Hemp Genotype Selection Coefficient to Improve Human Welfare Development

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Abstract*— The new concept of industrial hemp genetic selection examines the relationship levels of the Human Wellbeing Coefficient (K), and the variables grouped in three dimensions of promotion and protection human health with legality framework. The industrial hemp population was 47 hybrids. Eighteen samples were made for each hybrid during six weeks of evaluation. The Hewlett Packard 1100 HPLC equipment was used to analyze the fresh inflorescence samples during the 2019-2020 agricultural season in Montevideo, Uruguay. The hypothesis test, linear regression analysis, ANOVA, Decision Tree, PCA, HCA and Nearest Neighbor Analysis were used as statistics. The coefficient K has a statistically significant positive correlation with the variable Total Cannabidiols -TCBD (CC = ,978; p <.005). It also with the Total Tetrahydrocannabinols - TTHC variable (CC = ,936; p <.005) and with the Full Protection Projection -R1s variable (CC = ,979; p <.005). It is observed that the K coefficient is negatively correlated with the Coefficient of Promotion – R9. When K values increase, the probability of increasing the promotion and protection of human health is higher (R2=0,99; F(4,841) = 30.974.72; Sig = ,000). Hybrids whose K values >4.82 were selected. From the analysis of PCA and HCA, it has been possible to select the six best hybrids and were grouped into two categories. The first group of superior hybrids. This analysis confirmed the potency of the K Coefficient to select hybrids that contain the highest capacity to promote and protect human health.

Keywords*— Human wellbeing, hemp genetic material selection method, promotion and protection human health, cannabinoids , HPLC.

I. Introduction

The concept of the Human Well-being Coefficient (K) establishes a new method to select genetic material from the concentration of cannabinoids in the inflorescences of industrial hemp. The hypothesis is based on a relationship between the elements of promotion and protection of human health within the legal framework and the K coefficient. Along the same lines, the need arises to apply genetic selection to develop human well-being in the value chain. of industrial hemp. The present study complements the analysis of the concentration curve to identify the optimal harvest period of industrial hemp under controlled greenhouse conditions, which do not discriminate which group of individuals should be selected. This study works as a continuation for the development of the value chain.

The concepts of protection and promotion of human health and well-being are not new [1-4] and adaptation to protect health has existed as a form of evolution [5-8]. Even less new are the concepts of genetic selection of plants to generate human welfare [9-12]. For its part, the hemp industry requires compliance with the legal regulations imposed by each country so that this productive activity is carried out with good manufacturing practices [13-19]. In addition, the human brain has an endocannabinoid system that develops the nervous system and activity in the brain, which is useful for new therapies as it is interrelated with treatment status, disease progression, and severity of symptoms. [20-22]. However, what is important about the system is the ability to complement itself with the homeostasis of the brain and its physiology, as well as with regulatory enzymes and peripheral tissues [23-28]. In the case of the cannabinoids required by the endocannabinoid system of the human brain, these are produced in an agricultural field. Cannabinoids are present in the industrial hemp plant and their production determines the power of each one to provide well-being [29-33]. They are the cannabinoids of importance for the protection of health, being these associated with Cannabidiol (CBD) and provide protection related to sleep, pain, inflammation, anxiety and seizures [34-44]. In the case of Cannabinol (CBN), it is associated with promotion, since it refers to neuroprotection and appetite stimulation [45-50], as well as Cannabigerol (CBG) is related to health promotion, referring to digestive and antibacterial benefits [51-54].

Cannabidiol (CBD) protection is expressed within a known treatment range in all in vitro and in vivo assays performed and is associated with vibinilloid, serotonin, activated gamma, and adenosine receptors. Also, it may have receptor-independent bioactivity on cells [55-57]. In addition, CBD treated between 2.5 and 30 µM (30 min - 10 h) is useful for a wide variety of disorders [58-65]. Similarly, it increases the activity of the transport chain, mitochondrial biogenesis, the dynamics of the mitochondrial network and is involved in the differentiation of keratinocytes [66-71]. Cannabinol (CBN) is studied as neuronal protection, being a contribution to human health, as it is also understood as an attenuator of neurotoxicity and thus avoiding neurodegradation [72-74]. With this, its practical application is understood when generating the preservation of neuronal integrity and intervening on pathological proteins [75-76].

In addition, CBN stimulates hippocampal neurogenesis [77-78]. Also, CBN works effectively in the treatment of amyotrophic lateral sclerosis [79]. All these studies provide quantitative information on how CBN is used as a nutraceutical [80]. On the other hand, Cannabigerol (CBG) is currently labeled as the "mother of all cannabinoids" [81], and it is the one that decreases the presence of extrapyramidal symptoms (EPS) by reducing the expression of genes that regulate its production, which include gtfB, gtfC, gtfD, and ftf. This is important to allow antibacterial action [82]. Also, CBG has potential as an antimicrobial agent against pathogenic bacteria and fungi [83].

However, the legality of the productive activity of industrial hemp is limited by the Tetrahydrocannabinol (THC) content, according to the standards established by the countries [84-86]. Hence the importance of relating agricultural activity to the development of human well-being. This research aims to incorporate the elements of health

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promotion and protection within the legal framework that States provide to select genetic material and thus maximize human well-being. Therefore, the research questions are: Is it possible to find a coefficient to select industrial hemp genetic material that provides maximum human welfare? Is the Human Well-being Quotient related to aspects of promotion and protection of human health? And, is it possible to combine statistical tools that make it possible to separate groups of advanced genetic material that provide the greatest human well-being?

II. Methods and Materials

A. Factors, Dimentions and Variables Aspects

To find significance between the variables under study, SPSS v.25 was used. The population corresponds to the 47 asexual hybrids of industrial hemp, hypothesis testing, linear regression analysis, ANOVA, Tree, PCA and HCA were used as statistics. Three dimensions were designed for variable X "*Welfare factors with respect to legal framework*": (i) Health promotion (CBD), (ii) Health prevention (CBG & CBN); and (iii) Legal limit of the activity (THC); and one, for the variable Y "*Production of industrial hemp for the development of human well-being*": (i) Human Wellbeing Coefficient (*K*). Through the *K* Coefficient, a new model is proposed for the genetic selection of industrial hemp plants. The conceptual framework of the present investigation is reflected in Fig. 1 and Fig. 2. Table I, shows the variables for the promotion and prevention of human well-being within the legal framework of industrial hemp; and, the *K* Coefficient as also the dimension analyzed.

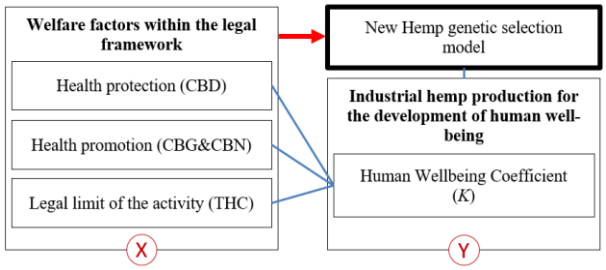

Fig. 1. Data analysis between variables. Source: Authors

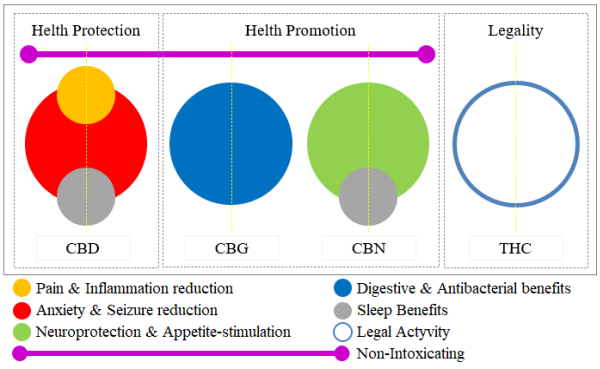

Fig. 2. Cannabinoids according to dimension analyzed. Source: Authors

Table I
Definition of Variables, Acronym and Source.

Dimension/Variable	Acronym	Source	Unit Measure
Protection			
Cannabidiol	CBD	AHP, 2014	%
Cannabidiolic acid	CBDa	AHP, 2014	%
Total Cannabidiols	TCBD	AHP, 2014	%
Full protection projection	R1s	Authors	θ_1
Promotion			
Cannabinol	CBN	AHP, 2014	%
Cannabigerol	CBG	AHP, 2014	%
Promotion coefficient	R9	Authors	ε
Legal Activities			
Tetrahydrocannabinol	THC	AHP, 2014	%
Tetrahydrocannabinolic acid	THCa	AHP, 2014	%
Total Tetrahydrocannabinols	TTHC	AHP, 2014	%
Full legality projection	R2s	Authors	δ_1
Dependent variable			
Human Wellbeing Coefficient	R17-P1	Authors	*K*

Source Authors

B. Human Wellbeing Coefficient (K)

The Human Wellbeing Coefficient (*K*) incorporates the components of health promotion and protection. It is designed to improve the genetic selection of industrial hemp plants.

The *K* coefficient identifies the plants that have the highest content of the 3 main groups of cannabinoids that benefit human health with a maximum of legality. It was calculated using (1):

$$\underbrace{K}_{\max\limits_{0<\delta_1<1}\ \delta_1} = 1/\left[\frac{\epsilon_1 + 1}{\epsilon_1 + \theta_1}\right] \qquad (1)$$

δ_1: Projection of full legality. Frame allowed by law. Expressed in % Total Tetrahydrocannabinol (TTHC). It is calculated using (2).
θ_1: Health protection for sleep problems, pain, inflammation, anxiety and seizures. Expressed in % of Total Cannabidiol (TCBD). It's calculated with using (3);
ε_1: Promotion coefficient. Expressed as the sum of Cannabinol (CBN) and Cannabigerol (CBG) per unit of Cannabigerol. t's calculated using (4).

Total Tetrahydrocannabinol data was normalized calculated with a linear regression respect the Tetrahydrocannabinolic acid content (R2=0,97; F(1,844) = 23.555,17; Sig= ,000). The coefficient K is calculated in plants containing Total Tetrahydrocannabinol <1%; With this, the plants that are below these levels were selected to be within the Peruvian legal framework (it can be modified to the Total Tetrahydrocannabinol content level of the countries where the *K* coefficient is required to be applied). It was calculated using (2):

$$\delta_1 = 0{,}035 + 0{,}913\ x_2 \qquad (2)$$

δ_1: % of Total Tetrahydrocannabinol
x_2: % of Tetrahydrocannabinolic acid.

Total Cannabidiol data was normalized calculated with a linear regression respect to the Cannabidiolic acid content (R2=0,99; F(1,844) = 82.749,97; Sig= ,000). It was calculated using (3):

$$\theta_1 = 0{,}054 + 0{,}912\ x_1 \qquad (3)$$

θ_1: % of Total Cannabidiol;
x_1: % of Cannabidiolic acid.

The promotion coefficient ε, was designed to identify the potential increase in total human health promotion per unit of neuronal promotion provided by industrial hemp. The coefficient is expressed in (4):

$$\varepsilon = (\beta_1 + \beta_2)/\beta_1 \qquad (4)$$

β_1: *Health promotion for digestive and antibacterial benefits. It is expressed in % of Cannabigerol -CBG;*
β_2: *Health promotion for neuroprotection benefits and appetite stimulation. It is expressed in % of Cannabinol – CBN.*

C. Sample collection and preparation

A total of 846 cannabis samples (fresh inflorescences) were collected from an authorized producer in Uruguay. Sample names were provided by licensed grower and different names may not necessarily represent different cultivars. The samples arrived in sealed and labelled craft paper bags. They were stored in a cold and dry environment facility prior to analysis. The raw material samples were treated in a humidity dryer for 24 h in a forced atmosphere at 35 °C. Next, the dried samples were crushed and homogenized. We proceeded to weigh 0,1 g of sample three times and place it in a 20 ml glass tube. The methanol was previously sonicated for 11 minutes at 60-65 °C and then 20 ml were used to add it to the homogenized and weighed sample; vortexed and passed through a 0.22 µm pore size nylon filter. Next, the filtrate was placed in a 2 ml vial that was then placed on the HPLC, starting the analytical technique.

D. HPLC Chromatographic Profiles.

Chromatographic methods are used to verify trace identification and the relative profusion of cannabinoids in the assayed sample [87-91]. For classification purposes, Total CBD corresponds to the potential that CBD in the acid state can be transformed into CBD without degrading. The same is true for Total THC. The samples were compared against a solution showing the main peak interrelated to the retention time [92]. The principles of standards identity, quality control and analysis are regulated by the Classification System of specific chemotypes for cannabis inflorescences [93].

E. HPLC systems and Analytic technique

For this study, a Hewlett Packard (HP) 1100 HPLC equipment was used, composed of the following elements: (i) Solvent degasser (G1322A), (ii) Tank pump (G1312A), (iii) WPALS (G1367A), (iv) Column compartment (G1316A), (v) HP 1100 series photodiode array detector (DAD) (G1315A) and (vi) C18 short column (Raptor ARC-18 2.7 µm 150 mm X 3.0 mm). The cannabinoid reference standards for CBN, CBG, CBD, Δ9-THC, CBDA; and, all standards were ≥98% pure. It was used: For the HPLC grade methanol extractions, for the mobile phase methanol and HPLC grade water; also, formic acid. All reagents were purchased from *Grupo Químico SRL* (Montevideo, Uruguay). The order of elution with a relative retention time per cannabinoid was as follows: CBDa = 1; CBG = 1.08; CBD = 1.14; CBN = 1.46; THC = 1.69; THCa = 1.96.

F. Method validation

For the validation of the method, the standards of the Association of Official Analytical Chemists were used [94]. To validate the reference standards, a PPM ≥ 950 was used as a coincidence factor and a PPI ≤ 1% as a standard deviation [95]. For the measurement of the calibration based on linearity ranges, concentration magnitudes of 5 to 1000 µg / mL were imputed for each cannabinoid standard used. Interday and intraday precision was calculated. The first, three days a week with 18 repetitions per day. The second, a validation sequence with 6 repetitions in a row (calculating the area of peaks expressed in mg / 100mg) [96-101].

G. Measured variables

The data collected included quantitative variables CBDa CBD, CBDa, TCBD, THCa, THC, TTHC, CBG and CBN. Forty-seven hybrids were evaluated for 42 days making a total of 846 HPLC analyzes of their inflorescences. The first week of measurement started when 50% of the plants contained inflorescence buds formed. Six consecutive weekly measurements were carried out on three different parts of each asexual hybrid totaling 18 repetitions for each hybrid evaluated.

H. Experimental Unit

The production of hemp inflorescences and their corresponding cannabinoid concentration evaluation were carried out under controlled conditions (greenhouse and laboratory). For the experiment, asexual reproduction was used as the propagation technique of the initial genetic material. Cuttings were collected from 5 plants of the cultivar Romalex registered in the National Registry of Cultivars (Uruguay). Each plant was grown in separate pots at a rate of 1.3 plants per m^2. Table 1, show the list of 47 evaluated hybrids, according to asexual collection material.

TABLE 1
LIST OF EVALUATED HYBRIDS, ACCORDING TO ASEXUAL COLLECTION MATERIAL

Mother code / Hybrid code				
PA-172-B	**PF-652-B**	**PG-411-A**	**PJ-062-A**	**PR-452-B**
1. 5583	11. 6215	19. 551	28. 5206	37. 545
2. 5585	12. 6298	20. 746	29. 5235	38. 634
3. 5595	13. 6302	21. 753	30. 5265	39. 1006
4. 5597	14. 7009	22. 5551	31. 5501	40. 1124
5. 5601	15. 7039	23. 5559	32. 5504	41. 1222
6. 5609	16. 7055	24. 5561	33. 5511	42. 1232
7. 5617	17. 7095	25. 5573	34. 5548	43. 1302
8. 5629	18. 7157	26. 5576	35. 6312	44. 1312
9. 5639		27. 5690	36. 6387	45. 2586
10. 5643				46. 2763
				47. 2814

Source Authors

I. Statistical design

Three samples were obtained for each of the 47 hybrids measured during 6 weeks. The data obtained were ordered in an X_{nxl} data matrix, where *n* is the number of rows (samples) and *l* is the number of variables in this investigation (cannabinoids levels and coefficients), resulting in a total of 846 samples and 37 measured variables (predictors). With the 30,456 data from this matrix. The statistical model $Y = X\beta + \varepsilon$ was used to analyze the linear regression [102, 113]. Three regression models were explored to determine the confirmation of the relationship between variable X: "*welfare factors with respect to legal framework* " and variable Y: "*industrial hemp production for the development of human well-being*". A tree-based classification model was created to group cases and predict values of the dependent variable (in this case: *K* coefficient) based on values of independent variables (predictors).

The confirmatory classification analysis was validated. The use of this tool has been used for data reduction and classification of variables by selecting a useful subset of predictors from a large set of variables and has been used to create a formal parametric model [103]. To determine the fusion of the categories that are not significantly different, the CHAID method was used. [104-105]. With the formal parametric model obtained during the classification of the tree process, the linear regression was calculated, the respective coefficients that make up the equation that best estimates the dependent variable (DV) [106, 114]. ANOVA was calculated with 95.0% confidence interval for B. With this validation, the *K* coefficient was calculated for each of the 47 asexual hybrids. The highest value obtained in the previous reduction

and classification process will serve as a reference for the selection of the genetic material evaluated. With this information, Principal Component Analysis (PCA) was carried out, since it is a useful and quick tool that serves as a guide to group hemp plants according to their chemical profile [107-109]. HCA has proven to be a powerful sample grouping tool to genetically classify individuals [110-112]. The mean value of the components was set at 1. Eigenvalues greater than unity were selected. The problem to be solved is to find a space with a smaller dimension that adequately represents the data [113]. To clarify the true value, the PCA was supplemented with HCA, using the method of Ward's linkage (with squared Euclidean distance and z-score standardization, KMO and Bartlett test). Finally, it was proposed to carry out the NNA. The level of sadistically significance used for this investigation, was set at 0.05. The hypothesis where it is possible to establish the Human Wellbeing Coefficient as a new way of genetic selection that incorporates the elements of promotion and protection of human health and to develop the cultivation of Hemp in that direction.

J. Instruments

For variable X, "*Welfare factors with respect to legal framework*" the following sources were used:

1. Hewlett Packard 1100 HPLC equipment and cannabis standards.
2. American Herbal Pharmacopeia - AHP, 2014. Cannabis Inflorescence: Standards Identity, Analysis and Quality Control.
3. Association of Official Agricultural Chemists - AOAC, 2012. Official methods of analysis of AOAC International, 2012.

Equation (1) was used for evaluated the variable Y, "*Industrial hemp production for the development of human well-being*" "; it is also, a dimension of the analyzed problem.

III. RESULTS

The forty-seven cuttings evaluated were collected from five plants registered in the National Registry of Cultivars of Uruguay. They were raised, under controlled greenhouse conditions, by an authorized producer in Uruguay. The sampling process lasted 42 days and began when the threshold of 50% of plants with flower buds was exceeded. For asexual hybrid, 18 samples (repetitions) were collected. The HPLC system was validated. The cannabinoid analysis was carried out under laboratory conditions. The collected data was entered into the $Matrix_{nxl}$ and analyzed. Hypothesis tests, linear regression analysis, ANOVA, decision tree, PCA, HCA and NNA were performed to find the statistical significance between variable X "*welfare factors with respect to legal framework*" and the variable Y "*industrial hemp production for the development of human well-being*".

In Table III, the summaries of Model (1) and Model (2) for the dependent variable *K* (Human Wellbeing Coefficient) were calculated. In Table IV, the ANOVA of *K* was calculated with respect to the selected predictors for both models. In Table V, the coefficients of the predictors of both models with respect to the dependent variable *K* were calculated.

TABLE III
SUMMARY[c] OF THE MODELS FOR THE HUMAN WELLBEING COEFFICIENT - *K*

Model	R	R square	R square adjusted	Standard error of the estimate
1	,99[a]	0,99	0,99	0,10
2	,99[b]	0,99	0,99	0,07

a. Predictors: (Constant), TTHC, CBN, CBG, CBD, THC, CBDa.
b. Predictors: (Constant), R1s, R9, TTHC, TCBD.
c. Dependent variable: Human Wellbeing Coefficient - *K* (R17-P1).
Source: Authors

TABLE IV
ANOVA[a] BETWEEN HUMAN WELLBEING COEFFICIENT AND WELFARE FACTORS WITH RESPECT TO LEGAL FRAMEWORK

	Model	Sum of squares	Gl	Quadratic mean	F	Sig.
1	Regression	532,27	6	88,71	9.646,61	,000[b]
	Residue	7,72	839	0,01		
	Total	539,98	845			
2	Regression	536,34	4	134,07	30.974,73	,000[c]
	Residue	3,64	841	0,00		
	Total	539,98	845			

a. Dependent variable: Human Wellbeing Coefficient – *K* (R17-P1).
b. Predictors: (Constant), TTHC, CBN, CBG, CBD, THC, CBDa.
c. Predictors: (Constant), R1s, R9, TTHC, TCBD.
Source: Authors.

TABLE V.
COEFFICIENTS[a] BETWEEN HUMAN WELLBEING COEFFICIENT AND WELFARE FACTORS WITH RESPECT TO LEGAL FRAMEWORK

	Model	Non-standardized coefficients		T	Sig.	95% confidence interval for B	
		B	Dev. Error			Lower limit	Upper limit
1	(Constant)	0,52	0,02	29,55	0,00	0,48	0,55
	CBDa	0,43	0,00	117,89	0,00	0,43	0,44
	CBD	0,02	0,02	1,07	0,28	-0,02	0,07
	THC	-0,00	0,20	-0,01	0,99	-0,40	0,40
	CBG	2,38	0,09	25,31	0,00	2,20	2,56
	CBN	-30,69	0,84	-36,78	0,00	-32,33	-29,06
	TTHC	0,10	0,06	1,54	0,12	-0,03	0,22
2	(Constant)	1,69	0,02	80,88	0,00	1,65	1,73
	TCBD	0,06	0,01	4,33	0,00	0,03	0,09
	TTHC	0,13	0,04	3,16	0,00	0,05	0,21
	R9	-0,96	0,01	-68,33	0,00	-0,99	-0,94
	R1s	0,39	0,01	28,66	0,00	0,37	0,42

a. Dependent variable: Human Wellbeing Coefficient - *K* (R17-P1).
Source: Authors.

The Multiple Linear Regression of Model (1) and (2), are capable of reducing the prediction error by almost 99% (square R value) in both cases when information from the predictors of each model is taken. As the value of F, in both models, they are statistically significant (R^2=0,99; $F(6,839)$ = 9,643.61; Sig = ,000); and, (R^2=0,99; $F(4,841)$ = 30.974.72; Sig = ,000 respectively). With these results, exist a relationship between the Human Wellbeing Coefficient - *K*. and the predictors of both models. In Figure 3, the standardized P-P normal regression residual was calculated by model.

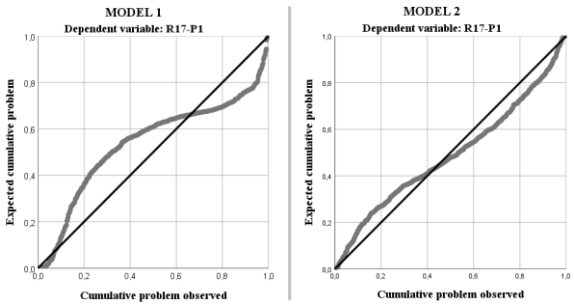

Fig. 3. Regression normal P-P plot standardized residual, by model.
Source: Authors

Model (3) was developed for later use in the *Decision Tree* classification process. Ordinary least squares regression was calculated to predict, in this Model (3), the value of the

dependent variable, Human Wellbeing Coefficient – *K* (R17-P1) for given values of Total Cannabinoids – TCBD. In Table VI, relationship between the dependent variable *K* and the independent variable – TCBD was calculated. In Table VII, ANOVA of dependent variable *K* was calculated. In Table VIII, the coefficients of Model (3) were calculated.

TABLE VI
SUMMARY[b] OF THE MODEL FOR THE COEFFICIENT *K*

Model	R	R square	R square adjusted	Standard error of the estimate
3	,98[a]	0,95	0,95	0,18

a. Predictors: (Constant), Total Cannabidiol – TCBD.
b. Dependent variable: Human Wellbeing Coefficient - *K* (R17-P1).
Source: Authors.

TABLE VII.
ANOVA[a] BETWEEN HUMAN WELLBEING COEFFICIENT AND TOTAL CANNABIDIOL

	Model	Sum of squares	Gl	Quadratic mean	F	Sig.
3	Regression	513,06	1	513,06	16080,73	,000[b]
	Residue	26,93	844	0,03		
	Total	539,99	845			

a. Dependent variable: Human Wellbeing Coefficient – *K* (R17-P1).
b. Predictors: (Constant), Total Cannabidiol – TCBD. Source: Authors

TABLE VIII
COEFFICIENTS[a] BETWEEN HUMAN WELLBEING COEFFICIENT AND TOTAL CANNABIDIOL

		Non-standardized coefficients				95% confidence interval for B	
	Model	B	Dev. Error	t	Sig.	Lower limit	Upper limit
3	(Constant)	0,48	0,03	17,11	0,00	0,42	0,53
	TCBD	0,47	0,00	126,81	0,00	0,47	0,48

a. Dependent variable: Human Wellbeing Coefficient - *K* (R17-P1).
Source: Authors

According to the results, the Multiple Linear Regression Model (3) fits the data since it is capable of reducing the prediction error by almost 95% (R squared value) when the information from the predictor TCBD is taken into account. As the value of *F*, in this case, is statistically significant (R^2=0,95; *F*(1,844) = 16.080,73; Sig=,000), for this Model (3), the most critical level, the t statistic of the regression coefficient is nonzero; therefore, there is a relationship between Total Cannabidiol – TCBD; and, the Human Well-being Coefficient - *K*. The positive relationship between the TCBD and the *K* coefficient is expressed in (5).

$$y = 0{,}476 + 0{,}473\,TCBD \qquad (5)$$

With equation (5), the value of the dependent variable *K* was calculated. It is statistically significant (R^2=0,95; *F*(1,844) = 16.080,73; Sig=,000), that, for each unit of TCBD, the coefficient *K* changes 0,47 times; and, is equal to 0,48 when TCBD is equal to zero. In Fig. 4, was calculated a clustered dispersion of TCBD by *K* Coefficient (R17-P1), according to starting genetic material.

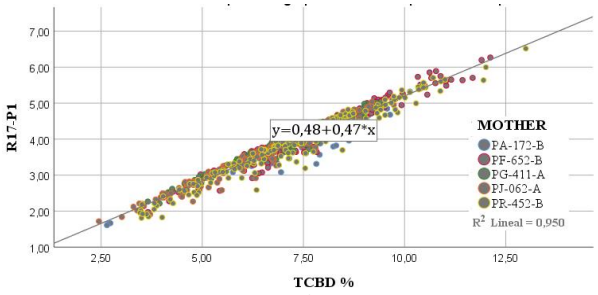

Fig. 4. Statically positive significance relationship between Total Cannabidiol -TCBD and Human Wellbeing Coefficient - *K*. Source: Authors

With the application of the *Decision Tree* procedure, Model (4) was created parametrically using the predictor selected from Model (3). To predict the *K* variable based on the TCBD values, 10 groups were classified since their relationship was statistically significant in the regression analysis. Values higher than those obtained in the tenth block of the *Decision Tree* analysis (in this case, *K*> 4.82), allowed the identification of the hybrids that obtained the greatest potential to provide protection and promotion of human health within the legal framework. (*F* = 904.3, Sig = .000). In Table IX, the *K* division values of the Model (4) was calculated.

TABLE IX.
DECISION TREE RESULTS CONSIDERING GROUPING OF COEFFICIENT *K*[a]

Node	Mean TCDB	Desv. Deviation	N	Percentage	*K*[b] Division values
0	7,28	1,65	846	100%	
1	4,22	0,69	84	10%	<= 2,79
2	5,55	0,56	85	10%	(2,79, 3,29]
3	6,31	0,43	85	10%	(3,29, 3,59]
4	6,84	0,44	84	10%	(3,59, 3,79]
5	7,24	0,37	85	10%	(3,79, 4,01]
6	7,62	0,33	85	10%	(4,01, 4,19]
7	8,00	0,33	84	10%	(4,19, 4,36]
8	8,34	0,31	85	10%	(4,36, 4,55]
9	8,77	0,36	84	10%	(4,55, 4,82]
10	9,96	0,90	85	10%	> 4,82

Growth method: CHAID.
a. Bonferroni adjusted. Statically significance: ,000.
b. Human Wellbeing Coefficient
Source Authors

Model (4) analyzed the statistically significant relationship between the *K* coefficient and the predictors under study, the *K* coefficient was calculated with the 846 samples collected from the 47 asexual hybrids. A factorial analysis was performed to explain the greater variance in the total variables intervened. Principal Component Analysis (PCA) was carried out, since it is a useful and quick tool that serves as a guide to group hemp plants according to their chemical profile. For the PCA, the correlation matrix was calculated with the predictors of Model (2). The variables were standardized before calculating the components. The mean value of the components was set at 1. Eigenvalues greater than unity were selected.

It was possible to establish a statistical significance (R2 = .99) between the values of K and the values of the "Welfare Factors With Legal Framework" (WFWLF). When K values increase, the probability of increasing the promotion and protection of human health is higher. In Table X, the correlation matrix derived from the PCA was calculated.

TABLE X.
CORRELATION MATRIX[a]

		TCBD	R1s	*K*[b]	TTHC	R9
Correlation	TCBD	1,00	0,99	0,98	0,96	0,01
	R1s	0,99	1,00	0,98	0,95	0,0.
	R17-P1	0,98	0,98	1,00	0,94	-0,17
	TTHC	0,96	0,95	0,94	1,00	0,01
	R9	0,01	0,03	-0,17	0,01	1,00
Sig. (unilateral)	TCBD		0,00	0,00	0,00	0,47
	R1s	0,00		0,00	0,00	0,43
	R17-P1	0,00	0,00		0,00	0,12
	TTHC	0,00	0,00	0,00		0,47
	R9	0,47	0,43	0,12	0,47	

a. Determinant = 9,839E-7
b. Human Wellbeing Coefficient – *K* (R17-P1)
Source: Authors

The *K* coefficient has a significant correlation with the variables Total Cannabidiols -TCBD (CC = .978; p <.005);

and, with the variable Total Tetrahydrocannabinols - TTHC (CC = .936; p < .05): Also, a strong positive with the Full Protection Projection - R1s variable (CC = .979; p < .05). It is observed that the coefficient *K* is negatively correlated with the Coefficient of Promotion – R9.

The variables were standardized before calculating the components. The first principal component was defined as the linear combination of the original variables that has maximum variance. Those components associated with eigenvalues lower than the mean variance were discarded. The mean value of the components was set at 1. Eigenvalues greater than unity were selected. In Table XI the total explained variance was calculated.

TABLE XI
TOTAL EXPLAINED VARIANCE

Component	Initial eigenvalues			Extraction		
	Total	% Var.	% Accum.	Total	% Var.	% Accum.
1	3,90	77,99	77,99	3,89	77,99	77,99
2	1,03	20,53	98,51	1,03	20,53	98,51
3	0,07	1,36	99,87			
4	0,07	0,11	99,98			
5	0,00	0,01	100,00			

Extraction method: Principal Component Analysis – PCA.
Source: Authors

According to the results, the PC1 explains 77% of the variability calculated on the data; and, PC2, explains it in 21%. The cumulative proportion of consecutive PCs was calculated at 98%. Eigenvalues greater than unity were selected. In Fig. 5, the variance explained by each component referred to each independent variable of Model (2) was calculated.

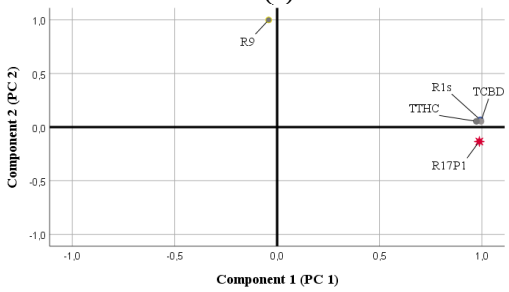

Fig. 5. Component chart. Source: Authors

The hybrids whose *K* values > 4,82 that were selected in the decision tree procedure are the same as those hybrids with eigenvalues greater than unity in the PCA. In Figure 6, the clustered dispersion of PC 2 (21%) by PC 1 (78%) was calculated according to genetic selection by means of the K Coefficient.

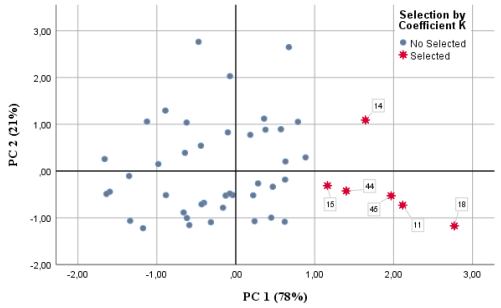

Fig. 6. Clustered dispersion of PC 2 (21%) by PC 1 (78%) according to Selection by Coefficient *K*. Source: Authors

Hierarchical Clustering Analysis (HCA) has proven to be a powerful sample grouping tool to genetically classify individuals. The identification of relatively homogeneous groups was analyzed using the hierarchical cluster procedure and was based on the predictors and dependent variable of the Model (2). From the analysis of PCA and HCA, it has been possible to select the six best hybrids. Also, with the predecessors of the Model (2), the NNA was carried out. The test scores of the hybrid with the highest Human Wellbeing Coefficient - *K*, were compared with those of the 5 closest neighbors. In Table XII, the k closest neighbors and their respective distances were calculated. The six hybrids were grouped into two categories. Category A, corresponds to the first group of superior hybrids with a distance less than 1; and, category B to the second with a distance greater than 1. The feature space plot and the quadrant map were calculated in Figures 7 and 8.

TABLE XII
NEAREST NEIGHBOURS AND DISTANCES, ACCORDING TO INITIAL FOCAL REGISTER

	Nearest Neighbours[a]	Nearest Distances	Selection Category[b]
Focal Registry	18. 7157		A
1	11. 6215	0.71	A
2	45. 2586	0.76	A
3	44. 1312	1.05	B
4	15. 7039	1.31	B
5	14. 7009	1.36	B

a. Number of cases evaluated and corresponded to the order of the list of hybrids described in Table 1.
b. According with de Human Wellbeing Coefficient – *K*.
Source: Authors

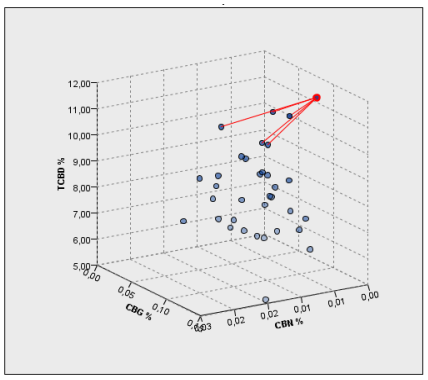

Fig. 7. Space of three predictors selected according to Coefficient *K*. Source: Authors

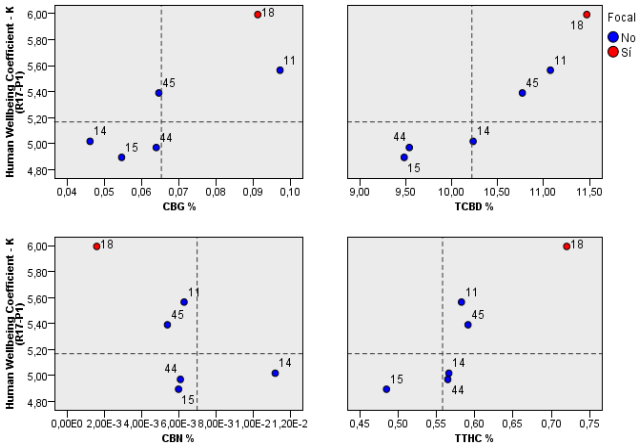

Fig. 8. Quadrant map: Target values by predictors for Initial focal record and nearest neighbours. Source: Authors

With this statistical analysis, records 18, 11 and 45 were selected and cataloged within the group of superior hybrids. A correlation was found with the previous HCA analysis in

which the same records stand out (see Fig. 8). In a second group, records 44, 11 and 15 were selected.

IV. Discussion

It has been possible to find a coefficient for selecting hemp genetic material that provides maximum human welfare [5-8]. The K coefficient is related to the dimensions of legality, protection, and promotion of human health based on industrial hemp [9-19, 84-86] and contributes to improving human well-being. This is how the K coefficient is linked to TCBD, CBG, CBN and TTHC [34-83]. The novelty raised in this study lies in the relationship between cannabinoid-producing agricultural activity and the requirement of the endocannabinoid system of the human brain. Although there are studies on the importance of the endocannabinoid system in the human brain [20-33], the *K* coefficient has managed to generate a new tool to understand the influence of legality, increase protection and promotion of health, as well as encourage multidisciplinary research that encompasses the agricultural field, health, or neurochemistry.

On the one hand, the combined use of statistical tools provides the ability to separate genetic material. In this way, the PCA allows the selection of material according to its chemical profile, and the use of HCA enables the genetic classification of the individuals with the best profile [110-113], as well as the separation of two groups by using NNA analysis. From another perspective, the strength of the study is that, as *K* values increase, the probability of increasing the promotion and protection of human health is greater. Likewise, it constitutes a coefficient to select superior hybrids within a determined population. Another strong point is the number of analyzes carried out, since they indicate the consistency of the work. In addition, this research will be useful for geneticists, agricultural producers, industrial producers, and the general population. On the other hand, the limitations found are related to the heterogeneity of the initial genetic material and the precision of the weight of the samples, as well as the storage conditions for their subsequent analysis. However, the *K* coefficient allows a broader view of both agricultural activity and the endocannabinoid system of the brain.

As a synthesis, the *K* coefficient allows selecting the individuals that contain the greatest potential to provide human well-being. Likewise, the present study has implications in the method of selection of genetic material and in the maximization of the potential that industrial hemp has to contribute to human well-being from the perspective of health. The next logical step is to establish gene banks that allow the production of quality seeds to expand the possibilities of the industrial hemp value chain, with the aim of generating improvements in agriculture and human health.

V. Conclusion

Based on the results, the new concept of Human Well-being Coefficient - *K* arises. To arrive at its construction, a method has been designed to incorporate variables grouped in the dimensions of promotion, protection, and legality, in terms of the productive activity of the industrial hemp. In this way, individuals with the potential to improve human well-being linked to the field of health have been identified. This is how it is verified that, by increasing the values of *K*, the probability of protection and promotion of human health also increases. In the same way, the highest values of the *K* coefficient generate an increase in the promotion of human health, with greater digestive and antibacterial protection per unit of neuronal protection. In addition, the HPLC as an instrument to identify data levels, and the articulation of certain statistical tools, allowed the selection of superior hybrids, as well as promising advanced material.

With the aforementioned, the relevance of the connection between agricultural production and the understanding of the endocannabinoid system of the human brain is highlighted. In the present work, it has been possible to include variables such as sleep problems, pain, inflammation reduction, anxiety, neuroprotection, appetite stimulation, antibacterial and digestive benefits, and legal activity. This highlights the versatility with which this study could be used and in the various areas in which it could be involved such as agriculture, genetics, neurosciences, chemistry, medical health, and mental health.

References

[1] Meyer Jr, A. F. (1955). The control of Occupatinal Helth Hazards: Concepts Of Engineerincg Application. *American Industrial Hygiene Association Quarterly*, 16(2), 131-133. https://doi.org/10.1080/00968205509344467

[2] Pryor, A., Townsend, M., Maller, C., & Field, K. (2006). Health and well-being naturally:'contact with nature'in health promotion for targeted individuals, communities and populations. *Health Promotion Journal of Australia*, 17(2), 114-123. https://doi.org/10.1071/HE06114

[3] Bunch, M. J. (2016). Ecosystem approaches to health and well-being: Navigating complexity, promoting health in social–ecological systems. *Systems research and behavioral science*, 33(5), 614-632. https://doi.org/10.1002/sres.2429

[4] Fleming, L. E., Leonardi, G. S., White, M. P., Medlock, J., Alcock, I., Macintyre, H. L., & Duarte-Davidson, R. (2018). "Beyond climate change and health: Integrating broader environmental change and natural environments for public health protection and promotion in the UK. *Atmosphere*. 9(7). 245.https://doi.org/10.3390/atmos9070245

[5] Cole, D. C., Eyles, J., Gibson, B. L., & Ross, N. (1999). Links between humans and ecosystems: the implications of framing for health promotion strategies. *Health Promotion International*, 14(1), 65-72. https://doi.org/10.1093/heapro/14.1.65

[6] McDaid, D., Curran, C., & Knapp, M. (2005). Promoting mental well-being in the workplace: a European policy perspective. *International review of psychiatry*, 17(5), 365-373. https://doi.org/10.1080/09540260500238397

[7] Ebi, K. L., Frumkin, H., & Hess, J. J. (2017). Protecting and promoting population health in the context of climate and other global environmental changes. *Anthropocene*, 19, 1-12. https://doi.org/10.1016/j.ancene.2017.07.001

[8] Aerts, R., Honnay, O., & Van Nieuwenhuyse, A. (2018). Biodiversity and human health: mechanisms and evidence of the positive health effects of diversity in nature and green spaces. *British medical bulletin*, 127(1), 5-22. https://doi.org/10.1093/bmb/ldy021

[9] Bellon, M. R. (1996). The dynamics of crop infraspecific diversity: A conceptual framework at the farmer level 1. *Economic botany*, 50(1), 26-39. https://doi.org/10.1007/BF02862110

[10] Rozin, P. (2005). The meaning of food in our lives: a cross-cultural perspective on eating and well-being. *Journal of nutrition education and behavior*, 37, S107-S112. https://doi.org/10.1016/S1499-4046(06)60209-1

[11] Fredrickson, B. L., Grewen, K. M., Coffey, K. A., Algoe, S. B., Firestine, A. M., Arevalo, J. M., & Cole, S. W. (2013). A functional genomic perspective on human well-being. Proceedings of the *National Academy of Sciences*, 110(33), 13684-13689. https://doi.org/10.1073/pnas.1305419110

[12] Kumar, H. (2020). Biotechnology: discoveries and their applications in societal welfare. Biotechnology business-concept to delivery. *Springer, Cham*, 3-44. https://doi.org/10.1007/978-3-030-36130-3

[13] Mead, A. (2019). Legal and regulatory issues governing cannabis and cannabis-derived products in the United States. *Frontiers in plant science*, 10, 697. https://doi.org/10.3389/fpls.2019.00697

[14] Krause, D., & Pullman, M. (2021). Fighting to survive: how supply chain managers navigate the emerging legal cannabis industry. *Journal of Supply Chain Management*, 57(3), 50-71. https://doi.org/10.1111/jscm.12238
[15] Kim, G. J., Hwang, S. J., & Berry, F. S. (2021). Explaining the strictness of medical marijuanaregulations in states. *The Social Science Journal*, 58(2), 186-205. https://doi.org/10.1016/j.soscij.2019.04.003
[16] Pusiak, R. J., Cox, C., & Harris, C. S. (2021). Growing pains: An overview of cannabis quality control and quality assurance in Canada. *International* Journal of Drug Policy, 103111. https://doi.org/10.1016/j.drugpo.2021.103111
[17] Wartenberg, A. C., Holden, P. A., Bodwitch, H., Parker-Shames, P., Novotny, T., Harmon, T. C., & Butsic, V. (2021). Cannabis and the environment: what science tells us and what we still need to know. *Environmental Science & Technology Letters*, 8(2), 98-107. https://doi.org/10.1021/acs.estlett.0c00844
[18] Hollenbeck, B., & Uetake, K. (2021). Taxation and market power in the legal marijuana industry. *The RAND Journal of Economics*, 52(3), 559-595. https://doi.org/10.1111/1756-2171.12384
[19] De Briyne, N., Holmes, D., Sandler, I., Stiles, E., Szymanski, D., Moody, S., ... & Anadón, A. (2021). Cannabis, Cannabidiol Oils and Tetrahydrocannabinol—What Do Veterinarians Need to Know? *Animals*, 11(3), 892. https://doi.org/10.3390/ani11030892.
[20] Schlag, A. K., O'Sullivan, S. E., Zafar, R. R., & Nutt, D. J. (2021). Current controversies in medical cannabis: recent developments in human clinical applications and potential therapeutics. *Neuropharmacology*, 108586. https://doi.org/10.1016/j.neuropharm.2021.108586.
[21] Wallach, J. (2021). Medicinal Cannabis: an overview for health-care providers. *Remington*, 75-101. https://doi.org/10.1016/B978-0-12-820007-0.00005-2
[22] de Lorimier, L. P., Hazzah, T., Amazonas, E., & Cital, S. (2021). Cannabinoids in Oncology and Immune Response. In Cannabis Therapy in Veterinary Medicine, *Springer, Cham*. doi:https://doi.org/10.1007/978-3-030-68317-7_10.
[23] Lu, H. C., & Mackie, K. (2021). Review of the endocannabinoid system. Biological Psychiatry. *Cognitive Neuroscience and Neuroimaging*, 6(6), 607-615. https://doi.org/10.1016/j.bpsc.2020.07.016
[24] Blanton, H. L., Barnes, R. C., McHann, M. C., Bilbrey, J. A., Wilkerson, J. L., & Guindon, J. (2021). Sex differences and the endocannabinoid system in pain. *Pharmacology Biochemistry and Behavior*, 173107. https://doi.org/10.1016/j.bpsc.2020.07.016
[25] Garani, R., Watts, J. J., & Mizrahi, R. (2021). Endocannabinoid system in psychotic and mood disorders, a review of human studies. *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, 106, 110096. https://doi.org/10.1016/j.pnpbp.2020.110096.
[26] Bourdy, R., Hertz, A., Filliol, D., Andry, V., Goumon, Y., Mendoza, J., & Befort, K. (2021). The endocannabinoid system is modulated in reward and homeostatic brain regions following diet-induced obesity in rats: A cluster analysis approach. *European journal of nutrition*, 1-13. https://doi.org/10.1007/s00394-021-02613-0
[27] Santoro, A., Mele, E., Marino, M., Viggiano, A., Nori, S. L., & Meccariello, R. (2021). The complex interplay between Endocannabinoid System and the Estrogen System in Central Nervous System and periphery. *International Journal of Molecular* Sciences, 22(2), 972. https://doi.org/10.3390/ijms22020972
[28] Iannotti, F. A., & Vitale, R. M. (2021). The endocannabinoid system and PPARs: focus on their signalling crosstalk, action and transcriptional regulation. *Cells*, 10(3), 586. https://doi.org/10.3390/cells10030586
[29] Delgado-Povedano, M. M., Callado, C. S. C., Priego-Capote, F., & Ferreiro-Vera, C. (2020). Untargeted characterization of extracts from Cannabis sativa L. cultivars by gas and liquid chromatography coupled to mass spectrometry in high resolution mode. *Talanta*, 208, 120384. https://doi.org/10.1016/j.talanta.2019.120384
[30] di Giacomo, V., Recinella, L., Chiavaroli, A., Orlando, G., Cataldi, A., Rapino, M., & Ferrante, C. (2021). Metabolomic profile and antioxidant/anti-inflammatory effects of industrial hemp water extract in fibroblasts, keratinocytes and isolated mouse skin specimens. Antioxidants, 10(1), 44. https://doi.org/10.3390/antiox10010044
[31] Duggan, P. J. (2021). The Chemistry of Cannabis and Cannabinoids. *Australian Journal of Chemistry*, 74(6), 369-387. https://doi.org/10.1071/CH21006
[32] Komarnytsky, S., Rathinasabapathy, T., Wagner, C., Metzger, B., Carlisle, C., Panda, C., & Varadharaj, S. (2021). Endocannabinoid System and Its Regulation by Polyunsaturated Fatty Acids and Full Spectrum Hemp Oils. *International Journal of Molecular Sciences*, 22(11), 5479. https://doi.org/10.3390/ijms22115479
[33] Addo, P. W., Brousseau, V. D., Morello, V., MacPherson, S., Paris, M., & Lefsrud, M. (2021). Cannabis chemistry, post-harvest processing methods. *A review. ndustrial Crops and Products*, 170, 113743. https://doi.org/10.1016/j.indcrop.2021.113743
[34] Bíró, T., Tóth, B. I., Haskó, G., Paus, R., & Pacher, P. (2009). The endocannabinoid system of the skin in health and disease: novel perspectives and therapeutic opportunities. *Trends in pharmacological sciences*, 30(8), 411-420. https://doi.org/10.1016/j.tips.2009.05.004
[35] Esposito, G., Filippis, D. D., Cirillo, C., Iuvone, T., Capoccia, E., Scuderi, C., & Steardo, L. (2013). Cannabidiol in inflammatory bowel diseases: a brief overview. *Phytotherapy Research*, 27(5), 633-636. https://doi.org/10.1002/ptr.4781
[36] Hammell, D. C., Zhang, L. P., Ma, F., Abshire, S. M., McIlwrath, S. L., Stinchcomb, A. L., & Westlund, K. N. (2016). Transdermal cannabidiol reduces inflammation and pain-related behaviours in a rat model of arthritis. *European Journal of Pain*, 20(6), 936-948. https://doi.org/10.1002/ejp.818
[37] Bragança, V. A., França, T. G., de Jesus, A. C., Palheta, I. C., Melo, F. P., Neves, P. A., & Borges, R. S. (2020). Impact of conformational and solubility properties on psycho-activity of cannabidiol (CBD) and tetrahydrocannabinol (THC). *Chemical Data Collections*, 26, 100345. https://doi.org/10.1016/j.cdc.2020.100345
[38] Fiani, B., Sarhadi, K. J., Soula, M., Zafar, A., & Quadri, S. A. (2020). Current application of cannabidiol (CBD) in the management and treatment of neurological disorders. *Neurological Sciences*, 1-14. https://doi.org/10.1007/s10072-020-04514-2
[39] Kienzl, M., Storr, M., & Schicho, R. (2020). Cannabinoids and opioids in the treatment of inflammatory bowel diseases. *Clinical and translational gastroenterology*, 11(1). https://doi.org/10.14309/ctg.0000000000000120
[40] Leen-Feldner, E. W., Bynion, T. M., Gournay, R., Bonn-Miller, M. O., & Feldner, M. T. (2021). Practical Considerations for Testing the Effects of Cannabidiol on Human Anxiet. *Journal of Anxiety Disorders*, 102429. https://doi.org/10.1016/j.janxdis.2021.102429
[41] Arout, C. A., Haney, M., Herrmann, E. S., Bedi, G., & Cooper, Z. D. (2021). A placebo-controlled investigation of the analgesic effects, abuse liability, safety and tolerability of a range of oral cannabidiol doses in healthy humans. *British journal of clinical pharmacology*, 1-9. https://doi.org/10.1111/bcp.14973
[42] O'Sullivan, S. E., Stevenson, C. W., & Laviolette, S. R. (2021). Could cannabidiol be a treatment for coronavirus disease-19-related anxiety disorders? *Cannabis and Cannabinoid Research*, 6(1), 7-18. https://doi.org/10.1089/can.2020.0102
[43] Murillo-Rodríguez, E., Machado, S., Imperatori, C., Yamamoto, T., & Budde, H. (2021). Natural Cannabinoids as Templates for Sleep Disturbances Treatments. In *Cannabinoids and Sleep*, Springer, Cham., 133-141. https://doi.org/10.1007/978-3-030-61663-2_9
[44] Meyer, E., de Mattos, B. A., Kirchhoff, F., & de Oliveira, R. M. W. (2021). Neuroprotective Effects of Cannabidiol Under Cerebral Ischemic Conditions. *Revista Brasileira de Farmacognosia*, 1-13. https://doi.org/10.1007/s43450-021-00199-6
[45] Chaturvedi, R. K., & Agrawal, D. C. (2021). Cannabis-Induced Neuroactivity: Research Trends and Commercial Prospects. Singapore: In *Medicinal Herbs and Fungi*. Springer, 159-185. https://doi.org/10.1007/978-981-33-4141-8_6
[46] Kladar, N., Čonić, B. S., Božin, B., & Torović, L. (2021). European hemp-based food products–Health concerning cannabinoids exposure assessment. *Food Control*, 129, 108233. https://doi.org/10.1016/j.foodcont.2021.108233
[47] Kaminski, N. E., & Kaplan, B. L. (2021). Immunomodulation by cannabinoids: Current uses, mechanisms, and identification of data gaps to be addressed for additional therapeutic application. *Advances in Pharmacology*, 91, 1-59. https://doi.org/10.2174/1381612827666210426092847
[48] Pugazhendhi, A., Suganthy, N., Chau, T. P., Sharma, A., Unpaprom, Y., Ramaraj, R., & Brindhadevi, K. (2021). Cannabinoids as anticancer and neuroprotective drugs: Structural insights and pharmacological interactions—A review. *Process Biochemistry*, 111, 9-31. https://doi.org/10.1016/j.procbio.2021.08.025
[49] Mouli, S., Ramesh, S., Govindarajulu, M., Almaghrabi, M., Fujihashi, A., Nadar, R. M., & Dhanasekaran, M. (2021). Balancing the Neuroprotective Versus Neurotoxic Effects of Cannabis. In: Agrawal D.C., Dhanasekaran M. (eds). Singapore: *Medicinal Herbs and Fungi*, Springer, 203-226. https://doi.org/10.1007/978-981-33-4141-8_8

[50] Maccarrone, M., Maldonado, R., Casas, M., Henze, T., & Centonze, D. (2017). Cannabinoids therapeutic use: what is our current understanding following the introduction of THC, THC: CBD oromucosal spray and others? *Expert review of clinical pharmacology*. https://doi.org/10.1080/17512433.2017.1292849

[51] Nachnani, R., Raup-Konsavage, W. M., & Vrana, K. E. (2021). The Pharmacological Case for Cannabigerol. *Journal of Pharmacology and Experimental Therapeutics*, 376(2), 204-212. https://doi.org/10.1124/jpet.120.000340

[52] Andriotis, E. G., Chachlioutaki, K., Monou, P. K., Bouropoulos, N., Tzetzis, D., Barmpalexis, P., & Fatouros, D. G. (2021). Development of Water-Soluble Electrospun Fibers for the Oral Delivery of Cannabinoids. *AAPS PharmSciTech*, 22(1), 1-14. https://doi.org/10.1208/s12249-020-01895-7

[53] Clarke, H., Roychoudhury, P., & Narouze, S. N. (2021). Other phytocannabinoids. Springer, Cham.: In *Cannabinoids and Pain*, 87-92. https://doi.org/10.1007/978-3-030-69186-8_12.

[54] Cole, D. C., Eyles, J., Gibson, B. L., & Ross, N. (1999). Links between humans and ecosystems: the implications of framing for health promotion strategies. *Health Promotion International*, 14(1), 65-72. https://doi.org/10.1093/heapro/14.1.65

[55] Ożarowski, M., Karpiński, T. M., Zielińska, A., Souto, E. B., & Wielgus, K. (2021). Cannabidiol in neurological and neoplastic diseases: Latest developments on the molecular mechanism of action. *International journal of molecular sciences*, 22(9), 4294. https://doi.org/10.3390/ijms22094294

[56] Xiong, Y., & Lim, C. S. (2021). Understanding the Modulatory Effects of Cannabidiol on Alzheimer's Disease. *Brain Sciences*, 11(9), 1211. https://doi.org/10.3390/brainsci11091211

[57] Chan, J. Z., & Duncan, R. E. 2021. Regulatory Effects of Cannabidiol on Mitochondrial Functions: A Review. *Cells*, 10(5), 1251. https://doi.org/10.3390/cells10051251

[58] Rimmerman, N., Ben-Hail, D., Porat, Z., Juknat, A., Kozela, E., Daniels, M. P., ... & Vogel, Z. (2013). Direct modulation of the outer mitochondrial membrane channel, voltage-dependent anion channel 1 (VDAC1) by cannabidiol: a novel mechanism for cannabinoid-induced cell death. *Cell death & disease*, 4(12), e949-e949. https://doi.org/10.1038/cddis.2013.471

[59] Huang, T., Xu, T., Wang, Y., Zhou, Y., Yu, D., Wang, Z., & Yan, C. (2021). Cannabidiol inhibits human glioma by induction of lethal mitophagy through activating TRPV4. *Autophagy*, 1-15. https://doi.org/10.1080/15548627.2021.1885203

[60] González-Mariscal, I., Carmona-Hidalgo, B., Winkler, M., Unciti-Broceta, J. D., Escamilla, A., Gómez-Cañas, M., & Muñoz, E. (2021). (+)-trans-Cannabidiol-2-hydroxy pentyl is a dual CB1R antagonist/CB2R agonist that prevents diabetic nephropathy in mice. *Pharmacological research*, 169, 105492. https://doi.org/10.1016/j.phrs.2021.105492

[61] Boehnke, K. F., Gagnier, J. J., Matallana, L., & Williams, D. A. (2021). Cannabidiol use for fibromyalgia: Prevalence of use and perceptions of effectiveness in a large online survey. *The Journal of Pain*, 22(5), 556-566. https://doi.org/10.1016/j.jpain.2020.12.001

[62] Liu, C., Li, H., Xu, F., Jiang, X., Ma, H., & Seeram, N. P. (2021). Cannabidiol Protects Human Skin Keratinocytes from Hydrogen-Peroxide-Induced Oxidative Stress via Modulation of the Caspase-1–IL-1β Axis. *Journal of Natural Products*, 84(5), 1563-1572. https://doi.org/10.1021/acs.jnatprod.1c00083

[63] Graczyk, M., Lewandowska, A. A., & Dzierżanowski, T. (2021). The therapeutic potential of cannabis in counteracting oxidative stress and inflammation. *Molecules*, 26(15), 4551. https://doi.org/10.3390/molecules26154551

[64] Li, J., Carvajal, R., Bruner, L., & Kaminski, N. E. (2021). The current understanding of the benefits, safety, and regulation of cannabidiol in consumer products. *Food and Chemical Toxicology*, 112600. https://doi.org/10.1016/j.fct.2021.112600

[65] Moltke, J., & Hindocha, C. (2021). Reasons for cannabidiol use: a cross-sectional study of CBD users, focusing on self-perceived stress, anxiety, and sleep problems. *Journal of cannabis research*, 3(1), 1-12. https://doi.org/10.1186/s42238-021-00061-5

[66] Valvassori, S. S., Bavaresco, D. V., Scaini, G., Varela, R. B., Streck, E. L., Chagas, M. H., ... & Quevedo, J. (2013). Acute and chronic administration of cannabidiol increases mitochondrial complex and creatine kinase activity in the rat brain. *Brazilian Journal of Psychiatry*, 35, 380-386. https://doi.org/10.1590/1516-4446-2012-0886

[67] Da Silva, V. K., de Freitas, B. S., da Silva Dornelles, A., Nery, L. R., Falavigna, L., Ferreira, R. D. P., & Schröder, N. (2014). Cannabidiol normalizes caspase 3, synaptophysin, and mitochondrial fission protein DNM1L expression levels in rats with brain iron overload: implications for neuroprotection. *Molecular neurobiology*, 49(1), 222-233. https://doi.org/10.1007/s12035-013-8514-7

[68] Gęgotek, A., Atalay, S., Domingues, P., & Skrzydlewska, E. (2019). The differences in the proteome profile of cannabidiol-treated skin fibroblasts following UVA or UVB irradiation in 2D and 3D cell cultures. *Cells*, 8(9), 995. https://doi.org/10.3390/cells8090995

[69] Casares, L., García, V., Garrido-Rodríguez, M., Millán, E., Collado, J. A., García-Martín, A., ... & Muñoz, E. (2020). Cannabidiol induces antioxidant pathways in keratinocytes by targeting BACH1. *Redox biology*, 28, 101321. https://doi.org/10.1016/j.redox.2019.101321

[70] Abyadeh, M., Gupta, V., Paulo, J. A., Gupta, V., Chitranshi, N., Godinez, A., & Mirzaei, M. (2021). A Proteomic View of Cellular and Molecular Effects of Cannabis. *Biomolecules*, 11(10), 1411. https://doi.org/10.3390/biom11101411

[71] Gruber, J. V., & Holtz, R. (2021). Purified Cannabidiol isolate does not inhibit active caspase-1 release in NLRP inflammasome-mediated UVB or ATP-activated keratinocytes or appear to reduce key inflammatory cytokines in UVB-irradiate keratinocytes. *International journal of cosmetic science*, 1-9. https://doi.org/10.1111/ics.12693

[72] Rodríguez-Cueto, C., García-Toscano, L., Santos-García, I., Gómez-Almería, M., Gonzalo-Consuegra, C., Espejo-Porras, F., & de Lago, E. (2021). Targeting the CB2 receptor and other endocannabinoid elements to delay disease progression in amyotrophic lateral sclerosis. *British Journal of Pharmacology*, 178(6), 1373-1387. https://doi.org/10.1111/bph.15386

[73] Filipiuc, L. E., Ababei, D. C., Alexa-Stratulat, T., Pricope, C. V., Bild, V., Stefanescu, R., & Tamba, B. I. (2021). Major Phytocannabinoids and Their Related Compounds: Should We Only Search for Drugs That Act on Cannabinoid Receptors? *Pharmaceutics*, 13(11), 1823. https://doi.org/10.3390/pharmaceutics13111823

[74] Paudel, P., Ross, S., & Li, X. C. (2021). Molecular Targets of Cannabinoids Associated with Depression. *Current Medicinal Chemistry*, 1-12. https://doi.org/10.2174/0929867328666210623144658

[75] Marsh, D. T., & Smid, S. D. (2020). Cannabis Phytochemicals: A Review of Phytocannabinoid Chemistry and Bioactivity as Neuroprotective Agents. *Australian Journal of Chemistry*, 74(6), 388-404. https://doi.org/10.1071/CH20183

[76] Abate, G., Uberti, D., & Tambaro, S. (2021). Potential and Limits of Cannabinoids in Alzheimer's Disease Therapy. *Biology*, 10(6), 542. https://doi.org/10.3390/biology1006054

[77] Valeri, A., & Mazzon, E. (2021). Cannabinoids and Neurogenesis: The Promised Solution for Neurodegeneration? *Molecules*, 26(20), 6313. https://doi.org/10.3390/molecules26206313

[78] Mahdi, O., Baharuldin, M. T., Nor, N. H. M., Chiroma, S. M., Jagadeesan, S., & Moklas, M. A. (2021). The Neuroprotective Properties, Functions, and Roles of Cannabis sativa in Selected Diseases Related to the Nervous System. *Medicinal Chemistry-Central Nervous System Agents*, 21(1), 20-38. https://doi.org/10.2174/1871524921666210127110028

[79] Oultram, J. M., Pegler, J. L., Bowser, T. A., Ney, L. J., Eamens, A. L., & Grof, C. P. (2021). Cannabis sativa: interdisciplinary strategies and avenues for medical and commercial progression outside of CBD and THC. *Biomedicines*, 9(3), 234. https://doi.org/10.3390/biomedicines9030234

[80] Richter, G., Hazzah, T., Hartsel, J. A., Eades, J., Hickory, B., & Makriyannis, A. (2021). Cannabis sativa: an overview. *Nutraceuticals*, 603-624. https://doi.org/10.1016/B978-0-12-821038-3.00038-0

[81] Russo, E. B., Cuttler, C., Cooper, Z. D., Stueber, A., Whiteley, V. L., & Sexton, M. (2021). Survey of Patients Employing Cannabigerol-Predominant Cannabis Preparations: Perceived Medical Effects, Adverse Events, and Withdrawal Symptoms. *Cannabis and Cannabinoid Research*, 1-12. https://doi.org/10.1089/can.2021.0058

[82] Aqawi, M., Sionov, R. V., Gallily, R., Friedman, M., & Steinberg, D. (2021). Anti-Biofilm Activity of Cannabigerol against Streptococcus mutans. *Microorganisms*, 9(10), 2031. https://doi.org/10.3390/microorganisms9102031

[83] Schofs, L., Sparo, M. D., & Sánchez Bruni, S. F. (2021). The antimicrobial effect behind Cannabis sativa. *Pharmacology Research & Perspectives*, 9(2), e00761. https://doi.org/10.1002/prp2.761

[84] Hädener, M., König, S., & Weinmann, W. (2019). Quantitative determination of CBD and THC and their acid precursors in confiscated cannabis samples by HPLC-DAD. *Forensic science international*, 299, 142-150. doi:https://doi.org/10.1016/j.forsciint.2019.03.046

[85] Romano, R., Aiello, A., De Luca, L., Sica, R., Caprio, E., Pizzolongo, F., & Blaiotta, G. (2021). Characterization of a new type of mead fermented with Cannabis sativa L.(hemp). *Journal of food science*, 86(3), 874-880. https://doi.org/10.1111/1750-3841.15614

[86] Sgrò, S., Lavezzi, B., Caprari, C., Polito, M., D'Elia, M., Lago, G., ... & Ferri, E. N. (2021). Delta9-THC determination by the EU official method: evaluation of measurement uncertainty and compliance assessment of hemp samples. *Analytical and bioanalytical chemistry*, 413(13), 3399-3410. doi:https://doi.org/10.1007/s00216-021-03283-x

[87] Merone, G. M., Tartaglia, A., Rossi, S., Santavenere, F., Bassotti, E., D'Ovidio, C., & Savini, F. (2021). Fast liquid chromatography-tandem mass spectrometry method for the simultaneous determination of phytocannabinoids in oily based preparations. *Journal of Pharmaceutical and Biomedical Analysis*, 114174. https://doi.org/10.1016/j.jpba.2021.114174

[88] Montilla, C. A. P., Schaiquevich, P. S., Guido, P. C., Caraballo, R. H., Valenzuela, G. R., Cruz, C. V., & Bournissen, F. G. (2021). An Ultrafast Ultrahigh-Performance Liquid Chromatography Coupled With Tandem Mass Spectrometry Method for Cannabidiol Monitoring in Pediatric Refractory Epilepsy. *Therapeutic Drug Monitoring*, 43(5), 712-717. Accessed 11 01, 2021. doi:doi: 10.1097/FTD.0000000000000846

[89] Muscarà, C., Smeriglio, A., Trombetta, D., Mandalari, G., La Camera, E., Grassi, G., & Circosta, C. (2021). Phytochemical characterization and biological properties of two standardized extracts from a non-psychotropic Cannabis sativa L. cannabidiol (CBD)-chemotype. *Phytotherapy Research*, 35(9), 5269-5281. https://doi.org/10.1002/ptr.7201

[90] Brighenti, V., Protti, M., Anceschi, L., Zanardi, C., Mercolini, L., & Pellati, F. 2021. Emerging challenges in the extraction, analysis and bioanalysis of cannabidiol and related compounds. *Journal of Pharmaceutical and Biomedical Analysis*, 192, 113633. https://doi.org/10.1016/j.jpba.2020.113633

[91] Analakkattillam, S., Langsi, V. K., Hanrahan, J. P., & Moore, E. (2021). Comparative study of Dissolution for Cannabidiol in EU and US Hemp oil products by HPLC. *Journal of Pharmaceutical Sciences*, 110(8), 3091-3098. https://doi.org/10.1016/j.xphs.2021.03.028

[92] Sarma, N. D., Waye, A., ElSohly, M. A., Brown, P. N., Elzinga, S., Johnson, H. E., ... & Giancaspro, G. I. (2020). Cannabis inflorescence for medical purposes: USP considerations for quality attributes. *Journal of natural products*, 83(4), 1334-1351. https://doi.org/10.1021/acs.jnatprod.9b01200

[93] American Herbal Pharmacopeia - AHP. (2014). Cannabis Inflorescence: Standards identity, Analysis and Quality Control. Monography, Scotts Valley, CA 95067: *AHP*. Accessed 11 01, 2021. https://herbal-ahp.org/online-ordering-cannabis-inflorescence-qc-monograph/

[94] Association of Official Analytical Chemists - AOAC. (2012). Official methods of analysis of AOAC International. Vol. 1. Rockville, MD 20850: Gaithersburg. *AOAC*. Accessed 11 02, 2021. http://www.eoma.aoac.org/

[95] Wiberg, K., Andersson, M., Hagman, A., & Jacobsson, S. P. (2004). Peak purity determination with principal component analysis of high-performance liquid chromatography–diode array detection data. *Journal of Chromatography A*, 1029(1-2), 13-20. https://doi.org/10.1016/j.chroma.2003.12.052

[96] Almeida, A. M. D., Castel-Branco, M. M., & Falcao, A. C. (2002). Linear regression for calibration lines revisited: weighting schemes for bioanalytical methods. *Journal of Chromatography B*, 774(2), 215-222. https://doi.org/10.1016/S1570-0232(02)00244-1

[97] Wang, M., Wang, Y. H., Avula, B., Radwan, M. M., Wanas, A. S., Mehmedic, Z., ... & Khan, I. A. (2017). Quantitative determination of cannabinoids in cannabis and cannabis products using ultra-high-performance supercritical fluid chromatography and diode array/mass spectrometric detection. *Journal of Forensic Sciences*, 62(3), 602-611. https://doi.org/10.1111/1556-4029.13341

[98] Citti, C., Battisti, U. M., Braghiroli, D., Ciccarella, G., Schmid, M., Vandelli, M. A., & Cannazza, G. (2018). A metabolomic approach applied to a liquid chromatography coupled to high-resolution tandem mass spectrometry method (HPLC-ESI-HRMS/MS): towards the comprehensive evaluation of the chemical composition of Cannabis medicinal extracts. *Phytochemical Analysis*, 29(2), 144-155. https://doi.org/10.1002/pca.2722

[99] Vaclavik, L., Benes, F., Fenclova, M., Hricko, J., Krmela, A., Svobodova, V., & Mastovska, K. (2019). Quantitation of Cannabinoids in cannabis dried plant materials, concentrates, and oils using liquid chromatography–diode array detection technique with optional mass spectrometric detection: single-laboratory validation study, first action 2018.11. *Journal of AOAC International*, 102(6), 1822-1833. https://doi.org/10.1093/jaoac/102.6.1822

[100] Krill, C., Rochfort, S., & Spangenberg, G. (2020). A high-throughput method for the comprehensive analysis of terpenes and terpenoids in medicinal cannabis biomass. *Metabolites*, 10(7), 276. https://doi.org/10.3390/metabo10070276.

[101] McRae, G., & Melanson, J. E. 2020. Quantitative determination and validation of 17 cannabinoids in cannabis and hemp using liquid chromatography-tandem mass spectrometry. *Analytical and bioanalytical chemistry*, 412(27), 7381-7393. https://doi.org/10.1007/s00216-020-02862-8

[102] Baños, R. V., Fonseca, M. T., & Alvarez, M. R. (2019). Multiple regression analysis using SPSS Statistics: A practical example. *Revista d'Innovació i Recerca en Educació*, 12(2), 1. https://doi.org/10.1344/reire2019.12.222704

[103] Abbas, S., Albarakati, W., Benchohra, M., & Nieto, J. J. (2016). Global convergence of successive approximations for partial Hadamard integral equations and inclusions. *Computers & Mathematics with Applications*, 10. Accessed 11 03, 2021. https://www.usc.gal/en/institutos/matematicas/publi_imat.html.

[104] Cantisano, G. T., Domínguez, J. F. M., & Depolo, M. (2008). Psychological contract breach and outcomes: Combining meta-analysis and structural equation models. *Psicothema*., 487-496. Accessed 11 03, 2021. https://reunido.uniovi.es/index.php/PST/article/view/8687

[105] Villardón, M. P. G., Villardón, J. L. V., Díaz, A. D., Galindo, P. V., & Alonso, M. C. P. (2010). An alternative to chaid segmentation algorithm based on entropy. *Revista de Matemática: Teoría y Aplicaciones*, 17(2), 179-197. Accessed 11 03, 2021. https://www.redalyc.org /articulo.oa?id=45326933005

[106] Kruk, C., Segura, A. M., Costa, L. S., Lacerot, G., Kosten, S., Peeters, E. T., ... & Scheffer, M. (2017). Functional redundancy increases towards the tropics in lake phytoplankton. *Journal of Plankton Research*, 39(3), 518-530. https://doi.org/10.1093/plankt/fbw083

[107] Al Bakain, R. Z., Al-Degs, Y. S., Cizdziel, J. V., & Elsohly, M. A. (2020). omprehensive classification of USA cannabis samples based on chemical profiles of major cannabinoids and terpenoids. *Journal of Liquid Chromatography & Related Technologies*, 43(5-6), 172-184. https://doi.org/10.1080/10826076.2019.1701015

[108] Isibor, P. O., Imoobe, T. O. T., Enuneku, A. A., Akinduti, P. A., Dedeke, G. A., Adagunodo, T. A., & Obafemi, D. Y. (2020). Principal components and hierarchical cluster analyses of trace metals and total hydrocarbons in gills, intestines and muscles of Clarias gariepinus (Burchell, 1822). *Cientific reports*, 10(1), 1-15. https://doi.org/10.1038/s41598-020-62024-9

[109] Łuczaj, W., Dobrzyńska, I., Wroński, A., Domingues, M. R., Domingues, P., & Skrzydlewska, E. (2020). Cannabidiol-mediated changes to the phospholipid profile of UVB-irradiated keratinocytes from psoriatic patients. *International journal of molecular sciences*, 21(18), 6592. https://doi.org/10.3390/ijms21186592

[110] Giupponi, L., Leoni, V., Pavlovic, R., & Giorgi, A. (2020). Influence of altitude on phytochemical composition of hemp inflorescence: A metabolomic approach. *Molecules*, 25(6), 1381. https://doi.org/10.3390/molecules25061381

[111] Chung, K., & Jung, H. (2020). Knowledge-based dynamic cluster model for healthcare management using a convolutional neural network. *Information Technology and Management*, 21(1), 41-50. https://doi.org/10.1007/s10799-019-00304-1

[112] Franchina, F. A., Dubois, L. M., & Focant, J. F. (2020). In-depth cannabis multiclass metabolite profiling using sorptive extraction and multidimensional gas chromatography with low-and high-resolution mass spectrometry. *Analytical Chemistry*, 92(15), 10512-10520. https://doi.org/10.1021/acs.analchem.0c01301

[113] Santos, R., Santos, P., Sharan, P. and, Rodriguez, C. (2021).. "Digital Agglomeration in the Improvement of the Human Development Index in Peru," *2021 IEEE 9th Region 10 Humanitarian Technology Conference (R10-HTC)*, Bangalore, India, 2021, pp. 01-07, doi: 10.1109/R10-HTC53172.2021.9641710.

[114] Marroquín, P., Peralta, P., Rodriguez, C. (2022). NEKO: Proposal of the first super-Agile methodology to improve work efficiency *5th International Conference on Computing and Informatics, ICCI* 2022, 2022, pp. 15–21. DOI: 10.1109/ICCI54321.2022.9756085