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Source: Mountain Research and Development, 38(1) : 24-34

Published By: International Mountain Society

URL: https://doi.org/10.1659/MRD-JOURNAL-D-17-00013.1

Mountain Research and Development (MRD)
An international, peer-reviewed open access journal
published by the International Mountain Society (IMS)
www.mrd-journal.org

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Prospects for Broader Cultivation and Commercialization of Copafam, a Local Variety of *Phaseolus coccineus* L., in the Brescia Pre-Alps

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The conservation and enhancement of agrobiodiversity have been promoted by local, national, and international institutions over the last few decades. In the context of that effort, this study focused on a little-known endangered landrace grown in the Brescia pre-Alps in Northern Italy: Copafam, a variety of runner bean (Phaseolus coccineus *L.). The agronomic characteristics of plants and the bromatological features of seeds harvested in 7 experimental fields, set up at different elevations in Northern Italy, were analyzed. Results showed that this landrace is most suitable for cultivation at higher elevations. As elevation increased from 110 to 1100 m above sea level, the plants became more vigorous and productive and the beans became larger and more digestible. This research not only increases knowledge of the specific landrace but can also serve as an example and stimulus for efforts to safeguard agrobiodiversity worldwide, because it suggests a number of strategies for the conservation and valorization of this particular mountain variety using existing legal and economic mechanisms for protecting agrobiodiversity.*

Keywords: *Mountain crops; agrobiodiversity; runner bean; Copafam; landrace; plant conservation; Lombardy; Italy.*

Peer-reviewed: *December 2017* ***Accepted:*** *January 2018*

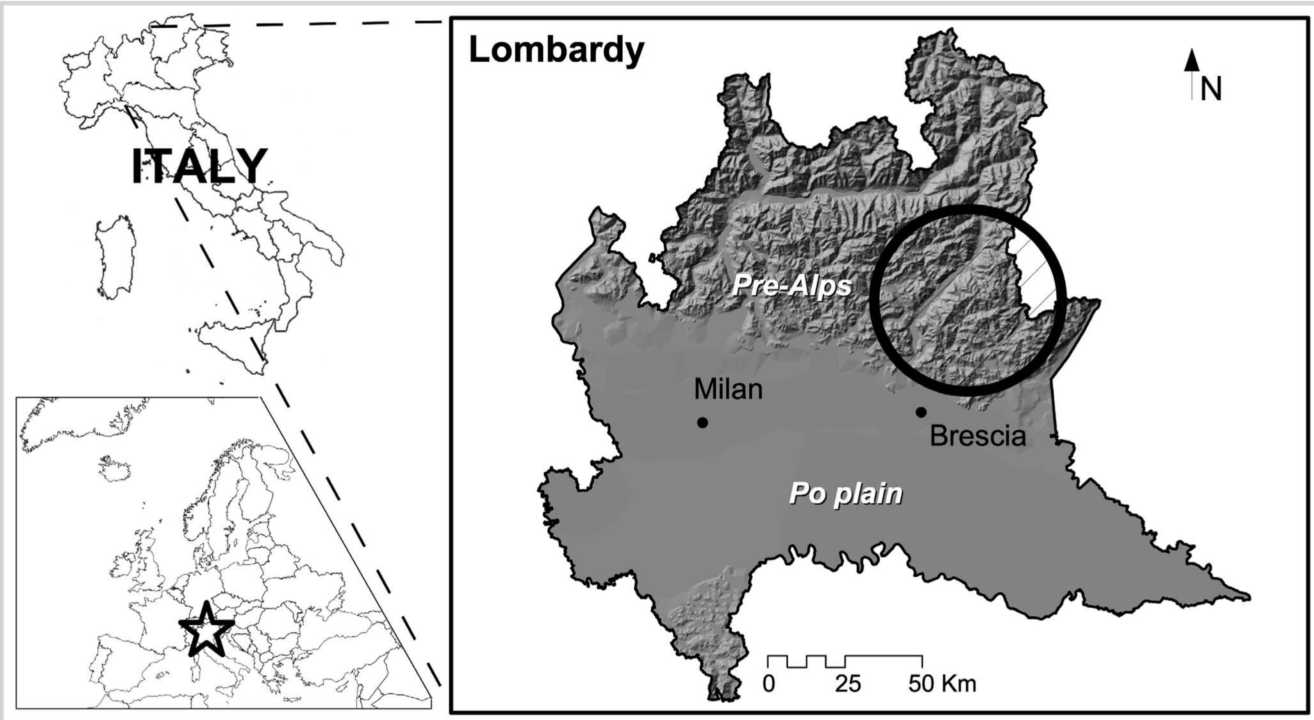
Introduction

The runner (scarlet) bean (*Phaseolus coccineus* L.) is an allogamous species of the Leguminosae family, native to Mesoamerica and widely cultivated in Europe (Giurcâ 2009) as both an ornamental plant and a food crop. It is a perennial plant with showy red or white flowers (depending on the variety) and edible pods and seeds that are larger and more digestible than those of the common bean (*Phaseolus vulgaris* L.) (Tutin et al 1968; Carlderon et al 1992; Labuda 2010). *P. coccineus,* like other bean species, was introduced in Spain in the 16th century and spread from there to other parts of Europe (Gepts and Debouk 1991; Zeven 1996; Perale 2001; Santalla et al 2004; Papa et al 2006; Spataro et al 2011). In Northern Italy, it is believed to have first been introduced in the territories of the Serenissima Republic of Venice. Nowadays, the runner bean is cultivated in several regions of northern and central Italy, mostly by small farmers who use it to prepare traditional dishes.

Molecular analysis by Acampora et al (2007), using sample accessions from different Italian regions, complemented earlier analyses (Negri and Tosti 2002; Sicard et al 2005), highlighting that Italian farmers, starting with the ancestral American runner bean, had bred their landrace (Camacho Villa et al 2005) through selection for seed size and seed coat color, but occasional gene flow maintained variability within landraces bred by different farmers in the same region. In addition, genetic studies (Spataro et al 2011; Rodriguez et al 2013) concluded that the European accessions belong to a different gene pool from the Mesoamerican one (the European continent could be regarded as a secondary diversification center for *P. coccineus*) and that the differentiation of the European gene pool could be due not only to selective pressure but also to genetic drift, differential gene flow with respect to Mesoamerica, and lack of introgression from wild forms.

A number of landraces of *P. coccineus* have been grown in Italy, with different seed sizes and seed coat colors. The landrace called Copafam, which is the focus of this study, is found in the pre-Alps of the Lombardy region in northern Italy, in particular in the Brescia pre-Alps (45°45′54″N, 10°24′10″E) (Figure 1) (Marazzi 2005). It was

FIGURE 1 Map of the region where Copafam can still be found (pre-Alps of the Lombardy region of Northern Italy). The black circle indicates the Brescia pre-Alps. (Map by Luca Giupponi)



once cultivated across a greater area but is now at risk of extinction as a result of depopulation and abandonment of mountain farms.

Copafam is a rapidly growing climbing herb that can reach 3 m in height and has many-flowered racemes (longer than leaves) and flowers with red corolla; sometimes, the wings and keel are white and, rarely, the whole corolla is white. The fruit is rough, about 15 cm long, and contains up to 5 seeds about 2.5 cm in length; the seed coat is pink, purple, beige, or white with brown or black streaks (Figure 2). According to hobby farmers in the province of Brescia, Copafam can be grown only in the mountains, because in the plains (the Po Valley), it would have problems both germinating and producing fruit. Few hobby farmers in the Brescia pre-Alps (and neighboring territories) grow Copafam and use it in the preparation of typical dishes (eg Copafam soup or pasta and beans). From an economic point of view, the runner bean (of which Copafam is a landrace) is the third most important bean species (after *P. vulgaris* and *P. lunatus*) (Santalla et al 2004), but Copafam is not available commercially; it is grown only for family use by hobby farmers who have never taken action to commercialize it. There is a risk that such local varieties and the traditions linked to them can be lost.

An estimated 75% of the genetic diversity of agricultural crops worldwide has been lost in the last decades (FAO 2004). The disappearance of local varieties is a great loss in terms of both agricultural and cultural or historical value. Therefore, local and international institutions and organizations have given special attention to the conservation of agrobiodiversity both in situ (through custodian farmers) and ex situ (in seed banks) (Love and Spaner 2007; Maxted 2013). At the international level, various conventions and treaties have aimed to improve agrobiodiversity conservation, including the Convention on Biological Diversity (CBD 1992, 2002; Bragdon 1996) and the International Treaty on Plant Genetic Resources for Food and Agriculture (FAO 2001), on which the Global Strategy for Plant Conservation is based (Sharrock et al 2014). International guidelines have been incorporated in national and local legislation in many countries.

In recent years, the Lombardy Region and the Italian Ministry of Agriculture, Food and Forestry have set up projects and established legislation aimed at safeguarding local and national agrobiodiversity, which is of fundamental importance for the Italian economy, given the wealth of varieties, breeds, and typical local products that exist throughout the country and the traditions linked to them (Barcaccia et al 2002; Cavagna et al 2012; Scarano et al 2014; Parisi et al 2016; Cassani et al 2017). In 2013, the Lombardy Region established a procedure for requesting the registration of local varieties in the European Register of Conservation Varieties (Spataro and Negri 2013) and is working to implement Italian law (1 December 2015 no. 194, known as the Cenni law) that provides for the conservation and enhancement of the biodiversity of agricultural and food interest. This law

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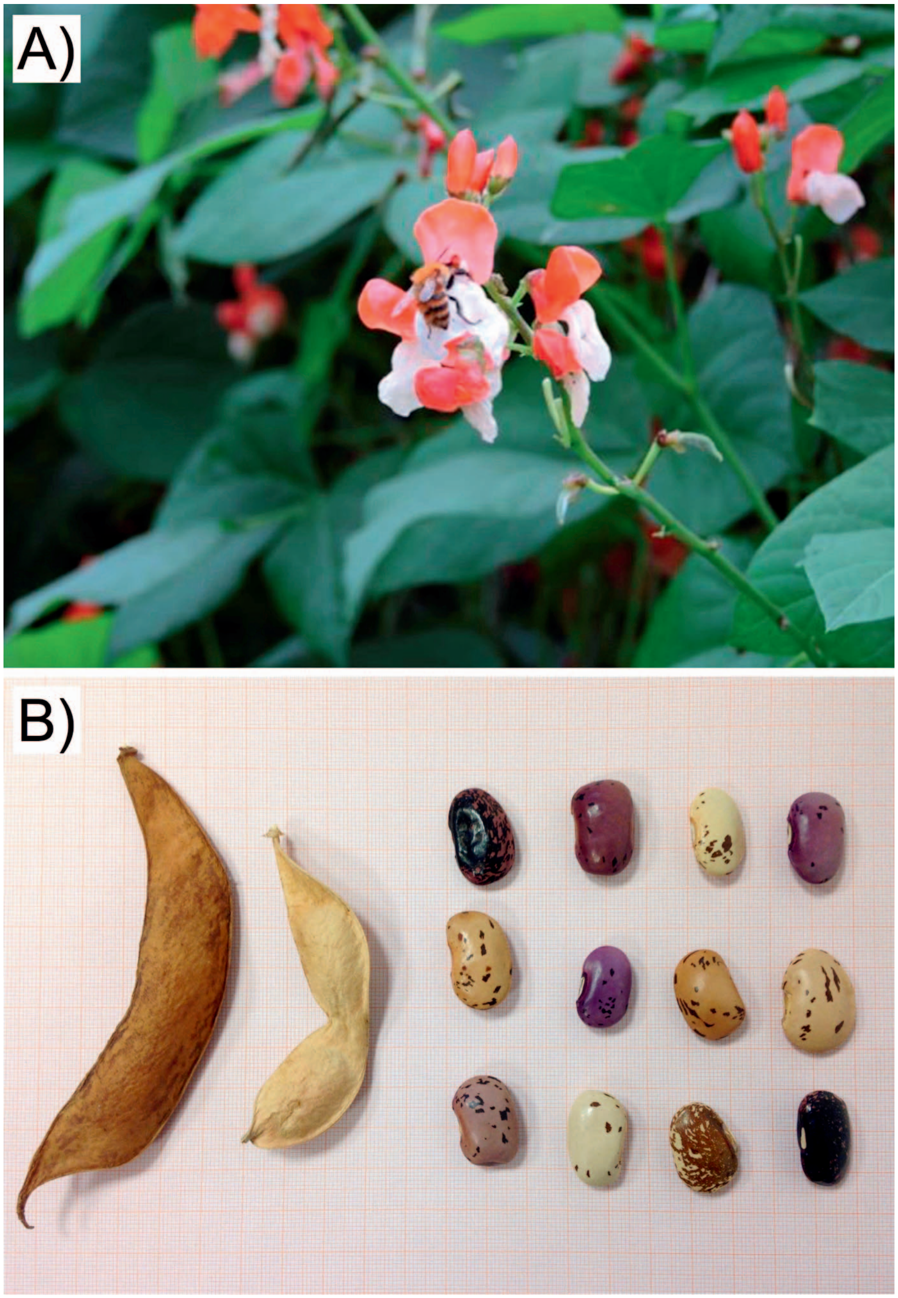
FIGURE 2 Copafam landrace. (A) Flowers; (B) pods and seeds. (Photos by S. Colombo and L. Giupponi)



provides support for efforts to conserve agrobiodiversity and establishes a registry of Italian agricultural food products such as the Copafam bean.

This study investigated the effect of elevation on the agronomic and bromatological characteristics of Copafam beans. The goals of the study were (1) to contribute to the knowledge of this little-known landrace, (2) to identify the elevations most suitable for its cultivation (and thus conservation), and (3) to explore its potential for inclusion in the Italian registry of agricultural products and for commercial exploitation by farmers in economically struggling mountain regions, such as the Lombardy pre-Alps.

Methods

Experimental fields

In spring 2015, 7 experimental fields of about 40 m^2 were set up at different elevations in the province of Brescia (Figure 3; Table 1). The fields were plowed and fertilized

FIGURE 3 Experimental field in Lavenone. (Photo by S. Colombo)

using bovine manure and then sown in the first half of May 2015 with seed sourced from a local hobby farmer. In each field, 50 seeds were sown at a 5-cm depth and 70 cm apart, in rows that were 100 cm apart (row length, 7 m; no. rows, 5). The fields were weeded and irrigated as needed throughout the production season (May to October). The soil and the plants were not treated with chemical pesticides or herbicides.

TABLE 1 Characteristics of the experimental fields.

Experimental field	Municipality	Elevation (masl)	N latitude (°)	E longitude (°)	Slope (°)	Aspect (°)
A	Toscolano Maderno	111	45.670654	10.646095	3	110
B	Rezzato	150	45.518086	10.335588	0	—
C	Vobarno	350	45.627147	10.490806	0	—
D	Agnosine	465	45.644821	10.361803	14	340
E	Pertica Bassa	785	45.751982	10.381688	9	200
F	Lavenone	1000	45.766666	10.406483	16	120
G	Lavenone	1100	45.770008	10.405506	7	110

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TABLE 2 Mean monthly and annual temperature and precipitation recorded by 4 weather stations in the Brescia pre-Alps at different elevations in 2015.

Month	Temperature (°C)				Precipitation (mm)			
	Weather station (elevation)				Weather station (elevation)			
	Brescia (125 masl)	Sarezzo (290 masl)	Bione (911 masl)	Treviso Bresciano (1147 masl)	Brescia (125 masl)	Sarezzo (290 masl)	Bione (911 masl)	Treviso Bresciano (1147 masl)
January	3.7	0.0	3.4	1.7	27.6	53.4	82.4	73.2
February	4.2	0.1	2.2	0.7	139.4	125.6	106.8	79.0
March	5.8	8.9	6.3	4.2	21.6	16.2	27.8	30.6
April	14.6	12.9	10.1	8.3	37.0	49.4	50.8	44.2
May	21.2	17.5	14.3	12.5	54.8	86.0	60.0	68.6
June	26.2	21.4	18.3	16.6	83.8	72.0	41.8	56.6
July	29.5	26.5	23.6	21.5	6.8	46.6	34.6	34.8
August	26.3	22.9	20.3	18.6	45.4	74.2	108.8	115
September	20.4	17.7	14.7	12.8	52.8	36.2	60.6	73.6
October	13.6	12.4	10.0	8.4	113.2	170.8	206	227.0
November	8.3	7.1	8.6	7.3	14.2	9.0	17.8	14.8
December	3.6	3.3	6.0	4.9	2.0	2.2	0.4	0.2
Annual[a)]	14.8	12.5	11.5	9.8	598.6	741.6	797.8	817.6

[a)] Annual values are averages for temperature and totals for precipitation.
Source: ARPA 2017.

Annual and monthly temperature and precipitation data were obtained from 4 weather stations at different elevations in the same geographical area (Table 2). In this area, the average annual temperature decreased by about 0.4°C per 100 m of elevation, while the average annual precipitation increased by about 16 mm.

Agronomic data collection

In each experimental field the following data were recorded:

- Germinability—number of germinated seeds compared with total number of seeds sown;
- Germination time—number of days from the sowing date to the date on which 2 or more seeds had produced a healthy sprout;
- Time to first flowering—number of days from sowing until the date on which more than half the live plants produced their first flowers;
- Plant height—average per field at 30 days after germination, with heights measured by hand using a measuring rod;
- Seed yield per plant—average per field, based on weight measured with analytical balance (PCE-BS 3000);
- 1000-seed weight—assessed by weighing (using analytical balance Precisa XB 220A) a sample of 100 seeds per field (or a smaller sample if the yield was too low).

Bromatological analysis

The seed samples collected in each field underwent bromatological analysis in the laboratory after grinding by a rotor mill (Fritsch Variable Speed Rotor Mill Pulverisette 14). The content of dry matter was calculated for each ground sample by weighing the samples fresh and after being dried in an oven at 105°C for 12 hours. The ash content was calculated with a similar procedure by incinerating the samples on a flame and then in an oven at 550°C for 5 hours. The raw protein content was calculated based on nitrogen content, which was determined using the Kjeldahl method; the digestion of the organic substance took place with sulfuric acid (96%) and copper oxide, and the distillation and titration of the sulfuric acid were carried out by automatic analyzer (Kjeltec Auto 1030 analyzer).

The percentage of ether extract (soluble substances and fat in ether) was determined by the Soxhlet method, using petroleum ether as the extractor, while the

TABLE 3 Agronomic data collected in each experimental field.

Experimental field	Germination time (days after sowing)	Germinability (%)	Mean plant height at 30 days (cm)	First flowering (days after sowing)	Seed yield per plant (g)	1000-seed weight (g)
A	7	89	140	28	0.5	680
B	8	83	200	31	0.1	630
C	9	96	160	26	0.2	640
D	11	92	140	31	0.2	660
E	9	100	75	34	47.1	1500
F	11	92	75	35	12.6	1600
G	11	92	65	35	10.2	1500
Average	9	92	122	31	10.1	1030
SE	2	5	51	4	17.1	472

determination of the crude fiber was carried out according to the Weende method, using the Ankom 220 analyzer for fiber. The neutral detergent fiber and the acid detergent fiber were determined for each sample using the Van Soest method and the Ankom 220 analyzer. Acid detergent lignin was determined gravimetrically as the residue remaining upon ignition after sulfuric acid (72%) treatment (Van Soest method). The percentage of nonfiber carbohydrate was calculated by subtracting the percentages of ash content, protein content, ether extract, and neutral detergent fiber from the total. Finally, hemicellulose content was calculated from the difference between neutral detergent fiber and acid detergent fiber, and cellulose content was calculated from the difference between acid detergent fiber and acid detergent lignin. Each analysis was performed in duplicate.

These analyses were also performed on the seeds of 2 commercial varieties of common beans (*P. vulgaris*), borlotti and cannellini (Szafranski et al 2005; Güzel and Sayar 2010), to compare their characteristics with Copafam.

Statistical analysis

Simple linear regression analysis was performed, using the least squares method, to identify linear relationships between elevation and other variables (dry matter, ash content, protein content, ether extract, neutral detergent fiber, acid detergent fiber, acid detergent lignin, nonfiber carbohydrate, hemicellulose content, cellulose content, germination time, germinability, mean height of plants at 30 days, time to first flowering, seed yield per plant, and 1000-seed weight). The significance of each linear model was evaluated by the *F*-test, and the coefficient of determination (R^2) was calculated to evaluate the goodness of fit of each model. The samples of beans from different experimental fields were also ranked using principal component analysis (considering the elevation of the fields and all previously mentioned agronomic and bromatological data) to highlight the main variables that differentiated the samples. Statistical analysis was conducted using the software R 3.3.2 (R Development Core Team 2015).

Results

Table 3 shows the agronomic data collected on the Copafam beans produced in the 7 experimental fields. Some agronomic values varied greatly from field to field. In particular, seed germination, seed yield, and seed weight were lower in the fields at lower elevations. However, the results of bromatological analysis (Table 4) were similar across all fields. The results for Copafam beans were similar to those for the other 2 varieties of beans (borlotti and cannellini) analyzed in this study. Our results were also similar to those provided for borlotti and cannellini in the food composition tables published by the Council for Research in Agriculture and Agricultural Economic Analysis (CREA 2009).

Table 5 shows the results of linear regression analysis of the relation between elevation and the agronomic and bromatological characteristics of the plants in the study plots; Figure 4 highlights 6 of these variables (acid detergent fiber, acid detergent lignin, germination time, mean height 30 days after germination, time to first flowering, and 1000-seed weight). As Figure 4 shows, an increase in elevation increased the time required for seed germination and for blooming of Copafam flowers, as well as the weight (and thus the size) of the seeds produced. In addition, an increase in elevation decreased the speed of growth and the amount of acid detergent fiber and acid detergent lignin. These results are substantiated by the results of principal component analysis (Figure 5).

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TABLE 4 Results of bromatological analysis of Copafam seeds from each experimental field and of 2 commercial varieties of common bean (*P. vulgaris* L.), borlotti and cannellini. Values of dry matter and ash content represent percentages of total weight; other values represent percentages of dry matter weight.

Experimental field	Dry matter	Ash content	Protein content	Ether extract	Neutral detergent fiber	Acid detergent fiber	Acid detergent lignin	Nonfiber carbohydrate	Hemicellulose	Cellulose
A	92.4	5.2	21.5	1.3	16.6	14.0	4.4	55.5	2.6	9.6
B	92.8	5.3	20.7	1.6	15.9	13.2	4.3	56.4	2.7	9.0
C	92.5	5.2	19.8	1.3	14.1	12.6	4.3	59.7	1.6	8.3
D	93.1	5.2	22.4	1.9	15.4	13.5	4.2	55.1	1.9	9.3
E	93.1	5.3	19.2	2.3	14.0	11.8	4.0	59.3	2.2	7.8
F	92.5	5.0	21.4	1.5	14.9	12.4	3.6	57.2	2.4	8.8
G	92.8	4.8	17.9	2.4	14.8	11.7	3.4	60.1	3.1	8.3
Average	92.7	5.1	20.4	1.8	15.1	12.7	4.0	57.6	2.4	8.7
SE	0.3	0.2	1.5	0.5	0.9	0.9	0.4	2.1	0.5	0.6
Borlotti	89.0	4.4	23.1	1.5	12.0	11.5	2.8	59.0	0.5	8.7
Cannellini	89.8	4.4	22.3	1.1	15.1	10.8	2.1	57.2	4.2	8.7

Discussion and conclusion

This research has provided a more complete picture of the agronomic and bromatological characteristics of the Copafam bean grown in the Brescia pre-Alps. Many characteristics of Copafam depend on the elevation at which it is grown, at least in the study area. In the fields at higher elevations, there was an increase in the size of the seeds (up to twice the size of those produced at low elevations) and a reduction of their fiber (acid detergent

TABLE 5 Results of the simple linear regression analysis (considering the elevation of the experimental fields as the independent variable) and of the *F*-test.

Variable	R^2	*F*-statistic	*P* value	Significance
Dry matter	0.048	0.255	0.635	Not significant
Ash content	0.543	5.929	0.059	Not significant
Protein content	0.252	1.684	0.251	Not significant
Ether extract	0.423	3.667	0.114	Not significant
Neutral detergent fiber	0.340	2.581	0.169	Not significant
Acid detergent fiber	0.689	11.100	0.021	$p < 0.05$
Acid detergent lignin	0.915	54.110	0.001	$p < 0.01$
Nonfiber carbohydrate	0.314	2.290	0.191	Not significant
Hemicellulose	0.077	0.419	0.546	Not significant
Cellulose	0.334	2.510	0.174	Not significant
Germination time	0.622	8.224	0.035	$p < 0.05$
Germinability	0.237	1.554	0.268	Not significant
Plant height at 30 days	0.823	23.260	0.005	$p < 0.01$
First flowering	0.689	11.050	0.021	$p < 0.05$
Seed yield per plant	0.260	1.759	0.242	Not significant
1000-seed weight	0.857	30.030	0.003	$p < 0.01$

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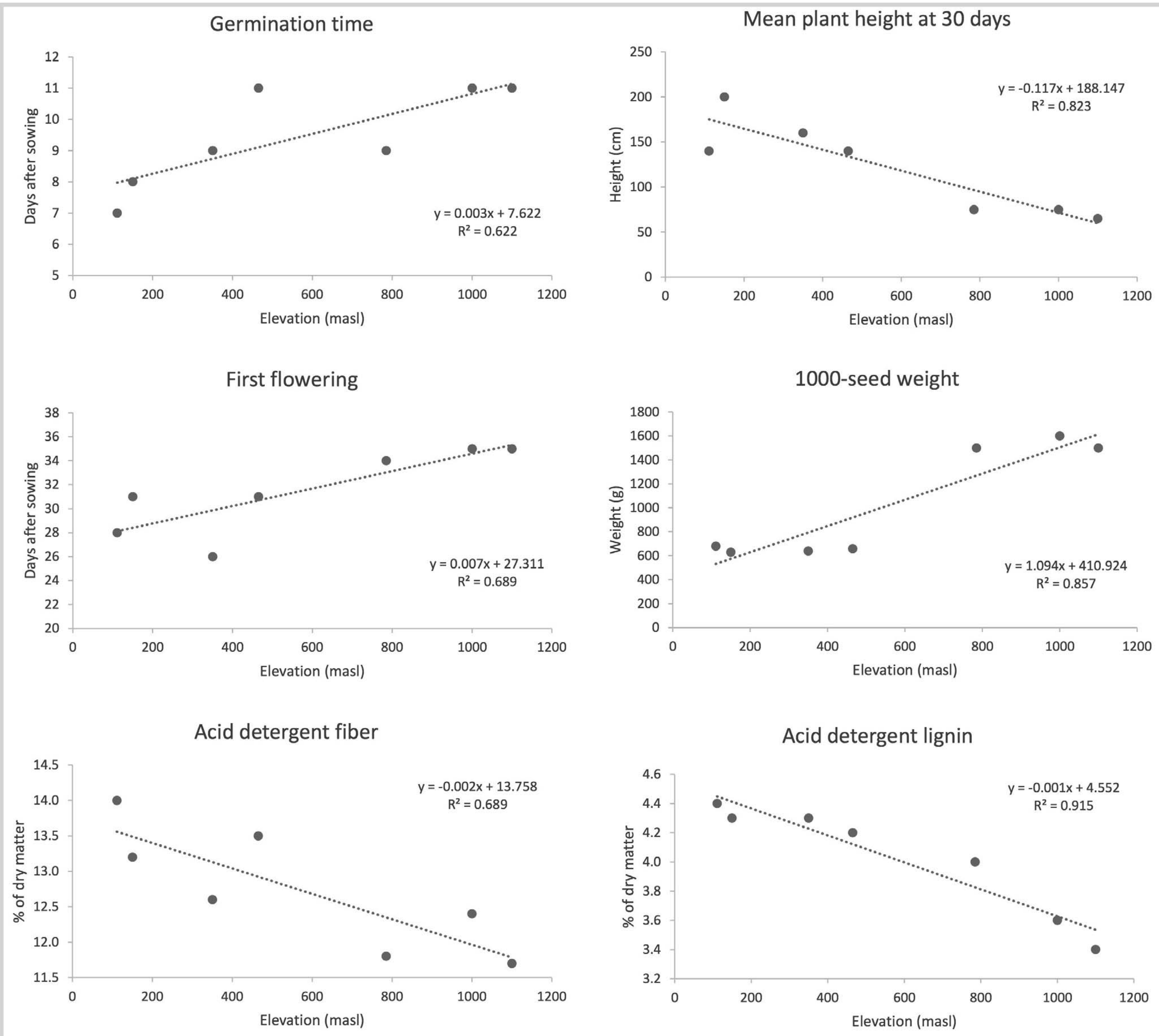
FIGURE 4 Trends of 6 variables by elevation. Each point represents a single experimental field.



fiber) and lignin (acid detergent lignin) content, making them more digestible. Observations in the field and the results of agronomic analysis (Table 3) showed some difficulty in seed germination, and even more difficulty in the production of beans, in the fields located at lower elevations—110 to 465 m above sea level (masl)—suggesting that Copafam does not tolerate high temperatures during the growing season and that, in the geographical context studied, it is well suited to be grown in the hilly and mountain belt (500–1100 masl). This confirms assertions by local hobby farmers that Copafam can be grown productively only in the hills and mountains, a feature common to some other varieties of *P. coccineus* grown in central and northern Europe (Poland, Germany, Britain, and the Netherlands) under temperatures that are prohibitive for growing common beans (Labuda 2010). Researchers (Hardwick 1972; Rodiño et al 2007) have found that *P. coccineus* is more tolerant of cold than *P. vulgaris*.

This research considered an elevation range of 1000 m (111–1100 masl) that includes the plain, hilly, and mountainous areas in which most local agriculture takes place. It would be interesting to analyze the performance of this cultivar at higher elevations to discover the extent to which the variables taken into consideration maintain the same trends and thus to identify the optimum elevations for cultivating this type of bean. It would also be interesting to conduct similar studies on more

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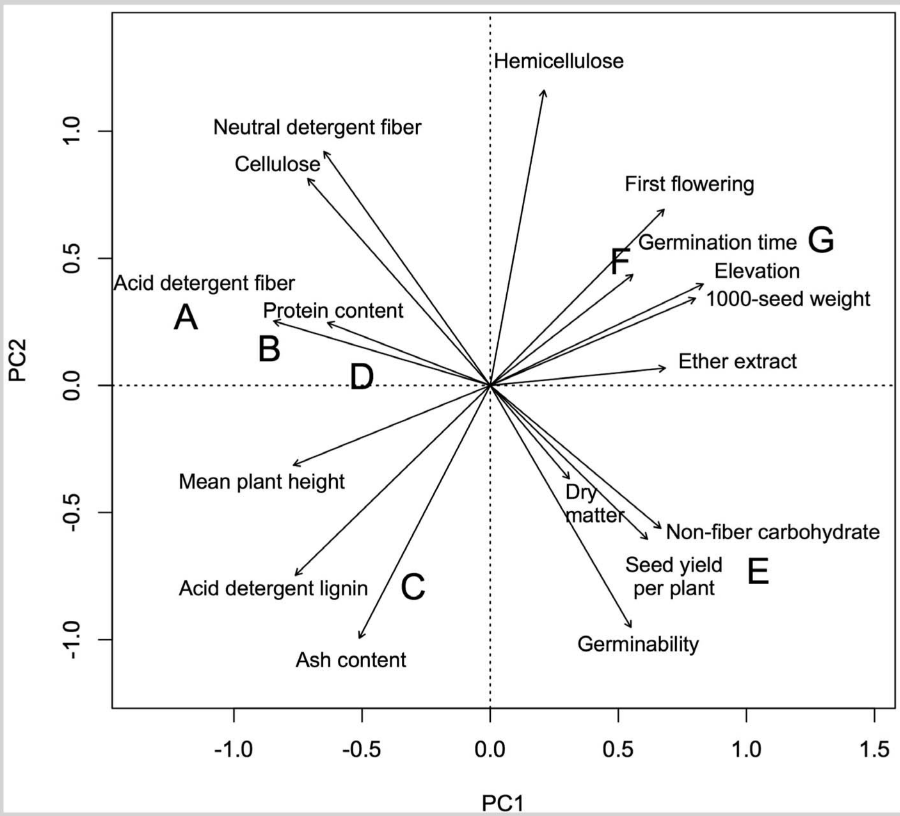
FIGURE 5 Principal component analysis ordination biplot of samples (capital letters) associated with variables (arrows). Total variance explained by first and second axis = 72.58% (PC1 = 54.54%; PC2 = 18.04%).



experimental fields throughout the Lombardy pre-Alps, monitoring environmental, production, and bromatological parameters over several years, to see whether some areas are more suitable than others for the cultivation (and conservation) of this landrace.

Copafam is not cultivated in the Po Plain (where the flat land would allow easier and larger-scale cultivation), which makes it a resource exclusive to pre-Alpine mountain territories. Therefore, it could improve the income of farmers and restaurateurs in the mountain areas of Lombardy, because consumers are increasingly interested in local agricultural products and traditional foods. This will happen only if the variety is expertly conserved, cultivated, and commercialized. This research can serve as a first step in safeguarding and enhancing Copafam: because more scientific data on the characteristics of this landrace are available, activities aimed at conserving and spreading knowledge about it can be initiated, complying with local, national, and international laws and objectives. Procedures will soon be started for the inclusion of Copafam in the list of the traditional food products of Lombardy and in the national register of biodiversity for food and agriculture. The Ge.S.Di.Mont. research center will compile a technical–scientific report on Copafam and will submit the request to the Lombardy Region for the inclusion of this landrace in the register. Action will also be taken to disseminate knowledge concerning Copafam through seminars and workshops to stimulate farmers and restaurateurs in the Lombardy pre-Alps to return to cultivating the variety and to offer local dishes to support the economy of marginal areas, following the example of existing efforts on behalf of other landraces in the area.

In the pre-Alps of Lombardy, farmers' groups and other organizations are already cultivating corn landraces (eg Spinato di Gandino, Rostrato Rosso di Rovetta, Scagliolo di Carenno, and Nero Spinoso) (Cassani et al 2017) registered in the European Register of Conservation Varieties (Spataro and Negri 2013) and have created high-quality food products, some of which are sold not only on the local market but also in nearby cities. The success of these ventures seems to be primarily due to the establishment of associations of farmers, restaurateurs, and traders who are interested in promoting the landraces (and products derived from them) but also due to special labels that identify and certify the quality and provenance of such products (such as the Municipal Designation of Origin and Slow Food brands) and the support such associations have received from local institutions and research centers that are concerned with the conservation of agrobiodiversity and the sustainable development of mountain regions.

In conclusion, this article, in addition to presenting knowledge regarding a little-known landrace, aims to be a stimulus for all those who are called to safeguard worldwide agrobiodiversity and in particular to encourage other researchers to commence similar research targeted not only at identifying and characterizing landraces and little-known traditional agricultural products but also at identifying a strategy for their valorization as an aid to sustainable development. Such research, which requires close cooperation among universities or research centers, other institutions, and local farmers, has never been so topical, given the increasing awareness that agrobiodiversity can be a resource for areas that are being abandoned, like those in the Alps (Giorgi and Scheurer 2015), and all areas whose agro-food specialties have not yet been fully exploited**.**

ACKNOWLEDGMENTS

We thank Dr. Simone Colombo and Dr. Stefania Bonacina for their work in the fields and the laboratory. This study was supported by a Lombardy Region–Ge.S.Di.Mont. Research Centre agreement (*Accordo di collaborazione fra Regione Lombardia e Ge.S.Di.Mont*. per *attività di ricerca scientifica ed applicata e di diffusione della conoscenza inerente il territorio montano lombardo,* art. 4 c.7 l.r. 22/2016).

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