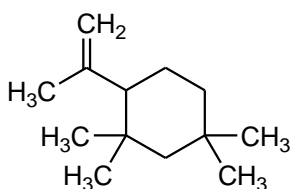


Ladies and Gentlemen: We got'em!

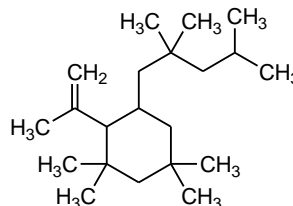
$C_{13}H_{24}$ and $C_{21}H_{40}$ RUBBER OLIGOMERS – NOW AVAILABLE AT TOXIKON!!

Toxikon Europe, a Leading Lab in providing Extractables and Leachables Services for the Pharmaceutical Industry, has developed a Procedure to obtain and analyze the Pure Rubber Oligomers $C_{13}H_{24}$ and $C_{21}H_{40}$ from Rubber Extracts.

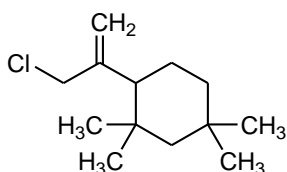
The Procedure to obtain the $C_{13}H_{24}$ and $C_{21}H_{40}$ Oligomers can be Considered as a Crucial First Step in Synthesizing the Brominated Oligomers $C_{13}H_{23}Br$ and $C_{21}H_{39}Br$, which are Considered as Potential Mutagenic Compounds.



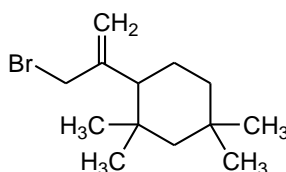
$C_{13}H_{24}$ Rubber Oligomer
1-Isopropenyl-2,2,4,4-tetramethylcyclohexane



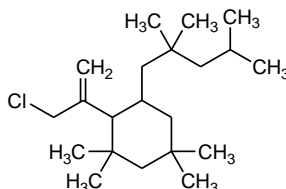
$C_{21}H_{40}$ Rubber Oligomer
1,1,5,5-Tetramethyl-2-(1-methylethenyl)-3-(2,2,4-trimethylpentyl)-cyclohexane



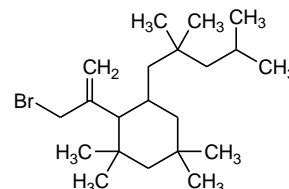
$C_{13}H_{23}Cl$ Rubber Oligomer
1-(1-Chloromethyl-ethenyl)-2,2,4,4-tetramethylcyclohexane



$C_{13}H_{23}Br$ Rubber Oligomer
1-(1-Bromomethyl-ethenyl)-2,2,4,4-tetramethylcyclohexane



$C_{21}H_{39}Cl$ Rubber Oligomer
1,1,5,5-Tetramethyl-2-(1-chloromethylethenyl)-3-(2,2,4-trimethylpentyl)-cyclohexane



$C_{21}H_{39}Br$ Rubber Oligomer
1,1,5,5-Tetramethyl-2-(1-bromomethylethenyl)-3-(2,2,4-trimethylpentyl)-cyclohexane

What were the Drivers to Develop a Procedure in order to Obtain the pure Rubber Oligomeric Compounds?

- All pharmaceutical butylrubbers (butyl, chlorobutyl, bromobutyl) do contain the $C_{13}H_{24}$ and $C_{21}H_{40}$ oligomers as impurities.
- Extractable and subsequent leachable studies have revealed, in certain cases, that $C_{13}H_{24}$ and $C_{21}H_{40}$ oligomers may leach out of the rubber into the drug product.
- For the $C_{13}H_{24}$ -Oligomer, limited scientific work has been performed to elucidate its chemical structure (NMR).
- Further, for the $C_{21}H_{40}$ -Oligomer, however, the chemical structure had not yet been determined experimentally. A structure was postulated, based upon the potential formation pathways from a butyl rubber.
- No scientific/experimental data has yet become available regarding the toxicity of these compounds.
- Obtaining these oligomers will now allow Toxikon Europe to synthesize the brominated oligomers (see below).

What has been Accomplished and what will be the Impact of these Analytical Standards for $C_{13}H_{24}$ and $C_{21}H_{40}$ on the overall Risk Assessments for Pharmaceutical Rubbers?

- After almost 2 years of research, the R&D team at Toxikon Europe has finally developed a procedure to isolate these compounds from rubber extracts via a. o. preparative chromatography.
- Both the $C_{13}H_{24}$ and the $C_{21}H_{40}$ oligomers were obtained with a purity of >90%.
- The chemical structures of both oligomers have been determined (NMR/HR-MS).
- The chemical structures of $C_{13}H_{24}$ and $C_{21}H_{40}$ can finally be included into each report of an Extractable or Leachable Study performed on butyl, chlorobutyl or bromobutyl rubbers.
- For the first time ever, the chemical standards for $C_{13}H_{24}$ and $C_{21}H_{40}$ can be used to perform accurate concentration measurements of these compounds as a leachable in the drug product.
- The $C_{13}H_{24}$ and $C_{21}H_{40}$ compounds are now included into the **TOX-RAY™** screener database in order to allow a rapid identification of the compounds when performing impurities testing or extractables testing.
- Toxikon Europe is preparing to perform AMES testing on both compounds in order to further evaluate their potential for Mutagenicity.
- The results of the AMES tests will allow Toxikon to monitor these compounds as leachables in drug products at appropriate threshold levels

Future Developments: Obtaining the Brominated Oligomers $C_{13}H_{23}Br$ and $C_{21}H_{39}Br$.

- Toxikon Europe is developing a “**Modified AMES**” test to determine the Mutagenicity of extracts of materials, obtained in a Controlled Extraction Study (CES).
- Preliminary results of this “**Modified AMES**” test have shown that CES extracts of bromobutyl rubbers are testing AMES positive, even at a mild extraction ratio, with a clear dose response effect.
- Structure Activity Relationship (SAR) assessments on the extractable compounds of a bromobutyl rubber have shown that the brominated oligomers $C_{13}H_{23}Br$ and $C_{21}H_{39}Br$ are compounds with mutagenic potential.
- Obtaining the Brominated Oligomers will allow Toxikon Europe to perform an AMES test (OECD 471) on both brominated oligomers in order to verify the presumptions of the Mutagenicity potential related to these compounds.
- If the AMES test confirms the conclusions of the SAR assessments, already performed on these compounds, it will be very likely that these compounds will need to be controlled as leachables at lower threshold levels.
- Having access to the brominated oligomeric standards $C_{13}H_{23}Br$ and $C_{21}H_{39}Br$ will allow Toxikon Europe to determine the presence and level of these compounds, leached from the rubbers into the drug product, at appropriate low threshold levels.
- The brominated oligomers are expected to be available at Toxikon Europe in Q3 of 2012.