



## Phenolic compound and herbicidal activity of *Rosa canina* L. leaf and stem extracts

Saad Inès\*, Jaouadi Rim, Elimem Mohamed, Ben Salem Amel, Rouz Slim

*Research Laboratory of Agricultural Production Systems and Sustainable Development,  
Department of Agricultural Production, Higher School of Agriculture of Mograne (ESAM),  
Mograne, Zaghouane, University of Carthage, Tunisia*

Article published on December 03, 2022

**Key words:** *Rosa canina*, Aqueous extracts, Flavonoids, Allelopathy, Bioherbicides

### Abstract

This study proposes the evaluation of the phenolic composition of leaves and stems extracts of *Rosa canina* (Dog rose) and their herbicidal abilities measured using assays of germination rate (G) and germination speed (GI) of *lactuca sativa* (Lettuce), *phalaris truncata* and *raphanus raphanistrum* seeds and assays of its seedling growth. Leaves and stems of dog rose were rich in total phenolics and total flavonoids with comparative contents. Whereas flavones/flavonols were abundant in leaves extracts and in traces in stems extracts. G and GI of target species seeds and root and shoot length of its plants were reduced from the lowest concentration of both extracts. Reductions of G and GI were more important in presence of leaves dog rose extracts compared to those noted with stems extracts. *Raphanus raphanistrum* was the most sensitive target species to both extracts. G and GI of this weed were canceled from 30g/l of leaves extract. Root and shoot growth of target species were more inhibited in presence of dog rose leaves extracts. Inhibitions reached 100% from concentrations of 40g/l, 60g/l and 90g/l respectively for wild radish, canary grass and lettuce. The findings allow us to demonstrate that leaves and stems from dog rose could be a prospective residual vegetable source of valuable phytochemicals, for future applications in the field of agroecology and biological weed control.

\*Corresponding Author: Saad Inès ✉ [saadines83@yahoo.com](mailto:saadines83@yahoo.com)

## Introduction

Wild rose, also called dog rose (*Rosa canina* L.) belongs to the Rosaceae family, which contains more than 100 species and grows mostly in Europe, Asia, North America, Africa and the Middle-East (Fascella *et al.*, 2019). In Tunisia, Dog rose was reported to grow throughout the northern and the backbone of Tunisia (Alapetite, 1979). Its Arabic name is Nesri. It is a shrub growing up to 4 m tall, with hooked thorns on floppy stems and single or double-serrated leaves which are divided into five or seven leaflets (Tabaszewska and Najgebauer-Lejko, 2020). Specimens of the plant are used as rootstocks for grafting. The wild plant itself is widely used to stabilize soil in land reclamation and specialized landscaping schemes (Koczka *et al.*, 2018). *Rosa canina* L. has been widely used in folk confectionery and remedy in Tunisia. It is traditionally used for the prevention and therapy of common cold, flu, gastrointestinal disorders and infections (Wenzig *et al.*, Nađpal *et al.*, 2016). Dog rose is also valued for both aesthetical reasons and because of the significant concentration of many active components (Tabaszewska and Najgebauer-Lejko, 2020). Many study focused on rose hip but few are interested with study focused on characterizing an unexplored source, namely leaf and stem of *Rosa canina* (kubczak *et al.*, 2020). Moreover, previous studies done on this plant report its phenolic compounds, and antioxidant activities (Ben Jemaa *et al.*, 2017; Ghazghazi *et al.*, 2010), antifungal activities and antimicrobial activities (Ghazghazi *et al.*, 2013; Polumackanycz *et al.*, 2020). No study, to our knowledge, focused on its herbicidal activity. This study aims to determine the chemical composition of aqueous extracts of both leaves and stems of *R. canina* as well as its allelopathic and herbicidal activities.

## Material and methods

### *Plant Material and Extracts Preparation*

The fully developed leaves and stems of *Rosa canina* L. were collected from Higher agronomic Institute of Mogran situated in Zaghuan in the North of Tunisia (Altitude: 158m-262m, Latitude: 36°23'-36°25', Longitude: 10°7'-10°10'). Collected organs of wild rose were rinsed then oven-dried at 40°C for 72 h and

grinded. 100 grams of each dried material were soaked in 1 L distilled water at room temperature for 24 h (Chon *et al.*, 2005). The extracts were filtered several times and kept at 4°C in the dark until use.

### *Phyto analytical Studies*

#### *Total phenolic content*

Total soluble phenolic compounds in the extracts were determined with Folin-Ciocalteu method. 100 µl of extract was added to 500µl of 1/10 diluted (in Milli-Q water) Folin-Ciocalteu phenol reagent and was incubated at ambient temperature and obscurity. After 5 min, 400µl of aqueous sodium bicarbonate (Na<sub>2</sub>CO<sub>3</sub>, 7.5%) was added and then allowed to stand for 90 min with intermittent shaking.

The absorbance was measured at 760 nm in a spectrophotometer (Singleton and Slinkard, 1977). A standard curve was prepared using gallic acid. The total phenolic contents were expressed in terms of gallic acid equivalents (mg/ g of dry mass) using calibration curve (R<sub>2</sub>= 0.971).

#### *Total flavonoid content*

Total flavonoid content was determined using the Chung *et al.* (2000) method. Briefly, 0.5mL of 2% solution of AlCl<sub>3</sub> in methanol was mixed with the same volume of extract. After mixing, samples were incubated in the obscurity for 30 minutes (Quettier *et al.*, 2000). A standard curve was prepared using Quercetine and absorption readings at 430 nm were taken against a blank. Results were expressed as mg Quercetine equivalent/g dry weight (mg QE/g dw) using Quercetine calibration curve (R<sub>2</sub> = 0.997).

#### *Total flavonones/flavanols*

Total flavonones/flavanols in the plant extracts were estimated using the method reported by Adedapo *et al.* (2008). 0.2 mL AlCl<sub>3</sub> (2%) ethanol solution and 3 mL NaNO<sub>2</sub> (5%) were added to 10 mg of diluted extracts. Absorption readings at 440 nm were taken after 2 h and 30 min incubation at 20°C. Total flavonones/flavanols content was expressed as mg Quercetin equivalents/g dry weight (mg QEs/g DW) using Quercetin calibration curve (R<sub>2</sub>=0.996).

### Bioassays

Aqueous extracts of leaves and stems of *R. canina* were diluted to give final concentrations of 10, 20, 30, 40, 50, 60, 70, 80, 90 and 100 g/l (Chon *et al.*, 2005). Extracts were tested on one crop (*Lactuca sativa* L.), one broadleaf weed (*Raphanus raphanistrum* L.) and one monocot weed (*Phalaris truncata* L.). Seeds were surface sterilized with diluted sodium hypochlorite for 15 min, then rinsed four times with deionized water (Chon *et al.*, 2005).

Two sets of Petri plates were prepared. In the first set, imbibed seeds were used to evaluate the effect of extracts on germination. The second set of pre-germinated seeds, with 1 mm root length, was used to evaluate the effect of extracts on root and shoot growth. The Petri dishes were placed in a growth chamber at 24/22°C for 14/10 h light and dark periods, respectively.

Treatments were arranged in a completely randomized design with four replications. Germinated seeds were counted at 24 h intervals during six days. Shoot and root length of receiver species were measured seven days after sowing the pre-germinated seeds in each Petri dish. Data were transformed to percent of control for analysis.

The index of germination GI was determined using the following formula (Chiapuso *et al.*, 1997):

$$GI = (N_1) * 1 + (N_2 - N_1) * 1/2 + (N_3 - N_2) * 1/3 + \dots + (N_n - N_{n-1}) * 1/n$$

Where,  $N_1, N_2, N_3, \dots, N_n$ : proportion of germinated seeds observed afterwards 1, 2, 3, ..., n-1, n days. This index shows the germination delay induced by the extract (Delabays *et al.*, 1998). The inhibitory or stimulatory percent was calculated using the following equation given by Chung *et al.*, (2001):

$$\text{Inhibition (-)/stimulation (+)\%} = \left[ \frac{\text{extract} - \text{control}}{\text{control}} \right] \times 100$$

Where Extract: parameter measured in presence of *R. canina* extracts and Control: parameter measured in presence of distilled water.

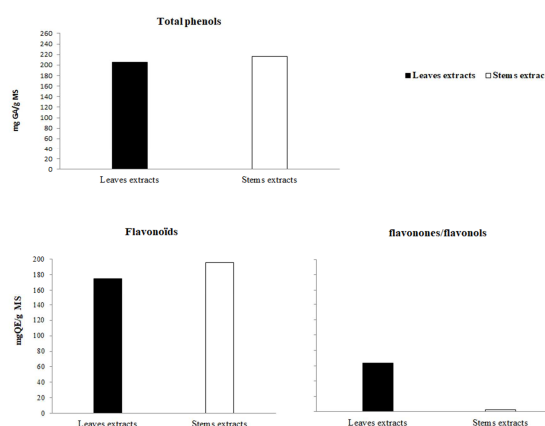
### Statistical analysis

The laboratory bioassays were conducted in a completely randomized design. All data were reported as mean  $\pm$  standard deviation (SD) of four replicates for biological activities and of five replicates for phytochemical analysis. ANOVA and a post hoc Duncan Multiple Range tests were performed for bioassays with IBM SPSS Statistics version 20, for Windows program, to analyze treatment differences. The means were separated on the basis of least significant differences (LSD) at the 0.05 probability level.

## Results and discussion

### Phytochemical screening

Total phenols contents in stems aqueous extracts of *rosa canina* (215mg GA/g MS) were comparable to those in leaves extracts (205mg GA/g MS) (Fig.1). These results are similar to those found by Kubczak *et al.* (2020) who stated that there was no significant difference in total phenolic compound contents between extracts of leaves and twigs of *Rosa canina*.



**Fig. 1.** Total phenols, total flavonoids, total flavones and flavonols contents in leaves and stems aqueous extracts of *Rosa canina*.

Phenolic compounds including tannins, flavonoids, phenolic acids, and anthocyanins proved to be a very important group of biologically active ingredients present in rosehip, but there are a few studies analyzing the content and composition of polyphenols in stems and leaves of *R. canina* (Koczka *et al.*, 2018). A few recent studies have indicated that stems from *R. canina* are also a good source of polyphenols (Ouerghemmi *et al.*, 2020; Riffault *et al.*, 2014).

Recently Ouerghemmi *et al.* (2020) showed that stem extracts from different *Rosa* species are an important source of phenolic compounds.

Total flavonoids content in stems aqueous extracts of *R. canina* were slightly higher than those in leaves aqueous extracts (196mg QE/gMS and 174mg QE/gMS, respectively) (Fig.1). These results proved richness of leaves and stems of this species in flavonoids and agreed with those found by Sytar *et al.* (2015). They found that *R. canina* proved to be the richest species between *Rosa* in flavonoids. However, Kubczak *et al.* (2020) announced that flavonoids accounted only about 9.5% and 5.5% of the phenolic compounds, respectively in leaves and stems aqueous extracts of *R. canina*.

They explicated that the result does not correspond exactly to the total flavonoid content of the extracts because  $AlCl_3$  reacts mainly with flavones, flavonols, flavanones and flavanols (Kubczak *et al.*, 2020).

Total flavonones/flavonols contents were important in leaves extracts (63mg QE/gMS) and were in traces forms in stems extracts (2.6mg QE/gMS) (Fig.1). Cunja *et al.* (2014) investigated the phenolic content in extracts from *Rosa* sp. The extract from *R. canina* was the richest in flavonols. Moreover, in a recent work, Ouerghemmi *et al.* (2020) explored polyphenolic composition of stem extracts from *R. canina* L. growing in Feija in North of Tunisia (Humid bioclimate) and founded that its profiles were dominated by flavonols which is in opposition with our results. In fact, these compounds were detected in our study in very small amounts compared with those in leaves extracts.

This discrepancy can be explained with the different solvents and methodologies used for the extractions (Dai and Mumper, 2010) and also by difference in regions where stems of dog roses were collected (Mograne-Zaghuan in our case, Sub-Humid bioclimate). Indeed, Ouerghemmi *et al.* (2016) showed that *R. canina* species collected in Zaghuan exhibited the lowest polyphenol content to respect to the corresponding species in the other studied

regions. Generally, plants growing in different altitude, latitude, longitude, soil and climatic conditions could show different phenolic content (Verma and Shukla, 2015; Koczka *et al.*, 2018).

#### *Phytotoxicity to germination*

##### *Effect of Rosa canina aqueous extracts on the germination rate of target species seeds*

Germination rate (G) and index (GI) of all target species seeds were affected by leaves and stems aqueous extracts of *R. canina*. Reductions in germination rates increased with increasing concentrations of extracts (Table 1 and 2).

Leaves aqueous extracts of dog rose were more toxic than its stems aqueous extracts for all target species (Table1). Germination rates of lettuce, wild radish and phalaris seeds, were canceled out when irrigated with leaves aqueous extracts at concentrations of 60g/l, 30g/l and 70g/l, respectively. While under the effect of stems aqueous extracts, germination rates were canceled from concentrations of 100g/l, 90g/l and 80g/l, respectively (Table 1).

*Raphanus raphanistrum* was found to be the most sensitive to *R. canina* leaf extract compared to *lactuca sativa* and *phalaris truncata*. From the concentration of 30g/l, seed germination of wild radish was reduced by 85% under the effect of leaves extracts against reductions of only 44% and 58%, respectively for the lettuce and phalaris seeds (Table 1).

##### *Effect of Rosa canina aqueous extracts on the germination index of target species seeds*

Germination index was slowed in all cases, and was more affected by leaves extracts than stems extracts (Table 2). From lowest concentration of leaves extracts, GI of *Raphanus raphanistrum* seeds were reduced by 87.5%. The respective values for *phalaris truncata* and *lactuca sativa* seeds were 74.8% and 22.8%, respectively. Under the same concentration of stems extracts, GI reductions were only 14% at means for lettuce and wild radish seeds, and 10% for phalaris seeds. (Table 2)

**Table 1.** Germination rate, expressed as% of control, of *lactuca sativa*, *raphanus raphanistrum* and *phalaris truncata* seeds, in presence of *Rosa canina* leaves and stems aqueous extracts.

	Extract Concentration (g/l)	Target species		
		<i>Lactuca sativa</i>	<i>Raphanus raphanistrum</i>	<i>Phalaris truncata</i>
Leaves extract	10	80,72 <sup>a</sup> ±6,72	16,66 <sup>ab</sup> ±28,86	33,89 <sup>bcd</sup> ±14,93
	20	75,72 <sup>a</sup> ±14,49	23,88 <sup>a</sup> ±8,97	70 <sup>a</sup> ±21,21
	30	56,9 <sup>ab</sup> ±5,94	15,55 <sup>ab</sup> ±14,82	41,39 <sup>bc</sup> ±13,54
	40	42,89 <sup>bc</sup> ±13,1	0 <sup>b</sup>	31,11 <sup>bcd</sup> ±16,61
	50	83,95 <sup>cd</sup> ±96,19	0 <sup>b</sup>	51,67 <sup>ab</sup> ±11,9
	60	23,95 <sup>cd</sup> ±5,13	0 <sup>b</sup>	30,56 <sup>bcd</sup> ±6,3
	70	17,76 <sup>d</sup> ±2,79	0 <sup>b</sup>	18,33 <sup>cde</sup> ±13,84
	80	6,38 <sup>d</sup> ±6,57	0 <sup>b</sup>	15 <sup>de</sup> ±11,18
	90	7,63 <sup>d</sup> ±2,64	0 <sup>b</sup>	13,06 <sup>de</sup> ±8,83
	100	0 <sup>e</sup>	0 <sup>b</sup>	0 <sup>e</sup>
Stems extract	10	92,37 <sup>a</sup> ± 5,65	74,17 <sup>a</sup> ±10,58	76,94 <sup>a</sup> ±10,81
	20	86,18 <sup>a</sup> ± 9,54	63,61 <sup>ab</sup> ±10,1	64,16 <sup>b</sup> ±8,29
	30	89,87 <sup>a</sup> ± 3,54	58,61 <sup>bc</sup> ±14,6	66,94 <sup>ab</sup> ±7,46
	40	88,75 <sup>a</sup> ± 7,39	61,11 <sup>ab</sup> ±11,78	59,16 <sup>b</sup> ±5,95
	50	84,93 <sup>a</sup> ± 6,98	55,55 <sup>bc</sup> ±6,8	48,88 <sup>c</sup> ±5,61
	60	21,51 <sup>b</sup> ± 2,06	44,72 <sup>c</sup> ±3,55	43,61 <sup>c</sup> ±4,11
	70	13,88 <sup>bc</sup> ± 1,94	29,16 <sup>d</sup> ±5,46	33,33 <sup>d</sup> ±4,08
	80	7,63 <sup>cd</sup> ± 2,64	23,61 <sup>d</sup> ±3,8	15,55 <sup>e</sup> ±5,61
	90	5,13 <sup>d</sup> ± 5,13	8,05 <sup>e</sup> ±4,67	0 <sup>f</sup>
	100	0 <sup>d</sup>	0 <sup>e</sup>	0 <sup>f</sup>

Means followed by same letters indicate non-significant differences between concentrations at p<0,05 (Duncan's test)

**Table 2.** Germination index, expressed as% of control, of *lactuca sativa*, *raphanus raphanistrum* and *phalaris truncata* seeds, in presence of *Rosa canina* leaves and stems aqueous extracts.

	Extract Concentration (g/l)	Target species		
		<i>Lactuca sativa</i>	<i>Raphanus raphanistrum</i>	<i>Phalaris truncata</i>
Leaves extract	10	77,14 <sup>a</sup> ±2,26	12,55 <sup>a</sup> ±21,74	25,2 <sup>bcd</sup> ±9,34
	20	49,64 <sup>b</sup> ±13,75	14,34 <sup>a</sup> ±6,27	57,71 <sup>a</sup> ±13,95
	30	33,3 <sup>c</sup> ±6,6	7,94 <sup>a</sup> ±7,83	29,6 <sup>bc</sup> ±14,37
	40	10,6 <sup>d</sup> ±7,48	0 <sup>a</sup>	24,98 <sup>bcd</sup> ±17,3
	50	11,36 <sup>def</sup> ±3,37	0 <sup>a</sup>	37,58 <sup>b</sup> ±13,85
	60	15,22 <sup>de</sup> ±3,87	0 <sup>a</sup>	23,58 <sup>bcd</sup> ±6,28
	70	10,61 <sup>efg</sup> ±3,9	0 <sup>a</sup>	11,45 <sup>cde</sup> ±8,28
	80	2,3 <sup>fg</sup> ±2,35	0 <sup>a</sup>	9,56 <sup>cde</sup> ±8,45
	90	0,55 <sup>fg</sup> ±0,7	0 <sup>a</sup>	5,3 <sup>de</sup> ±3,75
	100	0,24 <sup>g</sup> ±0,4	0 <sup>a</sup>	0 <sup>e</sup>
Stems extract	10	85,85 <sup>a</sup> ±3,33	85,97 <sup>a</sup> ±8,41	89,73 <sup>a</sup> ±15,24
	20	77,35 <sup>a</sup> ±12,04	86,92 <sup>a</sup> ±1,87	89,16 <sup>a</sup> ±3,48
	30	77,47 <sup>a</sup> ±12,55	76,25 <sup>a</sup> ±9,24	82,38 <sup>a</sup> ±7,52
	40	27,9 <sup>a</sup> ±10,05	74,19 <sup>ab</sup> ±12,32	77,56 <sup>a</sup> ±11,97
	50	50,54 <sup>b</sup> ±5,06	62,03 <sup>b</sup> ±12,36	51,4 <sup>b</sup> ±7,69
	60	13,76 <sup>c</sup> ±1,96	44,23 <sup>c</sup> ±9,53	39,65 <sup>b</sup> ±8,04
	70	6,92 <sup>cd</sup> ±1,79	24,5 <sup>d</sup> ±5,13	26,31 <sup>c</sup> ±4,45
	80	1,95 <sup>d</sup> ±0,76	14,93 <sup>de</sup> ±2,09	10,37 <sup>d</sup> ±2,26
	90	0,25 <sup>d</sup> ±1,47	6,03 <sup>ef</sup> ±4,01	0 <sup>d</sup>
	100	0 <sup>d</sup>	0 <sup>f</sup>	0 <sup>d</sup>

Means followed by same letters indicate non-significant differences between concentrations at p<0,05 (Duncan's test)

We noticed that the germination of the seeds is delayed or it stops in an advanced stage or it does not occur. Kruse *et al.* (2000) showed that when susceptible plants are exposed to allelochemicals, seed germination is delayed. With some seeds,

germination stops in the seed swelling stage. For others, germination stops at the beginning of the appearance of the radicle. For the two target species, the germination index (GI) was more affected compared to the germination rate.

It is well established that the germination index is the most sensitive indicator of allelopathic effects that may occur during the germination process (Ahmed and Wardle, 1994). Even if the rate of germination is not greatly affected, the delay in the germination of a species can have important biological and ecological consequences, as long as it can affect the ability of seedlings to establish under natural conditions (Chaves *et al.*, 2001) but also their competitive power for water and resources (Xingxinag *et al.*, 2009). Indeed, plants that germinate with reduced germination indices are generally small in size and poorly competent for environmental resources (Fallah Touzi and Baki, 2012).

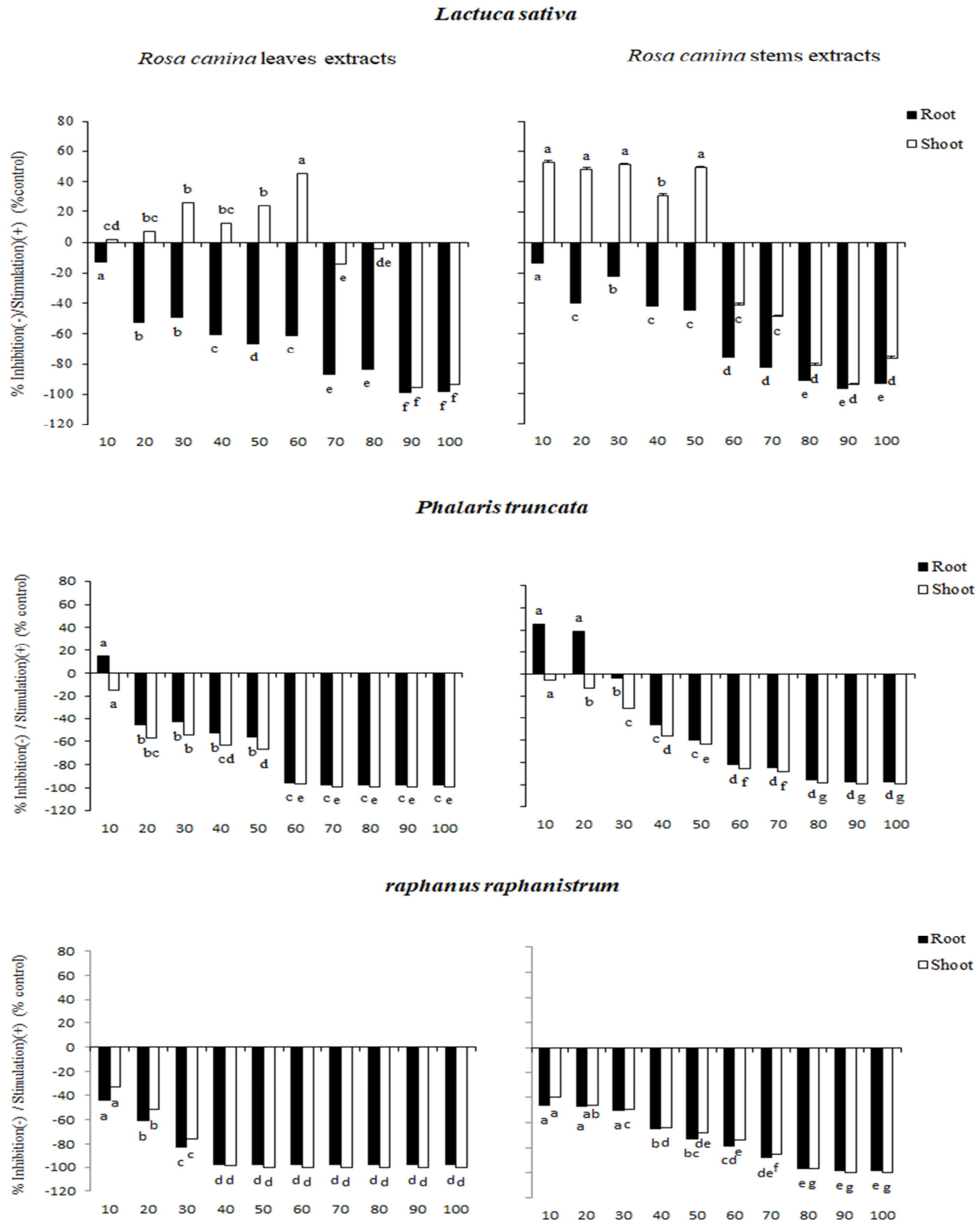
The inhibitions of germination rate and speed recorded in the present study increased with the concentration of the extracts. Moreover, the target species did not show the same sensitivity to the different extracts. In fact, *R. raphanistrum* was the most sensitive, particularly to *R. canina* leaf extracts. Also, *L. sativa* and *Ph. truncata* showed remarkable sensitivity to *R. canina* leaf extracts but in highest concentrations. This result underlines the specificity of allelochemicals, which can be selective in their actions and the target plants can be selective in their responses (Seigler, 1996) and it is in this selectivity that their exploitation must be sought for the control of weeds in the cultures.

On the other hand, our results showed that, in addition to the concentration effect which is a pretentious factor in the sensitivity of target species to allelochemicals, it is the organ effect which is also a factor of the same importance since we found that the leaf extracts of wild rose possess a high and remarkable allelopathic potential compared to its stems extracts, and this results from the concentration of allelochemicals in the leaves in higher quantities than at the stems. These results are confirmed by Djurdjevic (2004) who tested phenolic acids from aqueous extracts of *Allium ursinum* leaves and bulbs on several plants. They stated that bulb extract has a stronger inhibiting effect on germination than leaf extracts, so the content of allelopathic substances plays an important role in inhibition of germination. Indeed Hao *et al.* (2007) demonstrated

that the allelopathic potential can change with different parts of the plant, by the concentration of extracts and the fractions extracted. Ben Hammouda *et al.* (2001), also, demonstrated that barley leaf extract substances have a greater effect than its root extracts. Different plant tissues such as leaf, stem, root, seed and even flowers, can release certain amounts of allelochemicals into the environment (Chung *et al.*, 2000). All these studies as well as the present study showed that the sensitivity of the target species varied with extract concentration, organ nature and the species itself.

#### *Effect of Rosa canina aqueous extracts on the growth of target species plants*

*Rosa canina* aqueous extracts induced important reduction of roots and shoots lengths of all target species which varied with concentrations and organs (Fig. 2). Dog rose leaves and stems extracts induced reduction of lettuce root lengths at all concentrations which reached, respectively, means of 80% at 70g/l and 100% at 90g/l. Shoot lengths of lettuce were stimulated at concentrations between 10 and 50g/l under effect of leaves extracts (reached 49% at 50g/l) and between 10 and 60g/l under effect of stems extracts (reached 45% at 60g/l). Beyond these concentrations, shoots lengths were reduced and inhibitions reached 100% at 90g/l in presence of both extracts. For both weeds plants, dog rose extracts induced important reductions in roots and shoots lengths excepted roots of *phalaris truncata* irrigated with leaves extracts at 10g/l and with stems extracts at concentrations of 10g/l and 20g/l, where weak stimulations were noted (15% and 42% on average, respectively). Reductions of roots and shoots lengths of *phalaris truncata* reached 100% from concentrations of 60g/l and 80g/l of leaves and stems rose dog extracts, respectively. The corresponding values obtained for *raphanus raphanistrum* were 40g/l and 80g/l, respectively (Fig. 2). *Raphanus raphanistrum* proved to be more sensitive than *phalaris truncata*. Inhibitions of its roots and shoots lengths reached 80% and 50%, on average, from concentration of 30g/l of leaves and shoots of *rosa canina* extracts, respectively. At same concentration, corresponding values obtained for *phalaris truncata* were 48% and 17%, respectively.



**Fig. 2.** Inhibition (-) or stimulation (+) of roots and shoots lengths of *lactuca sativa*, *phalaris truncata* and *raphanus raphanistrum* plants irrigated with aqueous extracts (at different concentrations) of leaves and stems of *Rosa canina*, 7 days after germination. Means followed by same letters indicate non-significant differences between concentrations at  $p < 0,05$  (Duncan's test).

For growth tests, taking the initial growth of the species in the control as a reference point, the chemicals released from *R. canina* extracts had a negative effect on the growth of seedlings of all target

species. In most of the studied cases, both the length of the underground and above-ground organs was shorter than those in the control. The assessment of plant growth and development by determining the

length of the roots and the aboveground part of seedlings is more sensitive biotest than seed germination (Tatoj *et al.*, 2022).

In the presence of dog rose stems and leaves extracts at low concentrations, a stimulating effect on *Ph. truncata* roots and *L. sativa* shoots was noted. It is commonly believed in the subject literature that allelochemical substances released from various organs may inhibit or stimulate plant growth and development (Khanh *et al.*, 2008). Allelopathic compounds released in the aqueous extracts of *R. canina* leaves and stems limited the growth of seedlings to the greatest extent. Growth inhibitions were more pronounced in the presence of dog rose leaves extracts compared with stems extracts. Similar results were noted with *Festuca rubra* and *Raphanus sativus* seedlings watered with *Rosa gorenkensis* Besser organ extracts (Tatoj *et al.*, 2022). The authors noted that the greatest inhibitions were caused by extracts, prepared from leaves of *R. gorenkensis* Besser. Gniadzowska *et al.* (2004) said that the leaves are the organs in which the greatest amount of allelochemical compounds with a broad qualitative spectrum are accumulated. In our study, root growth of all target species were more affected than shoot growth. The roots of germinating seeds are more sensitive to allelochemical compounds than shoots, due to the first contact with the external environment after the seed coat has burst (Mazur, 2019). Growth reductions were more pronounced with higher concentrations of extracts. Mozdzen *et al.* (2022) also announced that the length of roots of *Festuca rubra* seedlings grown on aqueous extracts from the roots and stalks of *Rosa blanda* were significantly shorter than in the control. The inhibition of elongation growth was observed with the increase in the concentration of extracts (Mozdzen *et al.*, 2022).

In the case of *R. canina*, it is difficult to determine which chemical compounds had inhibitory effects on seed germination and seedling growth of target species, as there are no detailed data available on this subject. In fact, many reports describe *R. canina* hips (Abdallah, 2011; Lesjak *et al.*, 2016; Oguntibeju, 2018), flowers (Ben Jemaa *et al.*, 2017) and twigs and

leaves (Kubczak *et al.*, 2020) as an abundant source of antioxidants and correlated it with richness of this specie in phenolic acids and flavonoids (Kılıçgün and Altiner, 2010; Polumackanycz *et al.*, 2020; Tabaszewska and Najgebauer-Lejko, 2020). But few works treated herbicidal activities of dog rose parts extracts. Here we demonstrated that stems and leaves, can be considered potential sources of phenolic compounds, especially flavonoids. Probably, these compounds may be responsible for allelopathic property of this plant. Khang *et al.* (2016) evaluated the allelopathic potential of dehulled rice, rice, and hulls of rice on germination of weeds. The authors detected the presence of vanillin and vanillic acid (phenolic compounds), in root exudates of rice (donor plant) and in lettuce, barnyardgrass and radish (tested plants). They also demonstrated that most of the vanillin and vanillic acid treatments showed high inhibitory effects on germination rates and seedling growth of target species. Wu *et al.* (2002) showed that vanillic acid detected in wheat exudates was one of the most correlated factors in inhibiting ryegrass root length.

### Conclusion

In this study, we investigated extracts from dog rose leaves and stems. In general, the twice organs contained similarly amounts of total phenols and flavonoids whereas leaves extract was richer in flavonones/flavonols than stems extract. Both extracts caused reduction in germination rate and speed of seeds of target species and inhibited root and shoot length of its seedlings. Inhibitions were more pronounced in presence of leaves extracts which could be in relation with richness in flavonones/flavonols. The delay in germination of target species caused by allelochemicals contained in dog rose extracts can in turn be one of the solutions for controlling these weeds by shifting the stages of plant development with the main crop. Results allow concluding that stems and leaves of *R. canina* could present a new natural residual source of bioactive phytochemicals, to be employed as bioherbicides to control weeds. Further research into the chemistry of *Rosa canina* will identify and isolate allelochemicals responsible for this type of interaction. Also Further extensive characterisations (Insecticidal and nematicidal



activities) of leaf and stem phytochemical compositions are still required to better exploit this agrochemical waste in sustainable agriculture.

### Acknowledgements

The authors thank the High School of Agriculture of Mograne (ESAM), for their financial support.

### References

- Abdallah EM.** 2011. Plants: An alternative source for antimicrobials. *Journal of Applied Pharmaceutical Science* **1**, 16-20.
- Adedapo AA, Jimoh FO, Koduru S, Afolayan AJ, Masika PJ.** 2008. Antibacterial and antioxidant properties of the methanol extracts of the leaves and stems of *Calpurnia aurea*. *BMC. Complementary and Alternative Medicine* **8(53)**, 16-20.
- Ahmed M, Wardle DA.** 1994. Allelopathic potential of vegetative and flowering ragwort (*Senecio jacobaea* L.) plants against associated pasture species. *Plant and Soil* **164**, 61-68.
- Alapetite GP.** 1979. Flore De La Tunisie; Angiospermes–Dicotyledones; Apetales–Dialypetales. Programme Flore Et Végétation Tunisiennes Ed. Imprimerie Officielle de la République Tunisienne pp. 651.
- Ben Hammouda M, Ghorbal H, Kremer RJ, Oueslati O.** 2001. Allelopathic effects of barley extracts on germination and seedlings growth of bread and durum wheat. *Agronomie* **21**, 65-71.
- Ben Jemaa H, Ben Jemia A, Khelifi S, Ben Ahmed H, Ben Slama F, Benzarti A, Elati J, Aouidet A.** 2017. Antioxidant activity and A-Amylase inhibitory Potential of Rosa Canina L. *African Journal of Traditional, Complementary and Alternative Medicines* **14(2)**, 1-8.
- Chaves N, Sosa T, Escudero JC.** 2001. Plant growth inhibiting flavonoids in exudate of Cistus ladanifer and in associated soils. *Journal of Chemical Ecology* **27**, 623-631.
- Chiapuso G, Sanchez AM, Reigosa MJ, Gonzaez L, Pellissier F.** 1997. Do germination indices adequately reflect allelochemical effects on the germination process. *Journal of Chemical Ecology* **23**, 2445-2453.
- Chon SU, Jang HG, Kim DK, Kim YM, Boo HO, Kim YJ.** 2005. Allelopathic potential in lettuce (*Lactuca sativa* L.) plants. *Scientia Horticulturae* **106**, 309-317.
- Chung IM, Ahn JK, Kim JT, Kim CS.** 2000. Assessment of allelopathy potentiality and identification of allelopathic compounds on Korean local rice varieties. *Korean Journal of Crop Science* **45**, 44-49.
- Chung IM, Ahn JK, Yun SJ.** 2001. Assessment of allelopathic potential of barnyard grass (*Echinochloa crus-galli*) on rice (*Oryza sativa* L.) cultivars. *Crop Protection* **20**, 921-928.
- Cunja V, Mikulic-Petkovsek M, Stampar F, Schmitzer V.** 2014. Compound Identification of Selected Rose Species and Cultivars: An Insight to Petal and Leaf Phenolic Profiles. *Journal of American Society for Horticultural Science* **139**, 157-166.
- Dai J, Mumper RJ.** 2010. Plant phenolics: extraction: analysis and their antioxidant and anticancer properties. *Molecules* **15**, 7313-7352.
- Delabays N, Ançay A, Mermillod G.** 1998. Recherche d'espèces végétales à propriétés allélopathiques. *Revue Suisse De Viticulture Arboriculture Horticulture* **30**, 383-387.
- Djurdjevic L, Dinic A, Pavlovic P, Mitrovic M, Karadzic B, Tesevic V.** 2004. Allelopathic potential of *Allium ursinum* L. *Biochemical Systematics and Ecology* **32**, 533-544.
- Fallah Touzi A, Baki BB.** 2012. Allelopathic potential of *Brassica juncea* L. Czern. var. ensabi. *Pakistan Journal of Weed Science Research* **18**, 651-656.

- Fascella G, Angiolillo FD, Massimo M, Amenta M, Romeo FV, Rapisarda P, Ballistreri G.** 2019. Bioactive compounds and antioxidant activity of four rose hip species from spontaneous Sicilian flora. *Food Chemistry* **289**, 56-64.
- Ghazghazi H, Graca M, Hasnaoui B, Sebei H, Figueiredo AC, Pedro LG, Barroso LG.** 2010. Phenols, essential oils and carotenoids of *Rosa canina* from Tunisia and their antioxidant activities. *African Journal of Biotechnology* **9**, 2709-2716.
- Ghazghazi H, Aouadhi Ch, Hasnaoui B.** 2013. Activité antimicrobienne d'huile essentielle et d'extraits des feuilles de *Rosa canina*. *Revue F.S.B.* **XI**, 179-188.
- Gniazdowska A, Oracz K, Bogatek R.** 2004. Allelopatia- nowe interakcje oddziaływania pomiędzy roślinami (Allelopathy- new plant interactions). *Kosmos* **53(2)**, 207-217.
- Hao ZP, Wang Q, Christie P, Li XL.** 2007. Allelopathic potential of watermelon tissues and root exudates. *Scientia Horticultura* **112**, 315-320.
- Kılıçgün H, Altuner D.** 2010. Correlation between antioxidant effect mechanisms and polyphenol content of *Rosa canina*. *Pharmacognosy Magazine* **23(6)**, 238-241.
- Khang DT, Anh HL, Ha Pham TT, Phung Thi Tuyen.** 2016. Allelopathic Activity of Dehulled Rice and its Allelochemicals on Weed Germination. *International Letters of Natural Sciences* **58**, 1-10.
- Khanh TD, Xuan TD, Chung IM, Tawata S.** 2008. Allelochemicals of barnyardgrass-infested soil and their activities on crops and weeds. *Weed Biology and Management* **8(4)**, 267-275.
- Koczka N, Stefanovits-Bányai E, Ombódi A.** 2018. Total Polyphenol Content and Antioxidant Capacity of Rosehips of Some *Rosa* Species. *Medicines* **5**, 84.
- Kruse M, Strandberg M, Strandberg B.** 2000. Ecological effects of allelopathic plants. A review, Department of Terrestrial Ecology, Silkeborg, Denmark, Rep. No. 315.
- Kubczak M, Ainur B, Khassenova Bartosz Skalski, Sylwia Michlewska, Marzena Wielanek, Araylim N, Aralbayeva Maira K, Murzakhmetova, Maria amaraeva Maria Skłodowska, Maria Bryszewska, Maksim Ionov.** 2020. Bioactive Compounds and Antiradical Activity of the *Rosa canina* L. Leaf and Twig Extracts. *Agronomy* **10**, 1897; 1-20.
- Lesjak MM, Šibul FS, Anac GT, Beara IN, Mimica-dukic NM.** 2016. Comparative study of biological activities and phytochemical composition of two rose hips and their preserves: *Rosa canina* L. and *Rosa arvensis* Huds. *Food Chemistry* **192**, 907-914.
- Mazur A.** 2019. The role of seed coat in the germination and early stages of growth of bean (*Phaseolus vulgaris* L.) in the presence of chickweed (*Stellaria media* (L.) Vill.). *AUPC Studia Naturae* **4(1)**, 103-118.
- Oguntibeju OO.** 2018. Medicinal plants with anti-inflammatory activities from selected countries and regions of Africa. *Journal of Inflammation Research* **11**, 307-317.
- Ouerghemmi S, Sebei H, Siracusa L, Giuseppe R, Antonina S, Francesco C, Mariateresa C.** 2016. Comparative study of phenolic composition and antioxidant activity of leaf extracts from three wild *Rosa* species grown in different Tunisia regions: *Rosa canina* L., *Rosa moschata* Herrm. and *Rosa sempervirens* L. *Industrial Crops and Products* **94**, 167-177.
- Ouerghemmi S, Saija A, Siracusa L, Ruberto G, Dhaouadi K, Cimino F, Cristani M.** 2020. LC-DAD-ESI-MS and HPLC-DAD phytochemical investigation and in vitro antioxidant assessment of *Rosa* sp. stem pruning products from different northern areas in Tunisia. *Phytochemical Analysis* **31**, 98-111.

- Polumackanycz M, Kaszuba M, Konopacka A, Marzec-Wróblewska U, Wesolowski M, Waleron K, Bucinski A, Viapiana A.** 2020. Phenolic Composition and Biological Properties of Wild and Commercial Dog Rose Fruits and Leaves. *Molecules* **25(5272)**, 1-15.
- Quettier DC, Gressier B, Vasseur J, Dine T, Brunet C, Luyckx MC, Cayin JC, Bailleul F, Trotin F.** 2000. Phenolic compounds and antioxidant activities of buckwheat (*Fagopyrum esculentum* Moench) hulls and flour. *Journal of Ethnopharmacology* **72**, 35-42.
- Riffault L, Destandau E, Pasquier L, André P, Elfakir C.** 2014. Phytochemical analysis of *Rosa hybrida* cv. 'Jardin de Granville' by HPTLC, HPLC-DAD and HPLC-ESI-HRMS: Polyphenolic fingerprints of six plant organs. *Phytochemistry* **99**, 127-134.
- Seigler DS.** 1996. Chemistry and mechanisms of allelopathy interactions. *Agronomy Journal* **88**, 876-885.
- Singleton VL, Slinkard K.** 1977. Total phenol analysis: automation and comparison with manual methods. *American Journal of Enology and Viticulture* **28(1)**, 49-55.
- Sytar Oksana, Bruckova Klaudia, Hunkova Elena, Zivcak Marek, Konate Kiessoun, Brestic Marian.** 2015. The application of multiplex fluorimetric sensor for the analysis of flavonoids content in the medicinal herbs family Asteraceae, Lamiaceae, Rosaceae. *Biological Research* **48(5)**, 2-9.
- Tabaszewska M, Najgebauer-Lejko D.** 2020. The content of selected phytochemicals and in vitro antioxidant properties of rose hip (*Rosa canina* L.) tinctures. *NFS Journal* **21**, 50-56.
- Tatoj A, Mozdzen K, Barabaszkrasny B, Soltys Lelek A, Gruszka W, Zandi P.** 2022. Effect of *Rosa gorenkensis* Besser aqueous extracts on germination and early growth of native plant species. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca* **50(1)**, 1-20.
- Verma N, Shukla S.** 2015. Impact of various factors responsible for fluctuation in plant secondary metabolites. *Journal of Applied Research on Medicinal and Aromatic Plants* **2(4)**, 105-113.
- Wenzig EM, Widowitz U, Kunert O, Chrubasik S, Bucar F, Knauder E, Bauer R.** 2008. Phytochemical composition and in vitro pharmacological activity of two rose hip (*Rosa canina*) Preparations. *Phytomedicine* **15(10)**, 826-835.
- Wu H, Haig T, Pratley JE.** 2002. Biochemical basis for wheat seedling allelopathy on the suppression of annual ryegrass (*Lolium rigidum*), *Journal of Agriculture and Food Chemistry* **50**, 4567-4571.
- Xingxinag G, Li M, Zongjung G, Changsong L, Zuowen S.** 2009. Allelopathic effects of *Hemisterpa lyrata* on the germination and growth of wheat, sorghum, cucumber, rape and radish seeds. *Weed Biology and Management* **9**, 243-249.