates containing cationic and nonionic surfactants. *Colloids and Surfaces*, (5), 105–115.
- Lin, J. J., & Lin, S. F. (2003). Size, volume fraction, and nucleation of Stober silica nanoparticles. *Journal of Colloid and Interface Science*, (258), 159.
- Lyklema, J. (1995). The isoelectric point of bacteria as an indicator for the presence of cell surface polymers that inhibit adhesion. *Colloid Surfaces B-Biointerfaces*, 4, 191–197.
- Maas, W. (1971). Physical properties of ULV spray residues on biological targets. In W. MAAS (Ed.), *ULV Application and Formulation Techniques* (pp. 81–88). Amsterdam-The Netherlands: N.V. PHILIPS-DUPHAR.
- Madibela, O. R., & Kelemogile, K. M. (2008). Exposure of *Melia azedarach* fruits to *Eimeria* lowers oocyst output in yearling Tswana goats. *Small Ruminant Research*, 76, 207–210.
- Mahaffey, L., & Cranshaw, W. (2007). Some Insect / Disease Control Products Allowed for Use in Certified Organic Production.
- Martin, J. H. (1999). The whitefly fauna of Australia (Sternorrhyncha: Aleyrodidae), a taxonomic account and identification guide. *CSIRO Entomology Technical paper*.
- Martin, J. H., Mifsud, D., & Rapisarda, C. . (2000). The whiteflies (Hemiptera: Aleyrodidae) of Europe and the Mediterranean Basin. *Bulletin of entomological Research*, 90, 407–448.
- Mason, T. G., Wilking, J. N., Meleson, K., Chang, C. B., & Graves, S. M. (2006). Nanoemulsions: formation, structure, and physical properties. *Journal of Physics: Condensed Matter*, 18, 635–666.
- Matter, M. M., Gesraha, M. A., Ahmed, A. A. I., & Farag, N. A. (2002). Impact of neem and chinaberry fruit extracts on the pest/parasitoid (*Pieris rapae*/*Hyposoter ebeninus*) interactions. *Anzeiger für Schädlingskunde*, 75, 13–18.
- Maynard, D. N., & Cantliffe, D. J. (1989). *Squash silverleaf and tomato irregular ripening: new vegetable disorders in Florida*. (p. Fact Sheet VC–37). Florida.
- Mazlan, N., & Mumford, J. (2005). Insecticide use in cabbage pest management in the Cameron Highlands, Malaysia. *Crop Protection*, (24), 31–39.

- Mazumder, M. E. H., & Rahman, S. (2008). Pharmacological evaluation of Bangladeshi medicinal plants for antioxidant activity. *Pharmaceutical Biology*, 46, 704–709.
- McAuslane, H. J. (2012). Sweetpotato Whitefly B Biotype of Silverleaf Whitefly , *Bemisia tabaci* (Gennadius) or *Bemisia argentifolii* Bellows and Perring (Insecta : Hemiptera : Aleyrodidae) 1, (June).
- McWhorter, C. G. (1982). *The use of adjuvants. In: Adjuvants for Herbicides* (pp. 10–25). WSSA, Champaign, IL.
- McWhorter, C. G. (1993). Epicuticular wax on johnsongrass (*Sorghum halepense*) leaves. *Weed Science*, (41), 475–482.
- McWhorter C. G., & Barrentine, W. L. (1988). Spread of paraffinic oil on leaf surfaces of johnsongrass (*Sorghum halepense*) leaves. *Weed Science*, 37, 458–470.
- Miller, P., & P. Westra. (1998). How surfactants work. no.0.559. *Colorado State University Cooperative Extension, Crop Fact Sheet*. Retrieved from <http://www.ext.colostate.edu/pubs/crops,00564.html>
- Morales, D., Gutie, J. M., Garcı, M. J., & Solans, Y. C. (2003). A Study of the Relation between Bicontinuous Microemulsions and Oil / Water Nano-emulsion Formation, (17), 7196–7200.
- Mordue, A. J., & Nisbet, A. J. (2000). Azadirachtin from the neem tree *Azadirachta indica*: its actions against insects. *Annals of the Entomological Society of Brazil*, 29, 615–632.
- Morione, E., & Luis-Arteaga, M. (1999). *Viral diseases*. (J. C. van L. and Y. E. In R. Albajes, M. L. Gulino, Ed.) (pp. 16–33). Dordrecht, The Netherlands: Kluwer Academic Publishers.
- Mulla M.S., & Su T. (1999). Activity and biological effects of neem products against arthropods of medical and veterinary importance. *Journal of the American Mosquito Control Association*, 15(2), 133–152.
- Mulqueen, P. (2003). Recent advances in agrochemical formulation. *Advances in Colloid and Interface Science*, 106, 83–107.
- Muneef A.M., & Zuhair M.A. (2006). Effective of some Neem products (*Azadirachta indica* A. Juss) on growth and development of the beet armyworm (*Spodoptera exigua*). In *Ninth Arab Congress of Plant Protection, 19-23 November 2006*. Damascus, Syria.
- Muregi, F. W., Chhabra, S. C., Njagi, E. N. M., Lang’at-Thoruwa, C. C., Njue, W. M., Orago, A. S. S., Ndiege, I. O. (2004). Anti-plasmodial activity of some Kenyan medicinal plant extracts singly and in combination with chloroquine. *Phytotherapy Research*, 379–384.

- Murrell, J. N., & Jenkins, A. D. (1994). *Properties of liquids and solutions*. (J. Wiley & Sons, Eds.) (2nd. Editi., pp. 148–150). New York.
- Oliveira, M. R. ., Henneberry, T. J., & Anderson, P. (2001). History, current status, and collaborative research projects for *Bemisia tabaci*. *Crop Protection*, 20(9), 709–723.
- Osborne, L. S. (2008). Invasion and response: impacts of *Bemisia* on worldwide agriculture. *Journal Insect Science*, 8, 37–38.
- Osborne, L. S., & Landa, Z. (1992). *Biological Control of Whiteflies with Entomopathogenic Fungus*. *Florida Entomologists* (Vol. 75, pp. 456–471). Florida Entomologists.
- Pakpayat, N., Nielloud, F., Fortune, R., Tourne-Peteilh, C., Villareal, A., Grillo, I., & Bataille, B. (2009). Formulation of ascorbic acid microemulsions with alkyl polyglucosides. *European Journal of Pharmaceutics and Biopharmaceutics*, 72, 444–452.
- Palumbo, J. C., Horowitz, A. R., & Prabhaker, N. (2001). Insecticidal control and resistance management for *Bemisia tabaci*. *Crop Protection*, 20, 739–765.
- Pan, H., Chu, D., Ge, D., Wang, S., Wu, Q., Xie, W., ... Zhang, Y. (2011). Further spread of and domination by *Bemisia tabaci* (Hemiptera: Aleyrodidae) biotype Q on field crops in China. *Journal of Economic Entomology*, (104), 978–85.
- Patel, H. M., Jhala, R. C., Pandya, H. V., & Patel, C. B. (1992). Biology of whitefly (*Bemisia tabaci*) on okra (*Hibiscus esculentus*). *Indian Journal of Agricultural Sciences*[1] H. M. Patel, R. C. Jhala, H. V. Pandya, and C. B. Patel, *Indian Journal of Agricultural Sciences*, vol. 62, no. 7, pp. 497–499, 1992., 62(7), 497–499.
- Porras, M., Martínez, a., Solans, C., González, C., & Gutiérrez, J. M. (2005). Ceramic particles obtained using W/O nano-emulsions as reaction media. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, 270-271, 189–194. doi:10.1016/j.colsurfa.2005.06.018
- Porras, M., Solans, C., González, C., Martínez, a., Guinart, a., & Gutiérrez, J. M. (2004). Studies of formation of W/O nano-emulsions. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, 249(1-3), 115–118. doi:10.1016/j.colsurfa.2004.08.060
- Prabhaker, N., Toscano, N. C., & Coudriet, D. L. (1989). Susceptibility of the immature and adult stage of the sweetpotato whitefly (Homoptera: aleyrodidae) to selected insecticides. *Journal of Economic Entomology*, 82, 983–988.
- Priesner, B., & Corbett, B. (1994). *Bemisia tabaci*, 1(178), 1–7.
- Rahman, S. A., Sivapragasam, A., Loke, W. H., & Ruwaida, M. (2000). *Whiteflies in Malaysia* (p. 6). Universiti of Malaya, Kuala Lumpur.

- Rao, Q., Luo, C., Zhang, H., Guo, X., & Devine, G. J. (2011). Distribution and dynamics of *Bemisia tabaci* invasive biotypes in central China. *Bulletin of entomological research*, (101), 81–8.
- Rapisarda, C., & Garzia, G. T. (2002). Tomato yellow leaf curl *Sardinia* virus and its vector *Bemisia tabaci* in Sicilia (Italy): present status and control possibilities. *EPPO Bulletin*, 32(1), 25–29. doi:10.1046/j.1365-2338.2002.d01-22.x
- Rasdi, M., & Saiful, M. (2009). (Homoptera : Aleyrodidae) on Brinjal, (1986), 27–32.
- Rasdi, M. Z. (2005). Biology, distribution and effect of selected insecticides against whitefly (*Trialeurodes vaporariorum* Westwood and *Bemisia tabaci* Gennadius) on brinjal, *Solanum melongena* L. Universiti Teknologi MARA.
- Roditakis, E., Grispu, M., Morou, E., Kristoffersen, J. B., Roditakis, N., Nauen, R., Tsagkarakou, A. (2009). Current status of insecticide resistance in Q biotype *Bemisia tabaci* populations from Crete. *Pest management science*, (65), 313–322.
- Ronald F. L.Mau, & Jayma L.Martin Kessing. (2007). *Bemisia tabaci* (Gennadius). Retrieved January 25, 2011, from http://www.extento.hawaii.edu/kbase/crop/Type/b_tabaci.htm
- Rosano, H. L., Weiss, A., & Gerbacia, W. E. (1967). In Proceedings of the 12th International Congress on Surface Active Substances (p. 1,453). Moscow, USSR, September.
- Ruscoe, C. N. . (1972). Growth disruption effects of an insect antifeedant. *Nature New Biology*, 236, 159–160.
- Rybinski, W., Guckenbiehl, B., & Tesmann, H. (1998). Influence of co-surfactants on microemulsions with alkyl polyglucosides. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, (142), 333–342.
- Ryerse, J. S., Downer, R. A., Sammons, R. D., & Feng., P. C. C. (2004). Effect of glyphosate spray droplets on leaf cytology in velvetleaf (*Abutilon theophrasti*). *Weed Science*, (52), 302–309.
- Sadurni, N., Solans, C., Azemar, N., & Garcia-Celma, M. J. (2005). Studies on the formation of O/W nano-emulsions, by low-energy emulsification methods, suitable for pharmaceutical application. *European Journal of Pharmaceutical Sciences*, 26, 438–445.
- Saxena, R. C. (1993). Scope of neem for developing countries. In *Souvenir World Neem Conference* (pp. 30–36). Bangalore.
- Saxena, R. C., & Khan, Z. R. (1985). Effect of neem oil on survival of *Nilaparvata lugens* (Homoptera: Delphacidae) and on grassy stunt and ragged stunt virus transmission. *Journal of Economic Entomology*, 78, 1079–1082.

- Schmutterer, H. (2002). *The Neem Tree* (2nd ed., p. 829). Mumbai, India: The Neem Foundation.
- Schoonhoven, A. V. (1978). The use of vegetable oils to protect stored beans from bruchid attack. *Journal of Economic Entomology*, *71*, 254–256.
- Shadmany, M., Omar, D., & Muhamad, R. (2013). First Report of *Bemisia tabaci* (Hemiptera : Aleyrodidae) Biotype Q in Malaysia, *96*(1), 280–282.
- Shafiq, S., Faiyaz, S., Sushma, T., Farhan, J. A., Khar, A. K., & Ali, M. (2007). Design and development of ramipril nanoemulsion formulation. *Journal of Biomedical Nanotechnology*, *3*, 28–44.
- Shafiq, S., Shakeel, F., Talegaonkar, S., Ahmad, F. J., Khar, R. K., & Ali, M. (2007). Development and bioavailability assessment of ramipril nanoemulsion formulation. *European Journal of Pharmaceutics and Biopharmaceutics*, *66*, 227–243.
- Sharma, V. K., Vasudeva, R., & Howden, C. W. (1999). A survey of gastroenterologists' perceptions and practices related to *Helicobacter pylori* infection. *American Journal of Gastroenterology*, *94*, 3170–3174.
- Siddig, S. A. (1991). Evaluation of neem seed and leaf water extracts and powders for the control of insect pests in Sudan. In *Technical Bulletin* (p. 39). Agric. Res. Corp., Shambat Research Station.
- Solans, C., Esquena, J., Forgiarini, A., Uson, N., Morales, D., & Izquierdo, P. (2003). Absorption and aggregation of surfactants in solution, in Nano-emulsions: Formation, Properties and Applications. (K. L. Mittal & O. S. Dinesh, Eds.) (pp. 525–554). New York, NY: Marcel Dekker.
- Solans, C., Izquierdo, P., Nolla, J., Azemar, N., & Garcia-Celma, M. J. (2005). Nanoemulsions. *Current Opinion in Colloid and Interface Science*, *10*, 102–110.
- Sulek, M. W., & Wasilewski, T. (2006). Tribological properties of aqueous solutions of alkyl polyglucosides. *Wear*, *260*, 193–204.
- Sunder, S. (2006). The neem tree, Phytochemistry of neem ingredients, Uses of neem and its products. In *Neem for Organic Farming and Health* (pp. 8–10, 19–23, 70–71). J.V Publishing House.
- Syed, A. R., Sivapragasam, A., Loke, W. H., & Mohamad Roff, M. N. (2000). Whiteflies infesting vegetables in Malaysia. In *Plant Resource Management Seminar*. Sarawak, Malaysia.
- Syed, A. R., Sivapragasam, A., Loke, W. H., & Mohd. Roff, M. N. (2000). Whiteflies infesting vegetables in Malaysia Organized by. In *The plant resource management seminar* (pp. 38–44). Sarawak, Malaysia.: MAPPS, DoA Sarawak and SIAS.

- Syed Abdul Rahman, S.A. R., Sivapragasam, A., Loke, W. H., & Roff, M. N. (2000). *Whiteflies Infesting Vegetables in Malaysia*. (pp. 38–43). MARDI Serdang, CABI-SEARC.
- Tadros, T. F., Izquierdo, P., Esquena, J., & Solans, C. (2004). Formation and stability of nano-emulsions. *Advances in Colloid and Interface Science*, 108-109, 303–318.
- Taylor, P. (2003). Ostwald ripening in emulsions: estimation of solution thermodynamics of the dispersed phase. *Advances in Colloid and Interface Science*, (106), 261–285.
- Trotta, M., Gallarate, M., Pattarini, F., & Carlotti, M. E. (1999). Investigation of the phase behaviour of systems containing lecithin and 2-acyl lysolecithin derivatives. *International Journal of Pharmaceutics*, (190), 83–89.
- Tu, M., & Randall, J. M. (2003). *Adjuvants*. In: *Weed Control Methods Handbook. Tools and Techniques for Use in Natural Areas*. The Nature Conservancy. (T. M., H. C., & Randall J.M, Eds.) (pp. 1–25). Wildland Invasive Species Team.
- Usón, N., Garcia, M. J., & Solans, C. (2004). Formation of water-in-oil (W/O) nano-emulsions in a water/mixed non-ionic surfactant/oil systems prepared by a low-energy emulsification method. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, 250(1-3), 415–421. doi:10.1016/j.colsurfa.2004.03.039
- Van Gysel, A. B., & Musin, W. (2005). Methylamines. In *Ullmann's Encyclopedia of Industrial Chemistry*. Wiley-VCH Verlag, Weinheim.
- Varona, S., Martin, A., & Cocero, M. J. (2009). Formulation of a natural biocide based on lavender essential oil by emulsification using modified starches. *Chemical Engineering and Processing: Process Intensification*, 48, 1121–1128.
- Verkerk, R. H. J., & Wright, D. J. (1993). Biological activity of neem seed kernel extracts and synthetic azadirachtin against larvae of *Plutella xylostella* L. *Pesticide Science*, 37, 83–91.
- Wanamarta, G., & Penner, D. (1989). Foliar absorption of herbicides. *Review of Weed Science*, (4), 215–231.
- Wang, C. J., & Liu, Z. Q. (2007). Foliar uptake of pesticides—Present status and future challenge. *Pesticides Biochemistry and Physiology*, 87, 1–8.
- Warisnoicharoen, W., Lansley, A. B., & Lawrence, M. J. (2000). Nonionic oil-in-water microemulsions: the effect of oil type on phase behaviour. *International Journal of Pharmaceutics*, 198, 7–27.
- Wheeler, G. S., Slansky, F. J., & Yu, S. J. (2001). Food consumption, utilization and detoxification enzyme activity of larvae of three polyphagous noctuid moth species when fed the botanical insecticide rotenone. *Entomologia Experimentalis et Applicata*, (98), 225–239.

- Xu, J., Wang, W. L., & Liu, S. S. (2006). The occurrence and infestation of *Bemisia tabaci* biotype Q in partial regions of Zhejiang province. *Plant Protection*, 32, 121.
- Xu, Y. X., Hanna, M. A., & Josiah, S. J. (2007). Hybrid hazelnut oil characteristics and its potential oleochemical application. *Industrial Crops and Products*, 26, 69–76.
- Zielinska, K., Wilk, K.A., Jezierski, A., & Jesionowski, T. (2008). Microstructure and structural transition in microemulsions stabilized by aldonamide-type surfactants. *Journal of Colloid and Interface Science*, 321, 408–417.

