

In vitro antigenotoxic activity of probiotic *Lactobacillus plantarum* IMC 510 and IMC 513 against two different endocrine disruptors: bisphenol A and di-n-hexyl phthalate

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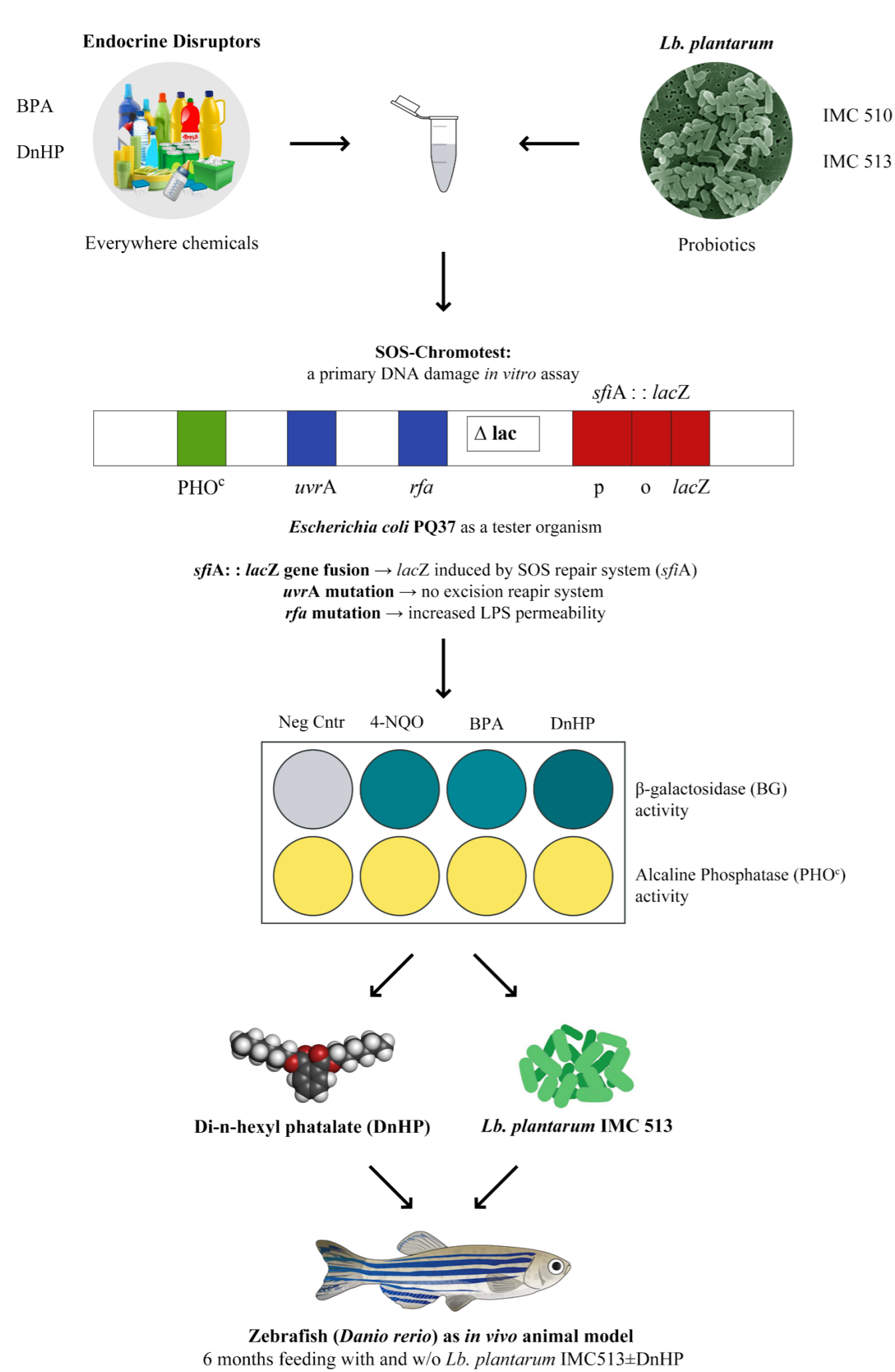
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INTRODUCTION

Nowadays, the interest in diets or dietary components able to influence the composition and the activity of the intestinal microbiota and consequently modulate the risk of genotoxicity and colon cancer is relevant in the scientific community. Within this topic, the microbial ability to make a protective role at gastrointestinal (GI) level by counteracting the biological activity of genotoxic compounds, and thus preventing the DNA damage, is thought important for reducing gut pathologies and colon cancer incidence and is considered a new tool for probiotics and functional foods.

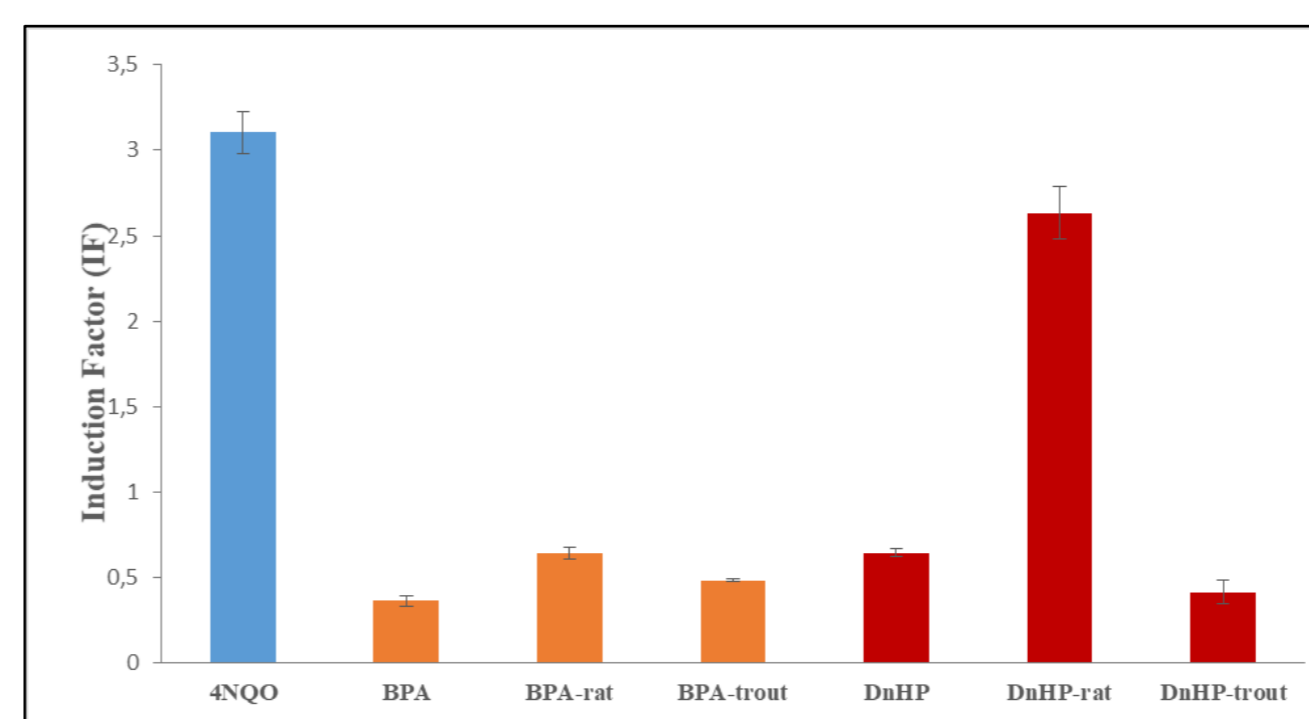
In the gut, a variety of genotoxic compounds can be found and among all of them, there is a group of pollutants present in the environment and in foods called endocrine disruptors (EDs). EDs are exogenous substances that alter the functions of the endocrine system through estrogenic and anti-estrogenic activity, which interfere with normal hormonal function in human and wildlife (1, 2).

In this perspective, the antigenotoxic activity of two probiotic *Lactobacillus plantarum* (IMC 510 and IMC 513, supplied by Synbiotec Srl, Camerino, Italy), previously tested against a model genotoxin (3) was evaluated against two different EDs: bisphenol A (BPA) and Di-n-hexyl phthalate (DnHP), commonly used in food packaging and/or in thousands of everyday products.



EVALUATION OF EDs GENOTOXICITY

Genotoxic activity of two different EDs: BPA and DnHP was evaluated by SOS response using *Escherichia coli* PQ37 as tester strain (4) with and without rat/trout S9 metabolic activation. The known mutagen 4-nitroquinoline-1-oxide (4-NQO), which produces DNA adducts, was used as reference genotoxic compound.



Genotoxicity, expressed through Induction Factor (IF), showed DnHP as the most genotoxic molecule

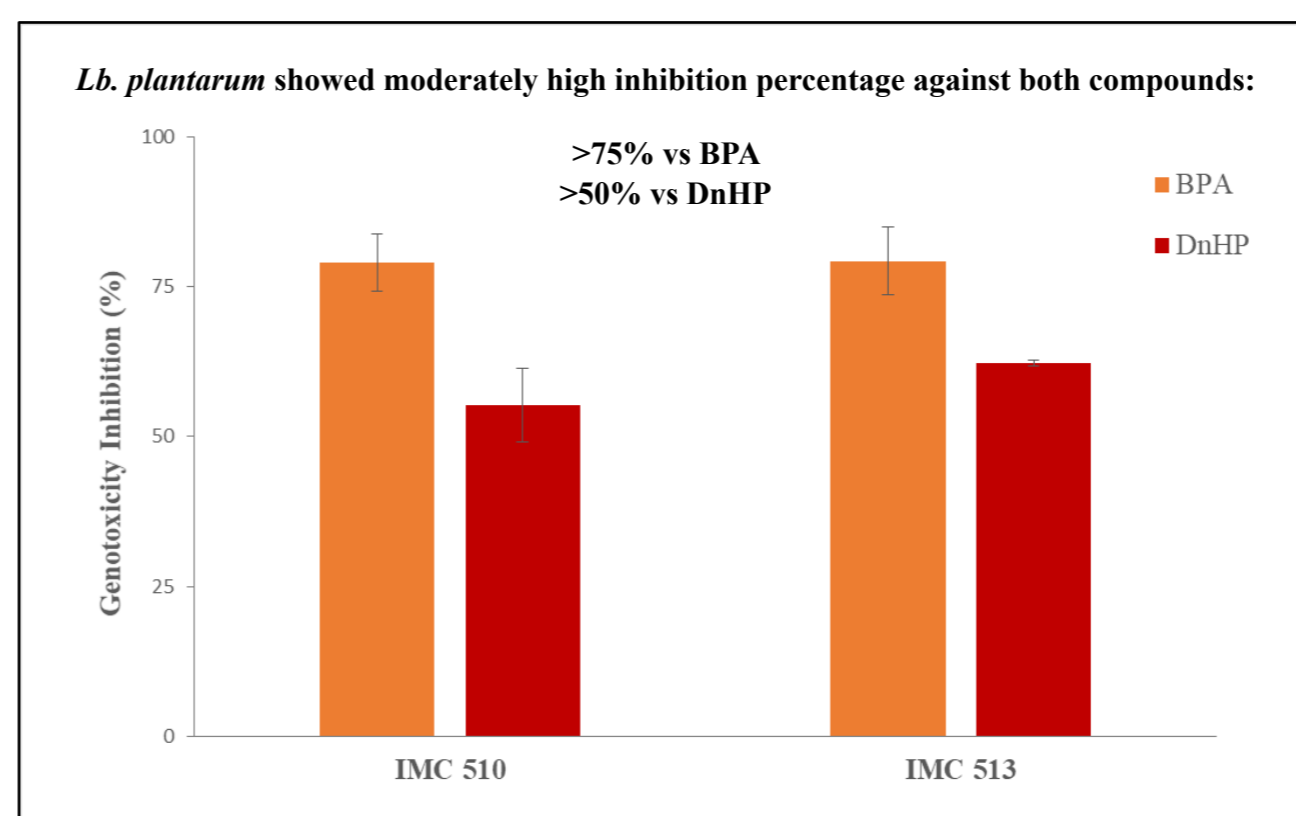
$$IF_{SOS} = R_S / R_{NC}$$

R_S = BG/PHO^c ratio of the sample
 R_{NC} = BG/PHO^c ratio of negative control

GENOTOXICITY INHIBITION

Short-term Biological Method: Strains inhibitory activity on genotoxicity (GI) induced by EDs, was studied using the SOS-Chromotest, a primary damage assay with *Escherichia coli* PQ37 (*sfiA::lacZ*) as the tester organism (3, 6).

$$GI (\%) = (IF_{SOS} / IF_{EDs}) * 100$$



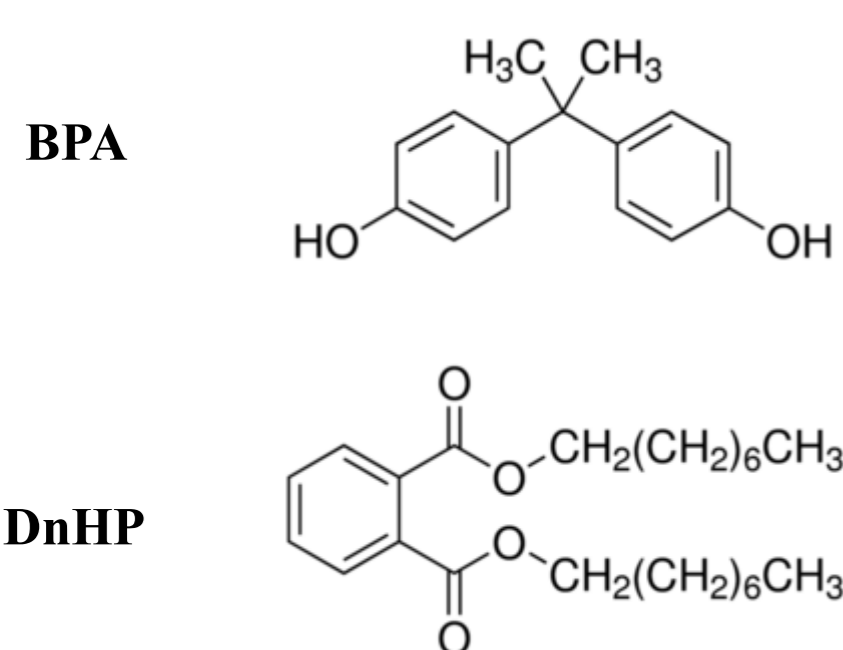
IF REDUCTION

IF _{EDs}	IMC 510	IMC 513
BPA	1.49±0.11	1.48±0.13
DnHP	1.62±0.08	1.53±0.01

Co-incubation of EDs with both *Lb. plantarum* probiotic strains showed appreciable reduction of the SOS induction factor of BPA and DnHP, which were both below the threshold level for genotoxicity (<2).

ENDOCRINE DISRUPTORS (EDs)

The endocrine disruptors investigated in the study for their genotoxic activity were Bisphenol A (BPA) and Di-n-hexyl phthalate (DnHP), commonly used in food packaging and/or in thousands of everyday products.



PROBIOTIC STRAINS

The microbes used in the study, kindly provided by Synbiotec s.r.l., are *Lb. plantarum* strains with documented probiotic activities, that were previously tested for their ability to inhibit a model genotoxin (1) and for other functional activities (2).

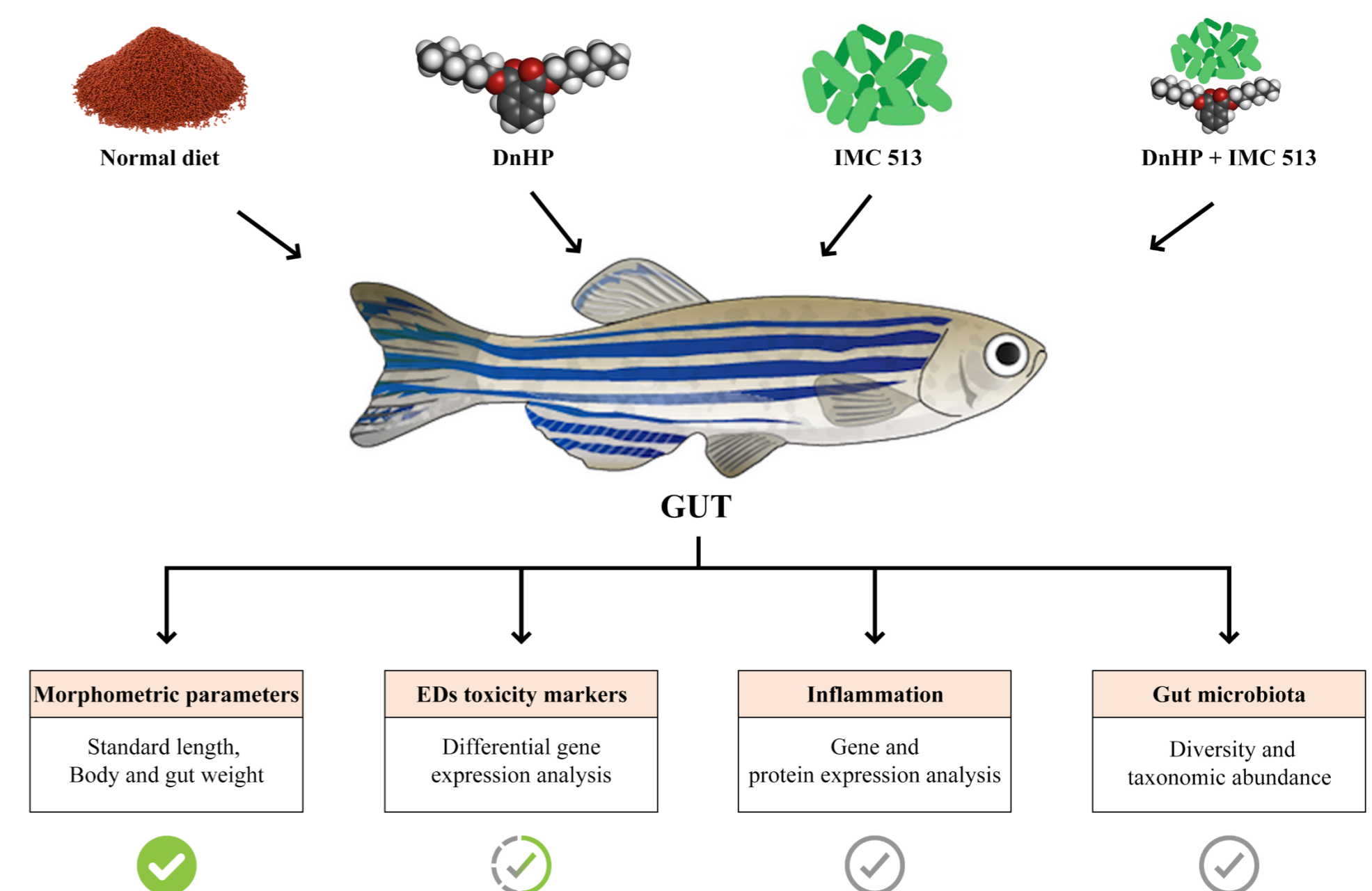
<i>Lb. plantarum</i> strains	Origin	Source
IMC 513	Human gut	Synbiotec s.r.l.
IMC 510	Human gut	Synbiotec s.r.l.

CONCLUDING REMARKS

- Co-incubation of EDs with both *Lb. plantarum* probiotic strains showed appreciable genotoxicity inhibition.
- The major genotoxicity on the tester organism PQ37 was induced by DnHP (metabolic activated by rat S9 microsomal enzymes).
- In vitro* results confirmed the ability of *Lb. plantarum* IMC 513 to inhibit the biological activity of two different environmental genotoxic compounds.
- Lb. plantarum* IMC 513 and DnHP were selected as a combined treatment to use in an *in vivo* experimental trail with zebrafish (*Danio rerio*).

IN VIVO EXPERIMENTAL TRIAL: Danio Rerio

The *Lb. plantarum* IMC 513 protective role in preventing DNA damage at GI level is under evaluation in the ongoing experimental trail with zebrafish (*Danio rerio*), daily fed with and without *Lb. plantarum* IMC 513 and DnHP for six months.



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