UNIVERSITA' DEGLI STUDI DI MODENA E REGGIO EMILIA
Department of Life Sciences

PhD SCHOOL OF AGRI-FOOD SCIENCES, TECHNOLOGIES AND BIOTECHNOLOGIES

"The meaning of bioactive compounds in Human Health"

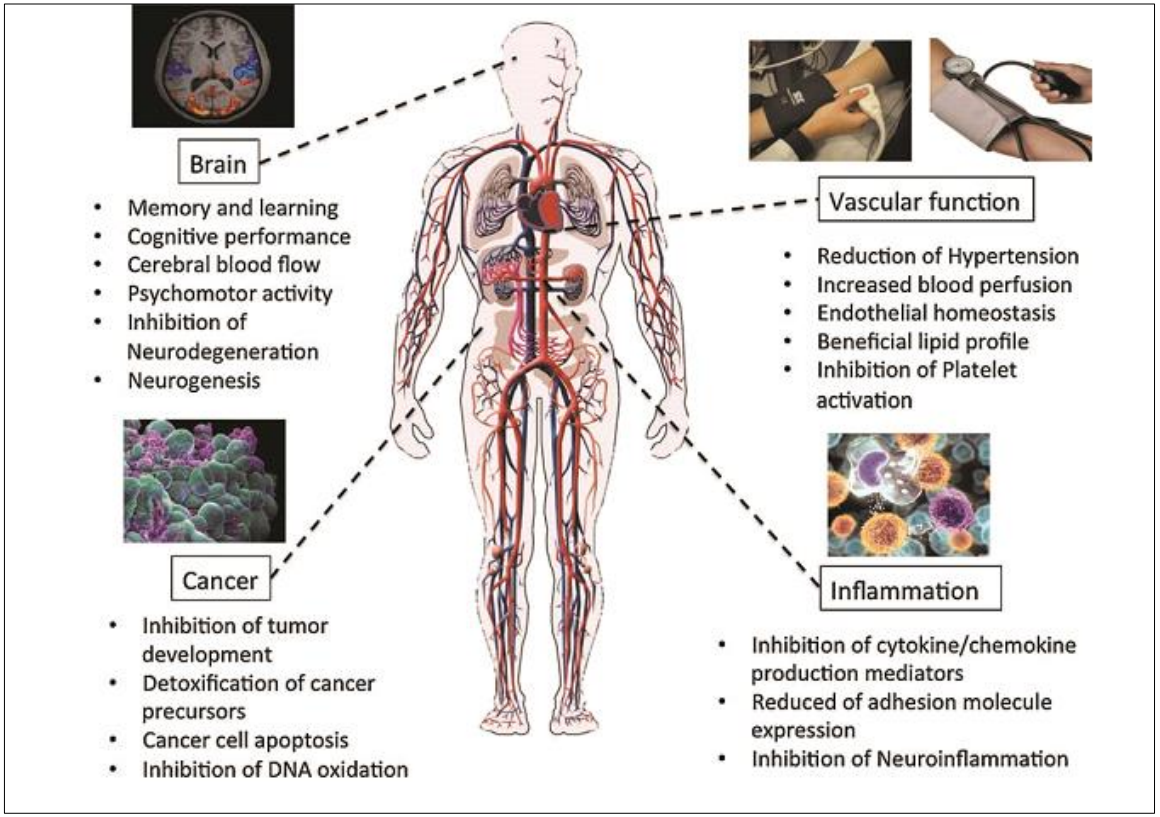
PhD student: Serena Martini
Tutor: Prof. Angela Conte, Dr. Davide Tagliazucchi

Polyphenols

Main sources are fruits, beverages such as tea, coffee, wine and fruit juices, chocolate and, to a lesser extent, vegetables, cereals and legume seeds

Polyphenols

- More than 8000 chemical structures
- Aromatic benzenoid (phenyl) ring, hydroxyl (-OH) groups
- Two main classes: **Flavonoids**
  Non Flavonoids



Polyphenols are abundant **micronutrients** in our diet, and evidence for their role in the prevention of **degenerative diseases** such as **cancer** and **cardiovascular diseases** is emerging. Their health effects depend on the amount consumed and on their **bioavailability**.

Cherry cultivars

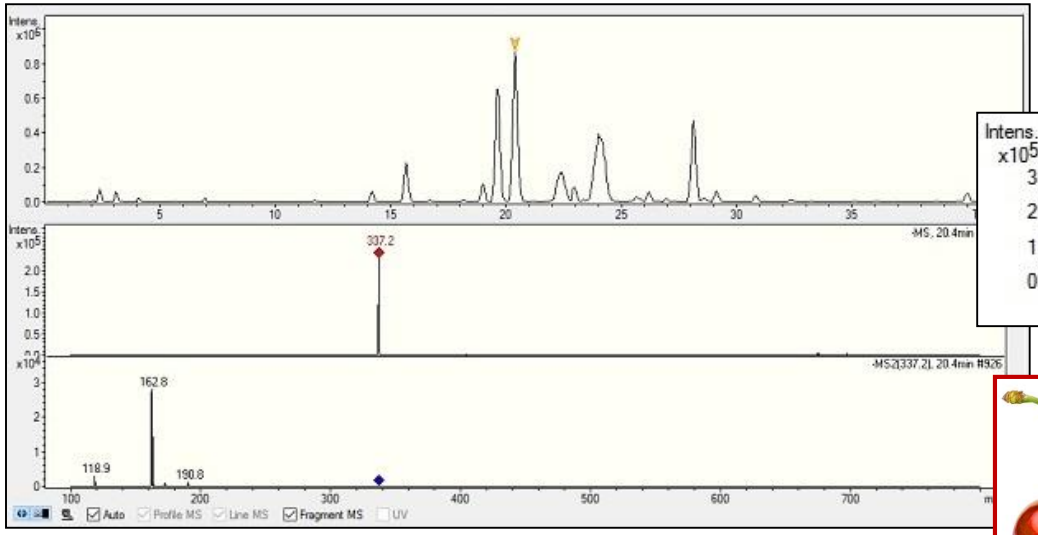
Light cherries

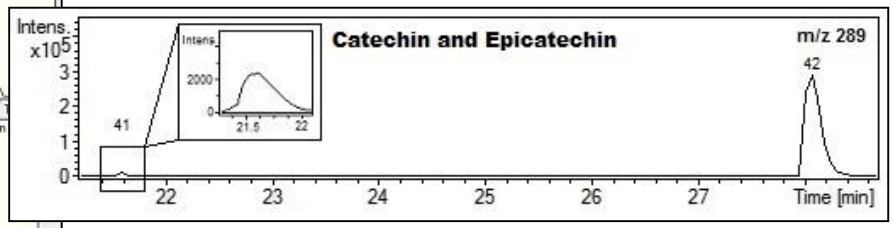
Dark cherries

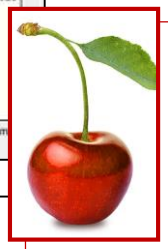
The objectives

1. • *In vitro* digestion of six cherry cultivars.
2. • C18 extraction of phenolic compounds from both digested and undigested cherries.
3. • Identification and quantification of cherry phenolic compounds by liquid chromatography-electrospray ionization-ion trap mass spectrometry (LC-ESI-IT-MS/MS).
4. • Evaluation of antioxidant activity ($ABTS^{+}$), superoxide anion ($O_2\bullet$) and hydroxyl (OH^{-}) radical scavenging activities and ferric reducing power (FRAP assay) and bioaccessibility.
5. • Cytotoxic and anti-proliferative activity of cherries polyphenols on human colon adenocarcinoma cell lines (Caco-2 and SW 480).
6. • Cytotoxic and anti-proliferative activity of major colon available phenolics and metabolites on Caco-2 and SW 480.

LC-ESI-IT-MS/MS identification







Tentative identification of 86 individual phenolic compounds in cherry cultivars

40 phenolic compounds identified for the first time in cherries.

Phenolic compounds profile and antioxidant properties of six sweet cherry (*Prunus avium*) cultivars

CrossMark

Serena Martini, Angela Conte, Davide Tagliazucchi *

Department of Life Sciences, University of Modena and Reggio Emilia, Via Amendola 2, 42100 Reggio Emilia, Italy

ARTICLE INFO

Article history:
Received 23 January 2017
Received in revised form 17 March 2017
Accepted 21 March 2017
Available online 22 March 2017

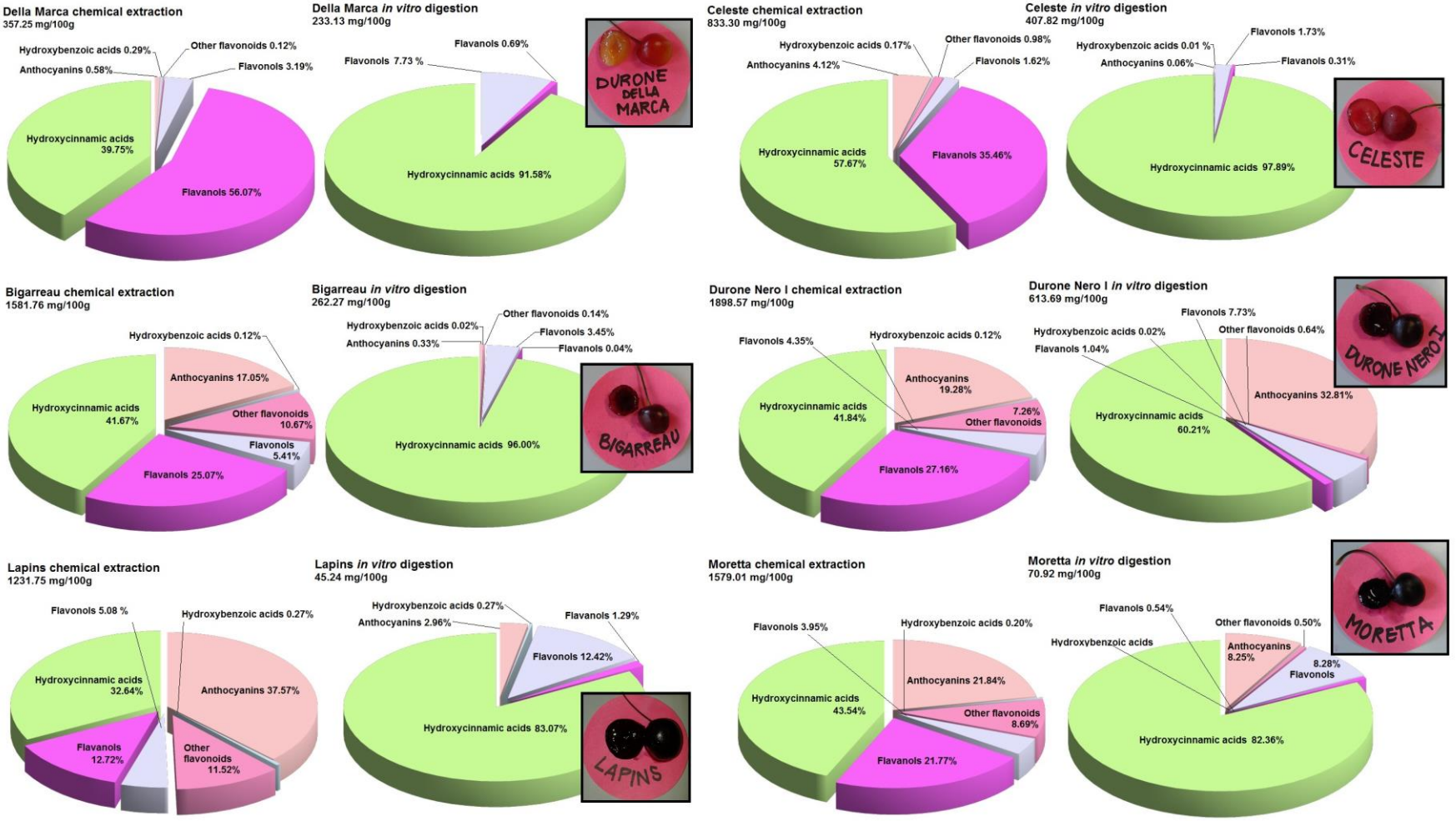
Keywords:
Cherries
Mass spectrometry
Polyphenols fingerprint
Metabolomics
Principal components analysis

ABSTRACT

Sweet cherry (*Prunus avium*) fruits are a nutritionally important food rich in dietary phenolic compounds. The aim of this study was to investigate the phenolic profile and chemometric discrimination of fruits from six cherry cultivars using a quantitative metabolomics approach, which combine non-targeted mass spectrometry and chemometric analysis. The assessment of the phenolic fingerprint of cherries allowed the tentative identification of 86 compounds. A total of 40 chlorogenic acids were identified in cherry fruit, which pointed out hydroxycinnamic acid derivatives as the main class of phenolics by number of compounds. Among the compounds detected, 40 have been reported for the first time in sweet cherry fruit. Hydroxycinnamic acids are also the quantitatively most represented class of phenolic compounds in the cherry cultivars with the exception of Lapins and Durone della Marca where the most representative class of phenolic compounds were anthocyanins and flavan-3-ols, respectively. This non-targeted approach allowed the tentative identification of the cultivar-compound relationships of these six cherry cultivars. Both anthocyanins and colorless phenolic compounds profile appeared to be cultivar-dependent. In detail, anthocyanins and flavonols patterns have the potential to be used for the determination of a varietal assignment of cherries.

Chemical quantification

Hydroxycinnamic and hydroxybenzoic acids, flavan-3-ols, flavonols, anthocyanins and other flavonoids obtained through chemical extraction or after *in vitro* digestion in six different cherry cultivars



Bioavailability

Proportion of a nutrient that is absorbed from the diet and used for normal body functions

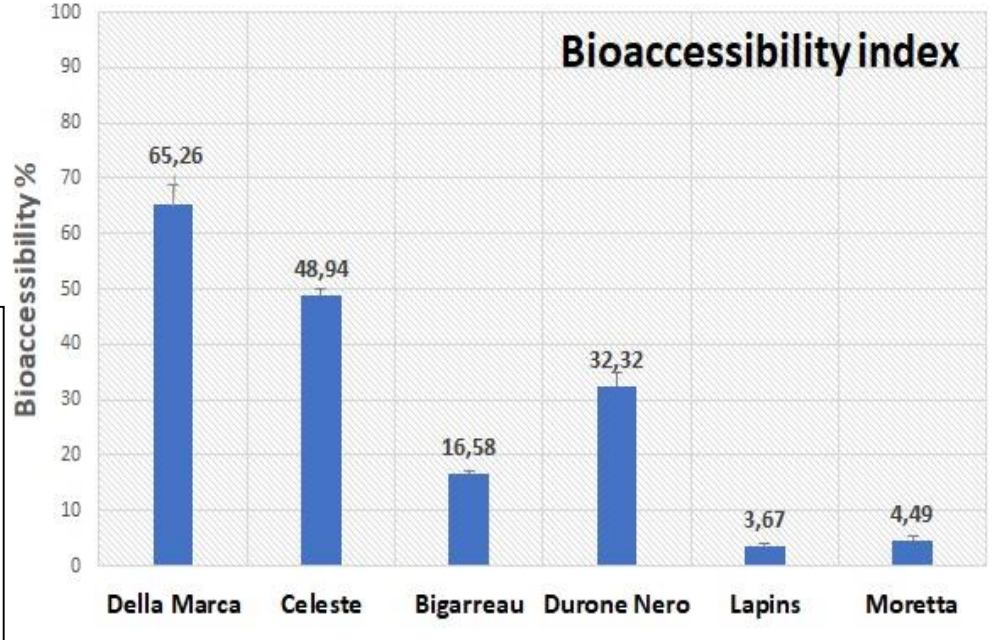
Bioaccessibility	Absorption	Bioactivity
Release of the nutrient from the physicochemical dietary matrix	Transfer across the gut wall (passing through the cells, in-between them or both) to the blood or lymphatic circulation	• systemic distribution • systemic deposition (stores) • metabolic and functional use • excretion (via urine or faeces)

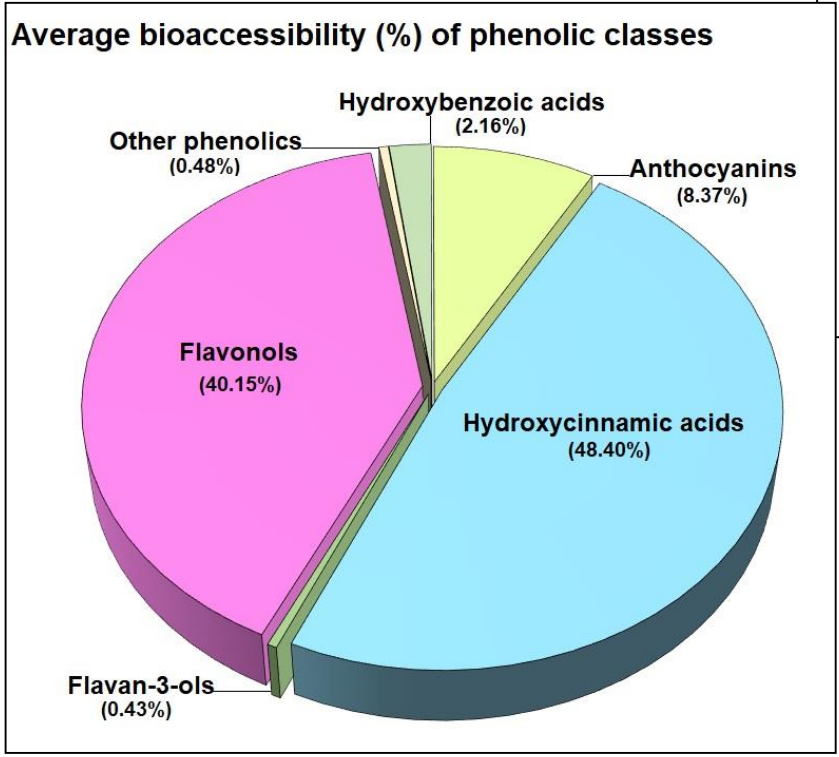
→ Food matrix

→ Molecular interactions

Bioaccessibility

The capacity of polyphenols to reach unmodified the intestinal tract after digestion, where the can carry out their antiproliferative activity against colon-rectal cancer cell.





The bioaccessibility is strictly related to the food matrix and to the cultivars themselves. Also, the phenolic classes display a different bioaccessibility.

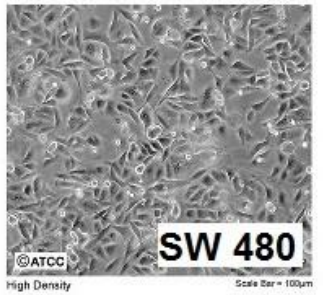
Cytotoxic and anti-proliferative activities

Caco-2 cell line

No cytotoxic and anti-proliferative effect.

SW-480 cell line:

digested and methanol polyphenol-rich extracts showed no cytotoxic activity. *In vitro* digested extracts showed higher anti-proliferative activity than the methanolic extracts in SW480 cell line.

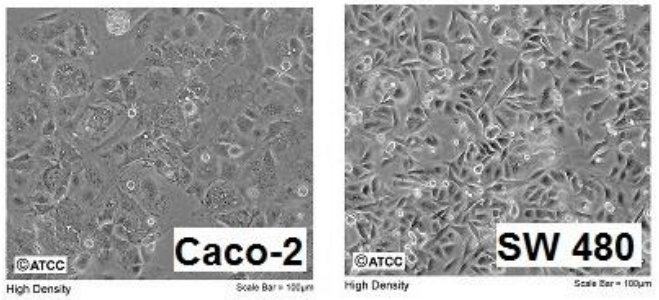


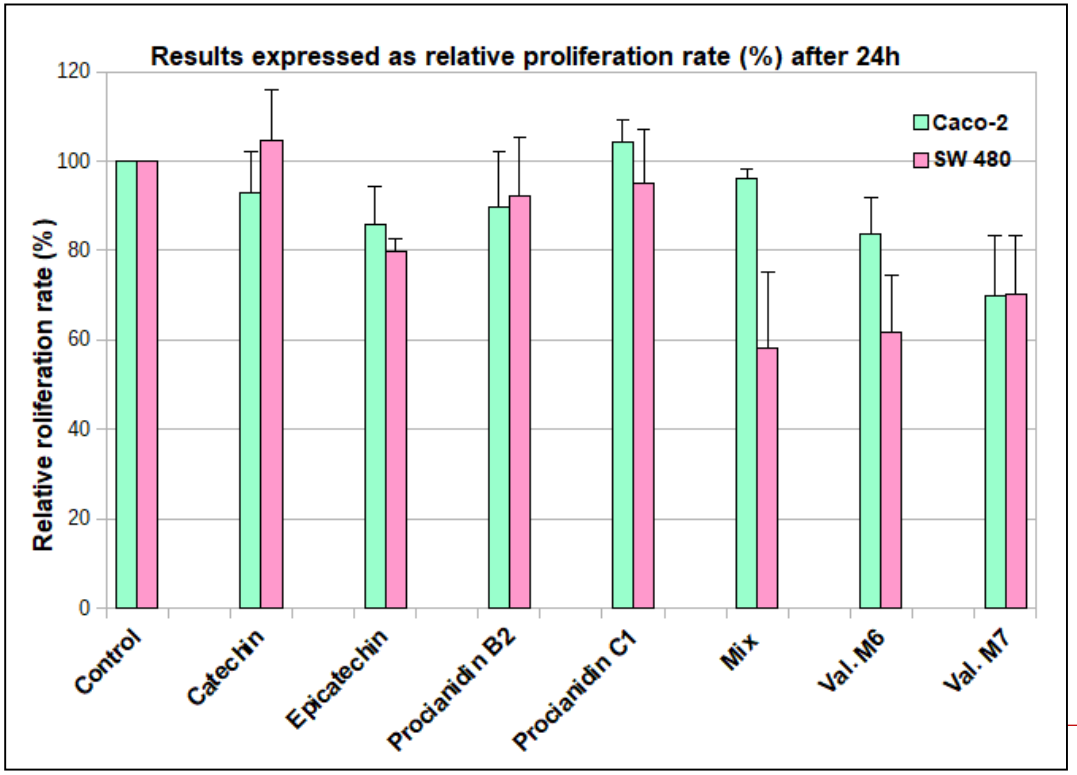
Cherry cultivars	SW 480 IC_{50} µg/mL	
	Chemical extracted	*In vitro* digested
Della Marca	15.43 ± 1.14	7.13 ± 1.05
Celeste	40.67 ± 1.02	23.75 ± 1.01
Bigarreau	7.76 ± 1.08	10.06 ± 1.00
Durone Nero I	17.11 ± 1,01	20.13 ± 1.01
Lapins	6.63 ± 1.07	1.02 ± 0.72
Moretta	11.58 ± 1.08	1.42 ± 0.56

Cytotoxic and anti-proliferative activities

Main metabolites of digested polyphenol-rich food

- Catechin
- Epicatechin
- Procyanidin B2
- Procyanidin C1
- M6 valerolactones
- M7 valerolactones





The absolute value obtained for each sample (concentration 25 mmol/L) is expressed as relative percent to the absolute value obtained for the untreated Caco-2/SW-480 cells and set at 100%.


Let one cherry leads to another!
Thank you
for your kind attention