RESCUE OF CACAO GENETIC RESOURCES RELATED TO THE NACIONAL VARIETY: SURVEYS IN THE ECUADORIAN AMAZON (2010-2013)

RESCATE DE LOS RECURSOS GENÉTICOS DEL CACAO RELACIONADOS CON LA VARIEDAD NACIONAL: EXPLORACIÓN EN LA AMAZONÍA ECUATORIANA (2010-2013)

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ABSTRACT

Traditional cocoa plantations in Ecuador are mostly composed of a complex mix of highly variable hybrid progenies, which has greatly reduced the population of native trees of the "Nacional" variety, to such a point that they are considered today as heading for extinction, which is increasingly worrying the international chocolate industry. Some years ago, we used genetic molecular markers to identify trees considered to be relics of the ancient original population of the "Nacional" variety, and some wild cocoa trees in a particular region of the southern Ecuadorian Amazon were identified as highly related to the "Nacional" variety. This paper presents the results of two surveys carried out in the southern Ecuadorian Amazon, in the Zamora-Chinchipe Province, in 2010 and 2013. The objective of these surveys was to search for, identify and rescue cocoa trees that might be the wild ancestors of the "Nacional" variety. In 2010, 83 mother trees were collected (budwood, pods and leaves) and 48 in 2013. They were preserved at the Granja Domono experimental farm, near Macas (Morona-Santiago province) and at the Tropical Experimental station Pichilingue, near Quevedo. The trees collected are currently being characterized for their genetic diversity, using molecular markers, and for the biochemical diversity of their beans.

Keywords: Cocoa, genetic diversity, Nacional cocoa variety, Zamora Chinchipe, Ecuador.

RESUMEN

Las plantaciones tradicionales de cacao están compuestas en su mayoría de una mezcla compleja de progenies híbridas con un alto grado de variabilidad, lo que ha reducido las poblaciones de árboles nativos de la variedad "Nacional", a tal magnitud que ahora se consideran en vía de extinción, lo que preocupa a la industria chocolatera mundial. Hace algunos años, se utilizaron marcadores genéticos moleculares para identificar árboles considerados como reliquias de la población original antigua de la variedad "Nacional", y algunos árboles silvestres de cacao de una región particular del sur de la Amazonía Ecuatoriana fueron identificados como altamente relacionados con la variedad "Nacional". Este artículo presenta los resultados de dos exploraciones realizadas en el sur de la Amazonia Ecuatoriana, en la provincia Zamora-Chinchipe, en los años 2010 y 2013. El objetivo de estas exploraciones fue buscar, identificar y rescatar los árboles de cacao que pudieran ser los ancestros silvestres de la variedad "Nacional". En el 2010, 83 árboles madres fueron recolectados (yemas, mazorcas y hojas) y 48 en el 2013. Las muestras fueron preservadas en la granja experimental Granja Domono, cerca de Macas (Provincia de Morona Santiago) y en la Estación Experimental Tropical Pichilingue, cerca de Quevedo. En la actualidad, los árboles recolectados están siendo caracterizados en su diversidad genética mediante la utilización de marcadores moleculares y también están siendo caracterizados en cuanto a la diversidad bioquímica de sus granos.

Palabras clave: Cacao, diversidad genética, variedad de cacao Nacional, Zamora Chinchipe, Ecuador.

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INTRODUCTION

Ecuador produces more than half of the fine and flavour cocoa marketed annually in the world. The South American country draws this advantage from a local variety that was massively planted by settlers in the lowland of Ecuador (in the upper reaches of the Guayas river) from the beginning of the 17th century. This variety was later named "Nacional".

Various hypotheses have been put forward as to the origins of the "Nacional" variety and its links with the indigenous peoples of Ecuador (Lerceteau *et al.* 1997), and all agree that the origins and uses of this variety date back much further than the arrival of the Spanish.

At the moment, traditional cocoa plantations in Ecuador are mostly composed of a complex mix of highly variable hybrid progenies, which has greatly reduced the population of native trees of the "Nacional" variety, to such a point that they are considered today as heading for extinction (Loor *et al.* 2009), which is increasingly worrying the international chocolate industry.

In this context, genetic molecular markers have been used to identify trees considered to be relics of the ancient original population of the "Nacional" variety (Loor *et al.* 2009). After analysis of wild material collected in Amazonia by Allen and Lass (1983), some wild trees were identified as highly related to the "Nacional" variety in a particular region of the southern Ecuadorian Amazon, considered now as its potential region of origin and domestication (Loor *et al.* 2012).

These discoveries are of paramount importance, be it scientifically, or economically and socially. Indeed, first and foremost, they enable a more systematic study of the variability of this variety in its natural environment, thereby making it possible to gain better knowledge of its origins and evolution. Then, by conserving and using these wild trees in breeding programmes, it will be possible to develop some new improved "Nacional" cocoa cultivars that can then be recommended for new plantations.

Given the situation and these prospects, the Ecuadorian Amazon holds a privileged position, as it is one of the regions in the world that shelters the greatest genetic diversity of the T. cacao species (Bartley, 2005), be it in its wild or domesticated state, which is reflected in the fact that representatives of all commercial types of cocoa can be encountered in the same zone, along with others which, although potentially rich in sensory terms, are still unknown or little used at local level. On the other hand, the galloping degradation of its primary forest is a cause for global concern, as is the obvious loss of many wild cocoa trees of great interest.

The aim of this paper is to present the results of two surveys carried out in the southern Ecuadorian Amazon following the work by Loor *et al* (2012), in order to search for, identify and rescue some cocoa trees that might be the wild ancestors of the "Nacional" variety. By setting up living collections of those trees, it will be possible to make use, in the breeding programs, of the genetic variability encountered in the Amazonian region, to increase fine cocoa production and maintain or increase its flavour quality.

MATERIALS AND METHODS

The survey zones

The region surveyed was a vast, heterogeneous territory identified as being the most likely centre of origin for the "Nacional" variety (Loor *et al.* 2012), comprising the eastern slopes of the Andes Cordillera and southern Ecuadorian Amazonia in Zamora Chinchipe province (see figure 1). Rainforest, range from 724 to 1188 meters above sea level.

In this region, there has been substantial environmental degradation, which has been transformed from an immense primary tropical rainforest into a region of large deforested zones, mainly due to cattle farming and the wood and mineral industries, where some small woodlands survive as the final and precarious remains of what was once a large virgin forest. It is precisely inside these forest remnants that some large and old cocoa trees can be found. Two surveys were successively undertaken in 2010 and 2013:

In 2010: The surveys were carried out in August and involved six cantons of Zamora-Chinchipe province: El Pangui, Yantzaza, Yacuambi, Centinela del Condor, Zamora and Nangaritza. The Shuar communities in the zone were informed in detail about the purpose of the surveys and involved in the plant collections.

Figure 1. Ecuador map (left), Zamora Chinchipe province (right) with the zones surveyed and the mother trees collected.

The first surveys took place in the valleys of the Yacuambi, Zamora and Nangaritza rivers (near La Saquea, La Paz, Zumbi, El Pangui, Paquisha, Bella Vista and Nankais). While, the second part of the survey was mainly conducted in the vicinity of the villages of Shaime and Nuevo Paraiso, along the Nangaritza, Numpatakaime and Shamatak rivers.

The collected material was transported to and preserved at the experimental farm Domono, near Macas (Morona-Santiago province) and some of the budsticks were sent to the Pichilingue tropical experimental station, near Quevedo.

In 2013: The surveys from the same province, involved the cantons of Palanda and Chinchipe, in the vicinity of Palanda and San Francisco del Vergel, then Zumba and Chito. The material collected was sown and grafted at Pichilingue in the days following the prospections.

Methodologies followed during the collections

During the two surveys, for each mother tree of interest, we collected:

- Young budsticks of a size suitable for side cleft grafting on rootstocks around 3 months old, a technique adopted by INIAP (Molina 2008; Anon. 2012), with paraffin applied to the tips and protected in isothermal bags (Lachenaud and Sallée 1993)

- Pods, where available, for sowing (as a priority) and, if sufficient, for micro fermentations. The fermented (in "Rohan trays"; Jiménez *et al.* 2011), dried cocoa was sent to CIRAD in Montpellier (France) for biochemical analysis of the aromatic compounds and, where appropriate, sensory analyses. The micro fermentations were carried out at Granja Domono and Pichilingue in 2010, and at Pichilingue in 2013.

- A few leaves, for further microsatellite molecular marker analysis of the diversity collected (Pugh *et al.* 2004), and the genetic kinship of the collected material with the 'Nacional' variety. The leaves were vacuum packed (in 2013) or dried (in 2010) and sent to CIRAD in Montpellier.

- For each tree encountered, the location was recorded using a GPS, along with some morphological data used as descriptors (architecture, pods, flowers), as well as sanitary and environmental data (habitat, stand size, topology and pedology).

Rescuing the planting material

As indicated, in 2010 the collected material was preserved, i.e. grafted and sown, at Granja Domono and Pichilingue, and then planted in plots; in 2013, it was all preserved at Pichilingue.

RESULTS AND DISCUSSION

2010 surveys

Pods could not be collected from all the mother trees as the climate in 2010 was particularly dry and the main harvest peak was earlier than expected. The exceptionally low level of the Nangaritza river also disrupted the scheduled survey plan, by preventing travel by water.

Table 1 presents the 83 mother trees collected, of which only 39 bore pods. The surveyed zones (Figure 1) were as follows:

Table 1. Identification, location (coordinates in degrees, minutes and seconds), elevation, habitat, morphology of the 83 mother-trees (accessions) collected in August 2010, and the stand to which they belonged. FT = a few trunks, ST = single trunk, T = tuft, BiT = bi-trunk, WB = witches' broom, FPR = frosty pod rot. In the "Observation" column, "W-B" means "White and purple beans". (N/A = not available)

Accession	Lat. S	Long. W	Elevation	Habitat	Stand	Architecture	Height	Pods harvested	Shape	Diseases	Observations
ZAMO 001	03.51.37	78.45.00	829	grazing land	3	FT	10	1	Nacional	WB, FPR	White beans
ZAMO 002	03.51.37	78.45.00	829	grazing land	3	FT	8	1	Nacional	WB, FPR	Purple beans
ZAMO 003	03.51.36	78.44.39	1003	Forest	Alone	T	9	3	Nacional	0	W-P beans
ZAMO 004	03.51.35	78.44.38	1033	Forest	Alone	T	8	1	Nacional	0	Purple beans
ZAMO 005	03.51.33	78.44.40	1033	Forest	Alone	FT	10	4	Nacional	0	Purple beans
ZAMO 006	03.51.33	78.44.40	1033	Forest	Alone	T	8	0	N/A	WB, FPR	
ZAMO 007	03.51.32	78.44.42	1015	Forest	Alone	FT	10	4	Nacional	0	White beans
ZAMO 008	03.51.31	78.44.43	1010	Forest	Alone	FT	13	0	Nacional	0	
ZAMO 009	03.51.28	78.44.45	1005	Forest	Alone	FT	15	1	Nacional	0	White beans
ZAMO 010	03.51.28	78.44.47	989	Forest	Alone	FT	16	1	Nacional	FPR	
ZAMO 011	03.51.25	78.44.49	985	Forest	4	ST	8	3	Nacional	WB	Purple beans
ZAMO 012	03.51.24	78.44.49	953	Forest	Alone	T	15	3	Nacional	WB	
ZAMO 013	03.51.22	78.44.47	949	Forest	Alone	FT	9	1	Nacional	Phytophthora	W-P beans
ZAMO 014	03.51.20	78.44.49	892	Forest	2	FT	12	5	Nacional	0	W-P beans
ZAMO 015	03.51.20	78.44.49	892	Forest	2	FT	9	3	Nacional	0	W-P beans
ZAMO 016	03.51.14	78.44.54	858	Forest	Alone	ST	8	4	Nacional	0	W-P beans

Continue...

PANG 001	03.32.56	78.36.38	858	grazing land	Alone	FT	8	1	Nacional	0	W-P beans
PANG 002	03.33.01	78.36.38	878	grazing land	Alone	BiT	N/A	3	Nacional	Phytophthora	W-P beans
PANG 003	03.33.03	78.36.42	888	grazing land	2	ST	10	0	N/A	N/A	
PANG 004	03.33.04	78.36.41	935	grazing land	2	T	15	1	Nacional	N/A	
PANG 005	03.33.04	78.36.42	942	grazing land	Alone	T	9	0	N/A	N/A	
PANG 006	03.33.05	78.36.44	947	grazing land	Alone	T	18	0	N/A	N/A	
PANG 007	03.33.05	78.36.45	945	Forest	Alone	T	8	1	Nacional	Phytophthora, WB	
PANG 008	03.33.05	78.36.47	956	Forest	3	T	8	1	Nacional	WB	W-P beans
PANG 009	03.33.05	78.36.46	956	Forest	3	FT	8	0	N/A	WB	
PANG 010	03.33.05	78.36.47	956	Forest	3	BiT	15	0	N/A	N/A	
PANG 011	03.33.05	78.36.47	956	Forest	Alone	T	18	0	N/A	N/A	
PANG 012	03.33.04	78.36.45	960	Forest	Alone	T	20	0	N/A	N/A	
PANG 013	03.33.03	78.36.47	992	Forest	Alone	T	12	0	N/A	N/A	
PANG 014	03.33.03	78.36.48	997	Forest	Alone	T	> 20	0	N/A	WB	
PANG 015	03.33.02	78.36.49	1024	Forest	10	FT	10	0	N/A	N/A	
PANG 016	03.33.03	78.36.49	1010	Forest	10	FT	10	1	Nacional	N/A	
PANG 017	03.33.03	78.36.49	1010	Forest	10	FT	8	2	Nacional	N/A	W-P beans
PANG 018	03.33.03	78.36.50	1011	Forest	10	BiT	> 20	0	N/A	N/A	
PANG 019	03.33.00	78.36.51	1015	Forest	Alone	FT	15	0	N/A	N/A	
PANG 020	03.32.58	78.36.51	1015	Forest	Alone	ST	15	0	N/A	WB	
PANG 021	03.32.58	78.36.51	1015	Forest	Alone	T	15	1	Nacional	WB	
PANG 022	03.32.53	78.36.50	972	Forest	Alone	T	15	1	Nacional	N/A	W-P beans
PANG 023	03.32.52	78.36.50	967	Forest	Alone	FT	10	18	Nacional	N/A	W-P beans
PANG 024	03.32.46	78.36.48	935	Forest	Alone	ST	8	5	Nacional	0	W-P beans
YACU 001	03.54.50	78.50.55	837	Friche	Alone	BiT	4	3	Nacional	Phytophthora	W-P beans
YACU 002	03.41.52	78.54.14	1064	grazing land	10	T	6	0	N/A	WB	
YACU 003	03.41.52	78.54.14	1064	grazing land	10	T	8	0	Nacional	WB	
YACU 004	03.42.03	78.54.17	1056	grazing land	Alone	ST	6	0	N/A	0	
YACU 005	03.42.03	78.54.17	1056	Ravine	2	T	8	0	N/A	0	
YACU 006	03.42.04	78.54.11	1085	Ravine	2	T	8	0	N/A	0	
YACU 007	03.42.02	78.54.10	1054	grazing land	Alone	FT	9	0	N/A	0	
YACU 008	03.42.02	78.54.08	1055	grazing land	Alone	ST	5	0	N/A	0	
YACU 009	03.42.03	78.54.08	1044	grazing land	Alone	ST	5	0	N/A	N/A	
YACU 010	03.42.03	78.54.04	1006	grazing land	Alone	T	18	0	N/A	N/A	
YACU 011	03.46.27	78.53.39	825	grazing land	Alone	FT	10	0	N/A	WB	
YACU 012	03.47.10	78.54.20	882	grazing land	3	ST	8	1	Nacional	WB	Purple beans
YACU 013	03.47.10	78.54.20	882	grazing land	3	ST	8	0	N/A	WB	
YACU 014	03.47.10	78.54.20	882	grazing land	3	ST	10	0	N/A	WB	
YACU 015	03.46.57	78.54.17	895	grazing land	3	FT	8	0	N/A	WB	
YACU 016	03.46.56	78.54.16	902	grazing land	3	FT	7	0	N/A	WB	
YACU 017	03.46.56	78.54.16	902	grazing land	3	ST	5	0	N/A	WB	
YACU 018	03.47.00	78.54.10	894	grazing land	Alone	FT	12	2	Nacional	N/A	W-P beans
YACU 019	03.47.00	78.54.09	887	grazing land	Alone	FT	10	0	N/A	N/A	
YACU 020	03.54.47	78.51.00	955	Fallow	Alone	BiT	8	0	N/A	WB	
YACU 021	03.59.50	78.51.00	873	Fallow	Alone	T	6	1	Nacional	0	W-P beans
BEVI 001	03.52.09	78.40.35	844	Forest	4	FT	9	3	Nacional	WB	Purple beans
BEVI 002	03.52.09	78.40.35	844	Fallow	4	N/A	8	3	Nacional	N/A	Purple beans
NANK 001	03.46.30	78.39.27	842	grazing land	8	BiT	15	3	Nacional	WB, Phytophthora	Purple beans
NANK 002	03.46.30	78.39.27	842	grazing land	8	FT	12	3	Nacional	N/A	Purple beans
NANK 003	03.46.30	78.39.27	842	grazing land	8	BiT	12	3	Nacional	0	Purple beans
NANK 004	03.46.37	78.39.29	854	grazing land	Alone	FT	15	3	Nacional	WB	White beans
NANK 005	03.47.06	78.40.27	842	grazing land	Alone	FT	12	5	Nacional	WB	W-P beans
NANK 006	03.48.39	78.41.07	838	Crop	Alone	N/A	12	3	Nacional	0	Purple beans

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PAQU 001	03.55.48	78.40.54	815	Fallow	Alone	FT	12	0	N/A	0	Pink flower peduncle
SHAI 001	04.20.24	78.39.03	924	Cleared zone	3	T	1.5	0	N/A	0	
SHAI 002	04.20.25	78.39.03	962	Cleared zone	3	ST	5	0	N/A	0	
SHAI 003	04.20.25	78.39.03	962	Cleared zone	3	ST	1.5	0	N/A	0	
SHAI 004	04.20.19	78.39.22	925	Forest	Alone	ST	5	0	N/A	0	
SHAI 005	04.19.52	78.39.52	802	Forest	Alone	FT	7	1	Nacional	0	
NUPA 001	04.22.21	78.39.47	914	Crop	4	FT	10	4	Nacional	WB	
NUPA 002	04.22.21	78.39.47	914	Crop	5	T	8	0	N/A	WB	
SHAM 001	04.19.57	78.41.19	925	Fallow	3	T	3	0	N/A	0	
SHAM 002	04.19.57	78.41.19	925	Fallow	3	T	12	0	N/A	0	
SHAM 003	04.19.57	78.41.19	925	Fallow	3	T	10	0	N/A	0	
SHAM 004	04.19.57	78.41.19	925	Fallow	Alone	ST	Nc	0	N/A	0	Red pods
GUAY 001	04.05.05	78.40.00	893	grazing land	3	T	12	0	N/A	WB	
GUAY 002	04.05.03	78.40.01	887	grazing land	3	FT	10	0	N/A	WB	

- Towards Zumbi, between the Zamora and Nangaritza rivers: accessions Zamo-001 to Zamo-016
- Towards El Pangui: accessions Pang-001 to Pang-024
- On the Yacuambi river, towards La Paz (around the Shuar village of Kiim): accessions Yacu-002 to 019, and towards La Saquea: accessions Yacu-001 and Yacu-020, 021
- On the Nangaritza river, towards Guazimi (accessions Guay-001 and 002), towards Paquisha (accession Paqu-001), towards Bella Vista (accessions Bevi-001 and 002) and towards Nankais (accessions Nank-001 to 004, along the Tses-entse river; Nank 005 and 006, on the roadside)
- Further upstream on the Nangaritza river, towards Shaime (accessions Shai-001 to 005)
- On the Numpatakaime river, towards Nuevo Paraiso (accessions Nupa-001 and 002)
- On the Shamatak river, not far from Shaime (accessions Sham-001 to 004).

The surveys and collections were fruitful, despite the end of the main harvest, apart from in the Bella Vista zone and especially the Nuevo Paraiso zone, where we only found a few cocoa trees.

The main traits of the places and collected trees were as follows:

- Often found on grazing land, sometimes in more or less degraded forests, on generally steep slopes and at a high average elevation (from 800 to 1,100 m). Some of the cocoa trees had recently been cut back by the farmers and were therefore greatly threatened; this situation could be explain by the land use change and expansion of intensive agriculture (Sala *et al.* 2006). For other hand, these changes of land could lead to habitat loss for more species and can even drive species decline and extinction (Vebrova *et al.* 2014).
- Even though many of the collected cocoa trees were not cultivated, it is impossible to reach any conclusion, at this stage, as to their wild nature. The elevations of our collections (up to 1,188 m) corresponded to the cultivation limits cited by Wood & Lass (1985).
- Age generally (according to appearance) old and height sometimes exceeding 20 m.
- Pods mostly of the "Nacional" shape, or similar follows. Almost all pods were green, ripening to yellow, but the color of seeds per pod was more variable than other character. This offers insight into relationships among the collected materials, and provide direct evidence of important commercial traits (Bartley 2005).
- Seeds of variable size and colour showed a great diversity inside and among fruits collected, pods with a mixture of white and purple seeds, pods with only white seeds follows, as reported by Allen and Lass (1983), or pods with seeds sometimes small or very flattened.
- Many diseases (witches' broom, frosty pod rot, black pod rot) and an often highly degraded physiological condition, with many deficiency symptoms and often numerous parasites.

The collected budsticks and pods (from 83 and 39 mother trees, respectively) enabled 1,370 grafts to be carried out and 1,106 open progenies were sown at Granja Domono and Pichilingue. Some bean micro-fermentations were carried out for 15 origins.

Photo 1. Cocoa fruits (pods and seeds) showing the diversity among some collected materials.

2013 surveys

Table 2 presents the 48 mother trees collected in 2013, of which 33 bore pod.

The surveyed zones (Figure. 1) were as follows:

- In the vicinity of Palanda:
  - Towards the La Florida archaeological site, along the Valladolid river: accessions PAL 1 to 7
  - At "La Mina": accessions PAL 8 and 9
  - At "La Cuesta de Santa Ana": accessions PAL 15 to 19
  - Near the town, along the Palanda river, accessions PAL 20 to 24.
- Towards San Francisco del Vergel: accessions PAL 10 (at Santa Clara del Vergel) and PAL 11 to 14
- In the vicinity of Zumba:
  - At "La Hoya del cacao": accessions PAL 25 to 28, and nearby, PAL 29 to 32
  - At Huamchunangui: accessions PAL 33 to 37
  - At La Gayusa: accessions PAL 38 to 40
  - At Isimanchi: accessions PAL 44 to 48
- Towards Chito: accessions PAL 41 to 43 (at La Fortuna, along the San Francisco river, on the border with Peru).

Unlike the collections in 2010, those in 2013 all involved cultivated trees, included in plots, or very close to dwellings. Some trees of the Trinitario type (red) were even found (but not collected). The pod shape was "Nacional" in 13 cases out of 35 (i.e. 37%), and Angoleta, Amelonado or "Other" in 22 cases follows. The general condition of the trees was better than those surveyed in 2010 (fewer diseases and deficiencies).

The collected budsticks and pods (from 48 and 33 mother trees, respectively) enabled 643 grafts to be carried out and 2,150 open progenies were sown at Pichilingue. Some bean micro-fermentations were carried out for 27 origins.

Table 2. Identification, location (UTM-UPS coordinates, in the 17M square of the World Geodetic System), elevation, habitat, morphology of the 48 mother trees (accessions) collected in May 2013. FT = a few trunks, ST = single trunk, T = tuft, BiT = bi-trunk, WB = witches' broom. The size of the stands and tree heights are evaluations. The elevations, provided by a GPS, are approximations.

Accession	GPS East	GPS North	Elevation	Habitat	Stand	Architecture	Height	Pods harvested	Shape	Diseases	Observations
PAL 1	707385	9487090	979	Crop	5	T	10	9	Nacional	Wb	
PAL 2	707385	9487090	979	Crop	5	T	8	3	Nacional	Wb	
PAL 3	707392	9487092	979	Crop	5	T	7	10	Other	Wb	
PAL 4	707392	9487092	979	Crop	5	FT	5	10	Nacional		
PAL 5	707472	9487355	985	Crop	10	ST	4	8	Angoleta	Wb	
PAL 6	707497	9487374	993	Crop	30	FT	8	5	Angoleta		
PAL 7	707497	9487374	993	Crop	30	FT	5	5	Amelonado		
PAL 8	707791	9488780	1015	Grazing land	2	FT	7	9	Other		
PAL 9	707769	9488812	1019	Grazing land	2	T	4	4	Other		
PAL 10	718399	9494627	1133	Crop	Nb	FT	10	4	Nacional		
PAL 11	716849	9482131	1181	Crop	30	FT	10	6	Nacional		
PAL 12	716850	9482109	1187	Crop	30	ST	6	7	Other		
PAL 13	716850	9482118	1188	Crop	30	FT	10	8	Other	0	Pale beans
PAL 14	716850	9482118	1188	Crop	30	FT	8	8	Nacional		
PAL 15	707384	9487961	1080	Crop	5	T	10	0		0	
PAL 16	707243	9488112	1132	Forest	3	T	12	0	Amelonado	0	
PAL 17	707273	9488134	1129	Crop	10	T	12	0		Wb	
PAL 18	707273	9488134	1129	Crop	10	T	12	0		Wb	
PAL 19	707259	9488113	1130	Crop	15	T		0		Wb	
PAL 20	707617	9486443	975	Crop	20	T	6	0		Wb	Pale flushes
PAL 21	707626	9486419	977	Fallow	20	T	15	3	Other	Wb	
PAL 22	707621	9486463	978	Fallow	20	T		0	Other	Wb	
PAL 23	707422	9485906	1063	Crop	3	T	7	0	Other	Wb	Red staminodes
PAL 24	707422	9485906	1063	Crop	3	T	5	1	Other	Wb	
PAL 25	708103	9460158	1150	Crop	4	T	8	7	Nacional	Phytophthora	
PAL 26	708105	9460145	1150	Crop	4	T	5	10	Nacional	0	
PAL 27	708092	9460177	1087	Crop	4	FT	6	1	Nacional	0	
PAL 28	708092	9460177	1079	Crop	4	T	7	0		0	
PAL 29	707586	9460322	1129	Crop	1	FT	6	2	Other	Wb	
PAL 30	707323	9460554	1105	Grazing land	3	FT	5	3	Angoleta	0	Deficiencies
PAL 31	707323	9460554	1105	Grazing land	3	FT	7	2	Other		
PAL 32	707323	9460554	1105	Grazing land	3	FT	8	0	Other		Deficiencies
PAL 33	703610	9454260	1044	Crop	45	T	6	1	Other	Wb	Almost dead
PAL 34	703610	9454260	1044	Crop	45	FT	8	2	Nacional	Wb	Pale flushes
PAL 35	703633	9454282	1044	Crop	45	FT	12	3	Nacional	Wb	
PAL 36	703564	9454042	1180	Crop	300	ST	8	7	Amelonado		
PAL 37	703572	9454029	1086	Crop	300	BiT	15	7	Angoleta		Pale flushes
PAL 38	704134	9454592	1028	Crop	30	T	12	4	Other	Wb	
PAL 39	704134	9454592	1028	Crop	30	T	10	5	Amelonado	Wb	Watery pod rot?
PAL 40	704122	9454641	1048	Crop	> 20	FT	6	5		Wb	Pink flushes
PAL 41	722464	9451467	865	Forest	10	FT	10	0			Steep slope
PAL 42	722464	9451467	865	Forest	10	ST	6	0			Pale flushes
PAL 43	722467	9451539	864	Forest	3	FT		0			
PAL 44	708584	9465793	880	Crop	3	FT	10	9	Nacional		
PAL 45	708584	9465793	880	Crop	3	FT	10	8	Nacional		
PAL 46	711490	9463787	724	Forest	10	T		1	Amelonado		
PAL 47	711473	9463789	731	Forest	10	FT	6	0		Wb	
PAL 48	709743	9454418	837	Crop	6	FT	10	0			

CONCLUSION

The outcome of these two survey and collection campaigns in Zamora-Chinchipe province proved positive, as it allowed rescue very old cacao trees that show morphological characteristics of the Nacional variety.

A total of 131 mother trees were collected and rescued in living collections at two strategic locations, under INIAP control: the experimental farm "Granja Domono" and the tropical experimental station "Pichilingue". More than 35% of the collected trees were of the typically 'Nacional' phenotype (pods and flowers), especially trees Zamo-014, 015, Pang-008, 024, Yacu-021, Nank-005, Pal 1, 2, 4, 10, 11, 14, 25, 26, 27, 34, 35, 44 and 45.

17% of the accessions collected had pale, sometimes white and even sometimes totally white seeds, like trees Zamo-001, Zamo-007, Zamo-009 and Nank-004.

The living collection at Pichilingue and Domono are cu-

rrently being characterized for its genetic diversity, using molecular markers, and for the biochemical diversity of the beans.

These surveys and collections prove essential for safeguarding the province's cocoa heritage, which is under serious threat from agriculture (mainly livestock farming) and diseases, and also for identifying possible ancestors of the 'Nacional' cocoa tree. Studies on the genetic variability of the collected material and its closeness to the 'Nacional' type may possibly reveal the need for further surveys in targeted sectors.

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