An in-vitro absorption screen with rosin/resins using everted rat intestinal sacs

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### Introduction

Rosin and Rosin derivatives are chemicals based upon rosin derived from pine trees and are used in a wide variety applications such as adhesives, pharmaceuticals, electronics, paper and as chewing gum and paints.

Rosin and its derivatives are referred to as UVCBs in regulatory programmes and indeed the composition of rosin is complex and variable. No single constituent is present at a concentration > 10%. When derivatives are made from rosin, the number of constituents increases significantly with thousands of potential component structures / isomers. Typical representative structures (not exhaustive) are given below for the acid and neutral fractions and for an ester derivative.

<table>
<thead>
<tr>
<th>Acid Fraction (45 – 65 %, 20 acids)</th>
<th>Neutral Fraction (5 – 15 %)</th>
<th>Rosin Pentaeerythritol ester</th>
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### Method

An ex vivo gut sac was prepared by gently emptying finally excited proximal small intestine over a glass cuv, with full sac isolation (30 min) mixing with TC199 media.

After incubation sacs were removed, washed in TC199 and blotted dry. Sacs cut open and serosal fluid collected. sacs weighed before and after fluid collection to accurately determine the volume of the medium in the sac.

- Everted gut sac was prepared by gently emptying finally excited proximal small intestine over a glass cuv, with full sac isolation (30 min) mixing with TC199 media.
- Rosin adduct esters were not (mono, di- and tri-esters) were not (mono-ester is not present in this product).

### Results – Intestinal Metabolism

Incubation of the three rosin derivatives investigated with proximal intestinal microsomes or cytosol did not show any detectable metabolite formation. Microsomal enzyme activity was confirmed using midazolam as a test substrate (rate of loss of parent calculated as 217 pmol/mg min or mg protein) and 4-nitrophyl acetate and 4-nitrophyl palmitate was used to confirm that cytosolic esterase and lipase activity was intact.

### Conclusions

No metabolism of Rosin, hydrogenated rosin or pentaeerythritol ester was observed by intestinal microsomes or cytosol. So, it is unlikely that metabolism influences absorption in the test system. No oxidation of the test materials was observed. There was absorption of acid fraction rosin constituents across the rat gut in the test system at a rate of 70-80%.

For pentaeerythritol ester, residual, unreacted rosin acids, that are also present in rosin, were absorbed. The di-, tri- and tetra-esters were not (mono-ester is not present in this product), suggesting that it is appropriate to read across from rosin and other lower molecular derivatives to the ester.

Such studies can be used to help select the most appropriate substances to test within a category and to give confidence in predictions across hypotheses, reducing the number of animals used in regulatory testing programmes.

### References