

Fatima El Amerany<sup>1,2,\*</sup>

<sup>1</sup>Laboratory of Sustainable Development and Health Research, Faculty of Science and Technology of Marrakech, Department of Chemistry, Cadi Ayyad University, PO Box 549, Marrakech 40000, Morocco;

<sup>2</sup>Interdisciplinary Laboratory in Bio-Resources, Environment and Materials, Higher Normal School, Department of Biology, Cadi Ayyad University, PO Box 575, Marrakech, 40000, Morocco.

\*Corresponding author: email: el.amerany.fatima@gmail.com; ORCID id: 0000-0003-3670-2648

## Abstract

In agriculture, chitosan has been known to exert a biostimulator effect on plant system; however, there is not enough information regarding its mechanisms on plant tissues as well as primary metabolism in fruit. The present work investigated the effect of chitosan isolated from *Parapenaeus longirostris* on tomato plants and fruit features. An increase in shoot growth was observed in plant treated with chitosan and this promotion was positively correlated with stomata aperture and xylem vessels growth. In addition, chitosan application to shoots affected fruit maturation by inducing the levels of sucrose and antioxidants (i.e., ascorbic acid) as well as fruit acidity. Thus, chitosan application to shoots could replace the use of fertilizers and enhance plant growth and fruit quality.

**Keywords:** Chitosan; mechanisms; fruit quality; fertilizers; plant development.

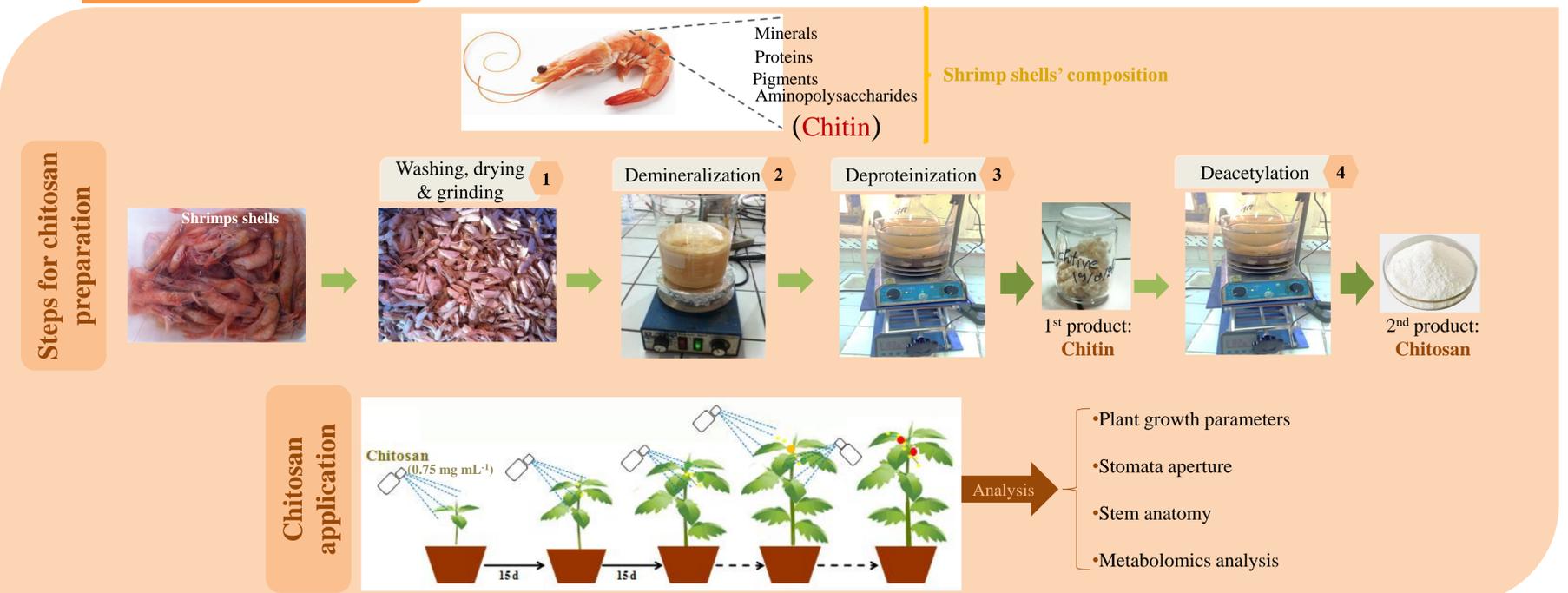
## Introduction

Over the past decades, the production of tomatoes in many countries in the world was affected by several factors, such as a rapid decline in the agricultural land area due to urbanization and water sources' pollution by fertilizers and pesticides [1,2]. The yield of tomato was also reduced because of an increase in climate changes, especially temperature rise, desertification, and soil's nutrient depletion [3]. To date, climate change is considered a global challenge with no limits. Among the causes that led to this phenomenon was the release of a large amount of untreated marine waste into the environment [4]. The accumulation of this waste caused the production of greenhouse gases and thereafter affected food security.

A growing interest has emerged recently to deal with these problems through the adoption of multiple appropriate and sustainable strategies. For instance, our previous study showed that the valorization of marine waste by producing biostimulants, such as chitosan, can reduce the pollution generated by this waste, replace the phytochemicals' use, and increase plant productivity [5]. However, the effect of foliar application of chitosan during seedling and fruit development on plant physiology, vascular bundles development, and the primary metabolism in fruits has not been yet studied.

Therefore, this work aimed to elucidate the effects of chitosan (deacetylation degree < 16%; molecular weight 318.53 kDa), isolated from *Parapenaeus longirostris* shells, on tomato growth and fruit features.

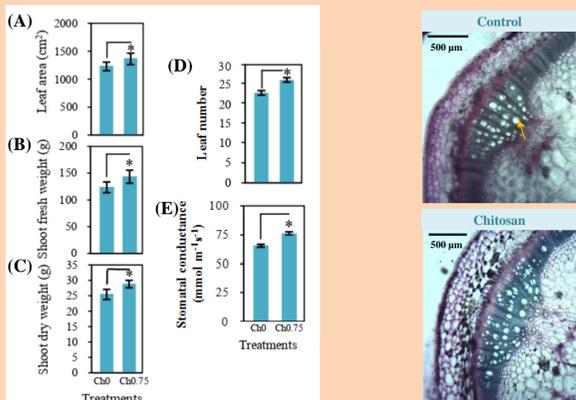
## Materials & methods



## Results

The plant growth parameters, such as leaf area, leaf number, and shoots fresh and dry weights, were significantly induced after applying chitosan to shoots by 10.93%, 14.81%, 16.09%, and 13.23%, respectively (Fig. 1). This promotion was positively correlated with stomatal conductance and xylem vessels' number (Fig. 2 and 3).

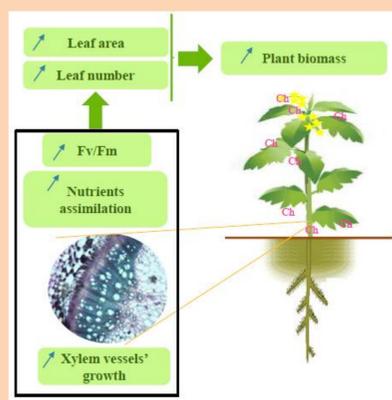
To get more details about how chitosan affects the metabolic pathways of the primary metabolism, targeted profiling for polar metabolites was conducted. Then, a putative model for the biosynthesis pathway of these metabolites was proposed. Fig. 4 showed that chitosan application to shoots induced the levels of metabolites that resulted from glycolysis, such as inositol phosphates, sucrose, ascorbic acid, and lactate. However, this application reduced levels of TCA cycle substrates and amino acids, especially arginine, methionine, and histidine.



**Fig. 1** Effect of chitosan on (A) leaf area, (B & C) fresh and dry weight of shoots, (D) leaf number, and (E) stomatal conductance. Data are expressed as the mean  $\pm$  SD (n=6). \* indicates significant differences between treatments following t test.



**Fig. 2** Cross section of tomato stems treated and non treated with chitosan (x100). Arrow indicates xylem vessel.



**Fig. 3** Summarized representation of the main effect of chitosan on plant growth

## Discussion

The data of this work showed that foliar spray of chitosan, isolated from shrimp shells, can boost the development of tomato plants. This effect might be due to better functioning of chloroplasts and the increase in gas exchange [6,7]. In addition, the improvement in plant growth appears to be in good agreement with stomatal conductance aperture and vascular bundles growth. The opening of stomata and increasing of xylem vessels number under chitosan application ensure strong assimilation of carbon dioxide, water, and mineral elements, which, therefore, induce the photosynthetic activity of the plant [5,8].

Regarding chitosan effect on fruit features, the model presented in Fig. 4 showed that the increase in the level of sucrose in fruits from plants treated with chitosan might be the result of chitosan degradation. While, the decrease in the level of citric acid and amino acids after chitosan application might be due to the redirection of carbon skeletons resulting from glycolysis towards other metabolic pathways, especially those of vitamin C and inositol biosynthesis as well as anaerobic metabolism. This is in agreement with the results obtained by Sajid et al. [9].

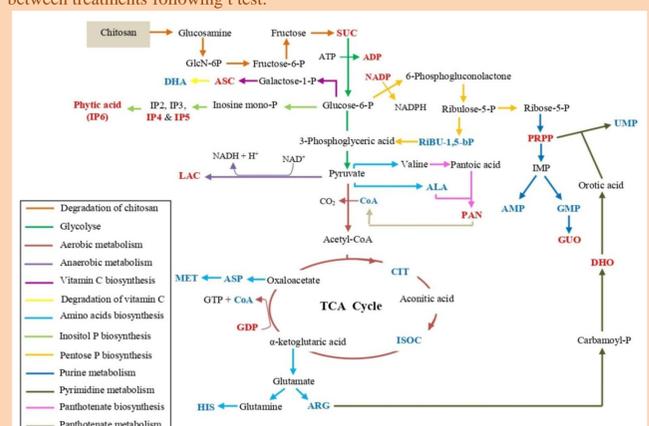
## Conclusion

The application of chitosan isolated from shrimp shells was beneficial for tomato growth and for improving fruit quality. The positive effect of chitosan is related to an increase in carbon dioxide fixation and nutrient assimilation and stimulation of a series of primary metabolic pathways of carbon and nitrogen metabolisms.

## References

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Conflicts of Interest: The author declare no conflict of interest.



**Fig. 4** Scheme summarizing the effect of chitosan on biochemical pathways in tomato fruits. The metabolites marked in red and blue colors represent increased and decreased metabolites, respectively.

**Abbreviations:** ADP, adenosine diphosphate; ALA, alanine; AMP, adenosine monophosphate; ARG, arginine; ASC, ascorbate; ASP, aspartic acid; ATP, adenosine triphosphate; CIT, citrate; CoA, coenzyme A; DHA, dehydroascorbic acid; DHO, dihydroorotate; GDP, guanosine diphosphate; GlcN-6P, glucosamine-6P; GMP, guanosine monophosphate; GTP, guanosine-5'-triphosphate; GUO, guanosine; HIS, histidine; IMP, inosine monophosphate; IP2, sum of inositol bisphosphates; IP3, sum of inositol trisphosphates; IP4, sum of inositol tetrakisphosphates; IP5, sum of inositol pentakisphosphates; IP6, inositol hexakisphosphate; ISOC, isocitrate; LAC, lactate; MET, methionine; NAD, nicotinamide adenine dinucleotide (oxidized); NADH, nicotinamide adenine dinucleotide (reduced); NADPH, nicotinamide adenine dinucleotide phosphate (reduced); PAN, pantothenate; PRPP, phosphoribosyl pyrophosphate; SUC, sucrose; RiBU-1,5-bP, ribulose 1,5-bisphosphate; UMP, uridine monophosphate.