

## Morpho-Agronomic Performances and Antioxidant Properties in Chilli Pepper Hybrids are highlighted by Multi-Scale Evaluation on Two Locations and Digital Fruit Imaging

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### Abstract

Chilli peppers are a vegetable crop that is commonly eaten fresh or dried as a spice. The nutritional value derived from the presence of beneficial healthy-related components, as well as the variety of applications, have boosted its popularity over the previous decade. Chillies are grown in Southern European nations using traditional cultivars and/or landraces that are well adapted to specific settings but do not always match industry needs, notably in terms of packing and processing. In this study, ten commercial hybrids were tested for productivity and phytochemical content, including carotenoids, capsaicinoids, ascorbic acid, and tocopherols, in two different environments. For the analysis of size, shape, and colour factors, automated instruments were used on fruits. In terms of productivity, the pepper materials were promising, however there was a reduced level of capsaicinoids and ascorbic acid found. The effects of genotype and environment on yield, fruit form, and capsaicinoids were found to be minor. The integration of phenomics data from several sources illustrates how hybrid breeding initiatives have focused on yield and morphology rather than quality as measured by phytochemical content.

**Keywords:** *Ascorbic acid; Capsaicinoids; Capsicum annum; Carotenoids; Colour By cielab; Fruit phenomics; Fruit Size And Shape; Yield-traits*

### Introduction

Chilli pepper is one of the most intriguing and widely consumed spice foods, owing to its great nutritional and health benefits in human diets. Today, the crop is grown on around four million hectares around the world, with a production of over 40 million tonnes. Chilli pepper cultivars differ greatly in terms of morphological traits and quantities of bioactive substances, particularly capsaicinoids, which contribute to the species' distinctive flavour and strong taste. Chilli fruits are versatile, serving as food in fresh, dried, or paste form (e.g., sauce, cream) or

for industrial application (e.g., as a colouring additive or a cosmetics agent). Both cultivated (*Capsicum annuum*) and other domesticated species of chilli pepper exist, including accessions such as the habanero (*C. chinense*), tabasco (*C. frutescens*), rocoto (*C. pubescens*), and aji kinds (*C. baccatum*). The majority of them are widely grown in the Caribbean, South America, and several Asian regions, where a mix of culinary and cultural variables has encouraged the use of extremely spicy varieties [1]. Cultivated *C. annuum* spicy variants, on the other hand, have a larger global market and are the most popular in Europe, where mild spicy variations are chosen by consumers [2]. After Asia, Europe is the second largest importer of chiles, and it also exports dried and packaged items. Given the growing popularity of spicy dishes, new cultivars must be developed and cultivated to meet the needs of both producers and consumers [3]. Farmers have chosen a variety of local types, which are currently farmed primarily on small and medium-sized farms. Because these materials are largely open-pollinated variations with off-types from high genetic diversity, they may still have some inherent genetic variability, even if they are well adapted to certain settings. For packing and processing, where the production chain requirements are more stringent [4,5], these limits are undesirable. We previously studied panels of chilli genotypes, including open-pollinated cultivars and landraces, for agronomic performance and nutritional characteristics, as well as the effect of genotype by environment interaction on attributes [6,7]. As seed firms' interest in developing new chilli cultivars has grown, improved uniform types with large yields, such as F1 hybrids, have been released. This study aims to expand the knowledge base on these types of varieties in order to: (a) better understand the characteristics that have largely influenced selection, (b) determine whether improved cultivars are more stable than landraces by estimating the G E effect, (c) investigate the possible performance gap between improved and unimproved cultivars, and (d) define additional chilli breeding goals.

As a result, ten exceptional hybrids (representing cherry, horn, and jalapeño morphotypes) were developed in two separate environments [8,9]. 18 agronomic and biochemical characteristics, as well as more than 40 morphometric and colour fruit factors, were phenotyped. Multi-trait analysis uncovers new information about chilli cultivars that can be used in future breeding operations. Ten commercial hybrids of grown pepper were chosen for fresh and dried consumption as plant material. Cherry ('Bomber' and 'Topik') and jalapeno ('Jalaprider' and 'Newpark') morpho-types with round and ovoid shapes, respectively, were included in the hybrids, while the remaining selection had a horn shape ('Anastar', 'Eris', 'Haruba', 'PH11421', 'Vulcan', 'Zigano'). Seeds were received from a variety of sources (Sativa, Esasem, United Genetics). Plants were planted in two locations: Battipaglia (BP) in the Campania Region's Sele Valley (40°37' N; 14°58' E, 65 m a.s.l.) and Montanaso Lombardo (ML) in the Lombardia Region's Po Valley (45°20' N; 9°26' E, 80 m a.s.l.). The two sites are separated by over 800 kilometres and have drastically different pedoclimatic characteristics. Total yield (grammes) [TY] of fully ripe fruits was measured using a manual calliper on ten fruits; average fruit weight (FW) (in grammes) was calculated by dividing the total yield by the number of fruits harvested; fruit length (FL) and fruit width (FD) (in centimetres) were measured using a manual calliper on ten fruits; and fruit shape index (FS) was calculated as the length/width ratio. Total yield (grammes) [TY] of fully ripe fruits was measured using a manual calliper on ten fruits; average fruit weight (FW) (in grammes) was calculated by dividing the total yield by the number of fruits harvested; fruit length (FL) and fruit width (FD) (in centimetres) were measured using a manual calliper on ten fruits; and fruit shape index (FS) was calculated as the length/width ratio [10].

### **Characterization of Chemical and Biochemical Compounds**

Fruits from each replication in both areas were analysed chemically and biochemically. After selecting fresh peppers with no visible faults, the plant material for the analyses was established. Peduncles were removed from selected fruits, which were then chopped along the longitudinal axis according to standard procedures and dried for 48 hours in a forced-air oven at 45 °C. The dried material was powdered at 4 °C in a Waring blender (Waring Commercial, Stamford, CT, USA) and kept in dark bottles at 20 °C until analysis. Chemical characteristics were assessed in a supernatant solution made by suspending 2 g of powder in 25 mL deionized water, stirring for 15 minutes, and then decanting. Fruits from each replication in both areas were analysed chemically and biochemically. After selecting fresh peppers with no visible faults, the plant material for the analyses was established. Peduncles were removed from selected fruits, which were then chopped along the longitudinal axis according to standard procedures and dried for 48 hours in a forced-air oven at 45 °C. The dried material was powdered at 4 °C in a Waring blender (Waring Commercial, Stamford, CT, USA) and kept in dark bottles at 20 °C until analysis. Chemical characteristics were assessed in a supernatant solution made by suspending 2 g of powder in 25 mL deionized water, stirring for 15 minutes, and then decanting. A Multi-Scale refractometer RFM 91 was used to determine the total soluble solid content (SSC), which was expressed in °Brix on 100 g of dried weight (°Bx dw) (Bellingham-Stanley Ltd., Kent, UK). A titroprocessor mod 682 equipped with a Dosimat 665 apparatus was used to determine the pH and titratable acidity (AC), both expressed in mEq percent dw (Metrohm, Herisau, Switzerland). Total carotenoids (TC) and their red (CR) and yellow (CY) fractions; ascorbic acid (AsA); capsaicin (CAPS), dihydro-capsaicin (DHC), nordihydro-capsaicin (NDHC); Scoville units (SHU); gamma-tocopherol (-toc), alpha-tocopherol (-toc); and gamma-tocopherol (-toc) The presence of carotenoids was determined using spectrophotometric methods, while the rest of the features were determined using High Performance Liquid Chromatography (HPLC). Previous papers provide details on the analytical techniques employed.

### **Digital Fruit Analysis**

To eliminate any bias due to light, a representative mass of fifteen mature fruits from each accession and the BP site were cut lengthwise and scanned with a CanoScan Lide 200 (Canon, Italy) at 300 dpi resolution in a dark room using a black background. Tomato Analyzer 3.0 (TA) [8] was used to analyse the photos that resulted. Thirty-eight fruit size and form parameters were assessed quantitatively. The colour of the fruit was measured using a handheld colorimeter (Minolta Chroma Meter CR-210; Minolta Corp., Osaka, Japan) to yield CIELab ( $L^*$ ,  $a^*$ ,  $b^*$ ) coordinates, as well as Chroma  $[(a^*)^2 + (b^*)^2]^{0.5}$  and Hue angle ( $\arctan b^*/a^*$ ).

### **Conclusion**

The potential of chilli hybrids in terms of agronomic and qualitative performance has been demonstrated in this paper. We assessed the properties of several types of chilli peppers using deep phenotyping, demonstrating how morphology, yield, and capsaicinoids are the key drivers of selection for high yielding and mild-pungent varieties. Instead, increased efforts are required to improve quality-related attributes. Further research using sensory analysis and panel tests could provide further insight into consumer preferences for the many chilli

genotypes now available. These data show how commercial variety breeding efforts may have prioritised high-yielding, morphologically stable genotypes above high-quality genotypes. The found substantial genotypic effect over the environment for agronomic traits appears to back this up. The comparable trend in capsaicinoids values across places shows that pungency should be introduced as an extra major goal in chilli pepper breeding for the market in order to generate somewhat spicy cultivars. In fact, the SHU levels found in this study were 30–50 times lower than those found in conventional hot peppers like habaneros (about 300,000 SHU). This reflects the Mediterranean diet's gastronomic and cultural applications. The high production paired with larger fruit weight is undoubtedly an advantage in hand harvesting, which is still extremely frequent with chillies, resulting in a higher final product yield for both the fresh and processed markets. As a result, the environment had little impact on the manipulation of these fruit characteristics. We created a more precise approach for shape attributes based on fruit scans to account for probable bias in hand measurements due to the curvature of fruits in the longitudinal section (in particular horn-shaped types). The fruit scan observations gave more detailed information, allowing for the measurement of more attributes than those that were visually recorded and manually collected. This demonstrates the utility of scans for phenotyping chilli pepper fruit. We identified similar trait correlations across locations, though BP had a higher number. This could be related to the different pedoclimatic conditions of the cultivation locations, as well as the quantitative nature of the qualities that are affected by environmental changes.

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