SWITCHING FROM JUVENILE LEAVES TO PHYLLODE OF ACACIA SALIGNA (LABILL.) H.L.WENDL. PROVED BY THE CONTROL OF SEEDLINGS AND STUMP SPROUT LEAVES

SONDES STAMBOULI-ESSASSI¹, ICHRAK SAMMOUD-BEN REJEB¹, SADOK BOUZID¹ AND FETHIA HARZALLAH SKHIRI^{2*}

¹Bioresources, Biotechnology & Climate Change Laboratory, Department of Biology, Faculty of Sciences of Tunis, University of Tunis El Manar, Tunis, Tunisia

²Genetics, Bioresources: Integrative Biology & Valorization Laboratory (LR14-ES06), Department of Biological Sciences, High Institute of Biotechnoloy of Monastir, University of Monastir, Monastir, Tunisia *Corresponding author's email: fethiaprosopis@yahoo.fr

Abstract

This work investigates the foliage's changes reported on seedlings and regenerated sprouts of *Acacia saligna* (Labill.) H.L.Wendl. cultivated in Tunisia. First, a tracking of leaves development on 50 three-month old heteroblastic seedlings was performed. The transition from juvenile foliage to mature leaves occurs through four several stages. The first two leaves were pinnately compound and stand opposite each other (stage 1). The following initiated five leaves are alternate and bipinnately compound (stage 2). The eighth leaf corresponds to a transition form (stage 3). After three months of growth, from the ninth leaf, seedlings start the initiation of phyllodes (stage 4). Second, in order to better understand the leaves' chronological switching and to examine the influence of the cutting height on the sprouting ability and on the biomass production, a total of 60 adult trees of *A. saligna* were randomly selected and cut at heights of 10, 30 and 50 cm aboveground level (20 trees each). The results indicated that the 50 cm stump height alowed a maximum rejuvenation expressed by the highest number of sprouts and juvenile bipinnately compound leaves; reminding those that have been developed on seedlings. Also, six-month old sprouts promoted the best transition phenomenon rate, the longest and the broadest phyllodes that are more vigorous than those developed on seedlings and adult trees.

Key words: Acacia saligna, Biomass, Heteroblasty, Juvenile, Sprouts, Stage.

Introduction

The Leguminosae or Fabaceae family is the third largest Angiosperm family, with an estimated 727 genera and 20,000 species (Lewis *et al.*, 2005). It has been traditionally classified into three widely accepted subfamilies, i.e., Caesalpinioideae DC., Mimosoideae DC. and Faboideae DC. (Anon., 2013). Mimosoideae is the second largest legume subfamily, with ca. 3,271 species grouped in 83 genera (Lewis *et al.*, 2005), from which we are interested in *Acacia* Mill. genus that is the largest one, estimated at more than 1,350 species (Maslin & McDonald, 2004).

Acacia saligna (Labill.) H.L.Wendl. (= A. cyanophylla Lindl. and of Mimosa saligna Labill.) (Le Floc'h et al., 2010) is native to Western Australia and planted widely around the world with an estimated 300,000 ha (Midgely & Turnbull, 2003). It is a bushy shrub, 2 - 5 m tall, branched near the base into several stems, but but it can also form a small tree of 5-9 m high, with a short main stem (Midgely & Turnbull, 2003). Acacia saligna occurs at low altitudes and in different soil types, especially on poor and calcareous sandy soils (Midgely & Turnbull, 2003). Acacia saligna is identified as one of three priority multipurpose species for arid and semi-arid zones by FAO's Silvae Mediterranea Network (Midgely & Turnbull, 2003). It is used extensively for sand dune fixation and the fight against erosion (Langkamp, 1987). In addition to its rapid growth and its drought tolerance, it has a high productive potential in wood, firewood, fodder and charcoal (Midgley & Turnbull, 2003). A. saligna was introduced in North Africa,

where several plantation programs had been established both to conserve soils and to ensure fodder availability particularly in dry seasons (Anon., 2010). Sylvo-pastoral plantations based on this species have been carried out for several years in the northern and central regions of Tunisia corresponding to the ecological zone of this species (El Euch, 2000). In Tunisia, this species produces important biomass (woody and leafy) (Derbel *et al.*, 2009) and its plant material is used in the compost manufacture (Ammari *et al.*, 2003). Its wood has been also successfully processed into particle board (Michaelides, 1979).

Despite all these advantages, *A. saligna* is relatively short-lived; about 10-20 years (Whibley & Symon, 1992). In Tunisia, in dry conditions, it does not exceed 10 years (Le Floc'h, 1988). So, it requires several rejuvenation cuts for a large biomass production to meet the livestock needs (Nasr *et al.*, 2013). *A. saligna* regenerates well and fodder biomass production is optimized by regular, annual harvesting; benefitting from fertilization on infertile soils (Midgely & Turnbull, 2003).

In Tunisia, the knowledge of the conditions and methods of fodder trees and shrubs propagation has been considerably improved for several years, but much remains to be done to increase their ligneous and leafy biomass production. Their sustained productivity and management on rangelands remains a serious and major challenge. Fodder production and quality are promoted by cutting management (time of first cutting, cutting height and frequency). Previous researches focused especially on the improvements in the biomass productivity by appropriate coppicing periods. According to the literature, cutting performed during the dormant period of plants results in the minimum stump mortality and the maximum new sprouts number (Culvenor, 1993). Mid and late summer and autumn are the most favorable seasons of coppicing promoting the best capacity to regrowth for *Quercus ilex* L., *Arbutus unedo* L., *Eurya japonica* Thunb., *Ilex pedunculosa* Miq., *Lyonia ovalifolia* (Wall.) Drude, *Mimosa* L., *A. auriculiformis* A. Cunn. ex Benth., *A. mellifera* subsp. *dellnens* (Burch.) Brenan and *A. nilotica* (L.) Willd. ex Delile (Dupuy & N'guessan, 1991; Ducrey & Turrel, 1992; Strohbach, 1999; Figueiroa *et al.*, 2006; Konstandinidis *et al.*, 2006; Imanishi *et al.*, 2010).

In fact, the regrowth was the lowest when the cutting of *Acacia* species was done in the rainy season (between January and April).

On the other hand, in regard to the leaf morphology, the juvenile leaves of the Australian phyllodinous Acacias (e.g., A. koa A. Gray, A. melanoxylon R. Br. and A. saligna) are bipinnately compound ones, but adult leaves are reduced to broad flattened petiole (phyllode without leaflets) (Purseglove, 1968) that have higher potential as a source of stock fodder (Shanta et al., 2015). Moreover, according to Maslin et al., (2003), around 70 taxa maintain compound leaves during their life cycle, but the majority of species (> 900 taxa) develop phyllodes. The switching from compound to transitional leaves and so to phyllodes is heteroblastic in Acacia and its timing is species dependent. However, juvenile leaves can be retained in certain species (Brodribb & Hill, 1993). The homology of the Acacia phyllode has been debated by the morphologists and taxonomists in the last two centuries. It is unique and its structure, development and the genetic processes involved in its formation are not well understood (Gardner et al., 2008).

In this study, we carried out an experimental field rejuvenation of the Tunisian *A. saligna*, in the aim of analyzing the effects of the cutting height on the sprouting capacity and identifying the best cutting height to be adopted in order to promote resprouting (maximum percentage of sprouted stumps), allowing to produce an important plant biomass. We also tracked the switching from juvenile leaves to phyllode by controlling the early developmental stages of three-month old seedlings and the shoot leaves obtained from stumps in order to better understand the *A. saligna* heteroblasty.

Materials and Methods

Seed germination and seedling establishment: Mature pods were collected in July 2016 on vigorous *A. saligna* trees of 15 years about, reaching 5 to 6 m in height and planted in the area of the Faculty of Sciences of Tunis, Tunisia (36° 49' N and 10° 09' E). Average annual temperature and precipitation in this area were 18° C and 448 mm, respectively. Three hundred uniformly shaped and sized seeds were manually removed from about 40 pods and cleaned with tap water, followed by distilled water. Seeds were placed in a container covered with mosquito net, under a small running tap water during 24 hours. Thereafter, they were treated under laminar air flow hood with chemical sterilants. Seeds were dipped first in 5% commercial bleach solution (sodium hypochlorite, NaOCI) for 15 min, then treated with 0.1%

mercuric chloride (HgCl₂) for 10 min. The sterilized seeds were rinsed three times with sterile distilled water, then 15 mL of sterile distilled water was added onto three layers of Whatman sterilized filter paper, lined on the bottom of a sterile Petri dish (90 mm). Twenty five swollen seeds were sown in each Petri dish and kept in a climatic growth chamber temperature-controlled at 28°C (8 h obscurity/16 h light) and 65% relative humidity. According to Whibley & Symon (1992), high temperatures between 28 and 30°C are necessary to ensure a maximum germination rate of A. saligna seeds. It corresponds to the most favorable temperature for the development of this tree in its natural environment. The filter paper was always kept wet with distilled water and the Petri dishes were covered to prevent the loss of moisture by evaporation. Seeds were kept under observation till complete germination. It was recorded daily and was considered complete once the radicle protruded about 2 mm in length. Twelve replicates of 25 seeds (= 300) per dish were examined till no new germination was observed. The total germination rate was calculated after 15 days (number of germinated seeds relative to total number of seeds) \times 100.

Seedlings transplantation and acclimatization: We have proceeded to the transplantation of two-week old seedlings, to larger containers consisting of 6 cm diameter and 750 cm³ volume Jiffy-pots (one seedling per pot), placed in the same conditions, containing peat substrate. Six-week old plantlets, considered as an independent autotrophic organisms capable of photosynthesis were transferred in 16 cm diameter and 2000 cm³ volume pots (5 plantlets per pot with a total of 50 pots), filled with a mixture of vegetal soil (2/3) and fine sand (1/3) then placed under a greenhouse (23°C day/18°C night temperature). To keep the substrate moist, the pots were irrigated twice a week with 100 mm of tap water.

Early vegetative growth of seedlings: A tracking of seedlings, during three months of growth, highlighting the chronological appearance of juvenile leaves and phyllodes was conducted. A total of 50 three-month old plantlets was randomly selected and four growth parameters have been controlled; the number of juvenile bipinnate leaves per plantlet and of leaflets pairs per penne and the number of phyllodes per plantlet as well as their length \times width (cm).

Stumps regeneration: In order to examine the impact of stump height on sprouting, 60 adult trees of *A. saligna* having 3-4 m of height belonging to one diameter class (20-30 cm), growing in the same study area and under the same environmental conditions, were cut at three heights 10, 30 and 50 cm aboveground (20 trees each). Each stump has been painted with a remarkable color. Stumps have been cut by using an electric forest saw. The cutting has been realized till late September 2016; corresponding to the end of the dry season in Tunisia. Five vegetative re-growth parameters were identified. Three months after coppicing, we counted the number of young sprouts as well as juvenile bipinnate leaves per stump and leaflets pairs per penne. Six

months after coppicing, the number of phyllodes per stump and their length \times width (cm) were reported. The rate of the transition phenomenon (average number of developed phyllodes per stump/average number of bipinnate leaves per stump \times 100) was calculated. In addition, we measured the length \times width (cm) of 30 representative phyllode samples collected randomly from *A. saligna* adult trees in the same area. Then, we compared their vigor with the phyllodes' developed on stumps after coppicing to evaluate the coppicing impact on the plant biomass production. We continued to track stump sprouts growth for one year.

Statistical analyses: Data collected from experiments were subjected to analysis of variance (ANOVA) using SPSS 13.0 for Windows package (SPSS Inc., Chicago, IL, USA). All the measured variables were transformed using arcsin square-root (arcsin $\sqrt{\times}$) before ANOVA. Averages were separated at the 5% significance level by a least significant difference test (Student's test).

Results and Discussion

Morphogenetic tracking of the foliar developmental stages during three months of seedlings growth: We kept track of A. saligna seedling leaf development, in particular, the transitions from juvenile to mature foliage, during 3 months, starting from seed germination. Epigeal germination started, after 3 to 4 days of seeds sowing (Fig. 1A, Fig. 1B). The highest germination percentage of $84.33 \pm 8.08\%$ was recorded 15 days after. According to Alshahrani (2018), in Acacias, germination rate depending on length, width, weight of the seed, and on the seed coat thickness. The germination was followed by the hypocotyl elongation and the appearance of the two fleshy cotyledonary leaves, in opposite arrangement, providing a food source for the growing seedlings (Fig. 1B, Fig. 1C, Fig. 2). Cotyledons turned yellow and fell 9-10 days after germination. Foliar switching has occured through four morphological developmental stages (Fig. 1C, Fig. 1F, Fig. 2).



Fig. 1. Switching from juvenile leaves to phyllode for *Acacia saligna* during the first 3 months of seedling growth. (A): Mature pods and seeds; (B): Hypocotyl elongation and cotyledonary leaves appearance one week after germination; (C): Three-month-old seedling showing the juvenile leaves developmental stages (from F1 to F9); (D): Bipinnately compound leaves observed on seedling two-month aged; (E, F): Transitional form of the leaves with the beginning of limb atrophy and the petiole flattening observed on seedling aged two months and half; (G): Phyllodes development with complete disappearance of the limb observed on three-month aged seedling.



Fig. 2. Schematisation of the passage from juvenile leaves to phyllodes during 4 stages for the seedlings of *Acacia saligna* cultivated in Tunisia. **Stage 1:** Development of the first (F1) and second (F2) pinnate compound leaves (pinnate: pinnate form). **Stage 2:** Development of the third bipinnate compound leaf (F3) and successively until the seventh (F7). **Stage 3:** Development of the eight (F8) transitional bipinnate leaf attached to a flattened petiole. **Stage 4:** Development of the ninth (F9) modified leaf; the first phyllode, a flattened petiole with a complete disappearance of the limb. (cot.l.: cotyledonary leaves; pcl.: pinnately compound leaves; ph.: phyllode).

Stage 1: One-month photosynthetically active seedlings produced the first and the second true leaves (F1 and F2, respectively). They were two opposite pinnately compound leaves attached at the same node (Fig. 1C, Fig. 2 stage 1), corresponding to the pinnate: pinnate mode. Several authors reported that the Acacia leaf ontogeny development proceeded through multiple stages (Miller & Miller, 2011). According to Vassal (1981), the majority of the Australian Acacia species seedlings exhibited either a pinnate: pinnate or a pinnate: bipinnate mode (the most common) for the two first true leaves. After germination the first true leaf, was pinnately compound in all cases, but the second one was either pinnately or bipinnately compound, but, in the less common form, the first two leaves were pinnate and opposite to each other (pinnate: pinnate). Miller & Miller (2011) confirmed that for the plants with the pinnate:pinnate phyllotaxy, first and second leaves, opened simultaneously and were opposite in arrangement, and concerning 37 Acacias taxa, the number of leaflet pairs on the first leaf and on the second one was always identical, in an average of 2.5 leaflet pairs per leaf. Also, in a comprehensive

examination of seedling development in *Acacia* section *Pulchellae*, the pinnate:pinnate form was found in all taxa (Guinet *et al.*, 1980). *A. strongylophylla* F. Muell., *A. murrayana* F. Muell. ex. Benth. and *A. pachyacra* Maiden & Blakely had also the pinnate:pinnate phyllotaxis (Ariati *et al.*, 2006). However, other previous studies have revealed that *A. victoriae* Benth., *A. synchronicia* Maslin, *A. pryrifolia* DC. and *A. dempsteri* F. Muell., had pinnate:bipinnate development (Miller & Miller, 2011).

Stage 2: The five next initiated leaves were bipinnately compound (F 3-7), each one consisting of a slender petiole that beared two pairs of opposite leaflets. The leaves arrangement became alternate from the third leaf (F3) (a single leaf is attached at a node) with a gradually elongation of the petiole (Fig. 1C, Fig.1D, Fig. 2 stage 2). The two-month old seedlings formed the first seven leaves. Their average number per plantlet was 7.02 ± 1.60 . This number was still invariable after three months of growth. The average number of leaflet pairs per penne was 4.20 ± 0.99 with a variation of 4 to 5 pairs.

Our observations were in complete agreement with Maslin et al., (2003), explaining that a typical progression goes from pinnate to bipinnate leaves in the first few seedling nodes. According to Miller & Miller (2011), the subsequent leaves in most Acacia seedlings were alternate. The maintaining of bipinnately compound leaves in mature plants has occurred multiple times in Acacia (Miller et al., 2003). Acacia foliage is highly variable and all species exhibit heteroblastic leaf development. Heteroblasty is where a plant may exhibit two or more leaf types during development (Wells & Pigliucci, 2000; Gardner et al., 2008). Conspicuous forms of heteroblasty are present in several genera such as Eucalyptus L'Hér. and Acacia Mill. (Goebel, 1913). In Eucalyptus occidentalis Endl., heteroblasty (a sudden vegetative change in the leaf's anatomy and function) is under genetic control (Java et al., 2010). In certain species, juvenile leaves can be retained, which is the case of heterophylly (Brodribb & Hill, 1993).

Stage 3: The two and a half-month old seedlings formed the eighth leaf (F8) with a compound limb that was gradually atrophied and a petiole that began flattening and took up the appearance of a leaf blade. It was the transitional form (Fig. 1C, Fig.1E, Fig. 1F, Fig. 2 stage 3). Transitional leaves are unique by having both pinnately compound leaves and a flattened petiole (Forster & Bonser, 2009).

Stage 4: Three-month old seedlings developed the ninth leaf (F9) with a petiole progressively flattened in a vertical plane, into linear, glabrous and greenish first phyllode with a complete disappearance of the limb (Fig. 1C, Fig. 2 stage 4). It is important to note that the three-month old seedlings haven't developed any more juvenile bipinnate compound leaves. They initiated only variable number of phyllodes. Their average number per plantlet was 3.92 ± 0.75 and their average length × width was $9.98 \times 1.5 \pm 0.29$ cm (Fig. 1G).



Fig. 3. Regeneration of *Acacia saligna* stumps after severe coppicing. (A): Development of juvenile bipinnate alternate leaves; (B, C): Petiole enlargement and transitional leaf forms; (D, E): Phyllodes polymorphism highly developed sprouts (one- year aged); (F): Spaced nodes and vigorous phyllodes.

Table 1. Relationship between A. saligna stump heights and sprouting ability 3 and 6 months after coppicing. Results are means \pm standard error. The values within a column with different letters for each parameter are significantly different (p<0.05) (Student's multiple range test) (n = 20).

Stump height (cm)	3 months after coppicing			6 months after coppicing		
	Number of sprouts/ stump	Number of bipinnate leaves/ stump	Number of leaflets pairs/ penne	Number of developed phyllodes/ stump	Phyllodes length × width (cm)	Rate of the transition phenomenon (%)
10	$35.85\pm7.45a$	$43.70\pm7.85a$	4.15 ± 1.09a	$23.70\pm4.41a$	13.55 × 2.0 ± 1.41a	54.23
30	$42.50\pm 6.69b$	$54.45\pm8.86b$	5.47 ± 1.58a	$33.25\pm7.27b$	14.95 × 3.4 ± 1.61a	61.06
50	$53.50 \pm 8.45c$	$60.20\pm5.83c$	$6.75\pm0.64b$	$44.90 \pm 5.52 c$	$\begin{array}{c} 19.05\times4.5\\ \pm1.47b\end{array}$	74.48

According to the literature, numerous plant species developed distinct juvenile and adult leaf forms. Species of Acacia are a model for this phenomenon (Taiz & Zeiger, 2006). For Miller et al., (2003) and Murphy et al., (2010), the transition from compound leaves to phyllodes varies among species, with 69 species maintaining compound leaves and never developed phyllodes. According to Brodribb & Hill (1993), the switching from transitional leaves to phyllodes is also heteroblastic in most species. Gardner et al., (2008) explained that bipinnate leaves are followed, in most species, by the transition to phyllodes, which can reach 25 cm long (Wood & Morris, 2007). Of the currently described species of Acacia, approximately 70 have bipinnate compound leaves at maturity and more than 900 have phyllodes (Maslin et al., 2003). According to Forster &

Bonser (2009), *A. implexa* Benth. is a heteroblastic species that develops compound (juvenile), transitional and phyllode (adult) leaves that differ widely in form and function. Hansen & Steig (1993) explained that true leaves of *A. koa* are beneficial in early establishment for rapid growth and for shade tolerance, whereas, phyllodes are better adapted to drought. Phyllodes are generally considered to be better adapted to hot, dry and high light conditions, whereas compound leaves are more suited to cool, wet and low light (Brodribb & Hill, 1993).

Stumps sprouting potential: Severe coppicing of tree trunk; at 10, 30 and 50 cm heights (20 trees per each height), promotes different regeneration degrees. The obtained results expressed for the different parameters analyzed, are summarized in Table 1.

All stumps sprouted (15 days after coppicing) from latent or dormant buds. Regeneration potential of each tree stumps various according to the height of cut. We report a positive relation between the stump height and the number of sprouts per stump. The average number of sprouts was recorded 3 months after coppicing. No regeneration was observed after this date. The highest average number was obtained with 50 cm stump height (Table 1).

Several species of trees and shrubs vigorously grow after the entire cut. Thus, the pruning of certain trees is very important to stimulate the development of new sprouts (Smith et al., 2012). In fact, sprouting play an important role in the development of hardwood trees because sprouts can provide a regeneration source with rapid growth supported by the root system of the parent tree (Knapp et al., 2017). According to Borde (2011), plants should have the vigor and the resources to make an important regrowth after severe pruning. In fact, for Borges Junior et al. (2004) cutting of A. mearnsii De Wild. trees at 60 cm promoted a high regrowth rate; 90% of the stumps developing new sprouts. According to Smit (2003), the leaf dry mass production tended to be higher when the cutting height was increased from 5 to 45 cm for A. erubescens Welw. ex Oliv. and Combretum apiculatum subsp. apiculatum Sond. In West Africa, a recent study conducted by Orou Matilo Timothee Bio et al., (2017) on A. auriculiformis A.Cunn. ex Benth., Eucalyptus camaldulensis Dehnh., and Tectona grandis L.f. showed that the stump dimensions was another important parameter which also had a significant effect on resprouting. They demonstrated that the capacity to regenerate of E. camaldulensis and T. grandis increased both the increase of stump diameter (40-60 cm). In opposite, for A. auriculiformis A. Cunn. ex Benth., when the stump diameter was larger than 35 cm, the sprouts' death risk increased (negative correlation). So, authors related this ability to regrowth for stumps to the capacity of new buds to mobilize and use the stored nutrients. However, for Xue et al., (2013), both stump height and dimensions have an effect on the sprouting rate. In fact, the cutting of Quercus variabilis Blume, the trunk diameter was more than 15 cm, at more than 30 cm height, maximized stump sprouting.

In Tunisia, only two studies have been conducted on this subject. Ammari et al., (1996) have defined an adequate exploitation technique for foliar biomass production of A. saligna, planted in the Center of Tunisia and under a semi-arid bioclimate. They demonstrated that the sprouting ability was significantly related to two parameters; the stump age and height of cut. They showed that at a young age, the stump adapted well to the coppicing. In fact, they obtained the best cutting response with the three-year old stumps. Moreover, on stumps coppiced at 60 cm, they recorded the best sprouts vigor and biomass rate production. More recently, Laamouri et al., (2002) have predicted the biomass production of A. saligna plantations, located at the North West of Tunisia. Their experimentation was conducted on 4 years old stumps, that all have been coppiced at 20 cm aboveground. However, the stump diameter varies from 1.8 to 13 cm. They demonstrated that the biomass

production was increased from 0.65 to 35.75 kg of green matter per stump and confirmed the positive correlation between the stump diameter and the sprouting ability.

Juvenile leaves development: Severe coppicing of *A.* saligna tree trunks had induced, directly after stump sprouting, during the first three months, the appearance of juvenile bipinnate leaves with a fine petiole (Fig. 3A) reminiscent from F3 to F7 leaves observed on seedlings. Their average number per stump reached, three months after coppicing, 43.70 ± 7.85 (10 cm aboveground). This value has increased gradually with the stump height and reached 54.45 ± 8.86 (30 cm) and 60.20 ± 5.83 (50 cm) (Table 1). The number of leaflet pairs per penne was variable. Their highest average number (6.75 ± 0.64) was recorded at 50 cm of stump height.

Transitional stage and phyllodes development: The bipinnate compound leaves development was stopped three months after the coppicing. From this date, it was noted the appearance of the transitional form leaves was carried on until 6 months after the coppicing and the juvenile character of the leaves disappeared progressively. Their petiole flattened and widened, indicating an intermediate stage between the juvenile leaves and the phyllode (Fig. 3B, Fig. 3C). The rate of the transition phenomenon increased with the stump height (Table 1). Six months after the coppicing, petioles of all the transitional leaves were transformed into enlarged phyllodes. Those were glabrous, light to dark green colored showing an important polymorphism (Fig. 3D, Fig. 3E). Their number, length and width varied according to the stump height. The highest average values for those parameters were recorded at a stump height of 50 cm aboveground (Table 1).

We compared the size (length and width) of the phyllodes obtained after rejuvenation with those developed on adult trees. The later ones had a diversity of size; from 7.4 \times 1.5 cm to 12.2 \times 3.0 cm. Thus, it was confirmed that, after six months of coppicing, whatever their height, stumps produced phyllodes longer and wider than those of seedlings (9.98 \times 1.5 \pm 0.29 cm) and of adult trees. In fact, Maslin & McDonald (2004) confirmed that A. saligna was a potential crop species characterized by its capacity to produce large quantities of biomass. The ability of a tree to coppice is the response strategy used by woody plants to recover some of the biomass initially lost after intensive cutting (Kaschula et al., 2005). During the tracking of the vegetative regrowth of sprouts from 6 to 12 months, we noticed that the new shoots from stumps showed long internodes. Phyllodes having an important area were all in alternate disposition along vigorous ramifications (Fig. 3F). Twelve-month old, regenerated A. saligna stumps had highly new branched stems.

Conclusions

Foliar development of *A. saligna* seedlings occurs through four important stages. Leaves switched from pinnately compound (the two first leaves) to bipinnately compound ones (from the third to the seventh leaf), then to transitional form (eighth leaf) and finally to phyllodes

(ninth leaf). After 3 months of growth, seedlings only initiated phyllodes. The stump height cutting, at 50 cm aboveground, promoted maximum sprouting and rejuvenation, expressed by the development of a juvenile bipinnately compound leaves, and allowing the production of an important plant biomass. When they were six-month old, phyllodes were initiated wider and longer than those developed on adult trees and seedlings. After one year, sprouts became vigorously branched.

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(Received for publication 8 December 2018)