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RESEARCH ARTICLE

BIOCHEMICAL EVALUATION OF FIVE WILD FOOD PLANTS USED DURING WELDING PERIOD IN SOUTHERN MALI.

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Abstract

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The fight against malnutrition has become one of the major concerns of many countries today. The most exposed are infants, older subjects and pregnant women. In sub-Saharan Africa, and particularly in Mali, studies have shown that wild food plants could help to combat effectively the malnutrition. The goal of this work was to contribute to the valorization of these picking products through a biochemical assessment of five wild food plants used during the welding period in southern of Mali. The plant material consisted of the fruits of *C. myxa*, *C. pinnata*, *D. microcarpum* and *S. birrea* and tubers *R. splendens*. The reactions in the tube was used to characterize the chemical groups. The monosaccharides were assayed by gas chromatography (GC) and total proteins by the Kjeldhal method. Total lipids were extracted by using soxhlet and quantified by the gravimetric method. The mineral elements were quantified by Atomic Absorption Spectrophotometry. The raw energy was evaluated by the calorimetric method of total combustion. The results showed that the wild food plants studied are rich in secondary and primary metabolites, in mineral and energetic elements. The maximum protein, carbohydrate and total lipid levels were obtained respectively in *Raphionachme splendens* (11.29%), *Sclerocarya birrea* (87.20%) and *Detarium microcarpum* (1.32%). GPC revealed relatively high levels of simple sugars, especially glucose (74.76% in *S. birrea*), which is the most commonly used sugar. The richness of these plants in nutrients, secondary metabolites and minerals could contribute to the fight against malnutrition.

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Introduction:-

In Mali, malnutrition is a public health problem, as in most countries in sub-Saharan Africa. It is one of the major causes of morbidity and mortality in children under five. The situation deteriorated with the crisis of 2012. In 2013, the fith Demographic and Health Survey (EDS-V) reported a national prevalence of 12.7% global acute malnutrition (GAM) and 5.1% severe acute malnutrition (SAM). . According to the results of the same survey, chronic malnutrition (CM) remains a concern with a national prevalence of 38.3% and underweight (PI) of 25.5%. In 2015, the Multiple Indicator Cluster Survey (MICS) showed respective rates of 13.5% MAG, 3.4% MAS, 30.4% MC and 25.0% PI. The results of the ENSAN surveys (2017) showed that food insecurity affects chronically nearly a quarter (24%) of Malian households. Given the extent of acute malnutrition in the country, joint integrated management programs for severe and moderate acute malnutrition have been established under the auspices of the Malian Government through the Ministry of Health. These programs have been operational for several years and have helped save the lives of many thousands of Malian children. Despite all these efforts, the SMART Mali data (2018) showed that the national prevalence of global acute malnutrition is 10%, which corresponds to a serious nutritional situation according to the world health organization (WHO) classification.

In order to break the vicious circle of malnutrition in Mali, numerous studies have been carried out in favor of the involvement of wild plant species. Research has shown the importance of wild plant species in human nutrition especially during times of scarcity and this around the world and mainly in Africa. In several African countries, wild food plants are an important source of income for local populations (Ayessou et al., 2009, Thiambiano et al., 2012, Guindo et al., 2014; Diarra et al., 2019). They also occupy a privileged place in the diet of rural populations (Soloviev et al., 2004, Aké Assi, 2014, Sène et al., 2018, Makkalao et al., 2015, Diarra et al., 2016). Lapante (2009) has shown that harvesting and consumption of wild fruits in Zimbabwe peak when food from crops is scarce. In West Africa, wild plants are the only recourse of populations during periods of hunger and famine (Chastanet, 2010, Diarra et al., 2016). The southern regions of Mali (Kayes, Koulikoro, Sikasso and Segou) have a very diverse flora especially wild food plants that are used during the welding period. Despite this wealth, statistics on malnutrition are worrying in these regions. According to the results of SMART Mali (2018) by region, the nutritional situation is considered precarious in the regions of Kayes, Koulikoro, Sikasso with prevalences between 5% and 10% of MAG, serious in the region of Ségou whose prevalence of global acute malnutrition exceeds the alert threshold of 10%.
Surveys performed in these areas indicate that among the most popular wild food crops are: *Cordia myxa* L., *Cordyla pinnata* (A.Rich.) Milne-Redh., *Detarium microcarpum* G. and Perr., *Raphionachme splendens* subsp . bingeri (A.Chev.) Venter and *Sclerocarya birrea* (A. Rich). Hochst. (Diarra et al., 2016). However, work on these food plants in these areas is mainly limited to inventories and ethnobotanical studies (Kouyaté et al., 2009, Diarra et al., 2016). To our knowledge, very few in-depth studies of biochemical composition have been conducted on these wild food plants used during the lean season in Mali.

In view of this situation, an evaluation of the biochemical characteristics of these products is necessary in order to contribute to the identification of individuals with high nutritional potential. That is why we initiated this work with the aim for contributing to the achievement of food self-sufficiency and the fight against malnutrition by proposing a table of biochemical composition of five wild food plants consumed in Mali in times of scarcity. The objective of this work was to study the biochemical composition of five wild plants used during the welding period in the southern regions of Mali.

Material and Methods:-

Plant material:-

Based on the literature, we selected five (05) food plants because of deficient of data on their nutritional quality. These species are *Cordia myxa* L., *Cordyla pinnata (*A.Rich.) Milne-Redh., *Detarium microcarpum* G. and Perr., *Raphionachme splendens* subsp. bingeri (A.Chev.) Venter and *Sclerocarya birrea* (A. Rich). Hochst.

The plant material consisted of the fruits of C. myxa, C. pinnata, D. microcarpum and S. birrea and tubers R. splendens.

Methods:-

Phytochemical Screening:-

Tube reactions were used for phytochemical screening (Wélé et al., 2018, Konaré et al., 2019, Togola et al., 2019). The research for alkaloids was done using Mayer and Dragendorff reagents. The reagents of Mayer and Dragendorff

show the presence of alkaloids through whitish and orange precipitates respectively. To characterize the tannins, an aqueous solution of $FeCl_3$ at 1% gives a greenish or blue-blackish coloration in the presence of the latter. The differentiation of the tannins (catechics and gallic) was obtained by the Stiasny reagent. Obtaining a yellow precipitate indicates the presence of catechics tannins. After filtration, in the presence of sodium acetate and a few drops of a 1% ferric chloride ($FeCl_3$) solution, a blue-black color appears, indicating the presence of gallic tannins.
The reaction with cyanidine allowed to characterize the flavonoids. An orange-pink color indicates the presence of flavones and a purplish pink color gave the flavonones, and a red color gives the flavonols or flavononols, collected on a layer of iso-amyl alcohol supernatant. Leucoanthocyanins were characterized by the same reaction with cyanidine but without adding the magnesium chips. The development of a red or purplish color indicates the presence of leucoanthocyanins. With the Liebermann-Burchard reaction, the presence of sterols and triterpenes is indicated by the appearance at the contact surface of the two liquids of a brownish-red or violet ring, the supernatant layer becoming green or violet. The presence of carotenoids was detected by the appearance of a blue color then becoming red in the presence of a saturated solution of antimony trichloride ($SbCl_3$) in chloroform. The appearance or not of UV fluorescence at λ = 366 nm was used to search for coumarins. We used the reagents of Baljet, Kedde and Raymond-Marthoud which respectively give an orange, purplish red, violet coloring in the presence of cardiotonic glycosides. The saponosides have been characterized by the persistent foam test and the mucilages using ethanol which gives a fluffy precipitate.

Dry matter and ashes content:-
The dry matter and ashes (total ash, hydrochloric ash and sulfuric ash) contents of our samples were determined by the protocol of the African pharmacopoeia (Traoré et al., 2019).

Determination of biomolecules:-
Lipids were extracted by the Soxhlet system and quantified by the gravimetric method (Thiombiano et al., 2014). Protein levels were determined by the Kjeldahl method taken up by Konaré et al. (2019). The mineral elements were assayed by inductively coupled atomic absorption spectrophotometry of an induction plasma (AES-ICP) according to the protocol described by Nikiema in 2018. The monosaccharides were identified and assayed by gas chromatography according to the protocol described by Gaël (2005). Gas chromatography is a rapid analysis technique that, due to its high sensitivity and its ability to separate complex mixtures allows to qualitatively and quantitatively assess the composition of different carbohydrate extracts. The monosaccharides released after acid hydrolysis of the polysaccharide are then analyzed by GC using the derivation procedure. This procedure consists in reducing the monosaccharides to alditols with sodium borohydride followed by their acetylation with acetic anhydride in the presence of N-methylimidazole. The volatile products obtained are then separated on capillary columns and then detected by GPC equipped with a flame ionization detector (FID). The stationary phases used for the separation of carbohydrate derivatives are apolar and grafted with siloxane derivatives.

Determination of energy values:-
The raw energy was determined after total combustion of the samples using an IKA® calorimetric bomb (Boudouma, 2007).

Data analysis:-
One-way analysis of variance (provenance) was performed on the assayed parameters. The one-way analysis of variance (ANOVA) using the Fischer comparison test at the 0.05 significance level was used to separate the means. All analyzes were done with the Minitab software (version 18.1).

Results:-

Phytochemical Screening:-

The characterization results for the chemical groups are shown in **Table 1** below.

Table 1:- Phytochemical Screening of samples

Samples / Chemical Groups	*C. pinnata*	*C. myxa*	*D. microcarpum*	*R. splendens*	*S. birrea*
Coumarins	+	+	+	+	+
Carotenoids	-	+	-	+	+
Flavonoids	-	-	-	-	-

Alkaloids	-	-	-	-	-
Saponosides	-	-	-	-	-
Catechechic tanins	-	-	+	-	-
Gallic tanins	-	-	-	-	-
Mucilages	-	-	+	-	-
Sterols and triterpenes	-	-	-	-	-
Cardiotonic glycosides	+	+	+	-	+
Anthocyanes	-	-	-	-	-
Leuco-anthocyanins	-	-	+	-	+

Dry matter and ashes content:-

The data from the analysis of the different organs of the selected plants are shown in **table 2**.

Table 2:-Dry matter and ashes contents and energy values of the samples ash hydrochloric ash sulfuric

Sapmles	Contents (g/100g)				Energy values (Kcal/kg MS 105°)
	Dry matter	Total ash	Ash hydrochloric	Ash sulfuric	
C. myxa	31.57 ± 0.78^{c}	1.00 ± 0.04^{e}	0.30 ± 0.03^{d}	0.02^{c}	1282.60 ± 46.10^{c}
C. pinnata	27.35 ± 0.11^{d}	4.52 ± 0.10^{c}	1.50 ± 0.03^{b}	0.14 ± 0.01^{bc}	1053.70 ± 65.78^{d}
D. microcarpum	94.54 ± 1.33^{a}	2.49 ± 0.07^{d}	0.86 ± 0.04^{c}	0.06 ± 0.01^{bc}	4037.50 ± 85.56^{a}
R. splendens	93.20 ± 0.83^{b}	12.06 ± 0.17^{a}	4.01 ± 0.27^{a}	4.00 ± 0.13^{a}	3698.60 ± 66.75^{b}
S. birrea	14.66 ± 0.10^{e}	5.80 ± 0.04^{b}	1.48 ± 0.07^{b}	0.19 ± 0.01^{b}	552.30 ± 27.51^{e}

The **figure 1** shows the correlation existing between the dry matter content and the energy value of the plants analyzed.

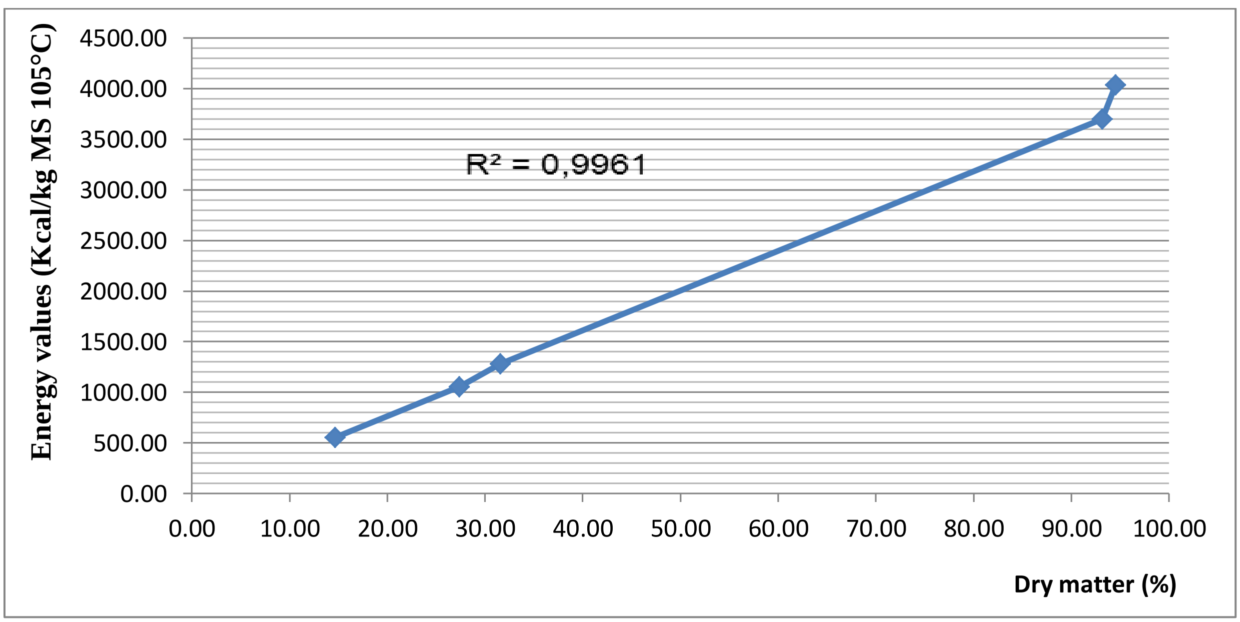

Figure 1:-Correlation between the dry matter and the energy value of our samples

Determination biomolecules contents:-

Contents of total of lipid, protein and carbohydrate:-

The results of the total lipid, protein and carbohydrate assay are shown in **table 3**.

Table 3:-Total protein, lipid and carbohydrate content of the samples (g / 100g)

Species	Total proteins (g/100g)	Total lipids (g/100g)	Total carbohydrate (g/100g)

C. myxa	1.93±0.13[b]	0.75±0.08[b]	84.80±3.28[a]
C. pinnata	1.68±0.10[bc]	0.20±0.04[c]	39.80±2.18[b]
D. microcarpum	2.12±0.20[b]	1.32±0.20[a]	77.30±3.45[a]
R. splendens	11.29±1.22[a]	0.51±0.10[b]	79.10±4.23[a]
S. birrea	0.43±0.06[c]	0.49±0.08[bc]	87.20±5.61[a]

Assays for monosaccharides:-

The sugars identified in the samples are shown in **table 4**.

Table 4:-Contents of monosaccharides (%) of our samples

Monosaccharides	Species				
	C. myxa	C. pinnata	D. microcarpum	R. splendens	S. birrea
Arabinose	-	-	-	6.62	-
Ribose	-	-	-	-	-
Rhamnose	-	-	-	-	-
Fucose	-	-	-	-	-
Xylose	-	-	-	-	-
Mannose	-	13.04	-	-	
Galactose	12.21	4.58	1.78	56.09	12.43
Glucose	72.44	8.58	42.79	16.34	74.76
Acide Glucuronique 1	0.11	-	32.78	-	-
Total carbohydrate	**84.80%**	**39.80%**	**77.30%**	**79.10%**	**87.20%**

Content of mineral:-

The analysis of eight minerals revealed relatively high levels of these elements in all the plants analyzed, as shown in **table 5**.

Table 5:-Determination of contents of mineral elements (mg / g of sample)

Plants	Na	K	Ca	Fe	Zn	Cu	Ni	Co
C. myxa	-	5.326	1.097	0.025	0.005	0.007	0.004	-
C. pinnata	0.020	3.001	0.241	0.012	0.007	0.001	0.002	-
D. microcarpum	0.022	3.850	0.932	0.012	0.010	0.002	-	0.001
R. splendens	0.016	1.559	0.238	0.042	-	0.002	0.002	0.002
S. birrea	-	1.666	0.379	0.005	-	0.003	0.003	0.001
Daily needs	**3-4 g**	**1-2g**	**0,8-1g**	**10-20mg**	**15mg**	**2-5mg**	**-**	**2 µg**

Discussion:-

Phytochemical screening showed our samples are very rich in secondary metabolites varied. The presence of carotenoids (which are the precursors of vitamin A) shows that our samples could be used in the fight against night vision disorders due to vitamin A deficiency. These carotenoids were found with *Cordia myxa*, *Raphionachme splendens* and *Sclerocarya birrea*. The alkaloids, saponins and anthracene derivatives are absent in the analyzed samples. Cyanogenic derivatives, which are substances, were also absent in all the samples analyzed. The mucilage found in some plants could improve the digestive transit allowing to fight constipation effectively.

Analysis of the data of table 2 shows that *R. splendens* tubers (93.20 ± 0.83%) and *D. microcarpum* fruits (94.54 ± 1.33%) are the richest in dry matter. These same species also have high energy values with 3698.60 ± 66.75 Kcal / kg DM for *R. splendens* and 4037.50 ± 85.56 Kcal / kg DM for *D. microcarpum*. This content obtained with *D. microcarpum* is greater than that obtained by Makalao et al. (2015) with fruits of the same species (92.75%).
A correlation study between the dry matter and energy content of the plants analyzed (figure 1) showed that the higher the content of the plants analyzed, the more raw energy it provides.

In general, the minimum and maximum ash contents were noted respectively in *C. myxa* and *R. splendens*. Total ash levels ranged from 1.00 ± 0.04% for *C. myxa* to 12.06 ± 0.17% for *R.* splendens. For the hydrochloric and sulfuric ash, 0.30 ± 0.03% and 0.02% were respectively obtained in C. myxa and 4.01 ± 0.27% and 4.00 ± 0.13% in *R.*

splendens. We recorded in *D. microcarpum* a content of 2.49 ± 0.07% which is lower than that obtained by Makalao et al. (2015) which was 3.51 ± 0.07%.

The ash content gives an idea of the content of inorganic elements, mainly mineral elements. Ash levels in condiments range from 1.63 g / 100 g to 8.53 g / 100 g in commonly consumed fruits (Oluyemi et al., 2006).
Thus, the concentrations of mineral elements varied from one plant to another. Some mineral elements were absent from the studied organs of our plants. It was sodium (Na) in the fruits of *C. myxa* and *S. birrea*; zinc (Zn) in the fruits of *R. splendens* and *S. birrea*; nickel (Ni) in the fruits of *D. microcarpum*; and cobalt (Co) in the fruits of *C. pinnata* and *C. myxa*.

All of our samples are highly potassium with contents ranging from 1.559 mg / g in C. myxa to 5.326 mg / g in *R. splendens*. These contents are relatively close to those obtained by Sène et al. (2018) who worked on the fruit pulp of S. birrea (2.53 - 3.85 mg / g). Then 1g of each sample would be enough to cover the recommended daily needs (1-2g). In view of their high potassium content, our studied species could be used in the treatment of heart failure. Iron levels in the samples varied from 0.005 mg / g to 0.025 mg / g respectively in the fruits of C. myxa and S. birrea. This favors possible exploitation in the fight against iron deficiency, which is very often the source of anemia. Sène et al. (2018) obtained in S. birrea fruit pulps a sodium content ranging from 1.45 mg / g to 1.55 mg / g, higher than our levels which ranged from 0.016 to 0.022 mg / g. On the other hand our calcium contents (from 0,238 to 1,097 mg / g) are superior to those obtained by Sène et al. (2018) that had levels between 0.521 and 0.944 mg / g.
The low levels of total ash in the organs of the food plants consumed are confirmed by relatively low concentrations of sodium, calcium and iron extracts. Sanogo et al. (2009) working on the traditional treatment of arterial hypertension and on the phytochemistry of two recipes used in the traditional treatment of arterial hypertension in Mali, have obtained the same relationship between total ashes and major minerals.

GPC revealed five types of monosaccharides in our samples namely arabinose, mannose, galactose, glucose and glucuronic acid 1. Monosaccharides are known for their energy intake and are increasingly involved in immunostimulant activity of plants.

For protein, the highest content was recorded in the fruits of *D. microcarpum* (2.12 ± 0.20%) and in the tubers of *R. splendens* (11.29 ± 1.22%). Note that Kouyaté et al. (2009) found 4.93% of protein in the fruit of *D. microcarpum*. Since we used the same method of determination, this difference can be explained by the fact that they soaked the fruits in distilled water for 15 hours whereas we did not soak our fruits before analysis. Sène et al. (2018) obtained with S. birrea fruit juices carbohydrate levels of 4.48% and protein of 0.45%; those are inferior to ours. By cons Makalao et al. (2015) had higher levels than ours with the fruits of *D. microcarpum* (6.12 ± 0.31 g / 100g MS).

Conclusion:-

This study showed that the five wild food plants consumed (fruits of *C. myxa*, *C. pinnata*, *D. microcarpum* and *S. birrea* and tubers *R. splendens*) during the welding period in the southern regions of Mali have various secondary metabolites. A significant correlation was found between the dry matter content of the analyzed plants and the raw energy. They have fairly high levels of total ash, mineral elements (sodium, potassium, calcium and iron). On the other hand, the very low levels of heavy metals (Co, Ni and Cu) show that these plants do not present significant risks for consumption. Their wealth in biomolecules (proteins, carbohydrates, lipids and monosaccharides) and energy show that they could contribute more to the fight against malnutrition especially during the lean season. However, it would be important to determine the composition of essential amino and fatty acids and vitamins. It would also be necessary to research and determine (if necessary) possible antioxidant, antihemolytic activities of these plants for a judicious operation.

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