Available online at www.ijpsdronline.com

International Journal of Pharmaceutical Sciences and Drug Research 2018; 10(2): 57-64



Research Article

ISSN: 0975-248X CODEN (USA): IJPSPP

(CC) BY-NC-SA

Evaluation of the Antioxidant Activity and the Cytotoxicity of Extracts of Capparis spinosa

F-Z Ennacerie¹, F Rhazi Filali^{1*}, N Moukrad¹, M Bouidra², A Bentayeb²

¹Department of Biology, Team Microbiology and Health, Laboratory Chemistry Biology Applied to the Environment, Faculty of Science, University Moulay Ismail, PB 11201, Zitoune, Meknes, Morocco ²Department of Chemistry, Team Physical chemistry condensed matter, Faculty of Science, University Moulay Ismail, PB 11201, Zitoune, Meknes, Morocco

Copyright © 2018 F Rhazi Filali *et al.* This is an open access article distributed under the terms of the Creative Commons Attribution-NonCommercial-ShareAlike 4.0 International License which allows others to remix, tweak, and build upon the work non-commercially, as long as the author is credited and the new creations are licensed under the identical terms.

ABSTRACT

The purpose of this study is to promote the plant spontaneous, harvested from the West of Morocco and streamline its use in quantifying the total phenolic content, assessing the antioxidant activity and testing the degree of toxicity on two types of eukaryotic cells plants and animals. The content, extracts aqueous and ethanolic flower buds and fruit, in phenolic compounds was determined according to the method Follin-Ciocalteu. The antioxidant activity was evaluated by two methods FRAP and DPPH. As for the cytotoxicity extracts, it has been evaluated by the test hemolysis and inhibition germination test of *Lepidium sativum* seeds. The different extracts of the two organs of this plant, have revealed richness in total polyphenols, especially those of the flower buds, as well as, an antioxidant activity, which is in the same order as that of vitamin C for the aqueous extract of flower buds. The effect hemolytic is shown positive for decoctat of flower buds. The ethanolic extract of fruits displayed an activity antimitotic expressed by the inhibition of elongation and growth seedlings of *Lepedium sativum*. The decoctat of flower exhibited an effect antigerminatif of moderate intensity which is reversible after rehydration of seed. *Capparis Spinosa* is a plant of quality pharmaceutical interesting for its activities antioxidant, antimitotic, healing, and for its wealth in phenolics compounds.

Keywords: Capparis spinosa, antioxidant activity, antimitotic, hemolysis, germination, Lipidium sativum seeds.

DOI: 10.25004/IJPSDR.2018.100202

Int. J. Pharm. Sci. Drug Res. 2018; 10(2): 57-64

*Corresponding author: Dr. F Rhazi Filali

Address: Department of Biology, Team Microbiology and Health, Laboratory Chemistry Biology Applied to the Environment, Faculty of Science, University Moulay Ismail, PB 11201, Zitoune, Meknes, Morocco

Tel.: +2120664488238

E-mail ⊠: fouzia.filali@yahoo.fr

Relevant conflicts of interest/financial disclosures: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Received: 08 January, 2018; Revised: 07 March, 2018; Accepted: 16 March, 2018; Published: 25 March, 2018

INTRODUCTION

With the aim of the valorization of the aromatic and medicinal plants spontaneous Mediterranean, as well as the rationalization of their use; our choice is focused on this species *Capparis spinosa* and precisely the flower buds (Capers) and fruit. *Capparis Spinosa* is a species which belongs to the family capparidaceae. It is shrub cultivated in the Mediterranean region and dry areas of Western Asia and Central. [1] At the National level, *Capparis spinosa* is spontaneous in several regions of Morocco such as Sidi Kacem, Fez, Taounate, Meknes, Marrakech and Safi. [2-4]

According to the National nutrient database (USDA), fruit of Capparis spinosa contain a variety of compounds with appreciable nutritional value, whose percentage is 5% for carbohydrates, 3% for dietary fiber, 2% for proteins and 0.9% for lipids, in addition to the vitamin C with a mean concentration of 4 mg/100 g weight. [5-7] As for the seeds of fruit, they contain sterols, carotenoids and tocopherols. [8] The pulp, of this plant is rich in phenolic compounds such as rutin, quercetin, vitamin C, tocopherols and carotenoids. [9] Concerning capers, they are rich in glucocapperin and mainly glucosinolate [10-11], flavonoids and the acids hydroxycinnamiques. [12]

View these values nutritious, flower buds "Capers" of this plant are consumed and sold in the Moroccan markets and exported to international countries. In addition they are used in agri-food as additive and stimulating sensory, gustatory, or natural preservative. [7] Moreover, fruit and capers of Capparis spinosa are very used in traditional medicine Moroccan. Besides in an ethnobotanic study preset, which we have made on the population of the province of Sidi Kacem located in the West of Morocco, it revealed many uses in traditional medicine to remedy infections skin, respiratory diseases and digestive illnesses. [6] Capparis spinosa is used also in pharmaceutical industry and modern medicine. [1-7] This plant has also attracted the attention of several researchers, for its many therapeutic virtues such as antimicrobial effect [13], antiinflammatory [4] and antioxidant activity [14] antiallergic, anti-histamine, [15-4] and antiviral roles. [11] These criteria allow Capparis spinosa to rank among the plants medicinal and aromatic the most appreciated. In order to contribute to the valorization of this plant, and to show the interest of its biological activities, and compare its qualities with those of other countries, fruits and flower buds of Caparis spinosa, harvested from the West of Morocco, were targeted in this study to illustrate its benefits and harms therapeutic and nutritional. In this study the total phenolic content was determined seen their interest biological important. The antioxidant activity of extracts ethanolic and aqueous was evaluated, and their cytotoxicity was estimated via two tests cytotoxic, the hemolysis and the phytotoxicity on Lepidium sativum seeds.

MATERIALS AND METHODS Plant material

Flower buds and fruits of *Capparis spinosa* are harvested in June 2015 from the Teghari region (Latitude:

34.412086, Longitude: -6.044096), located in the province of Sidi Kacem in Morocco. Its identification was carried out in the Plant Biotechnology and Molecular Biology Laboratory of the Faculty of Science, Meknes. [6] The flower buds and mature fruits were cleaned and then were dried in the shade at room temperature. The dry matter is then milled and kept away from moisture until the extracts are prepared.

Preparation methods of the extracts of the plant

Two types of extracts, decoctat at 10% and ethanolic, were prepared from the powder of flower buds and fruits of *Capparis spinosa* according to the method described by Ennacerie *et al.* [13]

Dosage of total phenolic content

Total phenolic content of extracts was determined; it was based on the method of folin ciocalteu method. The acid gallic was used as a standard. For achieving this test, volume 200µL of the ethanolic extract is mixed with 1 mL of reagent Follin ciocalteu diluted at 1/10. After 1 minute stirring, 0.8 mL sodium carbonate 7.5% is added. The mixture was incubated at the shed light and at room temperature for 30 minutes. Then, the absorbance is read at 765 nm against a blank using a spectrophotometer (Spectrophotometer UV-2005). The concentration of total phenolic compounds is expressed in mg equivalent of gallic acid per g (mg EAG/g). [16]

Antioxidant activity

Iron (III) Iron (III) Reduction Method

Antioxidant activity was determined by the ability of *Capparis spinosa* extracts to reduce ferric iron (Fe³⁺) to ferrous iron (Fe²⁺), which provides a blue-color complex readable by the spectrophotometer. The measurement of this activity was carried out according to the protocol described by Koné *et al.* [17] The absorbance of the reaction medium is read at 700 nm by the UV-VIS spectrophotometer (Spectrophotometer UV-2005). Control sample is similarly prepared by replacing the extract with distilled water. The positive control is represented by ascorbic acid. ^[18]

Reducing power of iron (%) = $[1 - (A1 / Ao)] \times 100$ With: - Ao: Absorbance of the negative control;

- A1: Absorbance of the extract

DPPH method

In the presence of free radical scavengers, purple DPPH (2.2 diphenyl 1 picryl hydrazyl) is reduced to 2.2 yellow diphenyl 1 picryl hydrazine. ^[19] The scanning activity of the DPPH radical was measured according to the protocol described by Lopes-Lutz *et al.* ^[20] The reading is performed by measuring the absorbance at 517 nm. The results are expressed as percentage of free radical inhibition (I%) according to the following formula:

I% = [1 - (Abs Sample / Abs Negative Control)] \times 100 The results obtained for each test compound are expressed relative to those obtained for vitamin C taken as reference. The 50% inhibition concentration (IC₅₀) was calculated from the percent inhibition graph as a function of the concentration of the test product.

Test cytotoxicity of extracts *Capparis spinosa* Biological material

Fruits and flower buds of *Capparis spinosa*, as previously reported are consumed by the population in the preparations culinary and medicine. For this reason tests cytotoxicity seem to be interesting to make sure the security of their consumption. Two tests were selected in this study for their simplicity and reliability, the first is carried out on human cells that are the red blood cells, and the second on the *Lepidium sativum* seed taken as example of cell plant.

The choice of red blood cells human is related to their importance in the maintenance of health and life of man. They are known for their sensitivity osmotic, their limited capacity resistance hemolysis towards the nature and the concentration of the components of the external environment. The *Lepidium sativum* seeds, can deduct the effect antimitotic or stimulating proliferation, of the substance tested by following the evolution of their germination and cell growing seedlings.

Evaluation of the hemolytic effect of *Capparis spinosa* **extracts** *in vitro*

Preparation of the erythrocyte suspension

The evaluation of the hemolytic effect of the extracts is carried out according to the method described by Ouedraogo *et al.* and Moukrad *et al.* [21-22] Fresh human blood samples are taken from a healthy donor, and are collected in sterile tubes containing sodium citrate as an anticoagulant with a volume for four volumes of blood. After centrifugation at 1500 rpm for 5 min, the supernatant is removed; the pellet is washed three times with phosphate buffered saline solution PBS (125 mM NaCl, 10 mM sodium diphosphate, pH 7.4). The last centrifugation persisted 10 min.

Preparation of extracts

The two extracts are diluted in phosphate buffered saline to obtain the following concentrations: 10, 5 and 1 mg/mL.

The hemolytic effect

In each tube, 1 mL of extract at different concentrations is introduced with $50\mu L$ of the erythrocyte suspension prepared. The tubes are mixed gently and left at room temperature and in the dark for 60 minutes. The negative control is prepared under the same experimental conditions, with the exception of the presence of the extract to be tested. After incubation, the tubes are centrifuged at 1500 rpm for 5 min. The observation of hemolysis is made directly to the naked eye; the number of repetition is three times for each concentration.

Effect of *Capparis spinosa* extracts on the germination of *lepidium sativum* seeds

Operating mode

20 seeds of *Lepidium sativum* are deposited in a Petri dish (50 mm in diameter), on a Whatman paper disc of the same diameter impregnated with 5 mL of the aqueous solution of the extract, with a concentration of 1 mg/mL. The seeds of *Lepidium sativum* are chosen from the same average size. Regarding the negative control, the disk of the Petri dish is impregnated with 5

mL of distilled water. Then all the dishes were incubated at 25°C and in the dark for one week. Every 24 hours the process of germination and elongation of the radicle is followed directly in the Petri dishes. The results of elongation of the radicle are expressed in mm. The test is considered valid if and only if the germination rate of the controls is greater than or equal to 90%. [23-24] For each test and for the control. These tests are made in triple.

Expression of results

Germination rate of the seeds of Lepidium sativum

The expected germination percentage of the control seeds varies between 90 and 100%. A seed is germinated when there is the opening of the seed and the emergence of a 3 mm stem. [25]

According to Côme, the germination rate (**Gr**) corresponds to the percentage of germinated seeds compared to the total seed sown, it is estimated by the following formula: [26-27]

$$G_r = (Ng / Ns) \times 100$$

Ng: Number of seeds sprouted.

Ns: Number of seeds sown.

Kinetics of germination

The kinetics of germination represents the variation of germination rate of the seeds of *Lepidium sativum* tested over time. Graphically, it is presented by a curve of percentage of germination as a function of time. It allows a precise vision of the germination evolution of a seed lot placed under specific conditions. [26-27]

Germination inhibition rate (GI%)

The ability of an extract or substance to inhibit seed germination is expressed by the following relationship: [24]

$$GI\% = (PG_{control} - PG_{test} / PG_{control}) \times 100$$

PG $_{control}$: Germination percentage of the control lot PG $_{test:}$ Germination percentage of the lot treated by extracts

Vigor of seedling Lepidium sativum

After determination of germination for seven days for each seed lot, a measure of the length of the radical is performed. This value is expressed as an average of extension of rootlets and the results are noted in mm. [24]

Vigor of the seedling = Percentage of germination × Length of seedlings

Reversibility of root growth of Lepidium sativum seeds

The evaluation of the toxicity of the extracts is also examined by observing the irreversibility of this phenomenon. The concentration of 1 mg/mL of the extracts is verified if it caused irreversible cell damage by inhibition, sowing the seeds in the presence of the extracts for two days and then rehydrating them in a medium containing only distilled water for 4 days. Phytotoxicity is considered null or negligible if the germination percentage of the seed of the control batch is between 90 and 100%, and the inhibition in the control dishes should be low compared to the tested groups. The vigor of the seedling of the control seeds

should be maximum and greater than that of the seeds tested

Statistical analysis

For statistical significance using the t-test, the probability value of p < 0.05 was considered statistically significant.

RESULTS

Dosage of total phenolic content

Total phenolic content of aqueous and ethanolic extracts of the two organs studied of *Capparis spinosa*, was determined according to the calibration curve of acid Gallic. The results are expressed in mg equivalent acid Gallic /g of dry plant (mg EAG /g dry matter) (Table 1).

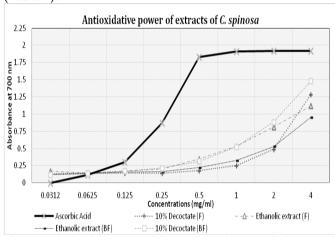
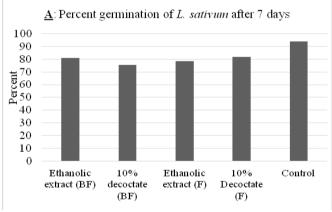


Fig. 1: Antioxidative power of flower buds (BF) and fruit extracts (F) of C. spinosa



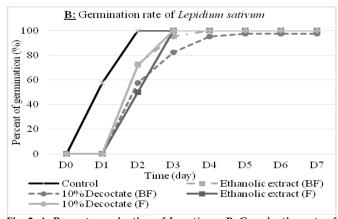


Fig. 2: A. Percent germination of *L. sativum*, B. Germination rate of *L. sativum* after 7 days of treatment with aqueous and ethanolic extracts of flower buds and fruits of *C. spinosa*

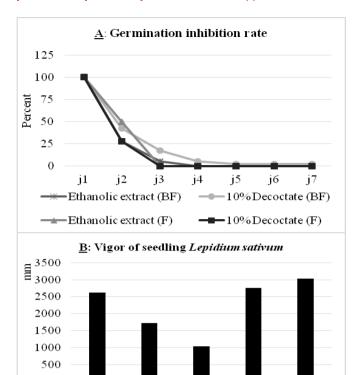


Fig. 3: Germination inhibition in percentage, B. Vigor of seedling of *L. sativum* after 7 days of treatment with aqueous and ethanolic extracts of flower buds and fruits of *C. spinosa*

Ethanolic

Decoctate extract (F) Decoctate

10%

Control

10%

(BF)

Ethanolic

extract

(BF)

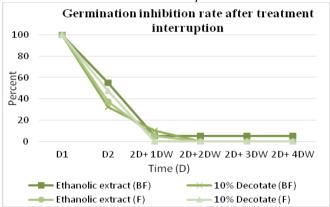


Fig. 4: Germination inhibition rate after interruption of treatment

Table 1: Total phenolic content of ethanolic and aqueous extract of flower buds and fruits of *Capparis spinosa* (mg EAG/g dry matter).

	Flower buds	Fruits
Ethanolic extract	72.59 ± 2.45	18.99 ± 1.84
10% Decoctate	49.89 ± 1.01	22.10 ± 0.57

Table 2: IC₅₀ of aqueous and ethanolic extracts of flower buds and fruits of Capparis *spinosa*

Produits	IC ₅₀ of flower buds (mg/ml)	IC ₅₀ of fruits (mg/ml)
Vitamin C	0.22	0.22
Ethanolic extract	0.52	0.61
10% Decoctate	0.25	0.4

According to these results, the total phenolic content of the flower buds is very high compared to that of fruits for both types of extracts. For flower buds, it is however higher in the alcoholic extract compared to that of the aqueous one.

Antioxidant activity

The results of the iron ion reduction activity by the extracts prepared from both Capparis spinosa organs are shown in figure 1. At a concentration of 0.5 mg/mL, the decoction of the flower buds and the ethanolic extract of the fruits have the same antioxidative power which is 0.37, as well as the two other extracts alcoholic of flower buds and aqueous of fruits, they also have the same value 0.24 relative to the reducing power of ascorbic acid which reaches 1.829. However, at a concentration of 4 mg/mL, the antioxidant effect increases for the four aqueous and ethanolic extracts of flower buds and fruits, and reaches respectively the values of the absorbance of 1.48; 0.95; 1.28 and 1.11. As for the antiradical activity of the four extracts of the two organs, it is expressed in IC50, and the results are grouped in Table 2.

The inhibition concentrations at 50% of free radicals, ethanolic extracts of flower buds and fruits, are almost the same size 0.52 and 0.61 mg/mL respectively, whereas for decoct, the values are in the order of 0.25 mg/mL for flower buds and 0.4 mg/mL for fruit.

Evaluation of the hemolytic effect of *Capparis spinosa* extracts *in vitro*

The result showed that the extract ethanolic of flower buds has an effect hemolytic low to both concentrations 10 and 5 mg/mL which is illustrated by a slight release of hemoglobin in the supernatant. However, to a low concentration of 1 mg/mL this extract has no hemolytic effect; erythrocytes retain their content hemoglobin intracellular. For decoct prepared from fruits and flower buds, no effect hemolytic has been detected. Cells have kept their normal form in all concentrations.

Effect of *Capparis spinosa* extracts on the germination of *Lepidium sativum* seeds

Germination rate of the seeds of Lepidium sativum

The germination capacity of *lepidium sativum* seeds after hydration with distilled water in control lots was 93.92%. It was affected by the concentration of 1 mg/mL of ethanolic and aqueous extracts of flower buds and *Capparis spinosa* fruits whose germination percentages are respectively 81.1; 75.4; 78.6 and 81.7% (Figure 2 A). According to these results the percentage of the most important germination is observed in the lot of the ethanolic extract of the fruits. While in the lot of the decoction of the flower buds the percentage of germination is lower.

Kinetics of germination

On other hand, the daily monitoring of the germination rate in the batches of the control and those treated with the extracts made it possible to notice a clear difference in the speed of the resumption of germination. For control seeds, germination is rapid and reaches 100% after 24 hours, with seed opening and the emergence of a 3 mm stem. Whereas for the lots treated with extracts, germination did not begin until after 48h. The germination rate reached 57.5%; 72% for aqueous extracts and 72%; 50% for alcoholic extracts of flower buds and fruits respectively. This rate does not exceed

90% from the fourth day for the batch treated by the decoction of flower buds, and for other lots this value is reached from the third day (Figure 2B).

Germination inhibition rate (GI%)

Inhibition of the germination process of *Lepidium* sativum seeds, during the 7 days of treatment with the extracts, depends on the type of the latter and the organ of *Capparis spinosa*. The number of non-sprouted seeds compared to the control decreases day after day but with a different pace. After 24 hours of exposure to the treatment, half of the seeds of the two batches of the aqueous extracts of the flower buds and the alcoholic extracts of the fruits did not germinate. While in the other two lots 25% of the seeds did not germinate. Inhibition of germination was insignificant in the third day generally for all batches, except that it was treated by the decoction of flower buds where it lasted 5 days (Figure 3A).

Vigor of seedling Lepidium sativum

In this experiment, after 7 days of incubation, the results reveal that the length of the radicle of the seedlings was reduced by nearly 66 and 10% respectively for the ethanolic and aqueous extracts of the fruits. And 43.5 and 13.71 % in case of ethalonic and aqueous extracts of flower buds (Figure 3 B).

Reversibility of root growth of Lepidium sativum seeds

In order to verify the continuity of the inhibitory effect of the growth or of the cell division of the extracts of the plant, The reversibility test (Figure 4) shows that the rehydration of the seeds of *Lepidium sativum* after two days of incubation in the extracts, gives a resumption of growth of rootlets in a normal way, so that in the second day the inhibition of germination was not detected.

DISCUSSION

Dosage of total phenolic content

Difference of total phenolic content or of distribution of secondary metabolites in the organs of the plant depends on the growth phase and climatic conditions (The high temperature, the sun exposure, drought, salinity), which stimulate the biosynthesis secondary metabolites such as polyphenols. [28-29] In referring to the results of the bibliography quantifying the polyphenols total in extracts of fruits of C. Spinosa, we noted that a difference of its values depends on the type of solvent and the geographic origin of the plant. However, Allaith found a content of 120 mg EAG/100 g fresh weight of fruits of C. spinosa Bahreïnienne. [7] As for Aliyazicioglu and its collaborators the content was 37.01 ± 0.03 mg EAG/100 g of extract of *Capparis spinosa* Turkish. [9] The comparison of literature founding, with these of our study, we notice that they found values of total phenolic compounds for the fruits of Capparis spinosa Moroccan are the richest.

Concerning flower buds, their total phenolic content for the methalonic extract was quantified by Bonina and collaborators which the result was 65.13 \pm 5.53 mg E

rutin/g. ^[12] This content is interesting, but it also remains low relatively to the results found in this study. Other research has confirmed the wealth of *Capparis spinosa* in polyphenols in other organs. Namely the leaves, by Proestos and colleagues who found 16 mg EAG/g of dry extract. ^[30] As for Arrar and collaborators, content was about 0.082 mg EAG/g extract dried. ^[31]

In general, this difference in values found is explained by the difference of the organs of the plant studied, and by the difference in standard used for the dosage polyphenols, by geographical factors and distinct treatment methods. However, the total polyphenol content of the two types of extract fruit and flower buds of Moroccan *Capparis spinosa* are the much more interesting.

Antioxidant activity

The values of this test show that this species *Capparis spinosa* has a good antioxidant activity, especially for flower buds in the form of decoction. This extract has more than half of reducing power of vitamin C at a concentration 4mg/mL. At this same concentration, comes a decoct of fruits in second class, then its extract alcoholic and at the end that of the flower buds.

Some researchers have rated this activity by the FRAP method. Allaith, tested extract methanolic of fresh fruits of this species collected from Bahrain, and found a total value average 9.059 ± 1.450 mmol TEAC / kg fresh weight. [7] Aliyazicioglu reported a value FRAP of $145.07 \, \mu$ mol Trolox/ $100 \, g$ dry weight for fruits dried of *Capparis spinosa* Turkish. [9] All these reported values, even if they come from fresh and dry samples with different standards, they confirm the antioxidant activity of this species.

As for the results of the DPPH method expressed in IC₅₀, they reveal power anti-oxidative important for the four extracts. In addition, the decocted of flower buds has the largest capacity for trapping of free radicals DPPH* and that is surrounding to that of the reference, vitamin C. Other extracts are also a power anti-radical rated of the largest in the lower as follows; the decocted of fruits, extracts alcoholics of capers and fruits. These results confirm so those obtained by the FRAP method. In general, referring to the bibliography, the Capparis spinosa species have shown antioxidant activity by both FRAP and DPPH methods in several studies by testing different parts of plant with different solvents. On the one hand, Fabri and collaborators confirmed this result for extract prepared from leaves and stems by ethyl acetate and butanol. [32] On the other hand, Fadili showed that fruits have high antiradical potency compered to leaves in ethyl acetate fraction. [29] As well as Germanò, Bonina and their collaborators, they reported that extracts of flower buds showed strong antioxidant activity with these both techniques and others. [33-12]

The antioxidant power of plants is often correlated with total phenolic content. This is clear, for both decoct and

the ethanolic extract of fruits, that are consistent with the results of Bonina, Meddour and their collaborators. [12-14] But this correlation is not valid for the alcoholic extract of the flower buds. This result is explained by the presence of other molecules in the ethanolic extract of flower buds capable of trapping free radicals such as tocopherols, carotenoids and glucosinolates. [33-34, 14]

Among the polyphenols responsible for the anti-radical power, flavonoids interact with many radicals. This type of molecule is present in high concentration in all parts of *Capparis spinosa*. [31-35] Flavonoids (Fi-OH) have low redox potentials, hence their thermodynamic ability to reduce oxidative free radicals such as superoxide, peroxyl and hydroxyl, by hydrogen transfer. [32]

Fi-OH + R > Fi- O + RH

We deduce from this study that the flower buds and fruits of *Capparis spinosa* have a significant antioxidant activity depending on their polyphenol content. They are also described as antibacterial activity and as synergistic effect on antibiotics. ^[13]

Evaluation of the hemolytic effect of *Capparis spinosa* extracts in vitro

The results of this test allow us to deduce that the ethanolic extract of the flower buds is the only one of the four extracts that has a toxic effect at a concentration of 10 mg/mL. It acted on the permeability of the erythrocyte membrane by interacting with the sterols of the membrane. [36-21] This interaction causes an increase in the permeability of the membrane to water accompanied by the entry of Na+ and the output of K+, hence the deformation of the cell and its lysis by releasing hemoglobin into the external environment. [37-22] The absence of toxicity in the other extracts can be explained by the difference of the phytochemical groups present in the two types of ethanolic and aqueous extract and the both organs of this plant. In addition, the type of solvent and the extraction techniques influence the chemical composition of the extracts. [38] Among the bioactive compounds affecting the stability of red blood cells, there are the saponins which have the properties of nonionic detergents, with cytotoxic activities such as hemolytic and antibacterial effects. [39] Therefore, a thorough study is needed to identify the molecules responsible for this effect and their mechanism of action on the globular membrane.

We found that the alcoholic extract of capers had high phenolic content. It also caused a hemolytic effect. So, we can suggest that the flower buds contain toxic phenolic compounds that have important antioxidant activity. Despite their richness in polyphenols and interesting anti-radical power, flower buds are not tolerated by human blood cells. A consumption of 10 mg/mL can cause adverse effects on health. However, flower buds are the consumed part of the plant in various Moroccan culinary preparations, and worldwide; prompting *in vivo* studies based on the

rationalization and modality of preparation and use of this plant.

Effect of *Capparis spinosa* extracts on the germination of *lepidium sativum* seeds

Germination is a transitional phase where the seed passes from a life slowed to a young autotrophic seedling. It is a very complex biochemical and physiological process that can be measured by several factors such as imbibition and respiration, [27] as well as the ability of a product to inhibit this phase is a complex process. Under natural conditions and in the presence of an exogenous stimulus such as water mainly, the seeds germinate. This germination is initiated by the synthesis of the amylase enzyme responsible for the degradation of the starch reserves constituting the albumin, thus releasing the energy necessary for the germination of the embryo. [40] Then, the embryonic growth phase that begins with the intervention of plant growth hormones such as auxin. [27] However, several hypotheses allow the explanation of the inhibition of the germination phenomenon by the extracts and which is based on the effect of the bioactive molecules that constitute them. On the one hand, they can act on the amylase enzyme by its inhibition or the occupation of its membrane site. On the other hand, they can mimic or be antagonistic to growth hormones or also inhibit their tissue actions. [41-²⁷ In addition, the difference in effect of the extracts is related to the variation in their chemical compositions either of the type of the molecules or also their concentration, knowing that the two most active extracts in this study are of two different organs and prepared by two different methods.

The decoction of flower buds showed a high content of phenolic compounds compared to the ethanolic fruit extract, and more antioxidant and hemolytic activity. It is deduced that the phenolic compounds are not the products directly responsible for the inhibition of germination, because this correlation is not found for the other extracts. So it can be due to other bioactive molecules that must be identified.

Regarding the vigor of Lepidium sativum seedlings, which is an index of the mitotic capacity of radicles, the rapid multiplication of cells induces longer root growth and vice versa. The calculation of this index showed that despite the strong inhibition of germination applied by the decoction of the flower buds, compared to the alcoholic fruit extract, their action on the elongation of the seedlings was contradictory. As a result, the decoction of flower buds has a direct effect on the germination process. While the alcoholic extract of the fruits affects the division and the cellular elongation. The latter, according to Muller they are sensitive to the presence of allelopathic compounds. [42-²⁷] These antimitotic and cell growth stimulating activities induced by the extracts studied previously can be considered as important characteristics of this plant, implying probably an anti-cancer and healing bioactive capacity, so a very promising therapeutic value which must be confirmed by more relevant studies.

The test of the reversibility of the growth of the radicles of the seeds of *Lepidium sativum* allowed us to deduce that the products tested, do not have a definitive effect of inhibition of the growth of the plant cells. The rootlets have resumed their growth in a normal way. This founding indicates the non-toxicity of these substances studied. According to previous research, the molecules responsible for the inhibition of seed germination and subsequent growth of *Lactuca sativa* seedlings are monoterpenoids, mainly those belonging to ketones and alcohols, followed by the group of aldehydes and phenols. [43] It can be deduced that the extracts studied contain one or more groups of monoterpenoids with a high rate in the decoction of flower buds which has also shown good antioxidant activity.

In conclusion, the results of this study confirm the therapeutic importance of *Capparis spinosa* and its pharmaceutical and nutritional value. The fruits and flower buds are rich in polyphenols. They have a very important anti-oxidant activity and possess an antimitotic and regenerative capacity of the tested cells, in addition to their antibacterial and antifungal activity. The benefits and harms of this plant are related to the methods of its preparation and the part and concentration of the plant used.

REFERENCES

- 1. Tlili N, Elfalleh W, Saadaoui E, Khaldi A, Triki S, Nasri N. The caper (*Capparis* L.): Ethnopharmacology, phytochemical and pharmacological properties. Fitoterapia. 2011; 82: 93–1010
- Inocencio C, Cowan RS, Alcaraz F, Rivera D, Fay MF. AFLP fingerprinting in Capparis subgenus Capparis related to the commercial sources of capers. Genet Resour Crop Evol. 2005; 52:137-44.
- Saifi N, Echchgadda G, Ibijbijen J. The morphological characterization of caper plant (Capparis ssp.) in North Morocco. J Food Agric Environ. 2010; 8:876–81.
- 4. El Azhary K, Tahiri Jouti N, El Khachibi M, Moutia M, Tabyaoui I, El Hou A, Achtak H, Nadifi S, Habti N, Badou A. Anti-inflammatory potential of *Capparis spinosa* L. in vivo in mice through inhibition of cell infiltration and cytokine gene expression. BMC Compl Alter. Med. 2017; 17:81
- United State Department of Agriculture, USDA National Nutrient Database for Standard Reference. Release, 2010, pp. 23
- 6. Ennacerie F-Z, Rhazi Filali F, Rahou A. Ethnobotanical study of medicinal plants used in traditional medicine in the province of Sidi Kacem, Morocco. Asian J. Pharm. Clin. Research. 2017; 10: 121–130.
- Allaith AA. Assessment of the antioxidant properties of the caper fruit (*Capparis spinosa* L.) from Bahrain. J. Asso. of Arab Univ. Basic and App. Sci. 2016; 19: 1–7.
- 8. Tlili N, Nasri N, Saadaoui E, Khaldi A, Triki S. Carotenoid and tocopherol composition of leaves, buds and flowers of *Capparis spinosa* grown wild in Tunisia. J Agric Food Chem. 2009; 57:5381–5.
- Aliyazicioglu R., Eyupoglu OE., Sahin H, Yildiz O, Baltas N. Phenolic components, antioxidant activity, and mineral analysis of *Capparis spinosa* L. Afr. J. Biotechnol. 2013; 12: 6643–6649.

F Rhazi Filali et al. / Evaluation of the Antioxidant Activity and the Cytotoxicity of Extracts of Capparis......

- 10. Matthäus B, Özcan M. Glucosinolate composition of young shoots and flower buds of capers (Capparis species) growing wild in Turkey. J Agric Food Chem. 2002; 50: 7323–7325.
- 11. Arena A, Bisignano G, Pavone B, Tomaino A, Bonina F. P, Saija A, Cristani M, D'Arrigo M, Trombetta D. Antiviral and Immunomodulatory Effect of a Lyophilized Extract of *Capparis spinosa* L. Buds. Phytother. Res. 2008; 22: 313–317.
- Bonina F, Puglia C, Ventura D, Aquino R, Tortora S, Sacchi A, Saija A, Tomaino A, Pellegrino ML, de Caprariis P. *In vitro* antioxidant and in vivo photoprotective effects of a lyophilized extract of *Capparis spinosa* L. buds. J. Cos. Sci. 2002; 53:321-335.
- 13. Ennacerie F-Z, Rhazi Filali F, Moukrad N, Ed-Dra A. Antibacterial synergistic effect of extracts of the organs of *Cappari spinosa* and in combination with antibiotics . Int. J. Adv. Res. 2017; 5(9): 1238-1247.
- 14. Meddour A, Yahia M, Benkiki N, Ayachi A. Etude de l'activité antioxydante et antibactérienne des extraits d'un ensemble des parties de la fleur du *Capparis spinosa* L. Leb. Sci. J. 2013;14:1.
- Trombetta D, Occhiuto F, Perri D, Puglia C, Santagati NA, De Pasquale A, Saija A, Bonina F. Antiallergic and antihistaminic effect of two extracts of *Capparis spinosa* L. flowering buds. Phytother Res. 2005; 19:29–33.
- Boizot N, Charpentier J-P. Méthode rapide d'évaluation du contenu en composés phénoliques des organes d'un arbre forestier. Cah. Tech. INRA. N°. spécial, 2006, pp. 79-82.
- 17. Koné WM, Kamanzi Atindehou K, Terreaux C, Hostettmann K, Traoré D, Dosso M. Traditional medicine in North Côted'Ivoire: Screening of 50 medicinal plants for antibacterial activity. J. Ethnopharmacol. 2004; 93(1):43–49.
- 18. Kiran CR, Rao DB, Sirisha N, Rao TR. Assessment of phytochemicals and antioxidant activities of raw and germinating Ceiba pentandra (kapok) seeds . J. Biomed. Res. 2015; 29(5):414–419.
- 19. Maataoui BS, Hmyene A, Hilali S. Activités anti-radicalaires d'extraits de jus de fruits du figuier de barbarie (*Opuntia ficus indica*). Leban. Sci J. 2006; 7(1):3–8.
- Lopes-lutz D, Alviano DS, Alviano CS, Kolodziejczyk PP. Screening of chemical composition, antimicrobial and antioxidant activities of Artemisia essential oils. Phytochemistry. 2008; 69:1732–1738.
- Ouedraogo Y, Nacouima O, Guissou I.P, Guede guina F.J. Evaluation in vivo et *in vitro* de la toxicité des extraits aqueux d'écorces de tige et de racines de mitragyna inermis (wilid).o.ktz (Rubiaceae). Pharm. Méd. Trad. Afr. 2001; 11: 13-29.
- Moukrad N, Rhazi Filali F, Zegaoui O, Guedegulna F. Microscopic observation of the effect of zno nanoparticles synthesized from different precursors on eukaryotic and prokaryotic cells. IJSR. 2013; 4 (5): 2319-7064.
- Bewley J. D. Seed Germination and Dormancy. American Society of Plant Physiologists. The Plant Cell. 1997; 9: 1055-1 066
- Moukrad N, Rhazi Filali F, Daou I, Zegaoui O. Phytotoxic activity of the zin oxyde nanoparticules synthesized from different precursors on germination and radicle growth of seed *Lepidium sativum*. Int. J.Scien. Res. Publ. 2014; 4(12):2250-3153
- Centre d'expertise en analyse environnementale du Québec, Inhibition de la germination et de la croissance chez les semences de végétaux. MA. 500 - GCR 1.0, Ministère de l'Environnement du Québec. 2003, pp.30.
- Côme D. Les obstacles à la germination (Monographie et physiologie végétale n° 6). Éd. Masson et Cie (Paris). 1970, pp.14, 24 and 27.

- Cherif R, Kemassi A, Boual Z, Bouziane N, Benbrahim F, Hadjseyd A, Gharib T, Ould el Hadj-Khelil A, SakeurM.L, Ould el Hadj M.D. Activités biologiques des extraits aqueux de Pergularia tomentosa L. (asclepiadaceae). Leb. Sci. J. 2016; 17(1): 25-35.
- 28. Falleh H, Ksouri R, Chaieb K, Karray-Bouraoui N, Trabelsi N., Boulaaba M, Abdelly C. Phenolic composition of Cynara cardunculus L. organs, and their biological activities. C. R. Biologies. 2008; 331: 372-379.
- 29. Fadili K, Zerkani H, Amalich S, Zair T. Etude phytochimique et evaluation de l'activite antioxydante des feuilles et des fruits du *Capparis spinosa* L. American. J. Inn. Res. Ap. Sci. 2017; 5(2): 108-118.
- Proestos C, Boziaris I.S, Nychas G.-J.E, Komaitis M. Analysis of flavonoids and phenolic acids in Greek aromatic plants: Investigation of their antioxidant capacity and antimicrobial activity. Food Chem. 2006; 95: 664–671
- 31. Arrar L, Benzidane N, Krache I, Charef N, Khennouf S, Baghiani A. Comparison between polyphenol contents and antioxidant activities of different parts of *Capparis spinosa* L. Phcog Commn. 2013; 3(2): 70-74.
- 32. Fabri RL, Nogueira MS, Braga FG, Coimbra ES, Scio E. Mitracarpus frigidus aerial parts exhibited potent antimicrobial, antileishmanial and anti oxidant effects. Bioresource Technology. 2009, 100: 428-433.
- 33. Germanò MP, De Pasquale R, D'Angelo V, Catania S, Silvari V. Evaluation of extracts and isolated fraction from *Capparis spinosa* L. buds as an antioxidant source. J Agric Food Chem. 2002; 50(5):1168-71.
- 34. Matthäus B, Ozcan M. Glucosinolates and fatty acid, sterol, and tocopherol composition of seed oils from *Capparis spinosa* var. spinosa and *Capparis ovata* Desf. var. canescens (Coss.) Heywood. J Agric Food Chem. 2005; 53: 7136–7141
- 35. Alzergy AA. Elgharbawy S MS, Ghyath SM, Mervat R M. Role of *Capparis spinosa* in ameliorating trichloroacetic acid induced toxicity in liver of Swiss albino mice. Life Sci J. 2015; 12(2): 26-39.
- Majester-Savornin B, Elias R, Diaz-Lanza AM, Balansard G, Gasquet M, Delmas F. Saponines of the ivy plant. Hedera helix and their leishmanicidic activity. Planta Med. 1991; 57: 260-262
- 37. Heinz A, Passow H. Role of external potassium in the calcium- induced potassium efflux from human red cell ghosts. J. Membrane Biol. 1980; 57:119-131.
- Shah H, Nisar M, Suhail M, Bacha N. Antimicrobial studies of the crude extracts from the roots of Chenopodium ambrosioides Linn. African. J. Microbiol. Res. 2014; 8(21): 2099–2104.
- Arabski M, Wegierek-Ciuk A, Czerwonka G, Lankoff A, Kaca W. Effects of Saponins against Clinical E. coli Strains and Eukaryotic Cell Line. J. Biomed. Biotech. 2012; 286216: 1-6.
- Regnault-Roger C, Philogene B. Bio-pesticides d'origine végétale. Jr, Vincent Ch. Ed. TEC and DOC, Paris. 2008, pp. 51-60
- 41. Feeny P. Plant apparency and chemical defense. Rec. Adv. Phyto. 1976; 10: 1-40.
- 42. Muller CH. Inhibitory terpenes volatilized from Salvia shrubs. Bull. Tor. Bot. Club. 1965, 92: 38-45.
- Rolim de Almeida L. F, Frei F, Mancini E, De Martino L, De Feo V. Phytotoxic Activities of Mediterranean Essential Oils . Molecules. 2010; 15: 4309-4323.
- 44. Tesoriere L, Butera D, Gentile C, Livrea MA. Bioactive components of caper (*Capparis spinosa* L.) from sicily and antioxidant effects in a red meat simulated gastric digestion. J. Agric. Food Chem. 2007; 55: 8465–8471.

HOW TO CITE THIS ARTICLE: Ennacerie F-Z, Rhazi Filali F, Moukrad N, Bouidra M, Bentayeb A. Evaluation of the Antioxidant Activity and the Cytotoxicity of Extracts of *Capparis spinosa*. Int. J. Pharm. Sci. Drug Res. 2018; 10(2): 57-64. **DOI:** 10.25004/IJPSDR.2018.100202