A   Introduction

1   Overview – Pesticides in Precision Agriculture

Pesticides are an important part of the technology used to control plant diseases and reduce losses caused by weeds and pests. Their use is a proven method of enhancing agricultural production and crop quality. Without pesticides it is estimated that as much as 45% of the world's food supply would be lost to pests: 30% to weeds, pests and diseases before harvest and another 15% between harvest and use. It is also estimated that losses would increase a further 10% if no pesticides were used at all; specific crop losses would range from zero to 100% (Pimentel *et al.*, 1992). Agricultural productivity has increased within the last 60 years due to improved crop cultivation methods, progress in breeding, fertilization, and irrigation as well as chemical plant protection. The global conventional agrochemical market has risen from $580 million in 1960 (County Natwest WoodMac, 1991) to about $30 billion in 2006 (Underwood, 2007). Herbicides are estimated to represent about 48% of the total agrochemical market, followed by insecticides (25%), fungicides (24%), and others (3%) making up the balance (Underwood, 2007). Remarkably, the herbicidal active ingredient glyphosate has turned into the largest selling agrochemical in the world amounting 10% of the total agrochemical market and about 20% of the herbicidal market. The global adjuvant market is estimated to represent about $1.5 billion globally (Underwood, 2007).

Commonly, farmers apply inputs such as pesticides or fertilizers uniformly in the field despite of great heterogeneities in soil type, weed density and distribution, nutrient availability, or disease pressure. However, this implies additional economical expenses and detrimental effects on the environment if agrochemicals are applied unnecessarily to disease- or weed-free areas.

The consideration of the aforementioned in-field variability is the main element of 'Precision Agriculture'. With development of new technologies such as global positioning system (GPS), concepts based on site-specific farming and variable rate technology, Precision Agriculture contributes significantly to reducing pesticide and fertilizer input on crop and environment. In this context, great advances have been made especially with camera-based weed recognition and classification in the field (Backes, 2005; Dammer and Wartenberg, 2007; Gebhardt, 2007). Based on this

information, it is possible to generate weed distribution maps and thereof site-specific application maps (Gerhards and Oebel, 2006).

2 Influencing factors on herbicide deposit formation

After a spray solution droplet has been retained on a plant surface, several physical and chemical processes influence droplet spreading, water evaporation from the droplet and the final form and nature of active ingredient (a.i.) deposition. All a.i.'s are distributed on targets by way of deposits that have structure. This structure is comprised of elements of different size, number, and sometimes concentration. These elements all contribute to create a dose that is the biologically effective unit of a.i. delivery. Deposit quality has been shown to have a profound influence upon the quantity of active ingredient required to control a pest (e.g. Downer *et al.*, 1999; Ebert *et al.*, 1999).

2.1 Physical processes during formation of herbicide deposits

The basic physical factors include those that determine the evaporation of spray liquid according to droplet and leaf dimensions, such as droplet size, weather conditions and formulants present (Baur and Pontzen, 2007). Evaporative loss of the carrier liquid (which is mostly water but can also be oil or a liquid fertilizer, e.g. liquid ammonium nitrate urea, LAU) already begins during transport of the droplet to the target surface (Hall *et al.*, 1993). After impaction, the droplet spreads along the water/leaf surface interface and evaporates via a mechanism of capillary flow (Fig. 1) wherein the liquid phase is abandoned preferentially at the edge of the drying droplet (Deegan *et al.*, 1997; Faers, 2007).

During evaporation, the a.i. precipitates out as a residue at the contact area between the droplet and the leaf surface resulting in an a.i. deposition pattern which may be smaller, the same, or larger than the original droplet footprint (Bukovac *et al.*, 2003). The physical nature and shape of the deposition area is closely related to the physicochemical properties of the spray solution (Falk, 1994) and the morphology, fine structure and chemistry of the plant surface (Stevens and Baker, 1987).
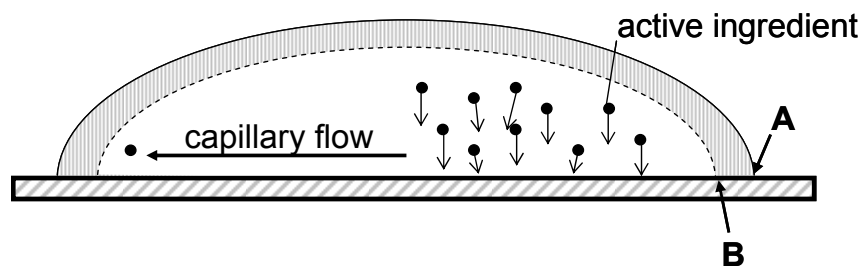
Figure 1: Schematic illustration of an evaporating droplet (modified after Deegan *et al.*, 2000; Faers, 2007). During evaporation, the air/water interface moves from the solid line to the dashed line, and the contact line will move from A to B. The active ingredient precipitates out as a residue at the contact line between droplet and leaf surface.

2.2 Active ingredient

Movement and metabolism of an a.i. is determined by the physicochemical properties of the compound and the physiological/biochemical characteristics of the target organism. The physicochemical properties which the formulation chemist may work with are therefore fixed by biological screening data, unless the a.i. is suitable for the synthesis of a salt (Stock and Davies, 1994). It becomes apparent that a.i.'s do not always have appropriate physicochemical properties to be formulated in their most biologically active form.

The most important factors which determine the feasibility of a formulation type are: physical form of the a.i. (Tab. 1), melting point, solubility characteristics in aqueous and organic media, and chemical stability (Scher, 1988).

Table 1: Physical properties of water-soluble pesticide formulations.

Formulation	Physical form of the a.i.	Physical form of formulation	Physical form in the spray tank
Emulsifiable Concentrate (EC)	Liquid	Solution	Emulsion (o/w)
Soluble Liquid (SL)	Liquid	Solution	Real solution
Soluble Powder (SP)/ Soluble Granule (SG)	Solid	Powder/ Granule	Real solution
Suspension Concentrate (SC)	Solid	Suspension	Suspension
Wettable Powder (WP)	Solid	Powder	Suspension
Water Dispersable Granule (WG)	Solid	Granule	Suspension
Oil Dispersion (OD)	Solid	Oil suspension	Emulsion (o/w)

Source: (Stock and Davies, 1994; Miller and Westra, 1998a; Anonymous, 2005)

Moreover, particular properties of the a.i. such as molecular weight, polarity and others influence the adhesion to plant surface, deposit formation, and/or diffusion into the leaf tissue.

All formulations from Tab. 1 are used for herbicides, whereas not all are suitable for foliar application (Miller and Westra, 1998a). Different formulations containing the same a.i. can lead to disparate deposit structures (Hess *et al.*, 1981; Rouanet *et al.*, 2001). The melting point also influences the physical form of an a.i. within a dried deposit. Baker *et al.* (1992) have reported in an extensive study with 26 a.i.'s (herbicides, fungicides, growth regulators, insecticides and model compounds) that chemicals with a melting point of greater than 200°C formed crystalline deposits whether formulated with or without surfactant. Chemicals with a melting point between 150° and 200°C formed crystalline deposits in the absence of surfactant but amorphous deposits in the presence of surfactant. Finally, chemicals with a melting point between 40° and 135°C formed amorphous deposits in both the presence and absence of surfactant.

Water solubility is closely related to the octanol/water partition coefficient (P) (Baker and Hunt, 1988), and the logarithmic value (log P) is used to describe the lipophilicity of a compound (Wang and Liu, 2007). Chemicals with log P < 0 are considered as hydrophilic, with log P 0.5 – 2.5 as chemicals of median lipophilicity and those with log P > 3 as lipophilic (Baker and Hunt, 1988; Wang and Liu, 2007). Experiments of Baker and Hunt (1988) have shown that the physical form of dried deposits varied with the polarity of the a.i., and uniform deposits without crystallization were obtained particularly for chemicals of median lipophilicity.

2.3 Adjuvants

An agrochemical active ingredient is often ineffective and of limited value to the end user if applied to the target surface alone. Therefore, so-called formulation additives and adjuvants, respectively, can directly be built into pesticide formulations or added as a tank-mix to alter spray solution flow rate, droplet size, dynamic surface tension, spreading over and wetting of plant leaf surfaces, uptake by plant foliage, pests and pathogens (Zabkiewicz, 2003). As a result, they increase effectiveness, ensure consistency, reduce rates, and/or minimize unwanted side effects such as spray drift (Green, 2001). They may constitute up to 30% of the pesticide formulation volume (Knowles, 2001). There are various definitions of the term 'adjuvant' mentioned in the literature, all basing upon the conclusion that an adjuvant is 'a substance without

significant pesticide properties added to an agrochemical or to a pesticide spray mixture to improve or modify the activity of the agrochemical (Anonymous, 1988; Underwood *et al.*, 2001). According to several parameters, adjuvants can be arranged in groups and usually they are classified by taking into account their chemical composition or objective of use. The main chemical classes, in order of commercial importance, are: surfactants, oils, polymers, polymer-forming compounds, and inorganic salts (Holloway, 1993). Among adjuvants, surfactants are probably the most important components for herbicides, as they are designed to improve the dispersing/ emulsifying, absorbing, spreading, and sticking properties and can speed up foliar uptake and improve biological effectiveness of a.i. (Kirkwood, 1993; Miller and Westra, 1998b). The term surfactants (<u>sur</u>face <u>act</u>ive <u>agents</u>) relates to their principal action at interfaces (Green and Hazen, 1998). According to Miller and Westra (1998c) surfactants can be divided into five major classes with respect to their chemical composition: non-ionic surfactants, crop oil concentrates, nitrogen-surfactant blends, esterified seed oils and organosilicone surfactants (Tab. 2).

Table 2: Surfactant classes according to their chemical composition.

Surfactant class	Chemical composition	Function
Non-ionic surfactants	Linear or nonyl-phenol alcohols and/or fatty acids	Reduce surface tension; improve spreading, sticking and herbicide uptake
Crop oil concentrates	Blend of paraffinic-based petroleum oils (80 - 90%) and surfactants (20 - 10%)	Reduce surface tension; improve spreading and herbicide uptake
Nitrogen-surfactant blends	Premix combinations of various forms of nitrogen (ammonium sulphate or 28% N) and surfactants	Reduce surface tension; improve spreading
Esterified seed oils	Fatty acids from seed oils reacted with an alcohol to form esters	Reduce surface tension; improve spreading, sticking and herbicide absorption
Organosilicone surfactants	Entirely silicone or blends of silicone with surfactants	Strong reduction of surface tension; improve absorption and rainfastness; facilitate stomatal and cuticular uptake

Source: Miller and Westra (1998c)

All surfactants are compounds that contain hydrophilic and lipophilic groups and the balance between the opposing effects can be determined (HLB – hydrophilic/lipophilic balance). Surfactants of low HLB value (1-10) are relatively lipophilic while those of high HLB (10-20) are relatively water soluble (Kirkwood, 1987). In most surfactants

the hydrophilic part consists of an ethylene oxide (EO) or a propylene oxide chain, and depending on the length of the EO chain the surfactant becomes a more hydrophobic (lower EO units) or a more hydrophilic (higher EO units) character (Maag, 1981; Wang and Liu, 2007).

2.3.1 Influence on deposit formation

The desired effects of surfactants on deposit formation are: uniform a.i. distribution and close contact with the leaf surface, prevention or delay in a.i. precipitation or crystallization, extended droplet drying time, and improved persistence and protection against losses (Matysiak, 1995). Indeed, surfactants play a significant role in residue formation and in determining the physical state of the residue by modifying the surface tension of the spray solution and by enhancing wetting of the plant surface and subsequent spreading during droplet impaction. Surfactants in the residue may serve as solvents, complexing agents and humectants (Bukovac *et al.*, 2003). As mentioned before, the a.i. and non-volatile spray additives precipitate out as a residue, varying in size, form and consistency, as the carrier phase and any volatile spray additives evaporate. The residue may be uniformly distributed over the footprint, as a number of individual masses within the footprint, often associated with specific morphological features that may be more easily wetted (e.g. over antiklinal walls, veins, base of trichomes, guard cells), or concentrated in an annulus at the periphery of the residue footprint (Bukovac *et al.*, 1995). The a.i. penetrates into the cuticle depleting the residue deposit at the residue:leaf surface interface (Bukovac *et al.*, 2003). Of particular interest is the often observed ring-like formation of deposits on diverse surfaces, which became popular with the explanation of the coffee-ring effect (Deegan *et al.*, 2000; Hu and Larson, 2006). In an evaporating droplet, the Marangoni flow, i.e. a surface tension gradient-driven convection based on temperature gradients, is the driving force in deposition of residues (Fig. 2).

Particles from the air/water interface are carried inward towards the top of the droplet and then downward where they can adsorb to the surface or be carried along the water/leaf surface interface to the edge where the particles again can either be deposited or recirculated along the air/water interface back towards the top of the droplet (Hu and Larson, 2006).
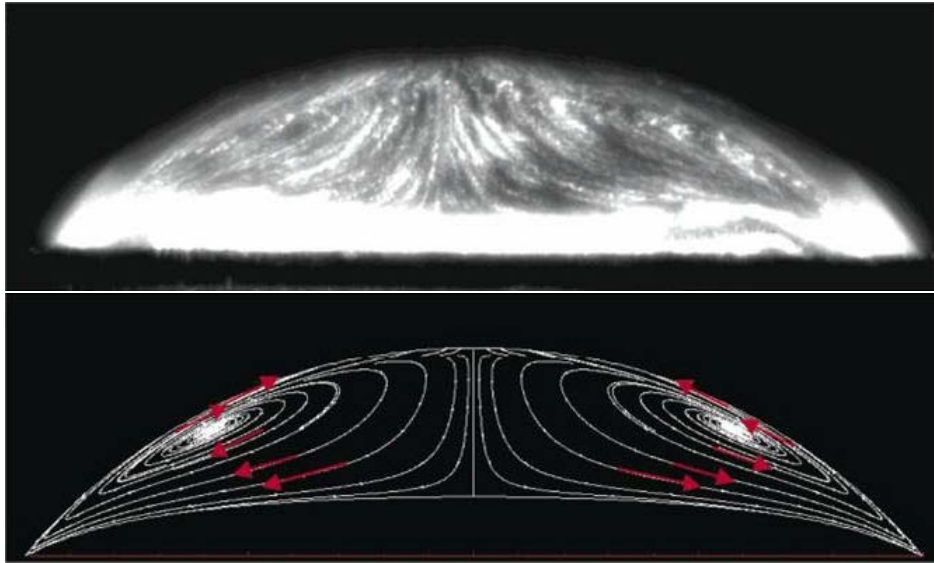
Figure 2: Marangoni flow field in a drying droplet (adapted from Hu and Larson, 2006).

In case of the ring formation, addition of a surfactant causes a suppression of the Marangoni flow. As consequence, the flow towards the edge transports suspended particles present, which makes the surface even rougher and the contact line even stickier resulting in pinning of the contact line at the droplet periphery (Deegan *et al.*, 1997; Faers, 2007).

Another deposition determining factor is the critical micelle concentration (cmc) of the surfactant. Beginning at a well defined concentration (cmc) of a surfactant, aggregates (micelles) are formed and substantial amounts of a.i. may partition into these micelles (Hoffmann and Ulbricht, 1981). Thus, the a.i. is unable to get into close contact to the leaf surface and penetration is diminished (Kirkwood, 1999).

2.3.2 Impact on uptake and biological efficacy

Surfactants added to foliar sprays can affect the uptake process by one or more of the following ways (Wanamarta and Penner, 1989; Uhlig and Wissemeier, 2000; Wang and Liu, 2007):

a)	increasing leaf wettability and spray retention on plant surfaces by reducing the surface tension;

b)	ensuring a greater spray contact between the droplets and the leaf surface by eliminating air films, especially on difficult-to-wet leaf surfaces;

c)   acting as humectant, delaying droplet drying through a hygroscopic effect and therefore lengthening the period of penetration;

d)   preventing or delaying crystal formation in the droplet residue;

e)   acting as co-solvent;

f)   increasing the direct entry through stomata;

g)   increasing the permeability of leaf cuticles;

h)   increasing the permeability of the plasmalemma; and/or

i)   facilitating movement through the cell wall by reducing interfacial tension between polar and apolar regions.

It has been proposed that surfactants increase the penetration of hydrophilic compounds by hydrating the cuticle, whereas they accelerate the uptake of lipophilic chemicals by increasing the fluidity (or reducing the viscosity) of the cuticular wax (Hess and Foy, 2000). It is generally agreed that the enhancement process is the net result of complex interactions between a.i., surfactant, and target species which are strongly influenced by surfactant concentration and composition, especially mean molar ethylene oxide (EO) content, and the physicochemical properties of the particular a.i. (Stock *et al.*, 1992). The presence of surfactants can increase the biological efficacy in a field at a given concentration of a.i. or may sometimes allow to reduce the a.i. rate per hectare as recommended without surfactants (Cronfeld *et al.*, 2001).

2.4   Leaf surface characteristics

The aerial surfaces of all plants are covered by a thin, continuous layer, termed the plant cuticle or cuticular membrane which minimizes water loss from the plant and also acts as a barrier to the penetration of foliage-applied chemicals (e.g. Riederer and Schreiber, 2001; Schreiber, 2005). This outer wall of the epidermal cells is composed of a framework of polymeric cutins with embedded cuticular wax and soluble waxes deposited on the surface as epicuticular wax (Kolattukudy, 1980). Cuticular waxes is a general term for complex mixtures of homologue series of long chain aliphatics like alkanes, alcohols, aldehydes, fatty acids, and esters with the addition of varying proportions of cyclic compounds like pentacyclic triterpenoids and hydroxycinnamic acid derivatives (e.g. Kolattukudy, 1980; Baker, 1982). Epicuticular waxes can appear in crystalline form (e.g. platelets, rodlets, scales, granules) or as an amorphous (smooth) layer (Barthlott *et al.*, 1998). Wax is primarily nonpolar (hydrophobic) but varies among species with regard to the degree of hydrophobicity, primarily due to

chemical composition of the wax. Hydrocarbons are highly hydrophobic, whereas alcohols and acids are less hydrophobic (Chachalis *et al.*, 2001a). Chemical composition, orientation and physical structure of the wax platelets may all affect wettability, absorption of a.i. and deposit formation (Kirkwood, 1987; Wanamarta and Penner, 1989; Falk, 1994). It is reported that ring-deposits rather appear on surfaces carrying contiguous arrangements of crystalline wax than on smooth leaf surfaces (Stevens and Baker, 1987) and that surfactants increase droplet spread more on crystalline than on smooth wax (Singh and Sharma, 2001). However, waxes cannot be considered as the main barrier for adsorption and diffusion of a.i. (Santier and Chamel, 1998; Schönherr, 2000). Leaf roughness and microstructure, i.e. presence, type and distribution of trichomes, presence of stomata as well as ridged antiklinal cell walls and veins, influence the distribution of a.i. on a leaf surface (Holloway, 1970; Hess and Falk, 1990) and affect the amount and quality of interface contact between the deposit and the leaf (Falk, 1994; Green, 2001). Additionally, droplet drying on plain wax surfaces may be faster than on others whose leaf surface probably provides protected compartments between the ridges and greater number of trichomes thus preventing rapid drying of the spray on the leaf surface (Singh and Singh, 1995).

The residue may be uniformly distributed over the droplet footprint, as a number of individual masses within the footprint, or concentrated in an annulus at the periphery of the residue footprint (Bukovac *et al.*, 1995). According to Hess *et al.* (1974), closely spaced trichomes might create air pockets beneath the droplets that would prevent leaf surface contact. As a rule, deposits on smooth leaf surfaces are circular in shape but extensions along ridged venations can occur (Baker, 1982). Nevertheless, the distribution of deposits is crucial for the penetration of a.i. and the stickiness of the deposit against wash-off by rain (Baur and Pontzen, 2007).

2.5 Influence of deposits on uptake of a.i.

It is known that uptake of pesticides into plant leaves is a diffusion process. Thereby, ionic compounds use aqueous polar paths of diffusion, whereas lipophilic molecules diffuse along the lipophilic wax and cutin domains (Schreiber, 2005). Concentration gradient of a.i. between the donor (leaf surface deposit) and the receiver (internal leaf tissues) is the driving force for the uptake process. Droplet spreading will dilute the amount of a.i. per unit of leaf surface (a.i. dose on leaf surface), thus reducing the concentration gradient (Liu, 2003). Also stomatal infiltration may occur, but only a

small proportion of stomata is involved in uptake processes (Eichert *et al.*, 1998; Eichert and Burkhardt, 2001).

3   Objective of the study

Many factors influence the performance of foliar-applied agrochemicals. Here, plant surface characteristics (e.g. micromorphology) and surfactants play a major role. In this study the target-oriented adaptation of the physicochemical properties from selected herbicides (glyphosate and glufosinate) and calcium leaf fertilizers to single weed species and horticultural crops is evaluated. A critical point in this context is the formation of a.i. deposits within a droplet footprint and the impact on penetration and biological efficacy. In the past, many studies and mathematical models overlooked the importance of shape and nature of a.i. deposits when investigating penetration of systemic compounds (Chachalis *et al.*, 2001b; Lamb *et al.*, 2001; Feng *et al.*, 2003; Mercer, 2007). Instead, uniform distribution of a.i. within the droplet spreading zone is assumed.

In this study a homologous series of rapeseed oil ethoxylates has been used as surfactants. The oligomere triglyceridethoxylates vary in their number of ethylene oxide (EO) units in the hydrophilic chain (Fig. 3) while keeping the lipophilic part constant: an average number of 5 (Agnique RSO 5®), 10 (Agnique RSO 10®), 20 (Agnique RSO 20®), 30 (Agnique RSO 30®), and 60 (Agnique RSO 60®) units has been employed. The use of the homologous RSO series allowed a systematic approach in varying the degree of hydrophilicity of the treatment solutions.
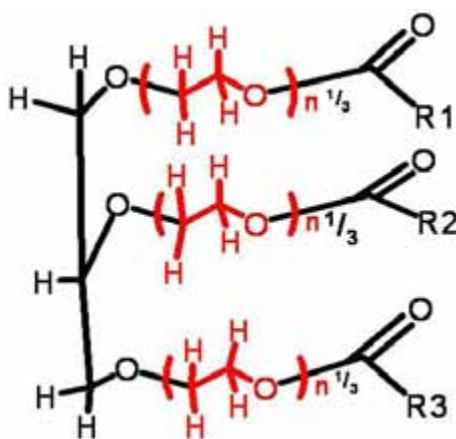

Figure 3: Schematic representation of the triglyceridethoxylate structure (Agrimul RSO®) with "n" units of ethylene oxide (EO).