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Research Article

Thermal Conversion of Pregeijerene into Geijerene in Essential Oil of *Chromolaena Odorata* L. King & Robinson (Asteraceae)

Esse Leon Wognin[1, *] , **Amenan Evelyne Tanoh**[2] , **Acafou Thierry Yapi**[2] ,
Kidjegbo Augustin Toure[1] , **Tomi Felix**[3] , **Zanahi Felix Tonzibo**[2]

[1]Preparatory School for the High Schools, Felix Houphouet Boigny National Polytechnic Institute, Yamoussoukro, Ivory Coast

[2]Training and Research Unit-Sciences of the Structures of Matter and Technology, Felix Houphouet Boigny University, Abidjan, Ivory Coast

[3]Chemistry and Biomass Team, University of Corsica, Ajaccio, France

Abstract

It is well-known that geijerene is mostly associated with pregeijerene. These terpenes are believed to be thermoconvertible by a type of sigmatropic transposition order supra or antarafaciale. Our contribution to the study of the molecular pair consisted on a combination of statistical and analytical approach to the chemical composition of *Chromolaena odorata* essential oil. A compilation of chemical compositions of the samples obtained by steam distillation and analyzed by GC, presented a variation of this binomial proportions in different countries. For Ivory Coast, the results of analysis of 71 samples showed a dominance of geijerene proportion. We carried out a two-stage sampling by chemical variability factors (effects of site and period). We chose, at random, three different samples we submitted separately to chromatographic analysis (GC) and spectroscopic analysis (^{13}C-NMR). It appears from this study that pregeijerene and geijerene were found in the essential oil with a relationship characterized by a polynomial degree 6 between these both molecules. Then during the chromatographic analysis, thermal conversion of pregeijerene into geijerene with the ratio 85.8% was observed. This result supported the idea of the coexistence of pregeijerene and geijerene in the essential oil of *Chromolaena odorata*. We intend to help clarify the kinetics and complete thermodynamics of this equilibrium.

Keywords

Chromolaena Odorata, Essential Oil Composition, Statistical Approach, Thermal Conversion, Geijerene, Pregeijerene

1. Introduction

It has been reported in literature that the geijerene is currently associated with the pregeijerene [1-8, 9-16]. In 1968, Jones and Sutherland have suggested that the pregeijerene isolated from *geijera parviflora* would be the precursor of geijerene [7]. In

addition, this precursor would result from of hedycaryol by the loss of an isopropylhydroxyl group [4, 9]. The exact stereochemistry of these two hydrocarbons was reported by Coggon et al., through X-ray crystallography. It is worth noting that, despite the presence of two asymmetric carbons, the geijerene is inactive [4, 13]. The thermal isomerization reaction is one of the most remarkable trans-trans-1, 5-cyclodecadieniques systems; it is a sigmatropic transposition [3, 3] supra or antarafaciale [4]. The pregeijerene would rearrange themselves into geijerene, then the geijerene would be an artifact resulting from pregeijerene by thermal transposition (Owolabi et al., 2010) [2]. Theoretical calculations undertaken by Owolabi et al. [2], have confirmed the hypothesis of the thermal conversion of pregeijerene to geijerene proposed by Jones and Sutherland and noted by AM Adio [2, 7, 12].

After these precursors, recently, several scientific works have been done to solve the problem of pregeijerene and geijerene. This work deals with the theoretical, spectroscopic and biological aspects of these both molecules. According to Owolabi, $\Delta_r G_0 = -4.16$ kcal obtained by calculations, would be compatible with almost complete conversion of pregeijerene to geijerene. Jared et al. [11], used the ^{13}C-NMR to elucidate the structures of these two molecules, basing their work on the numbering of Jones and Sutherland [2, 10, 7]. Furthermore, recent studies have shown that citrus roots infected with nematodes (*D. Abbreviatus)* product geijerene and pregeijerene [11]. The thermal conversion of pregeijerene into geijerene was studied. Taking account all stages of the production process, retention and chromatographic analysis of essential oil, only hypotheses have been put on the steps where the thermal conversion takes place: some have applied for the distillation step and other than the gas chromatography analysis in comparison with ^{13}C-NMR analysis [5, 7].

This study, consist to model and quantify the relationship between pregeijerene and geijerene in essential oil and their thermal conversion during gas chromatography.

2. Materials and Methods

2.1. Plant Material

Leaves from eight harvest localities were collected from May to November during three years (2007, 2008 and 2009) in

(1) Toumodi (6°55′ N, 5°03′ W) (9 Samples),
(2) Dougba (6°39'0" N, 5°7'60" W) (10 Samples),
(3) Yamoussoukro (6°49'0" N, 5°16'60" W) (17 Samples),
(4) Zambakro (6°43'60" N, 5°25'0" W) (9 Samples),
(5) Toumbokro (6°55'60" N, 5°30'0" W) (9 Samples),
(6) Attiegbakro (6°46'0" N, 5°7'0" W) (2 Samples),
(7) Tiebissou (7°9'47" N, 5°13'14" W) (6 Samples), and
(8) Tie-N'Diekro (7°25'16" N, 4°45'59" W) (9 Samples),

From 'Region du Belier' (6°49'60" N, 5°10'0" W) in the center of Ivory Coast (Figure 1) [17].
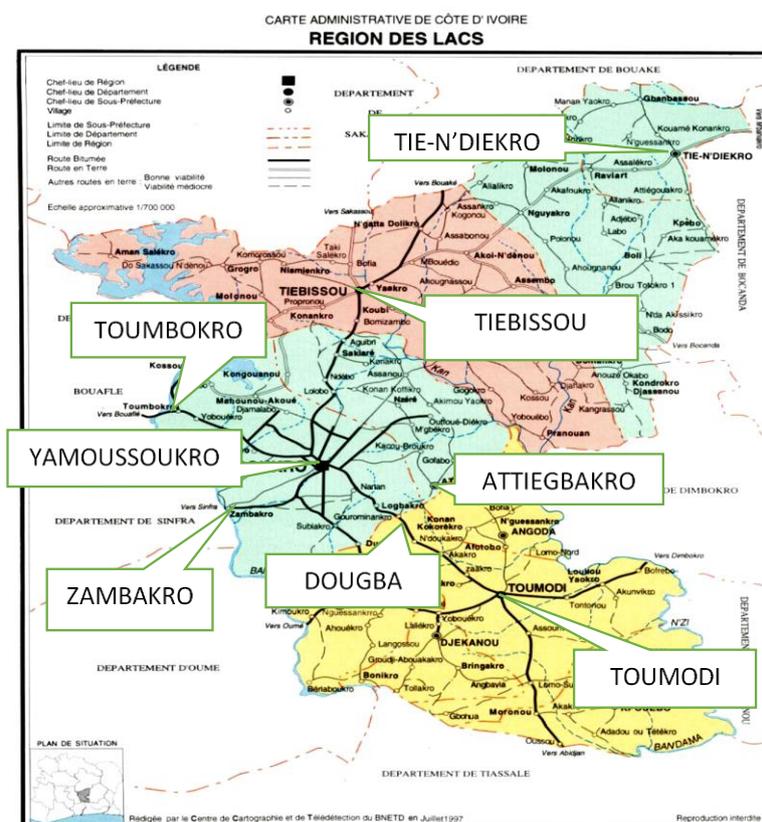
Figure 1. Sampling locations of Chromolaena odorata leaves from "Region du Belier" in Ivory Coast.

2.2. Essential-Oil Isolation

From May to November 2007 to 2009, fresh *Chromolaena odorata* leaves were collected at various sites in the "Belier Region" in the central part of Ivory Coast: Toumodi, Dougba, Yamoussoukro, Zambakro, Toumbokro, Attiegbakro, Tiebissou and Tie-N di ékro. For each site and each year, samples were repeated several times. These fresh leaves were placed in a pressure cooker containing water under a grid. The pressure cooker was connected to a Clevenger apparatus, and the device boiled for 03 hours. Condensation separated the essential oil from the aqueous phase. This is the hydrodistillation method. Extracted essential oils were dried over anhydrous sulphate magnesium and kept in the dark vials at +4 ℃ prior to analysis.

2.3. GC-FID Analysis

The oil samples were analyzed with a Perkin Elmer Clarus 500 apparatus equipped with FID and two fused-silica cap. columns (50mx0.22mm i.d., film thickness 0.25mm), an apolar BP-1 (polydimethylsiloxane) and a polar BP-20 (polyethylene glycol) column. The oven temperature was programmed rising from 60 to 220 ° at 2 ℃/min and then held isothermal at 220 ° for 20 min; injector temperature, 250 ℃; detector temperature, 250 ℃; carrier gas, He (0.8 ml/min); split ratio, 1/60. Retention indices (RIs) were determined relative to the tR of a series of n-alkanes with linear interpolation using the software Target Compounds from Perkin Elmer.

2.4. ^{13}C-NMR Analysis

The ^{13}C-NMR spectra were recorded with a Bruker AVANCE 400 Fourier Transform spectrometer operating at 100.63 MHz and equipped with a 5-mm probe, in CDCl$_3$, with all shifts referred to internal Me$_4$Si. The ^{13}C-NMR spectra of the oil samples were recorded with the following parameters: pulse width, 4 ms (flip angle 45 °); acquisition time, 2.7 s for 128 K data table with a spectral width of 25000 Hz (250 ppm); CPD (Composite Pulse Decoupling) mode decoupling; digital resolution, 0.183 Hz/pt. The number of accumulated scans was 2000–3000 for each sample, depending on the available amount of oil (when available, 45 –50 mg of essential oil in 0.5 ml of CDCl$_3$).

2.5. Identification of Components

The identification of the individual components was based
i) on the comparison of the GC retention indices (RIs) for the polar and apolar columns, determined relative to the retention time (tR) of a series of n-alkanes with linear interpolation, with those of reference compounds [18];
ii) for investigated samples, on the comparison of chemical shift values in the ^{13}C-NMR spectra of the essential oils with

those of reference spectra compiled in a laboratory-built library, following a computerized method developed in our laboratories, using home-made software. In the investigated samples, individual components were identified by ^{13}C-NMR at contents as low as 0.3–0.4%. The ^{13}C spectra were recorded with the following parameters: pulse angle 45 °; acquisition time = 2.7 s corresponding to an acquisition of 128 K with a spectral width (SW) of 24,000 Hz (approximately 240 ppm); relaxation time D1 = 0.1 s; digital resolution of 0.183 Hz/pt. To record the spectra of the essential oils (or chromatography fraction), a mass of 40 to 70 mg of essential oil or 9 to 50 mg of chromatography fraction was dissolved in 0.5 mL of CDCl3. The number of accumulations is between 2000 and 5000 for each recording. The signal data (FID) is multiplied before the Fourier transform by an exponential function (LB = 1.0 Hz). The following parameters were used for the spin echo experiments (JMOD programme) (5 mm probe): pulse duration = 8.6 μs (pulse angle 90°); repetition time between two experiments = 4.7 s; time [1/J(C,H)] calculated for a coupling constant J(C,H) = 140Hz: 7.1 ms. To record 2D NMR spectra involving heteronuclear correlations (HSQC, HMBC) or homonuclear correlations (COSY, NOESY), we used the pulse sequences defined by the Brücker programmes.

2.6. Quantification of Components

The relative contents of the oil constituents were expressed as percentage obtained by peak-area normalization without using correction factors.

2.7. Statistical Analysis

The correlation matrix was performed with Xlstat-Pro7.5.2 (Adinsoft, France) and the trendlines by excel of office 2013. The use of the solver of excel 2013 solved the equations. This solver has also helped to calculate the theoretical content of geijerene, using of the trend curves equations.

2.8. Determination of the Rate of Thermal Conversion from Pregeijerene into Geijerene

In chemistry, a quantitative or total reaction is a chemical reaction with an equilibrium constant very large in front of 1: $K \gg 1$. This is also equivalent to $\Delta_r G^0 < 0$ [19]. An incomplete reaction is one that cannot be complete. It has an associated equilibrium constant. It can also be a total reaction whose evolution has not yet had time to be completed. In this case, when the reaction is incomplete, the value of the rate of reaction may be less than the maximum value of the rate of reaction. To measure the evolution of the reaction in relation to the maximum evolution, we can therefore define a rate of reaction progress, a dimensionless quantity which represents the fraction of reactants that have reacted in relation to the

fraction that would have disappeared if the reaction had been complete. This rate of progress can be different from the rate of conversion α for each of the reactants. The conversion rate of reactant A is equal to the ratio of the number of moles of A remaining to the number of moles of A initially present [20]. We therefore write:

$$\alpha = \frac{n_{A transformed}}{n_{A initial}} = \frac{n_A^0 - n_A}{n_A^0} \tag{1}$$

We can also replace quantities of matter by concentrations.

The theoretical study of the thermoconversion of pregeijerene into geijerene meets all the above conditions [2]. This equilibrium being displaced in favor of geijerene, we have modeled the thermal conversion of pregeijerene to geijerene by a quantative reaction. The evolution table, which we have adopted, is presented as below:

$$\begin{array}{lccc} date & Pregeijerene & \rightarrow & Geijerene \\ t = 0 & [Preg]_{NMR} & & [Geij]_{NMR} \\ t \neq 0 & [Preg]_{GC} & & [Geij]_{GC} \end{array} \tag{2}$$
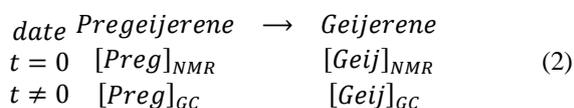
In this model, $[Preg]_{N.M.R}$ and $[Geij]_{N.M.R}$ represent respectively the initial concentrations of pregeijerene and geijerene. These are those obtained by ^{13}C-NMR, therefore, at low temperatures when thermoconversion is prohibited. Similarly, $[Preg]_{G.C}$ and $[Geij]_{G.C}$ represent the concentrations, at any given date, of pregeijerene and geijerene, therefore, at high temperatures when thermoconversion is permitted. These are those obtained by the G.C. The rate of conversion of pregeijerene is defined as follows, according to (1):

$$\alpha = \frac{[Pregeijerene]_{transformed}}{[Pregeijerene]_{initial}} = \frac{[Preg]_{N.M.R} - [Preg]_{G.C.}}{[Preg]_{N.M.R}} \tag{3}$$

3. Results

3.1. Presentation, Manipulation and Comparative Study of the Content of the Molecular Binomial in Samples

All the 71 samples were submitted to GC-FID analysis to determine retention index (RIs) on two columns of different polarity.

Among them, some samples, selected on the basis of their chromatographic profile, were also analyzed by ^{13}C-NMR. Therefore, the oil components were identified by comparison of their RIs and ^{13}C-NMR chemical shift values with those of authentic samples compiled in our laboratory-made libraries. For the study, only pregeijerene and geijerene contents were highlighted for eight locations of harvest: the predominant molecule geijerene is associated with pregeijerene as minority molecule (Table 1). Table 1 shows the proportions of the pregeijerene and geijerene molecules.

Table 2 compares the levels of the two molecules in essential oil samples from different countries.

Table 3 compares the percentages of the molecular pair after chromatographic analysis (GC) and after spectroscopic analysis (^{13}C-NMR).

Table 4 compares the decrease in pregeijerene with the increase in geijerene, for each validation sample.

3.2. Mathematical Modeling Between Content of Pregeijerene and Geijerene

All the trend line equations and their correlation coefficients for the effect of harvest location are summarised in Table 5.

Table 6 presents the model validation tests according to the two variability factors.

3.3. Determination of Conversion Rate of Pregeijerene into Geijerene

Table 7 shows the proportion of the thermal conversion of pregeijerene into geijerene to validate the sense of the chemical transformation observed during the GC analysis. It also shows the conversion rate of pregeijerene established clearly during the GC analysis.

4. Discussion

Table 1 shows the proportions of the pregeijerene and geijerene molecules. It can be seen that the proportion of geijerene is always higher than that of pregeijerene. During gas chromatography, some of the pregeijerene is converted to geijerene as a result of the temperature rise. This observation does not seem to be saw out elsewhere.

Table 1. Pregeijerene and geijerene contents from differents locations of Chromolaena odorata harvests.

| Compound name and class[a]	RI$_{apolar}$[b]	RI$_{polar}$[c]	Content (%)[d]							
harvesting sites			Toumodi	Dougba	Yamoussoukro	Zambakro	Toumbokro	Attiegbakro	Tiebissou	Tie-N'Diekro
number of samples			(9 sam-	(10 sam-	(17 sam-	(9 sam-	(9 sam-	(2 sam-	(6 sam-	(9 sam-

147

Compound name and class[a]	RI_{apolar}[b]	RI_{polar}[c]	Content (%)[d]							
			ples)	ples)	ples)	ples)	ples)	ples)	ples)	ples)
Geijerene	1134	1327	16.9±3.2	13.6±3.1	17.6±6.2	19.7±4.3	16.3±6.8	10.9±1.2	15.5±6.7	9.9±4.2
Pregeijerene	1276	1569	1.9±0.3	1.5±0.5	1.1±1.0	2.0±0.6	2.0±1.4	2.1±0.2	1.7±0.8	0.9±0.5

[a]Order of elution and contents determined on the apolar and polar column (BP-1 and BP-20); [b]RI_{apolar}: Retention index determined on the apolar column (BP-1); [c]RI_{polar}: Retention index determined on the polar column (BP-20); [d]Contents are given as mean±standard deviation.
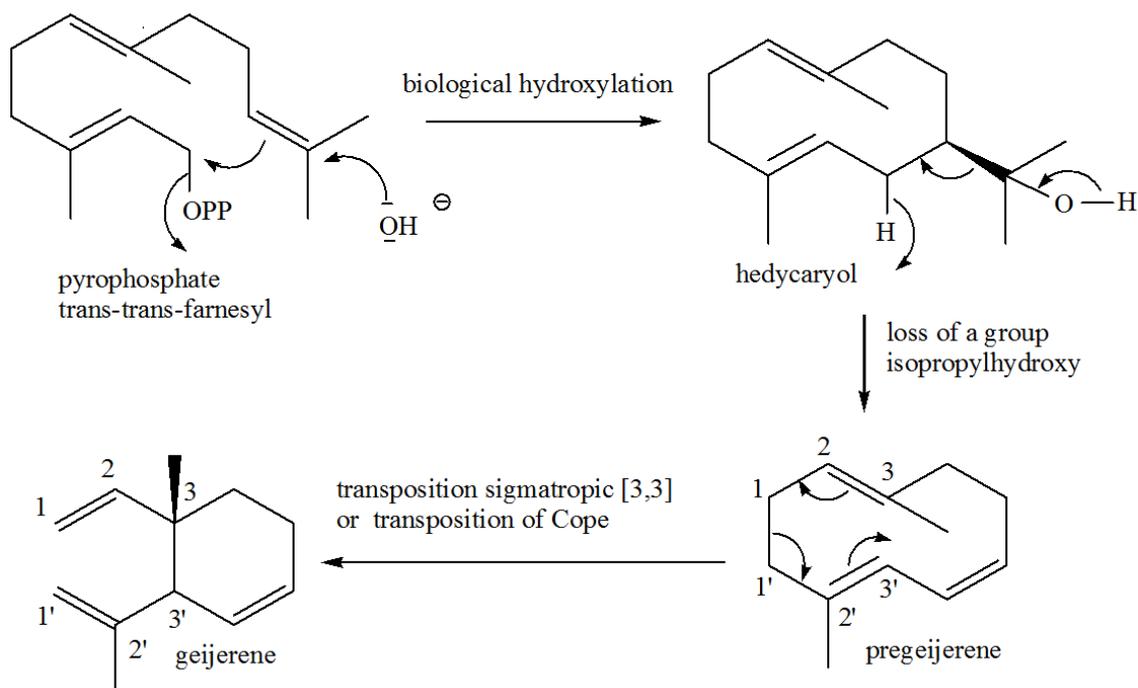
Scheme 1. *Biosynthesis of pregeijerene and Cope rearrangement between pregeijerene and geijerene [4].*
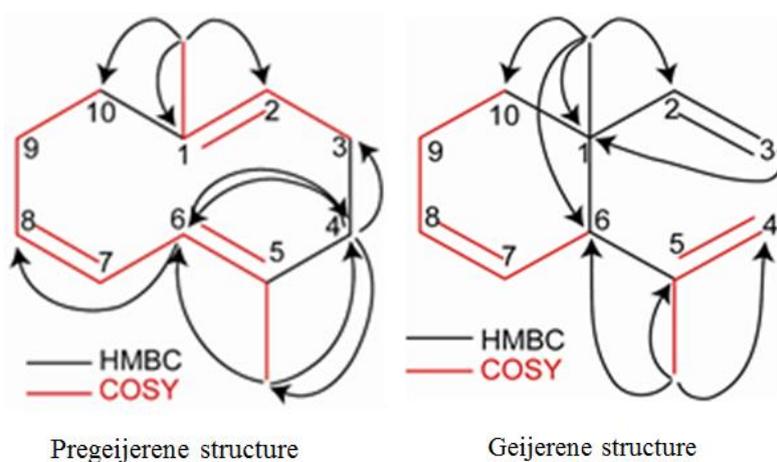
Scheme 2. *Pregeijerene and geijerene structures with Jones and Sutherland numbering [11].*

Table 2. *Average proportions of pregeijerene and geijerene in different countries.*

Countries	Benin_1	Benin_2	Togo_1	Togo_2	Nigeria	Congo_1 (forest)	Congo_2 (savannah)	India	Thaïland	Ivory Coast
Geijerene (y)	11.4	12.0	19.4	27.0	4.7	4.2	4.4	1.4	3.1	15.7
Pregeijerene (x)	19.9	14.6	10.5	0.1	2.8	14.8	33.3	14.2	17.6	1.5
Ratio (y/x)	0.5	0.8	1.8	270	1.7	0.3	0.1	0.1	0.2	10.5

x and y are the average percentages of molecules in the essential oil

Table 3. *Contents of pregeijerene and geijerene in the samples of validation.*

Samples	Components	GC		^{13}C-NMR	
		Content (%)	Ratio y/x	Content (%)	Ratio y/x
Toumbokro (2007)	Geijerene (y)	16.9	6.3	16.1	0.2
	Pregeijerene (x)	2.7		3.4	
Yamoussoukro (2008)	Geijerene (y)	19.3	9.7	13.7	0.6
	Pregeijerene (x)	2.0		7.6	
Tiebissou (2009)	Geijerene (y)	3.2	16	1.8	<0.2
	Pregeijerene (x)	0.2		< 0.3	

x: content of pregeijerene in the essential oil (%); y: content of geijerene in the essential oil (%)

Table 4. *Comparative study of the decrease in pregeijerene and the increase in geijerene for validation samples.*

Samples	Decrease in pregeijerene	Increase in geijerene
Toumbokro (2007)	16.1-2.7 = 13.4	16.9-3.4 = 13.5
Yamoussoukro (2008)	13.7-2.0 = 11.7	19.3-7.6 = 11.7
Tiebissou (2009)	1.8-0.2 = 1.6	>3.2-0.3 = 2.9

Table 5. *Model equations relating the proportions of pregeijerene and geijerene.*

Chemical variability factor	Dependent variables	Model equation	R^2
	Toumodi	$y = 10118x^6 - 108013x^5 + 477353x^4 - 10^6x^3 + 10^6x^2 - 10^6x + 291745$	0.9935
	Dougba	$y = 301.06x^6 - 2891.8x^5 + 11332x^4 - 23194x^3 + 26148x^2 - 15383x + 3695.1$	0.9968
	Yamoussoukro	$y = 36.113x^6 - 388.82x^5 + 1676.4x^4 - 3658.8x^3 + 4165.2x^2 - 2218.8x + 375.45$	0.9895
Site effect	Zambakro	$y = -220.69x^6 + 2375.4x^5 - 10433x^4 + 23840x^3 - 29716x^2 + 18991x - 4775.4$	0.9997
	Toumbokro	$y = -3.2201x^6 + 42.951x^5 - 217.58x^4 + 523.04x^3 - 605.67x^2 + 298.1x - 20.702$	0.6621
	Tiebissou	$y = -225.42x^5 + 1724.4x^4 - 4939.6x^3 + 6397.5x^2 - 3432x + 424.65$	1.0000
	Tié-N'Diékro	$y = 910.57x^6 - 4820.3x^5 + 9907.4x^4 - 10010x^3 + 5161.5x^2 - 1249.1x + 111.48$	0.9976

Chemical variability factor	Dependent variables	Model equation	R^2
	2007	$y = 79.807x^6 - 839.05x^5 + 3583.3x^4 - 7944.9x^3 + 9623.5x^2 - 6009.5x + 1512.5$	0.8573
Period effect	2008	$y = -16.767x^6 + 157.03x^5 - 588x^4 + 1115.8x^3 - 1115.8x^2 + 553.58x - 95.977$	0.9746
	2009	$y = -25.51x^6 + 153.29x^5 - 350.56x^4 + 379.95x^3 - 197.48x^2 + 52.704x - 1.5694$	0.9829
Organ effect	Feuille	$y = -7.10^8x^6 + 9.10^8x^5 - 4.10^8x^4 + 1.10^8x^3 - 10^7x^2 + 604570x + 6.7037$	0.9939

x: content of pregeijerene in the essential oil (%); y: content of geijerene in the essential oil (%)

Table 6. Results of the validation of polynomial models.

Factor of variability	Samples	Components	Real value of x and y (%)	Equation of the model	Calculated value of y (%)	Accuracy (%)
	2007	Geijerene	16.9	$y = 79.807x^6 - 839.05x^5 + 3583.3x^4 - 7944.9x^3 + 9623.5x^2 - 6009.5x + 1512.5$	18.31	8
		Pregeijerene	2.7			
Effetc of period	2008	Geijerene	19.3	$y = -16.767x^6 + 157.03x^5 - 588x^4 + 1115.8x^3 - 1115.8x^2 + 553.58x - 95.977$	18.3	5
		Pregeijerene	2.0			
	2009	Geijerene	3.2	$y = -25.51x^6 + 153.29x^5 - 350.56x^4 + 379.95x^3 - 197.48x^2 + 52.704x - 1.5694$	3.6	12.5
		Pregeijerene	0.2			

x: content of pregeijerene in the essential oil (%); y: content of geijerene in the essential oil (%)

Table 7. Conversion rate of the pregeijerene during the GC.

harvesting sites	Toumbokro	Yamoussoukro	Tiebissou
Average conversion rate α (%)	83.2	85.4	88.9
Average conversion rate (%)	85.8		

Indeed, a compilation of the chemical compositions of samples obtained by hydrodistillation and analysed by GC (Table 2) shows a variability in the proportions of this binomial in different countries. Average proportions were used for Ivory Coast.

Table 2 compares the levels of the two molecules in essential oil samples from different countries. Observation of this table shows that in Benin and Togo, the proportions of the two molecules are significant, while in Congo, India and Thailand, pregeijerene is higher than geijerene. However, our samples clearly show a dominance of the geijerene proportion, which is less noticeable in the Nigerian essential oil with relatively low proportions of both molecules. In addition, two-stage sampling per chemical variability factor (site effect and period effect) was carried out. Three different samples were selected at random and subjected separately to chromatographic analysis (GC) and spectroscopic analysis (^{13}C-NMR).

The results of the analyses, for the molecular pair, are shown in Table 3. This table compares the percentages of the molecular pair after chromatographic analysis (GC) and after spectroscopic analysis (^{13}C-NMR). This observation shows that the molecular pair is present in each of the samples, regardless of the method of analysis. Manipulation of this table resulted in Table 4, which compares the decrease in pregeijerene with the increase in geijerene, for each validation sample. We observe that, for each validation sample: - in GC, the percentage of geijerene is higher than that of pregeijerene; - in ^{13}C-NMR, the percentage of pregeijerene is higher than that of geijerene. In addition, for each validation sample (with the exception of Tiebissou where the exact value of geijerene could not be determined in NMR), the decrease in pregeijerene is equal to the increase in geijerene, when we change the values of these two molecules from NMR (cold) to GC (hot). We therefore suspect a thermoconversion of pregei-

jerene to geijerene.

We considered all the data from which a correlation between the pregeijerene and the geijerene is 0.581 (This value seems low, but it is significant, depending on the software used). Since the correlation between pregeijerene and geijerene levels is significant, we wanted to determine the mathematical model linking these levels from the trend curves. All the trend line equations and their correlation coefficients for the effect of harvest location are summarized in Table 5. The levels of the molecular pair in the three validation samples were used to validate the mathematical model obtained from the trend curves. Table 6 presents the model validation tests according to the two variability factors mentioned above. The calculated values of the geijerene percentage are of the same order of magnitude as the experimental percentages when the polynomial model is of degree 6. These models are validated: there is indeed a polynomial link of degree 6 between the pregeijerene and geijerene percentages.

Table 7 shows the proportion of thermal conversion of pregeijerene to geijerene in order to validate the direction of the chemical transformation observed in the GC analysis. It also shows the rate of conversion of pregeijerene clearly established in the GC analysis. It can be seen that at least 83% and at most 89% of the pregeijerene was converted to geijerene during the chromatographic analysis. These results make it possible to model, quantify and validate the thermal conversion, in part, of pregeijerene to geijerene during the chromatographic analysis. Overall, the trend function is a polynomial of degree 6 for all the harvesting sites, with the exception of Tiebissou. If we look at the correlation coefficients, we can see that pregeijerene and geijerene are strongly linked regardless of the harvesting site. However, the harvesting period allowed us to carry out our comparative study with a random selection of three validation samples (Table 3).

The thermal conversion of pregeijerene has been clearly established (scheme 1-2) [4, 7, 12]. The structures of pregeijerene and geijerene are known, as is the reaction mechanism for converting pregeijerene to geijerene (scheme 1-2) [4, 7, 10-12]. However, what is less well known is the stage at which the thermal conversion takes place, let alone the rate of conversion of pregeijerene to geijerene. Because of the high temperature at which the conversion takes place (120-170 °C), it is unlikely to be during steam distillation (which takes place at a temperature below 100 °C), as claimed by Bissangou and Ouamba [5].

5. Conclusions

Our contribution to the study of the thermal conversion of pregeijerene to geijerene in the chromatographic analysis is based on a statistical approach, using the essential oil of *Chromolaena odorata*. Therefore, we have carried out a two-stage sampling. The 71 samples were collected from eight sites during three successive years. These samples were analysed by GC-FID. A validation sample was submitted separately to GC and ^{13}C-NMR. The results show that, for the essential oil of *Chromolaena odorata* in Ivory Coast, during the chromatographic analysis in gas phase, 85.8% of pregeijerene is thermally converted to geijerene. This rate also translates the possibility of an equilibrium reaction for the duration of the rise in temperature during the chromatography. However, the comparative analysis of the results by GC and ^{13}C-NMR, reinforces our idea of the co-existence of the pregeijerene and the geijerene in *Chromolaena odorata*. This work has shown that geijerene, present in the chromatographic profile of Chromolaena odorata essential oil, is partly due to the thermal conversion of pregeijerene. So, geijerene is, in part, an artifact.

We intend to help clarify the kinetics and complete thermodynamics of this equilibrium.

Abbreviations

GC	Gas Chromatographic
^{13}C-NMR	Carbon-13 Nuclear Magnetic Resonance
FID	Flame Ionisation Detector
RI	Retention Index
CPD	Composite Pulse Decoupling

Author Contributions

Esse Leon Wognin: Conceptualization, Data curation, Formal Analysis, Investigation, Methodology, Software, Writing – original draft

Amenan Evelyne Tanoh: Conceptualization, Formal Analysis, Investigation, Methodology, Visualization, Writing – original draft

Acafou Thierry Yapi: Resources, Visualization

Kidjegbo Augustin Touré: Data curation, Formal Analysis, Methodology, Software, Supervision

Tomi Felix: Supervision

Zanahi Felix Tonzibo: Conceptualization, Supervision, Validation, Writing – original draft, Writing – review & editing

Conflicts of Interest

The authors declare no conflicts of interest.

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