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Resistance of tick *Rhipicephalus microplus* to acaricides and control strategies

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Abstract
Ticks are bloodsucking arthropods mandatory and represent a real problem for cattle. The aim of this review is to make an inventory of acaricide resistance and ways to fight against the tick *Rhipicephalus microplus*. The main classes of synthetic miticides are arsenical, organochlorine, organophosphates, amidines, macrocyclic lactones, growth regulators and phenylpyrazoles. Pyrethroids are the most widespread and used acaricide in the fight against ticks. Resistance to these classes acaricide was reported in all countries where the tick *Rhipicephalus microplus* is present. To deal with these ectoparasites, a variety of control methods are used. These include ecological, biological, and genetic approaches. Proper control of the ticks will allow farmers to save and limit the losses related to diseases transmitted by ticks. It is urgent to propose and develop new approach to fight against tick *Rhipicephalus microplus*.

Keywords: *Rhipicephalus microplus* resistance, control method, cattle

1. Introduction
Rhipicephalus microplus [1] is a cattle tick, whose development cycle is monoxenous. Although it is originally from India and Indonesia, this tick has shown easy adaptation to tropical and subtropical conditions [2]. It is one of the major obstacles to the development of cattle breeding. Historically, it was introduced in Eastern and Southern Africa from Southern Asia through Madagascar after the outbreak of rinderpest in 1896 [3]. In West Africa, it was discovered for the first time in Ivory Coast by imports of live cattle from Brazil in 2007 [4, 5] and recently in Benin, Burkina Faso and Mali [6]. An economic loss (reduction of weight, decrease of meat and milk quality) due to presence of tick is huge. In Brazil for example, losses due to its direct and indirect impact have been estimated to two billion US dollars in 2000 [7]. In Australia, they are estimated to 100 million Australian Dollars a year [8] and USD 48 million a year in Mexico [9]. Besides, tick can transmit pathogens such as viruses, bacteria and protozoa. The prevalence of tick borne diseases is 1.7% to 36% in the Caribbean [10], 6.25% in Zambia, and 25.0% in Zimbabwe [11]. A prevalence of 9% and 10.8% was recorded in Burkina Faso and Benin respectively [12]. The use of synthetic acaricide currently remains the only way to fight [13]. Introduced in 1895, Arsenical compounds were first used to control ticks [14], and in late 1930s, the first organochlorine compounds were introduced in the market to control ticks that became resistant to arsenic [15]. Synthetic pyrethroids were introduced in the 1970s [15] and they are currently adopted by most of the breeders. However, tick resistance, particularly *Rhipicephalus microplus* to this class of acaricide is common in many countries including Brazil [16, 17], Australia [18, 19], Argentina, New Caledonia, South Africa [20, 21], Benin, Cote d'Ivoire [4, 5], Burkina Faso and Mali [6]. Pyrethroids have their action site localized in the nervous system of arthropods [22]. Their molecular targets include the voltage-dependent sodium channel [23] and acetylcholinesterase (AChE). It is shown that occurrence of mutations in the genes encoding for these target proteins result in structural changes that intend to reduce their affinity with miticides. The detection of genes involved in ticks resistance and their mechanisms is essential to establish a sustainable strategy of control of the tick [24].

Material and methods
In this study, we made a summary of different research works carried out on tick resistance to

acaricide particularly *Rhipicephalus microplus* resistance and the control methods that are commonly used. In this review, we explained the concept of ticks' resistance to acaricide, the resistance mechanism particularly to pyrethroids which is the most used family of acaricide. Control methods were also investigated with a focus on methods that are environmental friendly and sustainable.

Concept of tick resistance to acaricide

Resistance is the occurrence, in a population, of individuals with the ability to tolerate doses of toxic substance that are lethal for the majority of individuals in a normal population of the same species [25]. The toxicity of acaricide is the result of their interaction with biological targets present in the tick. Several steps are needed before the acaricide exerts its action. It must first be in contact with the arthropod, enter his body, be converted in some cases into the active metabolite and then transported to the action site. Each of these steps is under the control of one or more genes. Any mechanism that changes one of these steps can therefore lead to a resistance [26, 27]. Ticks' résistance to acaricide may therefore result from several mechanisms, which can be for instance a change in absorption or excretion of the acaricide, a change in metabolic pathways that allow the acaricide degradation or finally a modification of its action site. The metabolic resistance is due to the increase of the enzymes activity in charge of miticides detoxification. These include sterases, oxidases cytochrome P450 and glutathione S-transferases [28]. The increase in activity of the enzyme may be due to a change in the conformation of the enzyme making it more efficient or due to it over production [29] that may sometimes be up to 12% of total protein of an individual [30]. The overproduction of enzyme may be due to a modification of a regulatory gene controlling the expression level of the enzyme [31, 32], an increase in the number of copies of the genes which code for these enzymes [33]; this is a complementary mechanism to the kdr mutation. As for the changes in the acaricide action site, they are due to point mutations occurrence in the genes coding for target proteins. This causes structural changes that reduce their affinity with miticides. These mechanisms can also cause cross-resistance to all miticides acting on the same site [34]. In most species, the mutation points are generally found on genes encoding for proteins, as that the amino acid change must not only reduce the affinity of the acaricide with the action site, but also preserve the initial functions of the protein to a level that is compatible with the survival of the tick. In most cases, it is likely that genes that confer resistance are already present at very low level in the tick population prior to the introduction of a new acaricide. The lack of standardized techniques for diagnosing the resistance or lack of resistance monitoring system appears to be the main challenge in countries where *Rhipicephalus microplus* is introduced. Actually, resistance suspicions occur when there is a treatment failure in the fight against cattle ticks. Although treatment failure is often the result of inappropriate preparation or application of acaricide, ticks persistence after frequent and correctly applied treatments indicate that resistance to the acaricide is permanent [35]. The intensive use of miticides led to the development of resistance to almost all major classes of miticides in many countries including Australia, Mexico, Brazil, New Caledonia, South Africa, USA etc. the failure of tick control is not always due to the resistance, but can be caused by factors such as faulty equipment, improper dose or use of outdated and therefore ineffective chemicals. Therefore, when the failure of the fight against ticks is observed, the suspected resistance should be confirmed before selecting a new acaricide. Once resistance is confirmed, breeders and livestock sector actors should be informed about the use of alternative chemicals for the control of resistant populations. Therefore, the sensitivity of resistant ticks potential replacement product must be tested [36]. In addition, since the cross-resistance is known to occur between compounds of the same class, but not always, more than one representative of each class to be tested. Overall control of the ticks should facilitate the detection of resistance at an early stage is essential to slow its spread early in avoiding resistant individuals selection. It is impossible to understand the mechanisms involved in the development of resistance of ticks without understanding the mode of action of acaricide.

***Rhipicephalus microplus* resistance to synthetic pyrethroids**

Synthetic pyrethroids have been introduced in the 1970s [15] and are currently widely used in our farms. Pyrethroids syntheses are highly effective insecticidal and acaricide, they are not very toxic to mammals and are highly biodegradable. Already in the late 1980s, resistance has been reported in Australia [37], Brazil [38] and in Mexico in 1994 [39]. Today pyrethroids resistance is extremely common and is prevalent in all countries where resistance studies were performed [16, 17]. The voltage-gated sodium channel is the target site for the activity of pyrethroids and has been studied in many species of arthropods. Three types of mutation in the sodium channel are associated with resistance to pyrethroids ticks [19, 40]. A study on *Rhipicephalus microplus* Mexico populations showed a substitution type of mutation in domain III, section 6, and position of nucleotide 2134. The replacement of thymine by adenine has transformed the amino acid, isoleucine to phenylamine [41, 42]. This led to the occurrence of a change of structure of acaricide receptor located on the sodium channel gene. As a result, ticks that are subjected to such changes become insensitive to pyrethroid. Thus, two other alternative types of mutations have been identified in the domain II in the segment 4, the sodium channel gene: The leucine is replaced by isoleucine, which results in the substitution of cytosine by adenine and still on the segment 4 we see the replacement of the amino acid glycine by valine due to the substitution at position 215 of guanine to thymine in the Australian population [43, 44].The mutation of domain II is also found in the population of Brazil [18], South Africa and Argentina [21]. This mutation gives tick acaricide resistance as permethrin, cypermethrin and flumethrin [43]. Domain III mutation of the sodium channel gene associated with resistance of *Rhipicephalus microplus* was noted on the stump of the US and Mexico [16, 45]. In Mexico, studies show that *Rhipicephalus microplus* is resistant to deltamethrin, cypermethrin and flumethrin [46]. The tick *Rhipicephalus microplus* resistance to Cypermethrin, flumethrin was also reported in Brazil [38]. The search for a new management approach is important for the control of ticks.

Tick resistance to other classes of miticides

Control of cattle ticks with chemical compounds began in the late nineteenth century with the use of arsenic. This class of compounds was followed by organochlorines, organophosphates, amidines, synthetic pyrethroids, phenylpyrazoles, macrocyclic lactones, growth regulators and spinosyns. The introduction of each new acaricide class was followed by the emergence of resistance [21]. Arsenical compounds were first used to control ticks, introduced in

1895 [14]. In 1896, a deep immersion based on arsenic has been used in Queensland, Australia, to control the tick by a local farmer. This deep immersion arsenic was quickly adopted in other areas such as USA, South Africa and Cuba and the disposal has become a widespread practice in Australia [47]. The immersion should be frequent due to the very short persistence [24] of arsenic [48]. After about 40 years of use, the first case of resistance to arsenic *Rhipicephalus microplus* was reported in 1936 [49]. Thus, the first organochlorine compounds were introduced in the market as acaricide in 1939 [15] to control ticks resistant to arsenic. More compounds became available in the mid-1940; organochlorines have a high efficiency, a long residual activity and a broad spectrum of action. Moreover, they had the advantage of being less toxic and less expensive than arsenic. The first case of resistance to organochlorine has been observed in Brazil in 1952 [50]. A decade later, in 1962, the use of all organochlorine was forbidden to fight against ticks, due to residues in meat, milk and the environment resulting from its poor biodegradability and affinity for lipids [15]. To contain the infestation of ticks, organophosphates began to be used as ectoparasiticides in the mid-1950. They were used to fight against ticks that have become resistant to organochlorines. Organophosphates were less stable and less persistent than organochlorines, organophosphates but some are very toxic to mammals [51]. Organophosphate resistance was first appeared in Australia in the mid-1960s [52]. In addition, organophosphate resistance was also observed on a Christmas tree Texas strain [53]. Thus amidines have been introduced for the control of ticks in the mid 1970 [54]. Today, amitraz is the main active ingredient in this class of acaricide. The resistance to amitraz appeared 4 to 10 years after its first use in different parts of the world and has been identified for the first time in the early 1980, Australia [54]. Since then, resistance has also been observed in Mexico, South America, South Africa and New Caledonia. However, in 2007, [40] reported that amitraz was still one of the most popular miticides to control ticks on cattle in Australia, Latin America and Southern Africa [21]. As for macrocyc1iques lactones, they were introduced to the market in 1981 [55] and are divided into two categories [56]: avermectin and milbemycin oxime. Macrocyclic lactones are systemically active against ticks and have a longer residual activity than synthetic pyrethroids and are active against a range of arthropods and nematodes [56]. However, the long wait for meat and milk can limit the use of this class in cattle. Resistance to avermectin (doramectin and ivermectin) was first reported in Brazil in 2001 [57] and was later also reported in Mexico ivermectin [58]. Nowadays, fipronil is the only compound of phenylpyrazoles use in livestock for the fight against cattle ticks and its use began in the mid-90 [59]. It has a long residual activity [56] and continues up to five weeks in the field [60]. Resistance to fipronil was reported for the first time in 2007 with Uruguay [61], and later in Brazil [62]. Growth regulators are one of the last classes of miticides, with the first compound representing the class, fluazuron available in the market acaricide since 1994 in Australia. The resistance has not been reported in the literature, but a case was presented in a poster session of Congress in 2010 [63]. A new strategy must be implemented to fight against ticks.

Control strategies

The fight against ticks is not only to limit the losses they cause, but also those due to diseases transmitted by ticks or associated with ticks. This struggle must be conducted in a concerted and rational manner based on the various genera and species present in the area, but especially in the light of understanding their respective biology. Thus, several control methods are used worldwide (Table 1).

Table 1: Control method against ticks

Methods	Processes	References
Fight With Trap	Trap and ticks removal from the host animal	host animals Products invasion against ticks [66 ; 81]
Traditional Method	Manual removal	[70 ; 71 ; 69 ; 72 ; 73 ; 74 ; 75 ; 76]
	Application of traditional products containing harmful substances to ticks	
Chemical Method	Use of synthetic miticides against ticks	[35 ; 2 ; 18 ; 61 ; 82]
Ecological Control	rotational grazing to control the infestation	[63 ; 66; 64 ; 65]
Biological Control	predators to fight against ticks	[67 ; 83, 84]
Genetic Control	Selection of animals resistant to ticks	[77 ; 79 ; 80 ; 85 ; 86 ; 87]

Several chemical methods of ticks control were developed, including the use of miticides such as organochlorines, organophosphates, macrocyclic lactones amidines and synthetic pyrethroids. At this level, it is equally important to note that misuse of these miticides leads to the development of resistance of ticks to acaricide. Chemical control is now widely used in our farms but it is very costly to farmers, who do not always have the means to face it. It is extremely difficult to control ticks and the diseases they transmit. Today controlling the infestation of cattle is based mainly on synthetic miticides. The indiscriminate use of these substances also poses pollution problems etc. There is therefore great interest in the development of alternative methods of control. Some are described briefly here.

- ***Ecological and biological control***

The ecological approach is to modify the habitat of the tick so as to cripple its reproductive cycle [64], based on a driving rotational grazing. Indeed, it amounts to prevent contact between ticks and host and thus to an interruption of the cycle. According to Barre [65] cited by [66], this process is conceivable that in the case of modern farming (intensification) because it is necessary to have large clean pasture land can be withdrawn for several weeks or months of animal feed [67]. Many parasites, parasitoids or predators may have a negative impact on the ticks present in the medium. But note that there is little use of these organisms test (bacteria, fungi, nematodes and insects) to reduce tick populations [68]. It was recently found that fungal spores (*Beauveria bassiana* and *Metarhizium anisopliae*) had some activity against *Rhipicephalus appendiculatus* and the tropical bond tick infesting rabbits and cattle on which they were sprayed [68] regulating ixodidienne and the population to which they are associated. This is the case of the ant *Solenopsis geminata* [67] predator of the tick *Rhipicephalus microplus.*

- ***Traditional methods***

The manual diptank is the most commonly used method by farmers in West Africa. It is a method that is easily applicable to a small population of animals. However, it turns out binding for high parasite load, which is the case of *Rhipicephalus microplus*. It is time consuming and labor intensive, especially when the infestation is observed in the beginning of rainy season (farm work), or end of the rainy season [66]. Breeders from the perspective of destroying ticks on animals, use a variety of plants containing active ingredients harmful to ticks such as [69]: *Actaea spicata*, a plant used externally in the form of lotions or mixed with fat pork; it heals and kills scabies mites. Studies on the mixture of essential oils of *Syzygium aromaticum* and *Cymbopogon citratus* gave a very satisfactory result on females engorged *Rhipicephalus microplus* [70]; the essential oil of *Azadirachta indica* share its bitter properties and its characteristic odor, has a potential acaricide [71]; *Ricinus communis* extract had acaricide effect on ticks [72], it helps to fight against many mites and is harmless to higher animals. Studies on *Tephrosia vogelii* in Benin [73], Zimbabwe [74] and Congo [75] revealed that the leaves, seeds and other parts of the plant contain active ingredients with acaricide properties. The leaves of *Mitracarpus villosus* and *Thevetia neriifolia* have larvicidal activity against *Rhipicephalus microplus* [76], the extract of *Annona squamosa* an acaricide property on the tick [77]. View the number of plants with activity against ticks, the use of plant extracts for control seems to be a promising alternative.

- ***Genetic resistance***

The external parasitism by ticks is a major constraint to the cattle. The use of genetically resistant animals is a complementary method to fight these ectoparasites. This alternative measure is introduced in Australia [78] to combat ticks. Studies are underway to highlight the breeds resistant to ticks. For example, resistance to *Rhipicephalus microplus* varies according to the breed but is usually more pronounced for example in Zebu cattle (*Bos indicus*) than in cattle (*Bos taurus*) [79]. In Australia, and Sahiwal breeds Braham (*Bos indicus*), respectively selected for meat and milk showed a natural resistance to *Rhipicephalus microplus* higher than *Bos taurus* breeds [80]. The selection of resistant animal breed tick occurs by crossing between animals in order to obtain a product having a natural immunity to these ectoparasites [81].

Conclusion

Ticks, for their direct impact and the diseases they transmit are a major constraint to the development of cattle breeding in infested cattle herds. The use of synthetic acaricide for control leads to a buildup of resistance. Control of *Rhipicephalus microplus* by the selection of resistant cattle or by the use of plant extracts appears to be a promising alternative to reduce the invasion of ticks.

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