PDA Pre-Conference Workshop

Impact of Pre-filled Syringe Packaging Components on Biopharmaceuticals

Vienna, November 06, 2017

Extractables and Leachables: Impact of Packaging Components on Therapeutic Proteins

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CONTENT

1. General E/L requirements for Pre-Filled Syringes
2. What makes Biologics (i.e. Therapeutic Proteins) Different?
3. Additional Concerns & Requirements for Biologics / Therapeutic Proteins
4. The PQRI – PDP Recommendations:
   Evaluating the risk of Leachable – Protein Interaction
5. Conclusion

1. General Extractable & Leachable Requirements for Pre-filled Syringe Systems (SVP's)

The selected
Container / Closure system
must be

"suitable for its intended use"

A C/C-system that is suitable for 1 Drug Product, may not be suitable for another Drug Product!

Suitability of Containers:

The Container / Closure system:

1. Should **Protect** the Drug Product
2. Should **not introduce toxic compounds** (safety)
3. Should be **Compatible** with the Drug Product
   - No Change in Drug Product
   - No Change in Packaging
4. Should **guarantee the Performance & Functionality** and guarantee the delivery of the drug/dose

C/C should **not introduce Toxic** Compounds:

- **Leachables** from the container closure
- **Leachables that undergo a physical/chemical change** in the drug product
- Leachables that **react with the API**
- **Toxicological Assessment** should address potential Safety Issues

1. GENERAL E/L-REQUIREMENTS – PRE-FILLED SYRINGES

PRIMARY PACKAGING *(not limitative)*	
YEAR	GUIDANCE / GUIDELINE / RECOMMENDATION...
<1999	21CFR 211.94(a) "DRUG PRODUCT CONTAINERS AND CLOSURES" *...not reactive, additive, absorptive to alter safety, identity, strength, quality or purity of drug...*
1999	"CONTAINER/CLOSURE SYSTEMS FOR PACKAGING HUMAN DRUGS AND BIOLOGICS" (DRAFT FDA-Guidance for Industry)
2003	EU COMMISSION DIRECTIVE 2003/63/EC, (§ 3.2.2.2 g) *CCS-information is part of the Market Authorization dossier.*
2005	"GUIDELINE ON PLASTIC IMMEDIATE PACKAGING MATERIALS" (EMEA Guideline) *Contains "Decision Tree" for different dosage forms*
2006	ICH Q8 "PHARMACEUTICAL DEVELOPMENT", § 2.4 CCS
2006	PQRI RECOMMENDATIONS – Safety Thresholds and Best Practices for Extractables & Leachables in OINDP
2014	USP <1663> (Extractables) & USP <1664> (Leachables)
2015	ICH M7: DNA Reactive Impurities in Pharmaceuticals
2017	PQRI – PDP/ODP RECOMMENDATIONS (soon to be released) Safety Thresholds and Best Practices for Extractables & Leachables in PDP/ODP

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1. GENERAL E/L-REQUIREMENTS – PRE-FILLED SYRINGES

Examples of Packaging Concerns for Common Classes of Drug Products

Degree of Concern Associated with the Route of Administration	Likelihood of Packaging Components – Dosage Form Interactions		
	High	Medium	Low
Highest	Inhalation Aerosols and Sprays	Injections and Injectable Suspensions; Inhalation Solutions	*Sterile Powders and Powders for Injection; Inhalation Powders*
High	Transdermal Ointments and Patches	Ophthalmic Solutions and Suspensions; Nasal Aerosols and Sprays	-
Low	Topical Solutions and Suspensions, Topical and Lingual Aerosols, Oral Suspensions and Solutions	-	Oral Tablets and Oral (Hard and Soft Gelatin) Capsules; Topical Powders; Oral Powders

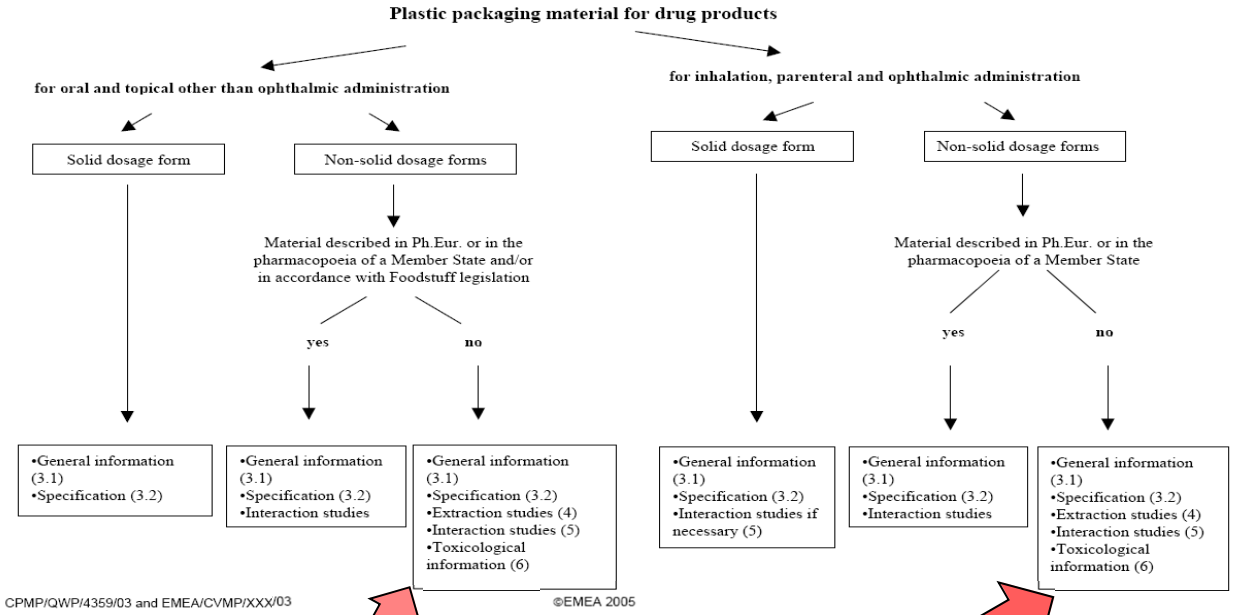
Revision of "Table 1" in USP <1664>,

Originally Included into the FDA Guidance for Industry (1999):

"Container/Closure systems for Packaging Human Drugs and Biologics"

"OTHER" DOSAGE FORMS:

LIKELIHOOD OF **INTERACTION** IS **HIGH**



E.P. COMPENDIAL TESTING IS REQUIRED BUT **NOT SUFFICIENT**.

ADDITIONAL REQUIREMENTS

1. **EUROPEAN PHARMACOPOEIA TESTS**
2. **EXTRACTION STUDIES**
3. **INTERACTION STUDIES (INCLUDING § 5.1 MIGRATION STUDIES)**

HOW to comply: Extractables Assessment

USP <1663> Monograph

"Assessment of Extractables Associated with Pharmaceutical Packaging/Delivery Systems"

This is an **INFORMAL** Monograph

PQRI – Parenteral &Ophthalmic Drug Products

Best Demonstrated Practice Recommendations: **Chemistry** & Toxicology

This is a **RECOMMENDATION**

These Documents ar either **INFORMAL** or **RECOMMENDATIONS**

Allow Flexibility in Design

What is the *intent*? => **Strategy** of testing

How to design the study for the envisioned intent? => **Tactics**

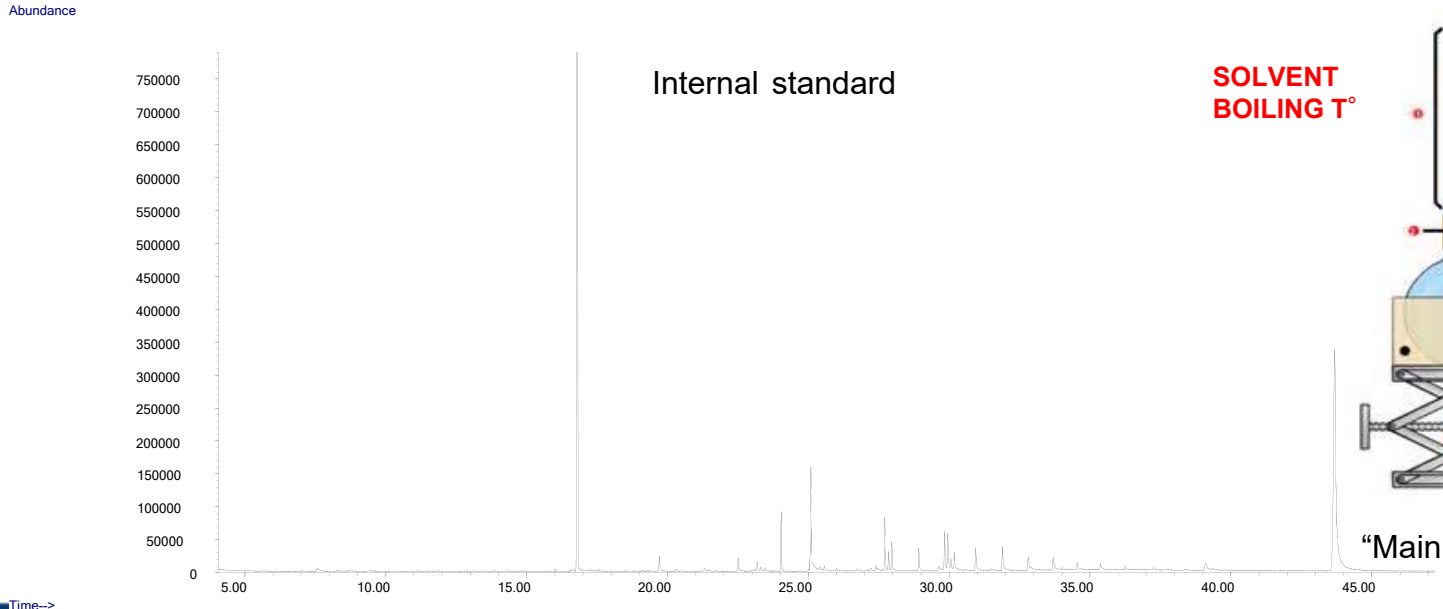
However, Justification is Needed!

Both **Identifying the Necessity** for an Extraction Study, as well as **Justifying the Design**, is the responsibility of the Holder of the NDA.

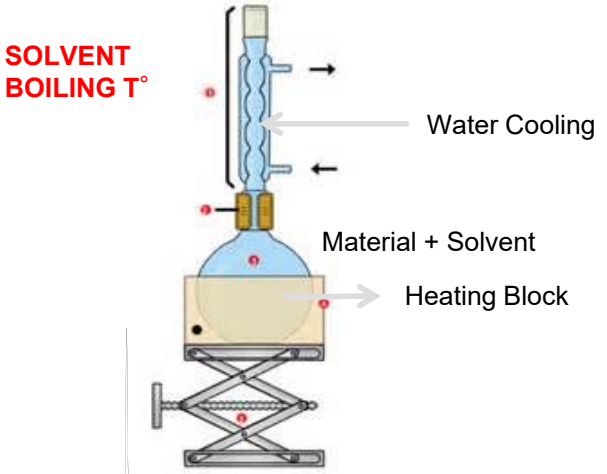
DEFINITIONS

EXTRACTABLES (from USP <1663>):

- Organic & Inorganic Chemical Entities
- released from
  - *a pharmaceutical packaging/delivery system*
  - *packaging component*
  - *packaging material of construction*
- into an extraction solvent under laboratory conditions



EXAMPLE: REFLUX



What is the **PURPOSE** of an Extraction Study?

- **Material Characterization** of the Packaging Components
- "**Impurities Profiling**" of the Materials
  - Identify as Many Compounds as Possible
  - Identify "Bad Actors" in the Materials
- **Early Risk Evaluation**: Potential *Patient Exposure* to Chemical Entities
- Allows to establish **Leachables – Extractable *correlations***
- **Identify & Justify Compounds** that may need to be **Monitored as Leachable**
  - Toxicity
  - Concentration in the Materials
  - Risk for Migration

What is the **PURPOSE** of an Extraction Study?

- Understand **the Effects of various Processes** on components
- Establish **Worst Case potential Leachables Profile**, when it is not scientifically possible to determine Leachables
- Use of **Extraction solutions** which are **"Compatible" with Screening** techniques: CLEAN SOLVENTS

- Typically Not as a Final Step in the Safety Assessment!

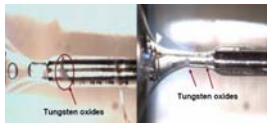
VARIABLES that may/will have an impact on the Study Design of an Extractable Study

- The **Classification & Specific Requirements** per Drug Product
  - Table 1 in FDA C/C-Guidance (1999)
  - Decision tree in the EMA-Guideline (2005)
- The **Composition of the DP**, in contact with the C/C system
- The **Type of contact** between the DP and the C/C system
  - Primary Packaging
  - Secondary Packaging (e.g. Needle Shield, Label,...)
- The **Types of Materials** used in te Manufacture of the C/C
  - E.g. Rubber versus Polyolefin for BFS
- The **Knowledge on the Composition** of Materials (from Vendor)
  - Additives, Catalysts, Oligomers, Colorants,...
- The **Use of the Data**
  - Only for this particular application, or also for other DP?
- Primary Packaging versus Manufacturing Equipment

Parameters To be Considered for an Extraction Study

- ✓ Extraction **Solvents**
- ✓ Extraction **Techniques**
- ✓ Extraction **Conditions** (Temperature, time)
- ✓ Extraction **Ratio's - Stoichiometry**
- ✓ *Analytical **Techniques***
  - *Screening Techniques*
  - *Targeted analysis for specific compounds*

1. GENERAL E/L-REQUIREMENTS – PRE-FILLED SYRINGES



(COATED) RUBBER

MONOMER REMAINDERS & POLYMER FRAGMENTS

FILLERS: *Clay, Talc, Carbonates…*

ANTIOXIDANTS: *BHT, Irganox 1010, Irgafos 128, ...*

CURING AGENTS: *S,S-Donors, Phenol-Formaldehyde…*

ACTIVATORS:

ZnO / Stearic Acid

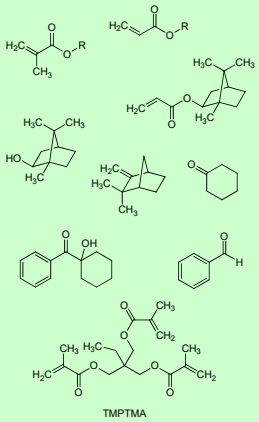
ACCELERATORS:

Carbamates, Sulfenamides…

OTHERS: *Pigments, Stabilizers, Release agents…*

NEEDLE GLUE

- POLYMER FRAGMENTS
- UV CURING-ACTIVATORS



TUNGSTEN (W)

A Tungsten pin is used in the production of glass pre-filled syringes to keep the syringe hub open (cavity where the staked needle is glued in)

GLASS BARREL

Major: Silica (SiO_2)

Alkali (e.g. Na_2O)

Minor:

K (K_2O),

B (B_2O_3),

Ca (CaO),

Al (Al_2O_3)

Colored glass: Fe_2O_3,TiO_2,CuO, Mn^{3+}

Sulfate (from dealkalization)

Silicone oil (provides lubricity)

COC/COP/PP BARREL

POLYMER FRAGMENTS SOLVENTS

ANTIOXIDANTS: *BHT, Irganox 1010, ...*

ACID SCAVENGERS: *Stearate,...*

LUBRICANTS: *Metal Stearate, FA Esters, ...*

WAXES

SLIP ADDITIVES: *Erucamide, Oleamide, ...*

PLASTICIZERS: *DEHP, ...*

RELEASE AGENTS

PIGMENTS

LABEL (ADHESIVE)

Permeable Plastic Materials (e.g. PP, …)

INK and ADHESIVE constituents in a complex composition, but at low concentrations

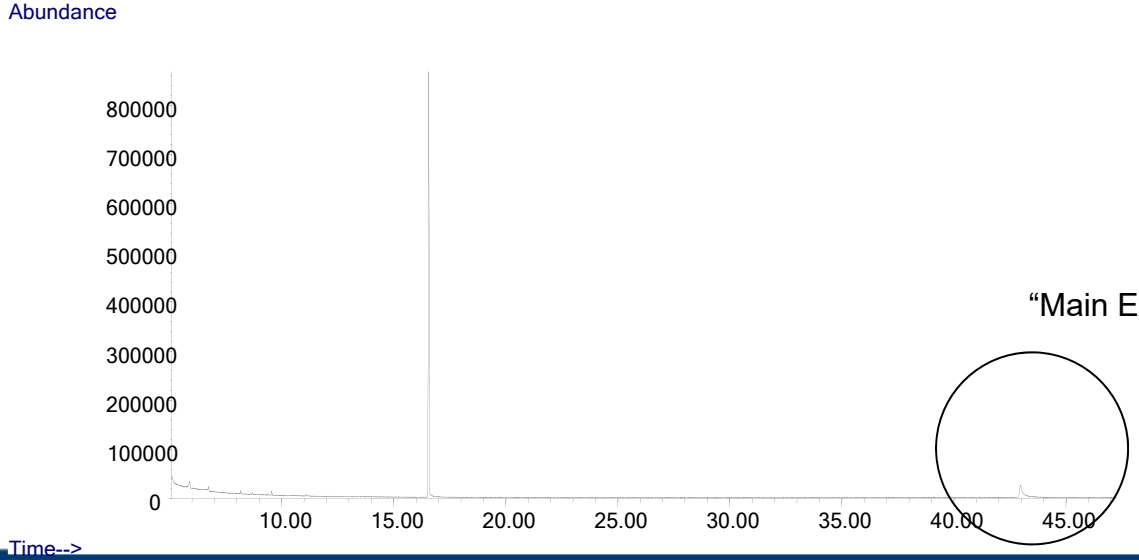
POLYMER FRAGMENTS, SOLVENTS, PHOTO-INITIATORS, STABILIZERS, TACKIFIERS, ...

e.g. Acrylates, PVA, NR, Benzophenone, Irgacure 184, Irgacure 651, Irganox 1010, Dehydroabietic Acid, DCHP, TBPP, Siloxanes, ...

1. GENERAL E/L-REQUIREMENTS – PRE-FILLED SYRINGES

LEACHABLES (from USP <1664>):

- Foreign Organic and Inorganic Chemical Entities
- present in a packaged drug product because they have leached into the packaged drug product from
  - *the packaging/delivery system*
  - *packaging component*
  - *packaging material of construction*
- under normal conditions of storage and use
- or during accelerated drug product stability studies



"Main Extractable" becomes a leachable!!

1. GENERAL E/L-REQUIREMENTS – PRE-FILLED SYRINGES

- Trying to assess the Leaching Behaviour
- Assess Potential Toxic Consequences = **SAFETY**
- Assess the Impact on **Drug Product QUALITY**
- Focus on Quantification of **"TARGET" COMPOUNDS**

  Known Polymer Additives Used *(and their degradation products)*
  Validation Package of Container Suppliers
  Extractables Study Information

- **"SIMULATED USE"** CONDITIONS

  Storage Time / Temperature / Humidity
  Conditions: Similar to **Stability Studies**
  Pharmaceutical Formulation as Contact Solution

- **QUANTITATIVE METHODS**

  Limit test, evaluated at eg the AET level or any other relevant level,
  Fully **Developed and Validated Methods** (ICH Q2B)

1. GENERAL E/L-REQUIREMENTS – PRE-FILLED SYRINGES

USP <1664> SOME REQUIREMENTS (although informal), ENFORCED BY THE FDA

- Evaluate **three batches** your to-be-marketed drug product for leachables
- Include assessments of **multiple time points** over the course of the stability study
- **Justify your selection of leachables** to be monitored in a "formal" leachable study,
- Use an **Analytical Evaluation Threshold** that corresponds to **5 µg/day**
  - *Find and Identify everything above the Qualification Threshold (QT) of 5 µg/day*
  - *Can only be achieved with a leachable study, run in "screening" mode*
- Provide detailed and quantitative (if possible) **extractables – leachables correlations**
- Include **secondary container closure system** components
- If you **cannot achieve the AET,** then this must be **justified** and the **methods' QL** will be used**.**
- The assessment should include include an assessment of **leached elemental impurities**
- Provide a **summary table**

From Presentation of D. Mellon (FDA, CDER) at the PQRI-PDA joint workshop on Container Closure, Devices and Delivery Systems, Washington 2 – 3 October 2017

2. What makes Biologics Different?

UNIQUE CHALLENGES OF BIOLOGICS

- **Administration** by injection is among those of highest concern
- **Likelihood of interaction** between packaging component and injectable dosage **is high**
- **Biologics are complex**
  - ✓ Large molecular weights
  - ✓ Abundance of binding sites on the surface (hydrophilic and hydrophobic)
  - ✓ Heterogeneous mixtures
- **Biologics are sensitive to structural modifications**
  - ✓ *Safety* considerations (immunogenicity)
  - ✓ *Efficacy* considerations (loss of activity, formation of neutralizing antibodies)
  - ✓ *Quality* considerations (protein aggregates, stability)

I. *Markovic (2014) regulatory Perspective on Extractables & Leachables in Biologics, ASTM E55 Workshop, May 21, 2014*
II. *Kim Li (2016) Predicting the risk of extractables and leachables (E&L) interacting with Therapeutic proteins, presentation at PEPTALK 2016*

E&L STRATEGY FOR BIOLOGICS MUST ADDRESS BOTH SAFETY AND QUALITY CONCERNS

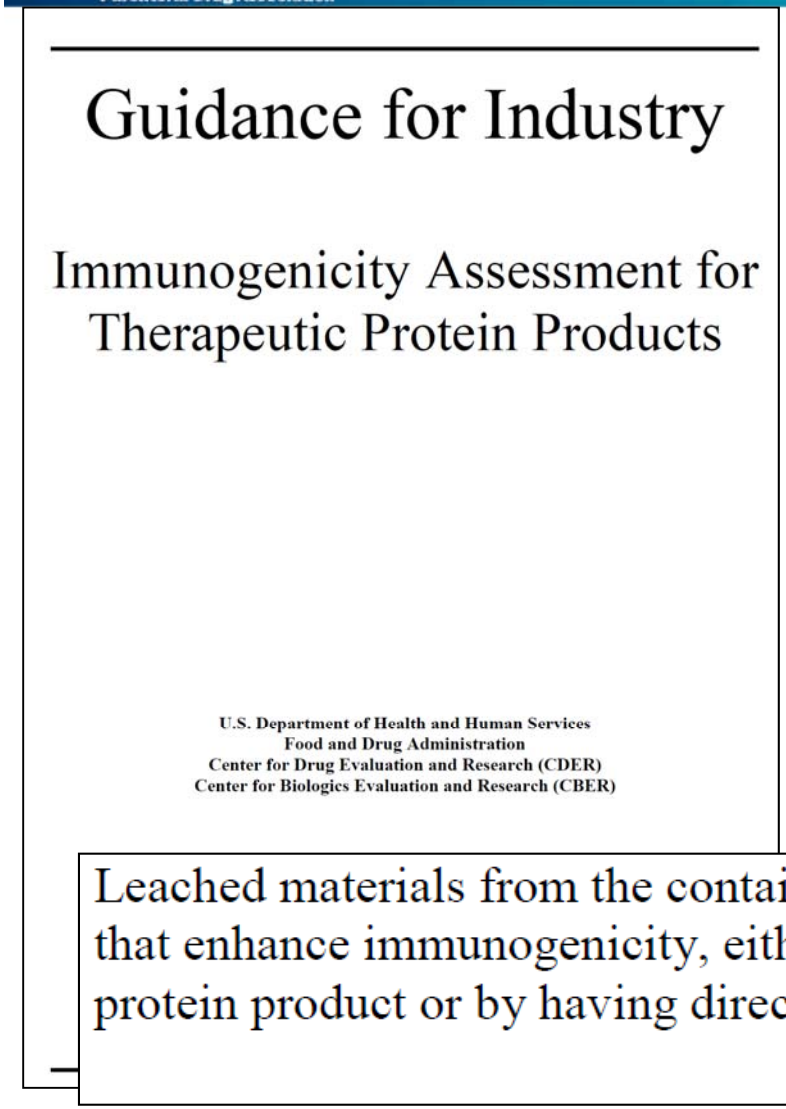
- The strategy can be applied to drug containers, drug delivery systems and single-use systems
- It should incorporate key ICH Q9 concepts, science- and risk based
- It should be phase appropriate, progressing from screening and selection of critical components to life cycle management of drug products

Evaluation of E/L should provide understanding of toxicity profile and likelihood of interaction with drug, excipient and/or package

I. *Kim Li (2016) Predicting the risk of extractables and leachables (E&L) interacting with Therapeutic proteins, presentation at PEPTALK 2016*

3. Additional E/L-Requirements - Biologics

3. ADDITIONAL REQUIREMENTS – THERAPEUTIC PROTEINS



Guidance for Industry

Immunogenicity Assessment for Therapeutic Protein Products

U.S. Department of Health and Human Services
Food and Drug Administration
Center for Drug Evaluation and Research (CDER)
Center for Biologics Evaluation and Research (CBER)

CONSEQUENCES FOR EFFICACY

– some of the concerns:

Development of "***Neutralizing Antibodies***" *(e.g. through chemically modified therapeutic protein product)* can **block the efficacy** of therapeutic protein products

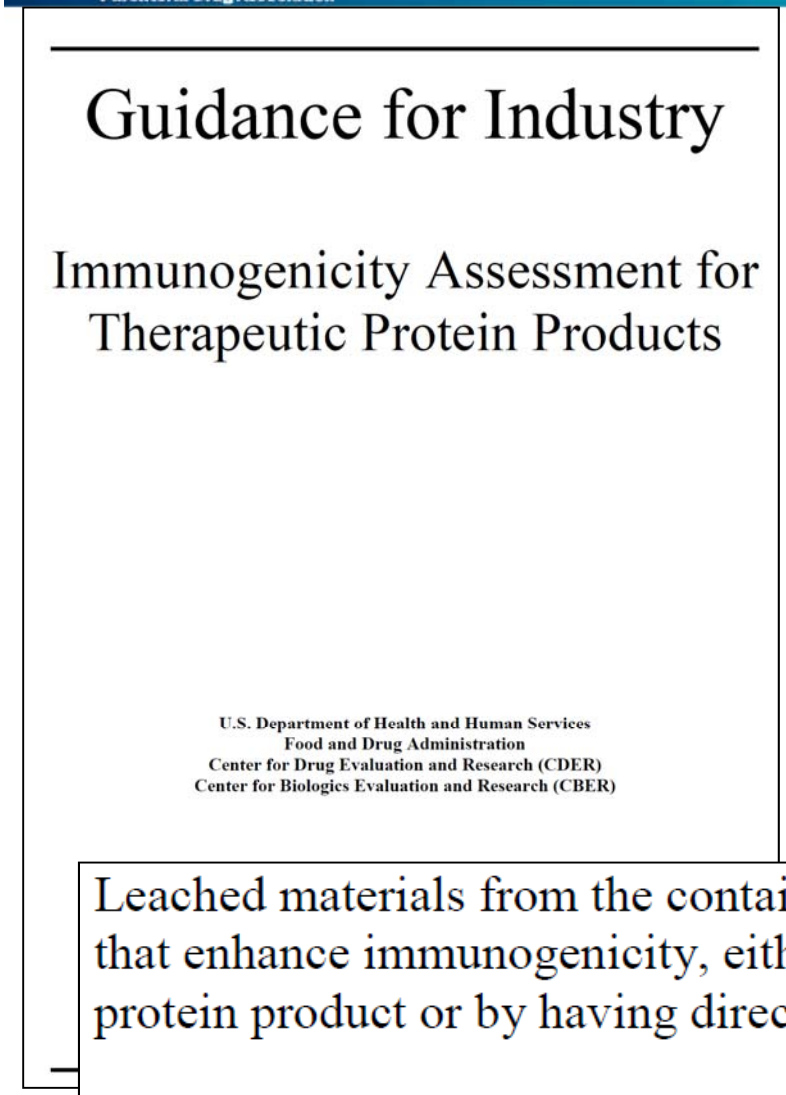
May also change the Pharmacokinetics

- Enhancing Clearance
- Or Prolonging Product Activity

> Leached materials from the container closure system may be a source of materials that enhance immunogenicity, either by chemically modifying the therapeutic protein product or by having direct immune adjuvant activity.

FDA Guidance for Industry, 2014

3. ADDITIONAL REQUIREMENTS – THERAPEUTIC PROTEINS



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CONSEQUENCES FOR SAFETY

– some of the concerns:

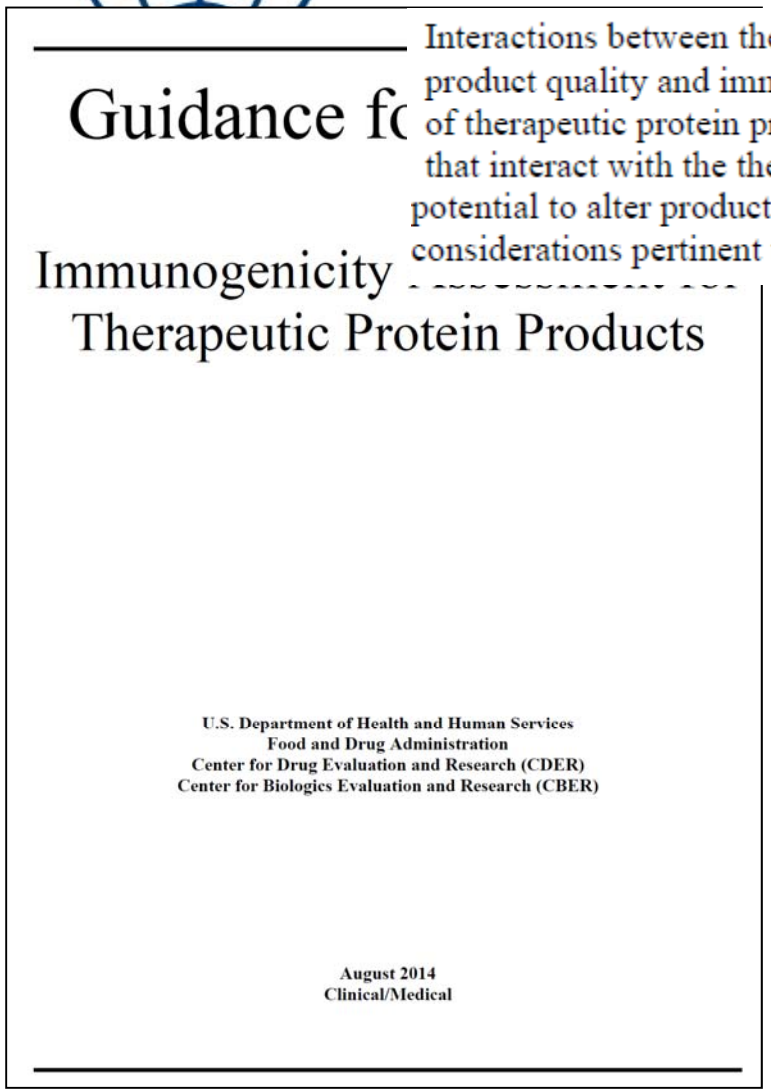
(e.g. "...through chemically modified therapeutic protein product...")

- Anaphylaxis (serious, accute allergenic reaction)
- Cytokine Release Syndrome
- "Infusion Reactions"
- Non-Acute Reactions
- Cross-reactivity to Endogeneous Proteins

Leached materials from the container closure system may be a source of materials that enhance immunogenicity, either by chemically modifying the therapeutic protein product or by having direct immune adjuvant activity.

FDA Guidance for Industry, 2014

2. WHAT MAKES BIOLOGICS DIFFERENT?



Guidance fc

Immunogenicity
Therapeutic Protein Products

U.S. Department of Health and Human Services
Food and Drug Administration
Center for Drug Evaluation and Research (CDER)
Center for Biologics Evaluation and Research (CBER)

August 2014
Clinical/Medical

Interactions between therapeutic protein products and the container closure may negatively affect product quality and immunogenicity. These interactions are more likely with prefilled syringes of therapeutic protein products. These syringes are composed of multiple surfaces and materials that interact with the therapeutic protein product over a prolonged time period and thus have the potential to alter product quality and immunogenicity. The following are other container closure considerations pertinent to immunogenicity:

FDA Guidance for Industry, 2014

"... Interactions are more likely with ***pre-filled syringes*** *of therapeutic protein products..."*

"... ***Materials that interact with the therapeutic protein product*** *over a prolonged time and thus* ***have the potential to alter product quality and immunogenicity..."***

3. ADDITIONAL REQUIREMENTS – THERAPEUTIC PROTEINS

- Glass and air interfaces can denature proteins and cause aggregation in glass syringes and vials.
- Glass vials have been known to delaminate at higher pH and with citrate formulations, potentially creating protein-coated glass particles, which may enhance immunogenicity of the therapeutic protein product (Fradkin et al. 2011). → Delamination
- Silicone oil-coated syringe components provide a chemical and structural environment on which proteins can denature and aggregate. → Silicone Oil
- Appropriate in-use stability studies should be performed to confirm that conditions needed to maintain product quality and prevent degradation are adequately defined. → In Use Stability Studies
- Leached materials from the container closure system may be a source of materials that enhance immunogenicity, either by chemically modifying the therapeutic protein product or by having direct immune adjuvant activity, including the following: → Directly – Indirectly leading to enhanced immunogenicity
    - Organic compounds with immunomodulatory activity may be eluted from container closure materials by polysorbate-containing formulations: a leachable organic compound involved in vulcanization was found in a polysorbate formulated product when the stopper surfaces were not Teflon coated (Boven et al. 2005). → Eprex - Case
    - Metals that oxidize and aggregate therapeutic protein products or activate metalloproteinases have been found in various products contained in prefilled syringes or in vials. For example, tungsten oxide that leached from the syringe barrel was reported to cause protein aggregation (Bee et al. 2009); and leached metals from vial stoppers caused increased proteolysis of a therapeutic protein because of activation of a metalloproteinase that co-purified with the product. → Tungsten Oxide Leading to Protein Aggregation

3. ADDITIONAL REQUIREMENTS – THERAPEUTIC PROTEINS

Sponsors should conduct a comprehensive extractables and leachables laboratory assessment using multiple analytical techniques to assess the attributes of the container-closure system that could interact with and degrade protein therapeutic products.

Because the United States Pharmacopeia *elastomeric closures for injections* tests do not adequately characterize the impact of leachables in storage containers on therapeutic protein products under real-time storage conditions, leachables must be evaluated for each therapeutic protein product in the context of its storage container under real-time storage conditions[8].

Testing for leachables should be performed on the product under stress conditions,[9] as well as under real-time storage conditions, because in some cases the amount of leachables increases dramatically over time and at elevated temperatures. Product compatibility testing should be performed to assess the effects of container closure system materials and all leachables on product quality.

4. The PQRI – P(O)DP Recommendations:

Evaluating the risk of Leachable – Protein Interaction

4. PQRI – THE RISK OF LEACHABLES - PROTEIN INTERACTIONS

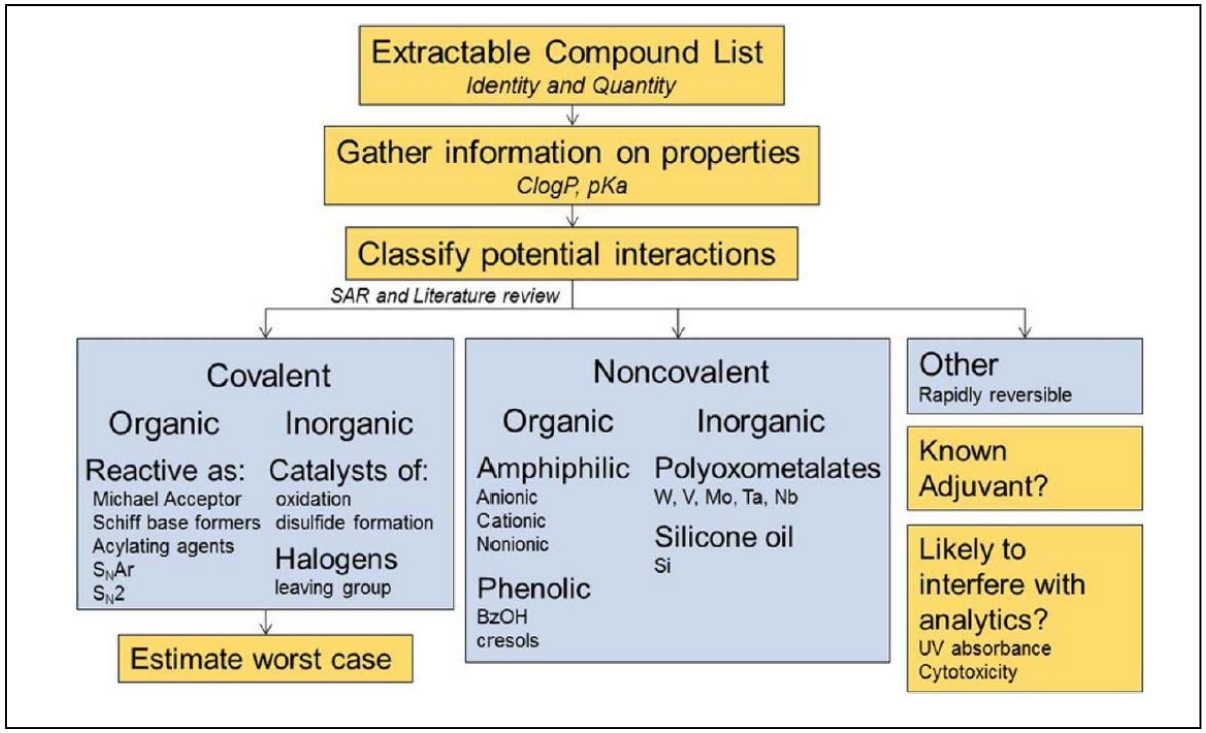
PQRI Product Quality Research Institute

The PODP Best Demonstrated Practice Recommendations – Chemistry and Toxicology

- **Toxicology** – Safety Concern Thresholds
- **Chemistry** – Recommendations for E & L Set-up on PDP
- For Parenteral DP:
  a "New" Issue – Compatibility Issues with Biopharmaceuticals

4. PQRI – THE RISK OF LEACHABLES - PROTEIN INTERACTIONS

Different Types of Interactions
Between Leachables and Therapeutic Proteins / Biologics
which can affect the Quality of the Biologic DP



Source: Kim Li et al. PDA J Pharm Sci and Tech 2015;69:590-619

4. PQRI – THE RISK OF LEACHABLES - PROTEIN INTERACTIONS

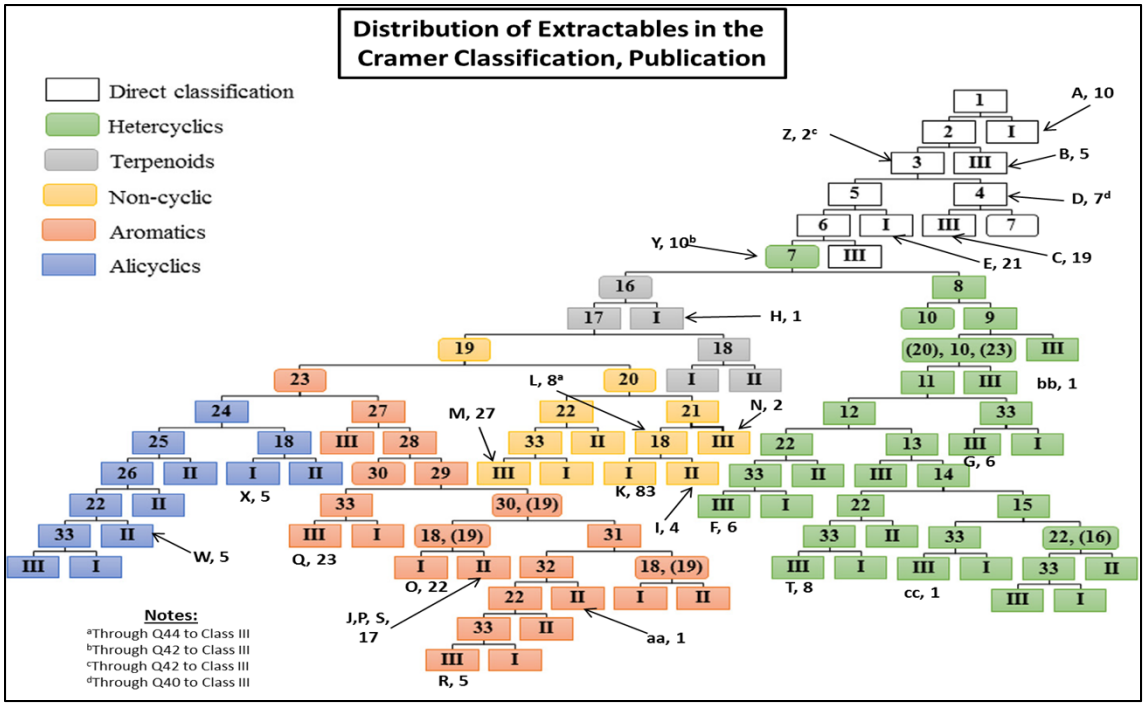
"Because of the irreversible nature of the protein modification, covalent binding presents a higher risk of affecting product quality attributes as compared to noncovalent binding"

A Partial List of Extractables that Could Induce Protein Modification via Covalent Binding

Agents or Mechanisms	Compounds
Michael acceptors	(2E,9Z)-Ethyl 12-oxoctadeca-2,9-dienoate 1-((2-Ethylheptyl)oxy)-**1**-oxopropan-2-yl (1-((2-ethylhexyl)oxy)-1-oxopropan-2-yl) maleate 1-((3-Butyl-4-methylcyclohexa-1,5-dien-1-yl)methoxy)-1-oxopropan-2-yl (1-((4-ethyl-3-methylbenzyl)oxy)-1-oxopropan-2-yl) maleate 1,6-Hexanedioldiacrylate 13-oxooctadeca-9,11-dienoic acid 1-Hydroxy-2-propyl methacrylate 1-oxo-1-(((2E,5E)-2-((Z)-prop-1-en-1-yl)octa-2,5-dien-1-yl)oxy)propan-2-yl (1-oxo-1-(((E)-2-((Z)-prop-1-en-1-yl)hept-2-en-1-yl)oxy)propan-2-yl) maleate 2,6 Di(tert-butyl)-4-hydroxy-4-methyl-2,5-cyclohexandien-1-one (BHT-OH) 2,6-di-tert-butyl-4-methylene-2,5-cyclohexandienone (BHT-quinone-methide) 2,6-Di-tert-butyl-p-benzoquinone (BHT-quinone) 2-Hydroxypropyl methacrylate 3-tert-Butyl-4-hydroxyanisole 4-ethyl 1-methyl 2-hexanoylsuccinate 7,9-bis(tert-butyl)-1-oxaspiro[4,5]deca-6,9-diene-2,8-dione (BODDD) Acrylic Acid Bis(1-((2-ethylhexyl)oxy)-1-oxopropan-2-yl) maleate Isomers Dibutylmaleate Dihexyl maleate (Methyl maleate) Isobornyl methacrylate Methacrylic acid (MAA) Tetraethylene glycol dimethacrylate Tetrahydrofurfuryl methacrylate

Source: Kim Li et al. PDA J Pharm Sci and Tech 2015;69:590-619

4. PQRI – THE RISK OF LEACHABLES - PROTEIN INTERACTIONS



Question:

Could we ever come to a situation where a **similar decision tree** could be established which **identifies the risk of interaction between a Leachable** and a Biologic DP, based upon

- **Physico-chemical properties** of the Leachable
- **Structural** evaluations of the Leachable

4. PQRI – THE RISK OF LEACHABLES - PROTEIN INTERACTIONS

- **Covalent** Interactions with **Organic Leachables** are **Predictable**
  (see examples in next slides)

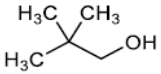
- **Covalent** Interactions with **Inorganic Leachables** and **Non-Covalent** Interactions are **more difficult to predict**.

  Information for Non-Covalent Binding should be based on:
  - ✓Literature
    - ✓*Silicone Oil*
    - ✓*Tungsten Oxide*
    - ✓*Cresols*
    - ✓*Metals, and metal oxides leading to protein oxidation/truncation*
  - ✓Case Studies
  - ✓Presentations
  - ✓Own Experiences
  - ✓...

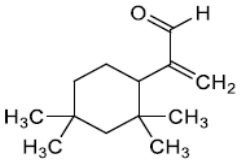
4. PQRI – THE RISK OF LEACHABLES - PROTEIN INTERACTIONS

EXTRACTABLES PROFILE

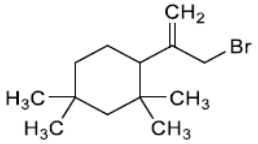
Cyclohexane
CAS# 110-82-7



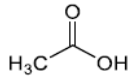
2,2-Dimethyl-1-propanol
CAS# 75-84-3



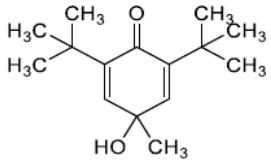
2-(2,2,4,4-Tetramethylcyclo-hexyl)acrylaldehyde
ToxID# 559



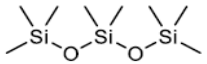
$C_{13}H_{23}Br$ Rubber Oligomer
ToxID 36/44/45



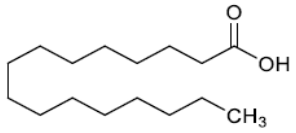
Acetic acid
CAS# 64-19-7



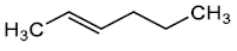
BHT-OH
CAS# 10396-80-2



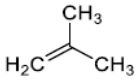
Octamethyl-trisiloxane
CAS# 107-51-7



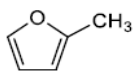
Palmitic acid
CAS# 57-10-3



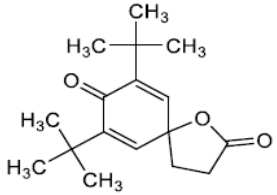
***trans*-2-Hexene**
CAS# 4050-45-7



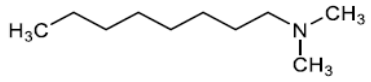
Isobutene
CAS# 115-11-7



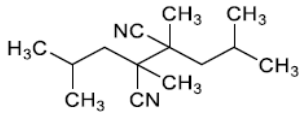
2-Methylfuran
CAS# 534-22-5



7,9-Di-*tert*-butyl-1-oxaspiro-[4,5]deca-6,9-diene-2,8-dione
CAS# 82304-66-3



***N,N*-Dimethyloctylamine**
CAS# 7378-99-6

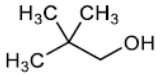


2,3-Dimethyl-2,3-diisobutyl succinonitrile
CAS# 80822-82-8

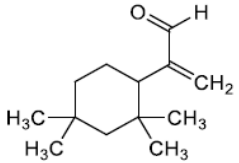
4. PQRI – THE RISK OF LEACHABLES - PROTEIN INTERACTIONS

EXTRACTABLES PROFILE

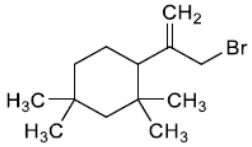
Cyclohexane
CAS# 110-82-7



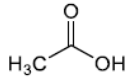
2,2-Dimethyl-1-propanol
CAS# 75-84-3



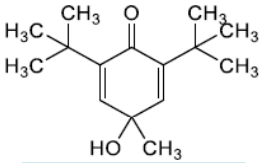
2-(2,2,4,4-Tetramethylcyclo-hexyl)acrylaldehyde
ToxID# 559
α,β-unsaturated carbonyl compound
Michael acceptor



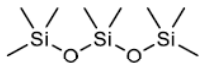
$C_{13}H_{23}Br$ Rubber Oligomer
ToxID 36/44/45
Alkyl halide
Alkylating agent



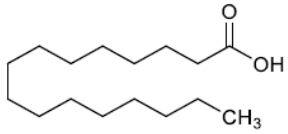
Acetic acid
CAS# 64-19-7



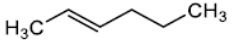
BHT-OH
CAS# 10396-80-2
Ketone
Schiff base former



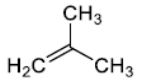
Octamethyl-trisiloxane
CAS# 107-51-7



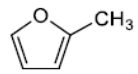
Palmitic acid
CAS# 57-10-3



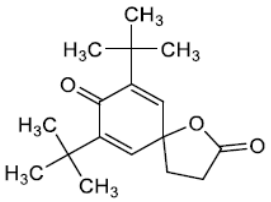
***trans*-2-Hexene**
CAS# 4050-45-7



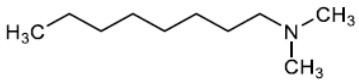
Isobutene
CAS# 115-11-7



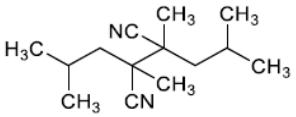
2-Methylfuran
CAS# 534-22-5



7,9-Di-*tert*-butyl-1-oxaspiro-[4,5]deca-6,9-diene-2,8-dione
CAS# 82304-66-3
Acylating agent



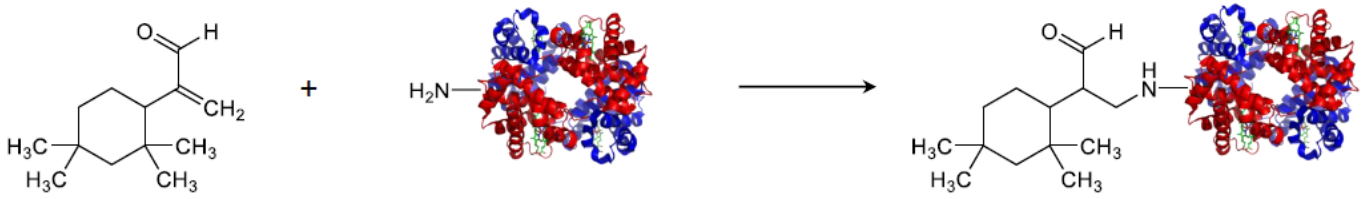
***N,N*-Dimethyloctylamine**
CAS# 7378-99-6



2,3-Dimethyl-2,3-diisobutyl succinonitrile
CAS# 80822-82-8

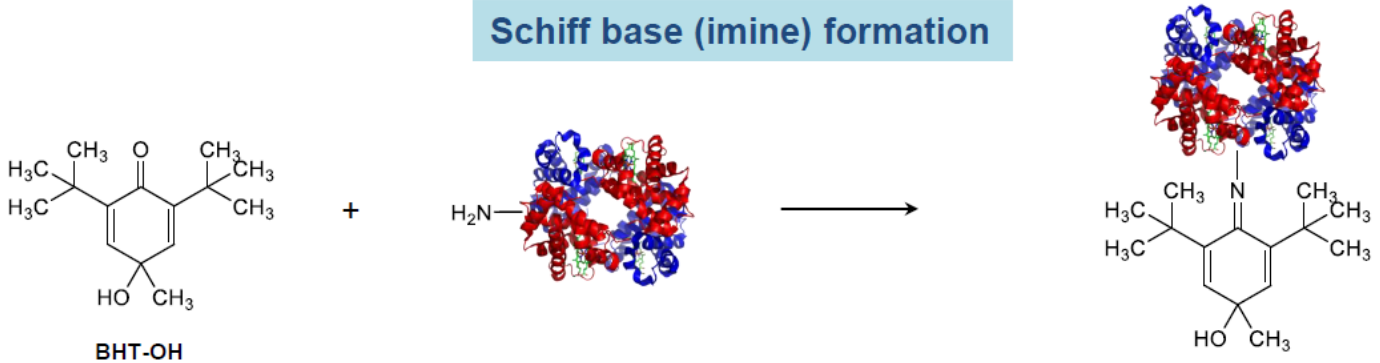
4. PQRI – THE RISK OF LEACHABLES - PROTEIN INTERACTIONS

Michael addition



2-(2,2,4,4-Tetramethylcyclo-hexyl)acrylaldehyde
ToxID# 559
α,β-unsaturated carbonyl compound
Michael acceptor

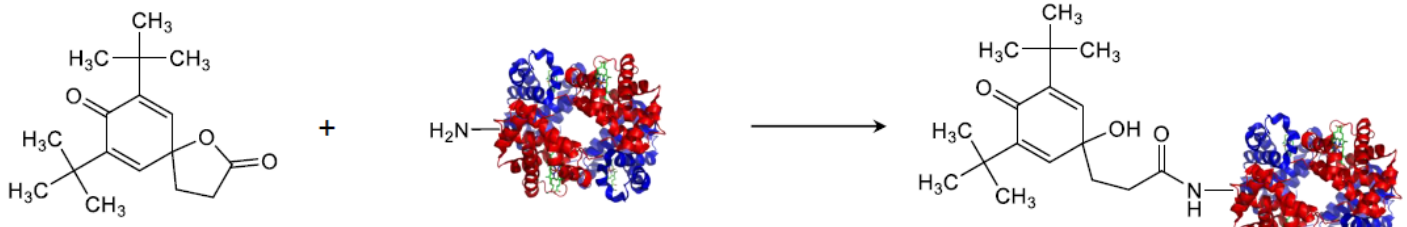
Schiff base (imine) formation



BHT-OH
CAS# 10396-80-2
Ketone
Schiff base former

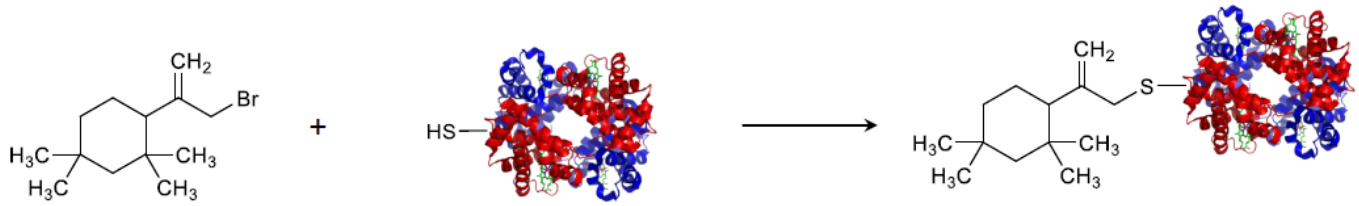
4. PQRI – THE RISK OF LEACHABLES - PROTEIN INTERACTIONS

Acylation



7,9-Di-*tert*-butyl-1-oxaspiro-[4,5]deca-6,9-diene-2,8-dione
CAS# 82304-66-3
Acylating agent

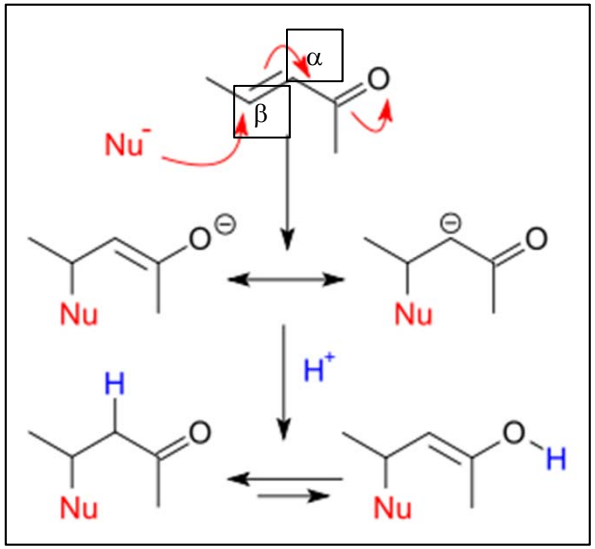
Alkylation



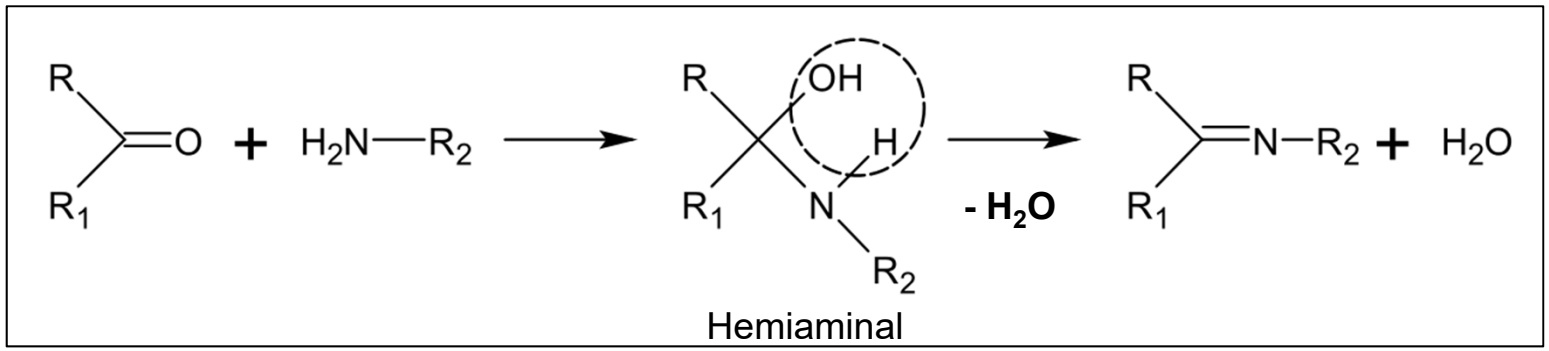
$C_{13}H_{23}Br$ Rubber Oligomer
ToxID 36/44/45
Alkyl halide
Alkylating agent

4. PQRI – THE RISK OF LEACHABLES - PROTEIN INTERACTIONS

Michael addition

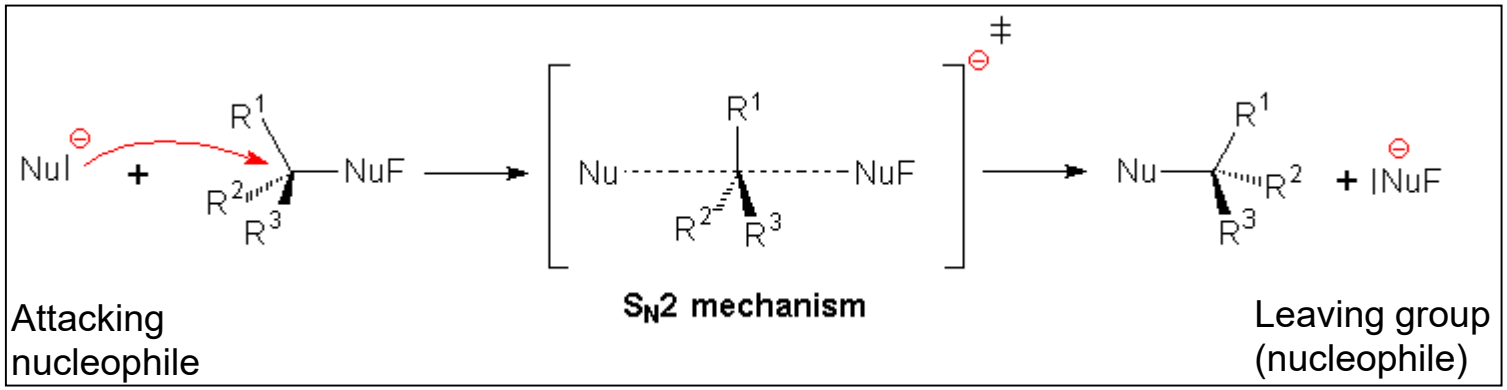


Schiff base (imine) formation



4. PQRI – THE RISK OF LEACHABLES - PROTEIN INTERACTIONS

Alkylation



5. CONCLUSION

5. CONCLUSION

Basis for the evaluation for E/L for a Therapeutic Protein: the same for a "Small Molecule" DP:

- ✓ However the **E/L- evaluation for a Biologic is augmented** with:
- ✓ Evaluation of **Tungsten (Oxide)**: could denaturate proteins
- ✓ Evaluation of **Silicone Oil**: could cause protein aggregation
- ✓ **Evaluation of the Extractables**: could they form *covalent bonds* with functional groups of the Protein?
  - Michael Acceptors
  - Schiff base formers
  - Acylating, Alkylating Agents
  - S_NAr, S_N2
  - ...
- ✓ **Evaluation Metals (elemental impurities)**: could they lead to interactions (eg oxidation) of the Protein? Literature?
- ✓ Other, **non-covalent** interactions are more **difficult** to assess

5. CONCLUSION

Basis for the evaluation for E/L for a Therapeutic Protein: the same for a "Small Molecule" DP:

- ✓ However the **E/L- evaluation for a Biologic is augmented** with:
- ✓ **Glass Delamination** *(although not strictly related to E/L)* can contribute to **Immunogenetic Responses**
- ✓ **Extraction Studies** should at least **bracket** both **hydrophilic ànd hydrophobic** properties of **Proteins**: multiple extraction solvents
- ✓ Evaluate the **impact of potential leachables on the Product Quality**:
  - Evaluate the Activity, Efficacy of the Therapeutic Protein.
  - Future:

    Interactions of Leachables with Proteins

    Immunogenicity Testing

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