



Free radical scavenging and antibacterial potential of two plants extracts (*Khaya senegalensis* and *Pseudocedrela kotschy*) used in veterinary pharmacopoeia in Benin

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ABSTRACT

In all developing countries like Benin, medicinal plants were the most widely used especially in rural areas to solve health problems in humans and in animal's life. But despite this use, all the mysteries of nature were not yet perceived by human. This paper reports the results of the chemical and biological studies of two plants, *Khaya senegalensis* and *Pseudocedrela kotschy* used by farmers in Benin in the treatment of gastro-intestinal diseases in livestock. Throughout the results, we noted in both samples the presence of several secondary metabolites such as saponins, catechic tannins, polyphenols, mucilages, flavonoids, anthocyanins, leuco anthocyanin, reducing compounds, sterols and terpenes. Concerning the extraction yield of metabolites, the binary water-ethanol (50/50) showed the best results compared to each of the two solvents used separately. Concerning the polyphenols content, it varied in the same plant depending of the extraction solvent nature. The test results of the scavenging activity showed a better one with the ethanolic extract of the stem bark of *P. kotschy* whereas the stem bark of *Khaya senegalensis* extracted with semi-ethanol was the most active. Both extracts displayed similar activities (CI50= 4 µg/ml) more pronounced than that of the reference compound used in this study, the BHA (CI50=4.8 µg/mL). The results of antibacterial activity of the ethanolic and hydroethanolic extracts of trunk bark of *P. kotschy* had bactericidal activity against *Staphylococcus aureus*, *Salmonella typhi*, *Escherichia coli* and *Klebsiella pneumoniae*. Concerning *K. senegalensis*, the ethanolic and hydroethanolic extracts showed an interesting antibacterial activity against *Salmonella typhi*, *Escherichia coli* and *Klebsiella pneumoniae*.

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Introduction

Herbal medicines were the most widely used especially in rural areas to solve problems of human and animal health in Saharan regions.

In Benin, livestock is one of the first activities undertaken by the man after agriculture to ensure food security and survival. Gastrointestinal infections are the major cause of mortality and economic losses in this sector. Very attached to their cattle, farmers can't stay indifferent to this kind of health problem and engages in the use of active principles from plants to fight against livestock diseases [1], [2]. This farmers' behavior was often linked to the affordable cost of herbal medicine and the resistance of some pests to synthetic products [3, 4].

Nowadays, the study of chemical composition of plant extracts became paramount. Indeed, the vegetable biomass was an inexhaustible source of molecules for therapeutic purposes. The continual development of phytochemical screening and biological investigations hardly contributed to the identification and isolation of bioactive compounds extracted from plants.

Deepening the plants knowledge became a priority for researchers to optimize their use in human and veterinary medicine.

Khaya senegalensis, family of Meliaceae (Syn: *Swietenia senegalensis*) was a tree averaging 35 meters in height, short and stocky. Its leaves are hairless, mostly pinnate, clustered towards the ends of branches with 3-7 pairs of opposite leaflets. The bark was dark gray and scaly [5], [6]. By against, *Pseudocedrela kotschy* (syn: *Pseudocedrela chevalieri*, *Cedrela kotschy*) is a tree that can reach 20 meters in height, cylindrical crown to ovoid, elongated, poorly developed. Its bark was gray and cracked [7, 8]. From the same botanical family these two plants are part of the therapeutic arsenal used by farmers to treat certain diseases of animals, such as gastrointestinal and diarrheal diseases, internal parasitism... etc [9, 10].

In the literature, very few works were devoted to the chemical and biological studies of *K. senegalensis* and *P. kotschy*. In Benin, these plants commonly used by traditional healers and farmers for their healing properties in the treatment

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of several pathologies had hardly been the subjects of scientific investigation. It was therefore appropriate to deepen the knowledge of the chemical and biological properties of the extracts of these plants.

The objectives of this work were to identify the metabolites present in these plants and to assess their polyphenolic compounds (total polyphenols, total flavonoids and condensed tannins) content. Antibacterial and antiradical activities of ethanolic, hydroethanolic and aqueous extracts of these two plants were also evaluated.

Materials and methods

Plant material

The plant material used in this study was constituted of the trunk bark of two plants belonging to family of Meliaceae, *Khaya senegalensis* and *Pseudocedrela kotschy* collected in January 2014 respectively from locality of Abomey-Calavi (Latitude: 6°27'0''N; Longitude: 2°21'0'') and Dassa (Latitude: 7°46'30.87''N; Longitude: 2°12'31.23''E) in Benin.

Biological material

Biological material was constituted of the reference strains of *Staphylococcus aureus* (ATCC27844), *Escherichia coli* (0157H7), *Salmonella typhi* (R0951401) and *Klebsiella pneumoniae* (ATCC35657). These strains had been provided by the National Health Laboratory of Ministry of Health in Benin Republic.

Methods

After plant material collection, the samples were dried in shade and then grinded to fine powder and stored for extract preparation.

Identification of secondary metabolites

Anthraquinones: To chloroform extract of each of these plants was added aqueous KOH 10%. After stirring, the presence of anthraquinones was confirmed by a shift of the aqueous phase to red [11].

Coumarins: 1g of plant powder was placed in a tube in the presence of few drops of water. The tubes were covered with the filter paper soaked in diluted 10% NaOH and were boiled. Any yellow fluorescence reflects the presence of coumarins after examination under UV at 365 nm [11].

Alkaloids: alkaloids were identified by Meyer test and confirmed by the Bouchardat test [12].

Sterols and terpenes: Terpenes and sterols have been demonstrated by the Liebermann - Burchard test [13].

Flavonoids: Flavonoids identification was made by the test of cyanidin [14].

Tannins: They were highlighted by the Stiasny test. This test consists to mix 5 ml of infused (5%) and 3 ml of Stiasny reagent. The resulting mixture was heated for 10 to 15 min. The appearance of a precipitate indicates the presence of catechic tannins. The filtrate was saturated with sodium acetate, the development of a blue blackish hue after adding 1% of FeCl₃ indicates the presence of gallic tannins [15].

Saponins: They were determined by the foam test; degree of dilution of aqueous discounted giving a persistent foam after shaking [11], [16].

Proteins: Proteins were identified by the biuret test which consists introducing into a test tube, a quantity of ethanolic extract was taken up in 2 ml of aqueous 20% NaOH which must add two drops of an aqueous solution at 2 % of CuSO₄. The appearance of a reddish or violet color indicates a positive reaction.

Polyphenols: The determination of compounds belonging to the group of polyphenols was made by reacting ferric chloride [16].

Essential oils: Volatile compounds were identified by the hydrodistillation method using a Clevenger -type extractor for two hours on average [17, 18].

Reducing compounds: Fehling's test solution involves boil 2 ml of aqueous extract and a few drops of Fehling's solution for 15 minutes. The appearance of a brick-red precipitate indicates the presence of reducing compounds [19].

Anthocyanins: To an infusion, add 5 ml of 10% H₂SO₄ and then 5ml of 50 % NH₄OH. The appearance of a red color that turned purplish blue in basic medium indicates the presence of anthocyanins [20].

Leuco-anthocyanins: 0.5 ml of 12 N HCl was poured into 3 ml of hydro- alcoholic extract. The acidified solution was brought to boiling water bath for 30 minutes. After cooling, the appearance of a purplish red color indicated the presence of leuco-anthocyanins [16] (Bruneton, 1993).

Mucilages: Introduce 1 ml of decoction 10% in a test tube and add 5 ml of ethyl ether. After ten minutes, obtaining a flocculent precipitate indicated the presence of mucilages [20] (Traoré, 2010).

Preparation of extracts

The technique used was that of maceration. 50g of each powder sample was introduced into a 500 ml flask containing 250 ml of extraction solvent (ethanol, water or ethanol - water 50/50). The vial was capped and allowed to stir continuously for 72 hours. After filtration, the extracts were evaporated to dryness at 60°C using a rotary evaporator Heidolph kind. The yield of extraction was calculated by the formula below.

$$R(\%) = \frac{\text{Mass of extracted}}{\text{Mass of plant material used}} \times 100$$

Determination of polyphenols compounds

Total polyphenols: The method of determination of total polyphenols according to Folin- Ciocalteu [21, 22] consisted to use a mixture of phosphotungstic and phosphomolybdic acid which was reduced during the oxidation of phenols in the mixture of tungsten blue oxide and molybden [23] (Ribéreau - Gayon, 1968). The absorbance was measured by a spectrophotometer (JENWAY 50/60 Hz) to 765 nm. Gallic acid was used as reference and the total polyphenol content in the extract was expressed in mg of gallic acid equivalents per gram of dry matter.

Total flavonoids: The method of aluminum trichloride (AlCl₃) was used to quantify the total flavonoids. This technique was based on the formation of the aluminum-flavonoids complex that had a maximum absorption at 500 nm [24, 25].

Condensed tannins: condensed tannins dosing was achieved by the method with vanillin sulfuric [26, 27]. The principle of this assay was based on the binding of vanillin aldehyde group on the carbon in position 6 of the ring of the catechol to form a red colored complex chromophore which absorbs at 510 nm.

Antiradical activity: The radical scavenging activity was evaluated by the DPPH method. The principle of this method was based on measuring the trapping of a free solution of DPPH radicals. This trapping was indicated by the disappearance of the purple color of DPPH. The tanks were left in the darkness for one hour and the absorbance measured at 517 nm [28, 29] (Brand- Willams et al., 1995; Agbangnan et al., 2013). The trapping percentage was determined by the formula:

$$P = \frac{(Ab - Ae)}{Ab} \times 100$$

P: percentage of trapping; Ab: absorbance of the white; Ae: Absorbance of the sample

Antibacterial activity

Preparation of microbial suspension

Preparation of the microbial suspension was obtained by emulsifying a pure colony of each strain in 5 ml of Mueller-Hinton broth according to the method cited by Yehouenou *et al* [30].

Principle

The Minimum Inhibitory Concentrations (MIC) and Bactericidal one (MBC) were determined using the micro dilution technique with Muller Hinton Broth [30-32].

An initial solution of extracts was prepared at the concentration of 400 mg.ml⁻¹ in the mixture of ethanol/water (40/60). 100µl of the medium Mueller- Hinton Broth were set down in every well of the microplaque. 100µl of the initial extract solution were set down in the first well. After homogenization in the well by aspiration-repression using a micropipette one got 200µl of an extract solution to 200mg/ml. 100µl of this new solution were appropriated and mixed in the MHB Muller Hinton Broth solution contained in the 2nd well and one pursued this set of dilution of reason two well by well until the 9th well, the remaining aliquot was thrown away. Finally 100µl of a soup of 18h of microbial culture (1.5106 CFU.ml-1 at scales 2 of McFarland) were added in every well. The 10th and 12th wells corresponded respectively to the positive witness and to the negative one and contained 100µl of MHB + 100µl of microbial culture medium for the positive witness and 150µl of MHB + 50µl of extract solution - mother to test for negative witness. The microplaque was covered with the aluminum paper and was placed during 18 hours in the incubator at 37°C. The MIC was estimated visually compared to the witnesses and every well was spread on the MHA gelose and placed to 37°C during 24 hours. The MBC corresponded to the smallest concentration of extract for which any microbial colonies development was not observed.

Results and discussion

The metabolites identified in the trunk bark of *K. senegalensis* and *P. kotschy* were shown in Table 1

Various secondary metabolites have been identified in the bark of the trunk of *K. senegalensis* and *P. kotschy* by a series of color reactions and precipitation more or less specific to each class of active ingredients. Among these secondary metabolites must include saponins, catechin tannins, polyphenols, mucilages, flavonoids, anthocyanins, leuco anthocyanins, reducing compounds, sterols and terpenes. The alkaloids were identified only at the trunk bark of *K. senegalensis* while quinones and coumarins were present only in the bark of *P. kotschy*. Within these two plants belonged to the same botanical family, there was a divergence about the occurrence of secondary metabolites. Mesbal *et al.* (1984) [33], identified in the bark of *K. senegalensis* harvested in Egypt coumarins which were absent in the sample from Benin. In the same plant harvested in Nigeria, Ishaku *et al* [34] identified tannins, polyphenols, flavonoids and cardiac glycosides, but they noted the absence of alkaloids and saponins which have been highlighted in the samples of Benin. To our knowledge, no investigation was conducted for the determination of secondary metabolites present in the trunk bark of *P. kotschy*. Akuodor *et al* [35] identified alkaloids, flavonoids, tannins, saponins, glycosides, carbohydrates, reducing compounds, terpenes and sterols in the leaves of *P. kotschy* harvested in Nigeria while Traoré [8] identified coumarins, catechic tannins, anthraquinones, saponins, mucilage, oses, leuco - anthocyanins, sterols and terpenes in the root bark of the same plant harvested in Mali. The variation of secondary metabolites could be related

to the time of the harvest, soil type, climatic factors or developmental stage of the plant [36, 37]. The richness of these two plants in secondary metabolites could explain their traditional use. The presence, for example of tannins and flavonoids in the bark of *K. senegalensis* and *P. kotschy* justifies the use of these plants in the treatment of diarrheal and gastrointestinal diseases in animals [38-40].

Yields of extraction

The yield of ethanolic, hydroethanolic and aqueous extracts were shown in Table 2

Yields ethanolic, hydroethanolic and aqueous extracts of bark trunk of *K. senegalensis* are respectively 14.8 % , 27.2 % and 16.0 % on the one hand , while those were for *P. kotschy* 13.2% , 23.6 % and 17.6 % in the one other. The hydroethanol solvent has to get better performance in these two plants from the same botanical family. These results show that the ethanol / water mixture (50/50) facilitates the extraction of plant metabolites.

Ishaku *et al* [34] obtained in yields respectively 4.15% and 5.19% of the yields of the aqueous and ethanolic bark trunk extracts of the same plant (*K. senegalensis*) harvested in Nigeria. These returns are almost three times smaller than those of ethanolic and aqueous extracts of Benin.

The time, place and the harvested plant age could influence the extraction yield [41]

Polyphenolic Compounds

Total polyphenols

The histogram in Figure 1 showed the total polyphenols contents expressed in mg of gallic acid equivalents per g of dry matter of the ethanolic, hydroethanolic and aqueous extracts of trunk bark of *P. kotschy* and of *K. senegalensis*.

These levels were respectively (3.272 ± 0.246); (3.328 ± 0.185) and (6.731± 0.018) mg EAG / g DM for ethanolic, semi-ethanolic and aqueous extracts of *K. senegalensis* and (6.849±0.326); (7.454±0.053) and (6.232±1.181) mg AG / g DM for those of *P. kotschy*. The total polyphenol content of the trunk bark of *P. Kotschy* extracts is substantially insensitive to the nature of the extraction solvent; by cons at *K. senegalensis*, the aqueous extract showed the highest polyphenol content. The total polyphenol content is high at the trunk bark of *P. Kotschy* and *K. senegalensis*. There is a change in the total polyphenol content in plants of the same botanical family. At the trunk bark of *K. senegalensis* collected in Ouagadougou, Karou [42] obtained a very low total polyphenol content of ethanolic extract (0.0472±0.0001) EAG mg / g D M compared to our results. Variability in levels of plant secondary metabolites from one region to another may depend on several factors. Among these, are the climatic and soil conditions (temperature, sun exposure, drought and salinity), storage conditions and the maturity of the plant [43, 44]

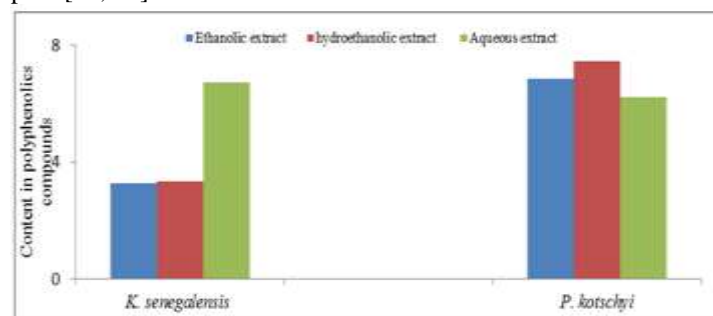


Figure 1: Total polyphenols Content in ethanolic, hydroethanolic and aqueous extracts of the trunk bark of *K. senegalensis* and *P. kotschy*

Secondary metabolites	<i>K. Senegalensis</i>	<i>P. kotschy</i>
Alkaloids	+	-
Polyphenols	+	+
Flavonoids	+	+
Anthocyanins	+	+
Leuco-anthocyanins	+	+
Anthraquinones	-	-
Free anthraquinones	-	-
Combined anthraquinones	p-heterosides	-
	p-heteroside with reduced genine	-
	p-heterosides	-
Reducing Compounds	+	+
Tannins	Gallic	-
	Catechic	+
Sterols and terpenes	+	+
Mucilages	+	+
Saponosides	+	+
Coumarines	-	+
Quinones	-	+
Proteins	+	+
Essential oil	-	-

Table 2: Extraction yields

Extracts	Yields(%)	
	<i>K. senegalensis</i>	<i>P. kotschy</i>
Ethanolic	14.8	13.2
Hydroethanolic	27.2	23.6
Aqueous	16	17.6

Table 3 : Minimum Inhibitory Concentrations and Minimum Bactericidal Concentration of extracts

Microorganism strains	Extracts	Concentrations (mg/ml)			
		<i>P. Potschy</i>		<i>K. senegalensis</i>	
		MIC	MBC	MIC	MBC
<i>S. typhi</i>	Ethanolic	3.125	6.25	1.56	3.125
	Hydroethanolic	1.56	3.125	0.39	0.78
	Aqueous	1.56	3.125	>100	>100
<i>S. aureus</i>	Ethanolic	1.56	3.125	0.78	>100
	Hdroethanolic	0.39	0.78	1.56	>100
	Aquous	6.25	12.5	3.125	6.25
<i>E. coli</i>	Ethanolic	0.39	0.78	1.56	3.125
	Hydroethanolic	0.78	1.56	3.125	6.25
	Aqueous	6.25	>100	0.39	0.78
<i>K. pneumoniae</i>	Ethanolic	12.5	>100	0.39	0.78
	Hydroethanolic	1.56	3.125	50	100
	Aqueous	12.5	25	25	>100

Total flavonoids

The figure 2 showed the levels of total flavonoids expressed as mg catechic equivalents per g of dry matter of the ethanolic, hydroethanolic and aqueous extracts of trunk bark of *K. senegalensis* and *P. kotschy*.

The total flavonoids content of the ethanol extract of the bark of the trunk of *K. senegalensis* are (54.733 ±1.462) mgCE/gDM while the hydroethanolic extract of (63.198±0.639) mg CE /gDM (90.468±16.907) mgCE/gDM for the aqueous extract. As for the stem bark of *P. kotschy* these levels are respectively (50.598 ± 0.685) mg CE / g DM (17.189 ± 0.091) mg EC / MS and g (42.407 ± 2.399) mg CE/gDM of the ethanolic, hydroethanolic and aqueous extracts. The nature of the extraction solvent influences the total flavonoids content of the trunk bark of *P. kotschy* and of *K. senegalensis*

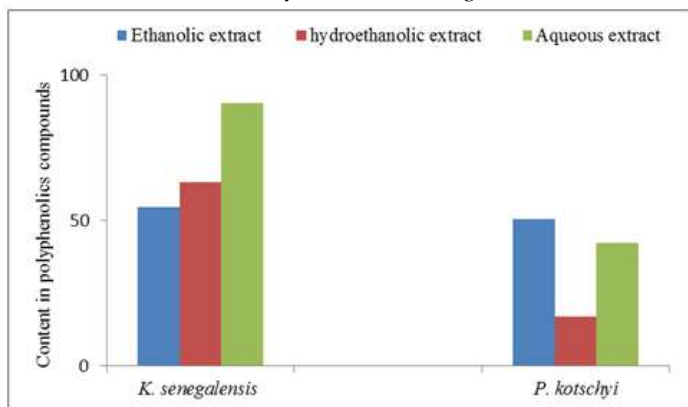


Figure 2. Total flavonoids content of ethanolic, hydroethanolic and aqueous extracts of trunk bark of *K. senegalensis* and *P. kotschy*

Condensed tannins

Figure 3 showed the levels of condensed tannins expressed as mg catechin equivalents per gram of dry matter ethanolic, hydroethanolic and aqueous extracts of bark trunk of *P. kotschy* and of *K. senegalensis*. These levels ranged from (73.581±0.658) mgCE/gDM (250.791±12.103) mg CE/g DM.

The aqueous extract of *P. kotschy* showed the lowest level followed by the ethanol extract of the same plant (130.977±2.105) mg CE /g D M. The highest content of tannins was observed for semi-ethanolic extract of *K. senegalensis* preceded by the hydroethanolic extract of *P. kotschy*. The content of condensed tannins is higher in the three extracts of *K. senegalensis* than the trunk bark of *P. kotschy*. The ethanol-water 50% facilitate the extraction of condensed tannins from the two samples.

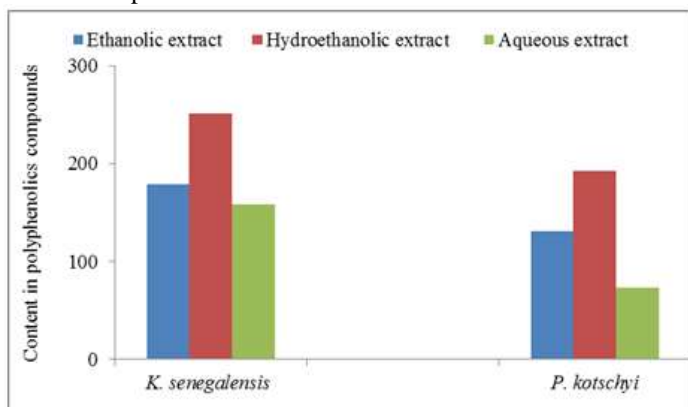


Figure 3. Content of condensed tannins of ethanolic, hydroethanolic and aqueous extracts of the trunk bark of *K. senegalensis* and *P. kotschy*

Antiradical activity of reference compounds and extracts from the trunk bark of the two plants (*K. senegalensis* and *P. kotschy*) Gallic acid, quercetin and butylated hydroxyanisol (BHA)

Figure 4 reflected the percentage of DPPH radical scavenging versus concentrations of gallic acid, BHA and quercetin.

These curves were used to determine the concentration of each standard compound that could trap 50% of DPPH free radicals. These concentrations were 0.9 µg / ml; 3µg/ml and 4.8 µg / ml for gallic acid, quercetin and BHA. Note that gallic acid had a scavenging activity more pronounced than quercetin and BHA. Note that gallic acid had a scavenging activity more pronounced than quercetin and BHA. These results were consistent with those obtained by Agbangnan *et al* [29] from the three molecules. This difference in activity observed at all three molecules would be bound to their reference structure.

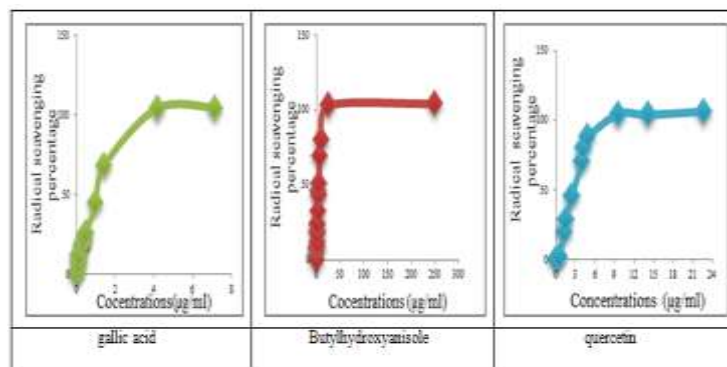


Figure 4: Antiradical activity of three reference compounds Extracts from the trunk bark of *K. senegalensis* and *P. kotschy*

Figures 5 and 6 reflected the percentage of free radical scavenging versus concentrations of ethanolic extracts, hydroethanolic and aqueous bark truncated *K. senegalensis* and *P. kotschy*.

An increase followed by a constant substantially equal to 100% was observed for the rate of free radical scavenging DPPH versus concentrations tested extracts. These curves allowed determination concentrations of the extracts to scavenge 50% of DPPH free radicals. These concentrations (CI₅₀) are respectively equal to 20µg/ml, 5µg/ml and 4µg/ml for ethanolic, hydroethanolic and aqueous bark truncated extracts of *K. senegalensis* compared to respectively to 4µg/ml, 6 mg / ml and 9µg/ml for extracts of the trunk bark of *P. kotschy*. The hydroethanolic of *K. senegalensis* and ethanolic of *P. kotschy* extracts showed better activity than that of BHA (CI₅₀=4.8 µg/ml) which is a synthesis of compounds antiradical. Both extracts are less active than gallic acid (CI₅₀=0.9µg/ml) and quercetin(CI₅₀=3µg/ml).

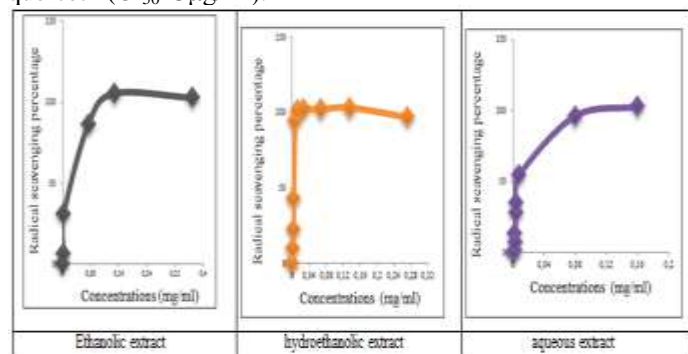


Figure 5: Antiradical activity of ethanolic extract of the stem bark of *K. senegalensis*

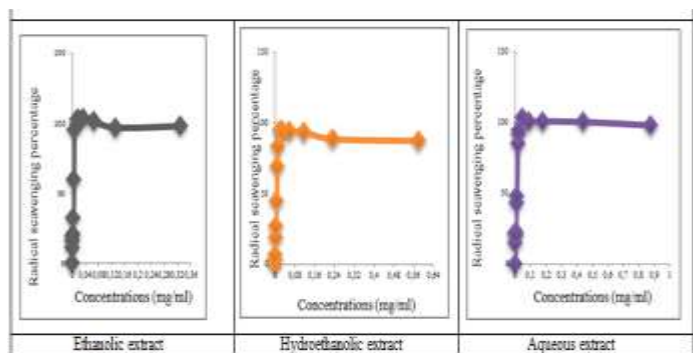


Figure 6 : Antiradical activity of extracts of the stem bark of *P. kotschy*

Antibacterial activity

The Minimum Inhibitory Concentrations (MIC) and Minimum Bactericidal Concentration of (MBC) extracts trunk bark of the of *P. kotschy* and *K. senegalensis* are listed in Table 3. Ethanolic, hydroethanolic and aqueous extracts of the stem bark of *P. kotschy* had bactericidal activity on *S. typhi* and *S. aureus*. The hydroethanolic extract showed the highest activity with CMB 0.78 mg / ml on *S. aureus* and 3.125 mg / ml on *S. typhi* followed by ethanolic extract with concentrations of 3.125 mg / ml for both strains. The aqueous extract, the less active on the two strains showed bactericidal activity against *S. Typhi* with a concentration of 3.125 mg / ml and 12.5 mg / ml against *S. aureus*. *E. coli* had bactericidal sensitivity with ethanolic and hydroethanolic extracts of the same plant, so that only the aqueous extract inhibited this strain at a concentration of 6.25 mg /ml. For *K. pneumoniae*, it shows insensitivity to the ethanolic extract of trunk bark of *P. kotschy* but sensitive to hydroethanolic and aqueous extracts respectively with MBC of 3.12 mg / ml and 25 mg / ml. For *K. senegalensis* extracts, ethanol extract inhibited *Salmonella typhi*, *S. aureus*, *E. coli* and *K. pneumoniae* respectively at concentrations equal to 1.56 mg / ml, 0.78 mg / ml, 1.56 mg / ml and 0.39 mg / ml. This extract is bactericidal with 3.125 mg / ml against *S. typhi* and *E. coli* (0.78 mg / ml) against of *K. pneumoniae*. Concerning the hydro ethanolic extract of *K. senegalensis*, it showed bactericidal activity against strains of *S. typhi*, *E. coli* and *K. pneumoniae* respectively at concentrations of 0.78 mg / ml, 6.25 mg / ml and 100 mg / ml, but inhibited the strain of *S. aureus*. Note that the aqueous extract of the trunk bark of *K. senegalensis* had no effect on *S. typhi* and *K. pneumoniae* but this extract displayed bactericidal activity with *S. aureus*. The antibacterial activity noticed on *S. aureus*, *S. typhi*, *E. coli* and *K. pneumoniae* with extracts from the bark of the trunk of *P. kotschy* and *K. senegalensis* could be explained by the presence of several major metabolites including flavonoids, tannins, terpenes, saponins and polyphenols [45], [46] either alone or in synergy between these compounds and other minority one identified. The insensitivity of *K. pneumoniae* face the ethanol extract of *P. kotschy* and aqueous extract of *K. senegalensis* could be explained by the selective action of extraction solvents face bioactive plant compounds studied.

Conclusion

Better understanding of the secondary metabolites of this study were in concordance to the biological activity of *P. kotschy* and *K. senegalensis* extracts used by farmers for the treatment of diarrheal diseases and gastrointestinal livestock in Benin. These two plants are rich in secondary metabolites such as polyphenolic compounds, mucilages, saponins, sterols and terpenes. Among the extraction solvents used, the binary ethanol-water (50 /50) has allowed a better extraction yield for these two plants. Assessment of phenolic content and biological

activities revealed that the hydroethanolic extract richest in phenolic compounds showed the highest scavenging activity from *K. senegalensis* extracts and the highest antibacterial activity from *P. kotschy* extracts on the four strains used in this study. Diversity in secondary metabolites and biological activities noted in extracts of these two plants could justify their use by farmers to treat diseases of livestock. It was therefore appropriate to guide future studies towards the isolation and characterization of bioactive molecules present in the active extracts investigated. It is therefore appropriate to guide future studies towards the isolation and characterization of bioactive molecules present in the active extracts investigated

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