# Review Points of Biological Safety Evaluation for Market Approval of Medical Devices

Pharmaceuticals and Medical Devices Agency
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# Review Points of Biological Safety Evaluation for Market Approval of Medical Devices

Introduction

The review points related to biological safety evaluation among the evaluations required for medical device regulatory applications in Japan are organized and published together with the information of international standard ISO 10993-1 or JIS T 0993-1.

- These review points are to show necessary evaluation items, etc. for medical devices shown in the scope of application specified in order to contribute to the improvement of efficiency in preparation of materials and acceleration of reviews when making approval/certification applications.
- These review points show the concept of review based on the current scientific knowledge, and should be reviewed and revised as needed according to future advances in science and technology.

1. Scope of application

These review points cover biological safety evaluation in compliance with ISO 10993-1 or JIS T 0993-1 when making an application for marketing approval and certification of medical devices in Japan (including partial change approval applications and partial change certification applications; hereinafter referred to as "application for marketing approval, etc.").

2. Biological safety evaluation of medical devices

In Japan, biological safety evaluation of medical devices requires identification of potential risks to the final product from a biocompatibility perspective. Particularly, for medical devices for which an application for marketing approval, etc. is scheduled to be made, biological safety evaluation in accordance with international standard ISO 10993-1 or JIS T 0993-1 is required. In the biological safety evaluation, it is necessary to take into account the materials used in the medical device to be evaluated, processing aids or additives that may remain, and residues after sterilization after clarifying the information such as the manufacturing process of the medical device, intended clinical use, contact sites of human body, and contact duration in advance.

However, since medical devices distributed in Japan and overseas have various types of products with different contact risks, it is considered unrealistic to require uniform biological safety evaluation for all medical devices. Therefore, ISO 10993-1 or JIS T 0993-1 suggests the evaluation methods with the clinical contact risks (contact form, contact site, or contact duration) of the target medical devices classified into some categories, rather than uniform evaluation of all medical devices as the same risk.

Accordingly, this document is based on the evaluation in accordance with ISO 10993-1 or JIS T 0993-1, and the test standards in the ISO 10993 series and describes specific examples of biological safety evaluation required at the time of the application for marketing approval in Japan. Please note that this document reflects ISO 10993-1/FDIS as of May 2025.

3. Contact risk and biological effect evaluation of medical devices

3-1. Categorization by duration of contact of medical devices

**A. Limited exposure:**
A medical device that has a total exposure period of less than or equal to 24 h.

**B. Prolonged exposure:**
A medical device that has a total exposure period of more than 24 h but not more than 30 d.

**C. Long-term exposure:**
A medical device that has a total exposure period of more than 30 d.

Note 1: Among these categories, medical devices with brief contact duration (lancet, hypodermic needle, etc.), whose single or cumulative contact duration is less than about 1 minute, are handled as "medical devices with brief tissue contact." In principle, a biological safety evaluation is not required for medical devices with brief contact. However, it should be noted that if there is a possibility that coating agents, lubricants, etc. that may remain in body tissues even after the medical device has been removed from the body, it is necessary to consider a biological safety evaluation (including the conduct of studies).

Note 2: For medical devices composed of multiple components, the worst-case should be evaluated for the longest contact duration among components or components classified by contact duration.

Note 3: For a medical device with multiple contact with the body, the duration should be calculated as the total contact duration for a single patient.

Note 4: For a medical device to be used continuously for a long period of time, the duration of contact should be selected from the total number of days when the same product is repeatedly used from the first use of the medical device containing the component. For detailed information on the concept, etc., refer to the latest ISO 10993-1. However, if the contact risk is low, the duration of clinical use per time is short, and the product is made of a non-medicinal/non-absorbable and non-degradable material, it is possible to evaluate by the cumulative duration of contact, not on a daily basis.

For example, since single-use feeding tubes that are replaced throughout the treatment period are used for several hours per day for more than one day, they may be considered as devices with Limited- and prolonged exposure when the days of use are integrated. However, if it is obvious from literature, etc. that the product has been widely used in clinical practice and that no toxicological problem has occurred with the material at the level of generic name, it is acceptable to derive the contact duration by adding cumulative clinical use time.

On the other hand, in clinical use, for hollow fiber artificial dialyzers that come into contact with blood and are expected to be used for more than 10 years per patient, or products that come into contact with circulating blood when used in combination with drug coating or drugs, it is necessary to make cumulative addition of contact duration on a daily basis conservatively.

Note 5: For products that are polymerized in the body or whose final product is absorbed/degraded, it is necessary to consider the constituents that come into contact with the body, constituents that are being degraded, constituents that are completely polymerized, and the state of the product that leads to subsequent degradation/absorption.

3-2. Categorization by contact site of medical devices

**(1) Non-contacting medical devices:**

In principle, biological safety evaluation is not required because these devices do not come into direct or indirect contact with the body. The term "body" includes users operating the product. For example, when a user wears gloves, gown, mask, and clothes during clinical use of the product and the product is not in direct contact with skin or mucosal membranes, it can be interpreted as "non-contacting."
(Example) Diagnostic software, X-ray generators for medical imaging, *in vitro* diagnostic medical devices, etc.

**(2) Medical devices in contact with intact skin (Table 1):**

Devices which have only direct contact or indirect contact with intact, uncompromised skin. If it is known that there is no significant biological risk even if no test is conducted, the rationale, etc. should be described in the STED, etc.

Testing/evaluation is not necessary when the risk to intact skin is clearly low or there is obviously no serious biological risk although the device (keyboard, switch button, dial, touch screen, etc.) may come into direct contact with the skin.

● Medical devices in contact with skin
(Example) Electrodes, fixation tapes, compression bandages, blood pressure cuff, pulse oximeter (finger insertion part), massagers (if the part of the product comes in contact with intact skin), etc.

Table 1. Biological effects for medical devices in contact with intact skin

| Contact duration | Cytotoxicity | Sensitization | Irritation | Systemic Toxicity | Local effects after tissue contact | Genotoxicity | Carcinogenicity | Hemocompatibility |
|---|---|---|---|---|---|---|---|---|
| A Limited (≤ 24 hours) | ○ | ○ | ○ | | | | | |
| B Prolonged (> 24 hours to ≤ 30 days) | ○ | ○ | ○ | | | | | |
| C Long-term (> 30 days) | ○ | ○ | ○ | | | | | |

*In subsequent revisions after ISO 10993-1:2018 the term "implantation" will be changed to "local effects after tissue contact" but will continue to be referred to as "implantation" in this guidance

**(3) Medical devices in contact with intact mucosal membranes (Table 2):**

(Example) Contact lenses, urinary catheters, intravaginal and intra-intestinal devices, endotracheal tubes, some dental prostheses and orthodontic devices, etc.

Table 2. Biological effects for medical devices in contact with intact mucosal membranes

| Contact duration | Cytotoxicity | Sensitization | Irritation | Systemic Toxicity | Local effects after tissue contact | Genotoxicity | Carcinogenicity | Hemocompatibility |
|---|---|---|---|---|---|---|---|---|
| A Limited (≤ 24 hours) | ○ | ○ | ○ | | | | | |
| B Prolonged (> 24 hours to ≤ 30 days) | ○ | ○ | ○ | ○ | ○ | ○ | | |
| C Long-term (> 30 days) | ○ | ○ | ○ | ○ | ○ | ○ | ○ | |

*In subsequent revisions after ISO 10993-1:2018 the term "implantation" will be changed to "local effects after tissue contact" but will continue to be referred to as "implantation" in this guidance

**(4) Medical devices in contact with either breached or compromised surfaces (skin or mucosal membranes) or internal tissues other than circulating blood (Table 3):**

- Medical devices in contact with either breached or compromised surfaces (skin/mucosal membranes).

  (Example) Dressings, patches used for skin burns, etc.

- Medical devices in contact with bone, dentin, internal soft tissues or organs.

  (Example) Laparoscopes, arthroscopes, drainage systems (drain tubes), dental filling materials, skin staples, products in contact with bone or pulp, etc.

- Medical devices implanted in tissue or bone.

  (Example) Orthopedic pins and plates, artificial joints, breast implants, gastrointestinal stents, ligation clips, pacemakers, intrauterine devices, etc.

Table 3. Biological effects for medical devices in contact with either breached or compromised surfaces (skin or mucosal membranes) or internal tissues other than circulating blood

| Contact duration | Cytotoxicity | Sensitization | Irritation | Systemic Toxicity | Local effects after tissue contact | Genotoxicity | Carcinogenicity | Hemocompatibility |
|---|---|---|---|---|---|---|---|---|
| A Limited (≤ 24 hours) | ○ | ○ | ○ | ○ | | | | |
| B /Prolonged (> 24 hours to ≤ 30 days) | ○ | ○ | ○ | ○ | ○ | ○ | | |
| C Long-term (> 30 days) | ○ | ○ | ○ | ○ | ○ | ○ | ○ | |

*In subsequent revisions after ISO 10993-1:2018 the term "implantation" will be changed to "local effects after tissue contact" but will continue to be referred to as "implantation" in this guidance

**(5) Medical devices in contact with circulating blood (Table 4):**

● Medical devices in direct or indirect contact with the circulating blood.
(Example) Solution administration sets (extension tube, etc.), blood administration sets, temporary pacemaker electrodes, haemoadsorbents, etc.

● Externally communicating medical devices in contact with circulating blood.
(Example) Intravascular catheter, oxygenator, extracorporeal oxygenator tube and accessories, dialyzer, dialysis tube, blood adsorber, etc.

● Medical devices implanted in a blood vessel, heart, etc. and in contact with circulating blood.
(Example) Components of pacemaker electrodes and leads within the cardiovascular system, artificial blood vessels, heart valves, vascular stents, vascular grafts, internal drug-delivery catheters, etc.

Table 4. Biological effects for medical devices in contact with circulating blood

| Contact duration | Cytotoxicity | Sensitization | Irritation | Systemic Toxicity | Local effects after tissue contact | Genotoxicity | Carcinogenicity | Hemocompatibility |
|---|---|---|---|---|---|---|---|---|
| A Limited (≤ 24 hours) | ○ | ○ | ○ | ○ | | ○ | | ○ |
| B Prolonged (> 24 hours to ≤ 30 days) | ○ | ○ | ○ | ○ | ○ | ○ | | ○ |
| C Long-term (> 30 days) | ○ | ○ | ○ | ○ | ○ | ○ | ○ | ○ |

*In subsequent revisions after ISO 10993-1:2018 the term "implantation" will be changed to "local effects after tissue contact" but will continue to be referred to as "implantation" in this guidance

Refer to Attachment A for specific examples of medical devices by category.

4. Categories and examples of selection of biological safety tests

4-1. **Medical devices in contact with intact skin**

**[Example 1]**

- To be evaluated: "Surface electromyograph (EMG) electrode (Class I)"/New application
- Biological effects for medical device categorization:
  Because of skin contact/limited exposure (within 24 hours), the biological effects required for the category are "cytotoxicity," "sensitization," and "irritation."
- Decision to conduct the test:
  It is necessary to check the adhesive pad part of the application site in a body for any problem such as irritation. Since there are no in-house sales results (e.g.: approved, certified), it was decided to perform "cytotoxicity test," "sensitization test," and "irritation test" for the evaluation.

**[Example 2]**

- To be evaluated: "Touch panel" of components of ultrasonic device/New application
- Biological effects for medical device categorization:
  Because of skin contact/limited exposure (within 24 hours), the biological effects required for the category are "cytotoxicity," "sensitization," and "irritation."
- Decision to conduct the test:
  It can be judged to be a non-contacting part because it is touched while wearing gloves in the medical care setting when touching the touch panel part. In addition, when touched directly with fingers, etc., there are no adverse events of palms/fingers reported due to contact with the screen made of materials similar to those of generally available electric appliances. Based on the above, biological safety evaluation by conducting the test is considered unnecessary.

4-2. **Medical devices in contact with intact mucosal membranes**

**[Example 1]**

- To be evaluated: Application for partial changes to "Long-term Foley catheter (Class III)"/coating agent "X"
- Biological effects for medical device categorization:
  Because of mucosal membranes contact/long-term exposure, the biological effects required for the category are "cytotoxicity," "sensitization," "irritation," "systemic toxicity," "implantation," "genotoxicity" and "carcinogenicity."
- Decision to conduct the test:
  Because the coating agent used for a pre-approved product with the same generic name was changed to a new coating agent "X," a new biological safety evaluation was required. In addition, the catheter with the coating agent "X" (final product) was used as the sample to be evaluated.

Among the items that need to be evaluated, the tests of "cytotoxicity," "sensitization," and "irritation" will be performed, but the tests of "acute systemic toxicity," "sub-acute systemic toxicity," "implantation," "genotoxicity," and "carcinogenicity" will be omitted.

The reason for omission is that the catheter part that accounts for the majority of this product is the same as the company's pre-approved product, and the biological safety of this product including the pre-change coating agent has already been assured.

Next, the coating agent "X" is the same before and after change at the generic name level of the material, there is no difference in the main material, and the difference is that the ZZ compound to be used as a catalyst is changed to YY compound. However, even if the catalyst is changed to YY, there is no change in the base material of the coating agent, and it has been evaluated separately in performance tests A and B that the performance is not significantly different from that of the pre-approved product.

About XX products to be changed have already been used overseas annually. Among the reported adverse events, there have been no events suspected to be genotoxic or carcinogenic, systemic toxicity, or events suspected to affect the area around the implantation site in clinical use, which may raise concerns about biological safety.

Based on the above, in addition to the "cytotoxicity test," which is considered to be most sensitive in detecting hazards, the local toxicity of extractable materials including the coating agent "X" was determined in the "sensitization test" and "irritation test." Since these tests confirmed the safety of the final product, the "implantation test" was omitted.

**[Example 2]**

- To be evaluated: "Single-use prescription colored contact lens (Class III)"/New application
- Biological effects for medical device categorization:
  Because of mucosal membranes contact/long-term exposure (considered cumulative, repeated use for "more than 30 days"), the biological effects required for the category are "cytotoxicity," "sensitization," "irritation," "systemic toxicity," "implantation," "genotoxicity" and "carcinogenicity."
- Decision to conduct the test:
  Additional biological safety evaluation was required because the materials of this product are different from those of the pre-approved product. "Sub-acute systemic toxicity," "sub-chronic systemic toxicity," "chronic systemic toxicity," and "implantation" were evaluated by "ocular irritation tests in rabbits" under the assumption of actual use. In addition, for "irritation," ocular irritation was evaluated in the " ocular irritation tests in rabbits." "Acute systemic toxicity" can be replaced with the evaluation for ocular irritation, and was evaluated in the above ocular irritation tests in rabbits .

  Based on the above, "sensitization test," "genotoxicity test," and " ocular irritation tests in rabbits" were performed to evaluate the biological safety. Since this product is a material that contacts the ocular mucosal membranes, "colony formation

method by direct contact method" and "colony formation method by medium extraction method" were performed for "cytotoxicity test" (Notes 6 and 7). All of them were final products containing preservative solution, and the maximum amount of additives such as coloring agents included in the scope of application was added. For ring-shaped colored lenses, the test was performed using samples with the minimum distance from the lens edge to the outer diameter of the ring-shaped colored part.

Note 6: When a stricter hazard detection is required such as when a new material, etc. is used or a significant change is made in the manufacturing process, it is necessary to separately examine whether the required test type is appropriate.

Note 7: If a new application for reusable soft contact lenses is to be evaluated, the biological safety evaluation of the lenses treated repeatedly 30 times with chemical disinfectant is also required. However, they are not positioned as performance evaluation tests or use simulation tests, and please note that GLP (Ministerial Ordinance on Good Laboratory Practice for Nonclinical Safety Studies of Medical Devices) will be applied.

4-3. **Medical devices in contact with either breached or compromised surfaces (skin or mucosal membranes) or internal tissues other than circulating blood**

**[Example 1]**

- To be evaluated: Application for partial change of the material X of "Negative pressure wound therapy system (Class III)"/"Dressing" to the material Y due to the change of supplier.
- Biological effects for medical device categorization:
  Because of medical devices in contact with compromised skin/mucosal membranes or internal tissues other than blood/prolonged exposure, the biological effects required for the category are "cytotoxicity," "sensitization," "irritation," "systemic toxicity," "implantation," and "genotoxicity."
- Decision to conduct the test:
  Because the materials, manufacturing process, and impurities, etc. in the manufacturing process of this product are different from those of the pre-approved product, new tests were performed for "cytotoxicity," "sensitization," "irritation," "sub-acute toxicity," "implantation," and "genotoxicity."
  For "systemic toxicity," only the "sub-acute systemic toxicity test" was performed and no "acute systemic toxicity test" was performed, considering that the dressing to be evaluated was not expected to have a large amount of exposure to systemic blood flow within 24 hours during the contact duration in clinical uses and was a non-absorbable material, and that the material to be changed is widely used in medical devices, although it is at the level of generic name.

4-4. **Medical devices in contact with circulating blood**

**[Example 1]**

● To be evaluated: "Extracorporeal membrane oxygenation (Class III)"/New application
● Biological effects for medical device categorization:
Because of externally communicating medical devices in contact with circulating blood/limited exposure (within 24 hours), the biological effects required for the category are "cytotoxicity," "sensitization," "irritation," "systemic toxicity," "genotoxicity" and "haemocompatibility."
● Decision to conduct the test:
Among the components of this product, the blood tank is the same as that of the pre-approved product XX, and the biological safety is assured at the time of application for approval of XX. Therefore, the material (final product), excluding the blood tank, was evaluated in the biological safety study in this application.
Of the biological effects required for the category, the "cytotoxicity," "sensitization," "irritation," and "haemocompatibility" were tested using the products to be evaluated. For the test items to be performed in the "haemocompatibility test," "haemolysis," "coagulation" "platelet activation," "complement system," and "haematological evaluation" were selected. In these tests, the test methods were established in consideration of the duration of use expected in the actual use of this product. For "acute systemic toxicity" and "genotoxicity," the chemical analysis of the extractable of the product to be evaluated showed only the substances below the AET (Analytical Evaluation Threshold), and thus no test was performed.

Note 8: For test items to be performed for "haemocompatibility test," a thrombosis risk should be examined and selected based on the characteristics of the device with reference to ISO 10993-4. In addition, it should be noted that haemocompatibility assessment may be required based on the expected duration of use for devices, etc. expected to have a high risk of thrombus formation, such as cardiopulmonary bypass systems.
In the *in vitro* thrombogenicity test, it is necessary to complete the test within a few hours during which the property of blood does not change. If the actual clinical use time is obviously longer, methods other than *in vitro* studies should be considered.
Note 9: Refer to Attachment B for biological safety evaluation method using chemical analysis results.

**[Example 2]**
● To be evaluated: "Single-use blood collection needle (Class II)"/New application
● Biological effects for medical device categorization:
Because of externally communicating medical devices in contact with circulating blood/limited exposure (within 24 hours), the biological effects required for the category are "cytotoxicity," "sensitization," "irritation," "systemic toxicity," "genotoxicity" and "haemocompatibility."
● Decision to conduct the test:
This product is a single-use blood collection needle that comes in contact with circulating blood. However, the maximum duration of contact during clinical use is within several minutes, and it is a limited exposure. The material that comes into direct contact with blood is metal stainless steel (SUS304, etc.), and the adequate

safety is assured by the official standard: XXX. In addition, there is no effect of substances for manufacturing that may be attached to the surface (outer surface) of the blood collection needle at the time of puncture.

Also, for elution from constituent materials such as tube portion that comes in indirect contact, almost no chemical substance is likely to be eluted in the body during the expected clinical use period (including the worst-case). Since the intended use of this product is "blood collection (taking from blood vessel using a needle, not injection into blood vessel with a needle)," the chemical substance in this product is quite unlikely to be eluted in the body during the clinical use.

Based on the above reasons, it is judged that the biological safety has been evaluated, and thus no test will be performed.

Note 10: The overall biological safety risk assessment method should be conducted in accordance with ISO 10993-1.

**[Example 3]**

● To be evaluated: "Artificial kidney dialysis machine (Class III)"/New application (when the generic name of the material is new)

● Biological effects for medical device categorization:
Because of externally communicating medical devices in contact with circulating blood/long-term exposure (considered cumulative, repeated use for more than 30 days), the biological effects required for the category are "cytotoxicity," "sensitization," "irritation," "systemic toxicity," "implantation," "genotoxicity," "carcinogenicity," and "haemocompatibility."

● Decision to conduct the test:

In artificial kidney dialysis machine, although multiple components come into indirect contact with the patient, the effect of chemical substances that may come into contact with the patient's body through dialysate or dialysis membrane on the body is small, and it is considered that there is no direct blood contact, compared to other externally communicating medical device even through cumulative contact. Furthermore, for materials, etc. used as components of the device, toxicological information is collected from precedents of use of in-house pre-approved products, multiple chemical toxicity databases, SDS, etc. in accordance with ISO 10993-1.

Therefore, among systemic toxicity tests, tests after the sub-acute systemic toxicity test, genotoxicity test, and carcinogenicity test are omitted by evaluating the equivalence of biological safety based on the actual use status of the artificial kidney dialysis device and the equivalence of the extractable test results specified in the approval standards. As described above, the implantation test does not need to be evaluated because the device does not come into direct contact with blood. For the haemocompatibility test, only the material induced haemolytic toxicity will be tested because the device does not come into direct contact with blood.

Based on the above, "cytotoxicity test," "sensitization test," "irritation test," and "haemocompatibility test (haemolysis)" were performed to evaluate the biological safety.

Note 11: It should be noted that if any new toxicity is observed in materials, etc. used for an individual component, a separate evaluation needs to be performed in accordance with the risk assessment specified in ISO 10993-1.

4-5. **Medical device in contact with as a breathing gas pathway medical device**

**[Example 1]**

- To be evaluated: "Single-use ventilator breathing circuit (Class III)"/New application
- Biological effects for medical device categorization:
  Tubes, connectors, etc. that simply pass-through gases with only indirect contact (exhalation/inhalation) and do not directly contact the human body are not handled as devices with body contact.
- Decision to conduct the test:
  No test was performed because the materials of this product are identical to those of the pre-approved product (or certified product) XX at the generic name level and there is no novelty of the material.

Note 12: For materials that have been sufficiently used as gas indirect materials and have accumulated knowledge, it is acceptable to describe the generic name level in the application form.

Note 13: For materials, etc. used for individual components, if special events such as degradation, freeing, elution of substances, and generation of new substances are expected to occur by reaction with gas, drug, water, etc., please note that they need to be separately evaluated in accordance with the risk assessment in ISO 10993-1.

Note 14: ISO 18562 for particulate matter in breathing gas pathway medical devices, volatile organic compounds and extractables with condensation shall be treated as reference standards.

*References: Tripartite Consultation (Bulletin) No. 202301 (September 13, 2023) "Handling of Gas Indirect Materials as Revisions to JIS T 0993-1: 2020"

4-6. Concept for the biological safety evaluation of packaging materials

Various substances are used in packaging materials for medical devices. When a medical device is marketed as a final product, for instance, in which a part of packaging materials comes into contact with a medical device and may contact a body during clinical use (e.g., adhesive), a biological safety evaluation for the packaging materials is also required. In addition, when the product is supplied to the medical setting, etc. in the form that the product is stored in the preservation fluid, attention should be paid to the necessity of evaluation of the absence of toxic substances due to a combination of the product with the preservation fluid and the storage container and the absence of physical degeneration of the product during clinical use.

When evaluating the biological safety of a preservation fluid, test samples should be selected considering the characteristics of the product and the actual status of clinical use, such as whether the final product containing the preservation fluid or the preservation fluid alone should be used and how the storage period should be set. Furthermore, it is desirable to evaluate substances that may pose a risk of contact with

the body in packaging materials with reference to ISO 10993-18 or ISO 10993-17 where necessary.

5. Individual biological safety tests

The information presented below provides general knowledge about individual tests that are required to perform a biological safety evaluation, as well as precautions to be taken when filing an application for marketing approval. When actually filing an application for marketing approval, etc., refer to the latest version of ISO 10993 series, which is a test specific standard.

In addition, if you want to confirm individual tests or individual applications in advance, it is desirable to proactively utilize the PMDA consultation. (For application for certification, consult with the certification authority.)

5-1. Cytotoxicity test　Reference standard: ISO 10993-5

The cytotoxicity test is an *in vitro* test using mammalian cultured cells and one of the tests required for evaluation of the biological safety of all medical devices. The cytotoxicity test is a test to detect cytotoxicity of a chemical substance extracted from a test sample with high sensitivity, and an extraction solvent is used as a medium. It is performed to capture the potential toxicity screening of test samples. It is therefore difficult to perform the test in combination with other tests (e.g., irritation and sensitization tests).

While colony formation has been considered a typical test method, several test methods (e.g. XTT method, MTT method, etc.) have been developed in conjunction with standard updates in ISO 10993-5. For some products, it may be necessary to select a direct contact test to better reflect clinical use. For example, for a substance essentially cytotoxic such as cyanoacrylate, it is necessary to conduct a preliminary test by confirming the dilution range of the test solution in consideration of the contact risk during clinical use.

When a test is conducted by a new test method in an application for marketing approval, etc., it is desirable to explain in an application form, etc. whether the sensitivity, reproducibility, dose dependence, etc. of the test are appropriate and whether the test method is appropriate in real-world clinical use to be evaluated.

5-2. Sensitization test　Reference standard: ISO 10993-10

The sensitization test specified in ISO 10993-10 is intended to determine whether chemical substances released from a medical device may cause a risk of delayed allergy (= sensitization risk) in humans. It is performed regardless of the duration of contact of the medical device. The test design specified in ISO 10993-10 does not detect immediate allergic reactions caused by oral or inhalation exposure.

For extraction solvents, since there are various options such as organic solvents and physiological saline solution, it is necessary to make a comprehensive judgment based on the novelty of materials in the product to be evaluated and the risk and duration of clinical contact. It should be noted that the scientific validity of the selected solvent needs to be added to the materials for the application for marketing approval, etc.

Positive control tests in the same species should be conducted as GLP studies, either on a case-by-case basis or periodically (at least approximately once every 6 months) to

ensure reproducibility and sensitivity of the test procedure. Please note that if a periodic positive control test fails, the sensitization test will be considered invalid after the test where the most recent valid positive response was observed.

5-3. Irritation test　Reference standard: ISO 10993-23

The irritation test evaluates inflammatory reactions caused by inflammatory mediators released from living cells damaged by irritative causative substances present in or on the surface of a medical device. It is performed regardless of the contact duration of medical devices that directly or indirectly contact with the body. ISO 10993-23 lists *in vitro* tests using reconstructed human skin models, *in vivo* tests using rabbits and tests specific to eye irritation.

For the substance with abundant background data on the irritation test, it is possible to apply the tolerable contact level (TCL), which is the toxicological information specific to irritants (Reference: ISO 10993-17).

Ocular study with rabbit eyes is required for contact lenses. Ocular study with rabbit eyes is specified in ISO 9394, and the effects on ocular tissues are evaluated based on the results of macroscopic and histopathological observations. Therefore, if the target test sample is a contact lens and an ocular study has been performed, there is no need to conduct an eye irritation test with an extract.

5-4. Systemic toxicity test　Reference standard: ISO 10993-11

The systemic toxicity test is conducted in mammals to check whether substances extracted from a medical device cause systemic adverse biological responses in organs and tissues through the systemic circulation, lymphatic system, or cerebrospinal fluid, or show systemic problematic reactions. The route of administration is intravenous or intraperitoneal. The test design will be selected from the 4 types including acute, sub-acute, sub-chronic, and chronic, depending on the duration of body contact and exposure in view of the clinical use. The assessment of systemic toxicity requires a stepwise approach. The test may not be necessary for devices in contact with intact skin or in limited duration contact with mucosal membranes.

Although it is indicated as "systemic toxicity" in the category table, it is also possible to select what test design should be selected for which contact risk by referring to the subtable in Table 5 in consideration of the contact duration in clinical use. When conducting the test, the test plan should be established in accordance with ISO 10993-11. For the medical device intended for implantation, it is possible to conduct a test combining sub-acute to chronic systemic toxicity test with implantation test design.

Table 5. Types of systemic toxicity tests required for each category assessment

| | Test type / Duration of contact | Acute | Sub-acute | Sub-chronic | Chronic |
|---|---|---|---|---|---|
| Intact mucosal membranes | Temporary | | | | |
| | Short- and medium-term | ○ | ○ | | |
| | Long-term | ○ | ○ | ○ | ○ |
| Compromised skin/mucosal membranes | Temporary | ○ | | | |
| | Short- and medium-term | ○ | ○ | | |
| | Long-term | ○ | ○ | ○ | ○ |
| Blood | Temporary | ○ | | | |
| | Short- and medium-term | ○ | ○ | | |
| | Long-term | ○ | ○ | ○ | ○ |

There is a major difference between "acute systemic toxicity test" and "single-dose toxicity test," which is a toxicity test of a drug. While a single-dose toxicity test is intended to confirm the change of toxicity from exposure to the test substance at a high dose, an acute systemic toxicity test is intended to confirm that there are no substances with acute systemic toxicity in solutions extracted from the final product, etc. in terms of detection of hazards.

However, for the products in the categories other than blood contact, if (1) a large amount of chemical substances that are in contact with the body or that may be eluted from the product are not supposed to be exposed to the systemic blood flow within 24 hours from the time of clinical use of the product to be evaluated, (2) the product is made of a non-absorbable and non-degradable material, and (3) the materials, etc. are substances that are widely used in medical devices or acute toxicity evaluation is publicly known from literature, etc., the evaluation can be performed by conducting a sub-acute systemic toxicity test without conducting an acute systemic toxicity test with the reasons described in (1) to (3) above.

For the repeated administration period in sub-acute and other tests, it is necessary to select an appropriate period along with the life of animals to be used, assuming the contact duration in the clinical use to be evaluated.

However, even if there is a long-term exposure risk and sub-chronic systemic toxicity and chronic systemic toxicity tests may be required for the product, the evaluation can be performed based on the results of sub-chronic systemic toxicity without performing chronic systemic toxicity, if, for example, the gross findings, haematological findings

(including urinalysis findings) and histological observations of the animals at the end of the sub-chronic systemic toxicity test period clearly show they are stable based on the results observed during the middle of the test or if it can be judged that the degradation and absorption reaction is clearly stable and no more changes are expected histologically.

In addition, in systemic toxicity risk assessment, toxicological screening limit (TSL), tolerable intake (TI), and threshold of toxicological concern (TTC) can be applied (Reference: ISO 10993-17).

5-5. Implantation test (local effects after tissue contact) Reference standard: ISO 10993-6

The implantation test to determine local effects after tissue contact (Note 15) is specific to medical devices. It is intended to histopathologically confirm the effects of local tissues due to the physical presence of implanted medical devices or substances leached into tissues. For the implantation sites to be evaluated, it is necessary to consider organs and tissues in which the product is implanted and may have an impact under actual clinical use. (Example: If a stent to be implanted in a blood vessel is evaluated, the test sample is implanted in a blood vessel for evaluation.)

As with the systemic toxicity test, the test may not be necessary for devices in contact with intact skin or in limited duration contact with mucosal membrane tissue.

When performing the test in combination with the systemic toxicity test, observation items of both tests should be included and attention should be paid to the necessity of handling matters such as the setting of the control group and the appropriateness of the amount of implantation.

If an absorbable or degradable material is to be evaluated, tissue observation is required, including at least three timepoints: An early phase of degradation immediately after implantation; the middle phase of degradation, in which the disintegration or fragmentation of the product is most severe due to degradation; and the terminal phase of degradation, in which the sample is completely absorbed or the degradation and absorption reaction has reached a plateau (the reaction curve seems to be halted).

Note 15: In subsequent revisions after ISO 10993-1:2018 the term "implantation" will be changed to "local effects after tissue contact" but will continue to be referred to as "implantation" in this guidance

5-6. Genotoxicity toxicity test Reference standard: ISO 10993-3

Genotoxicity is a test to determine whether a substance extracted from a medical device has genotoxicity (mutagenicity) that acts on the genetic material in the body, causing mutations. If a test shows genotoxicity, it is highly likely that this may have an effect on carcinogenicity when there is an effect on somatic cells, and on reproductive development when there is an effect on germ cells. Therefore, if the genotoxicity test determines that there is an obvious problem, the presence of a genotoxic type of carcinogenic substance in the medical device should be suspected.

Since genotoxicity appears after chemical substances are released into the systemic circulation, it may be possible to omit the evaluation of genotoxicity for medical devices

in contact with the circulating blood within a limited period of time until chemical substances are released or medical devices that have long-term exposure with tissues other than the circulating blood.

Although there are several tests to confirm genotoxicity, in general genotoxicity evaluation of materials of medical devices require at least two tests that detect gene mutations and chromosomal aberrations (*See ISO 10993-3 for details and concepts of the test).

As with sensitization tests, it should be noted that the selection of an extraction solvent needs to be judged comprehensively based on the novelty of the material in the product to be evaluated, the risk of clinical contact and contact duration, etc., and that an organic solvent alone is not necessarily essential. It should be noted that the scientific validity of the selected solvent needs to be added to the materials for the application for marketing approval, etc.

In addition, in genotoxicity risk assessment, toxicological screening limit (TSL), tolerable intake (TI), and threshold of toxicological concern (TTC) can be applied (Reference: ISO 10993-17).

5-7. Carcinogenicity test　Reference standard: ISO 10993-3

This test predicts whether or not substances extracted from the target of evaluation exhibit carcinogenicity. Carcinogens searched for are largely classified into mutagenic and non-mutagenic carcinogens. However, unlike drugs intended to be repeatedly administered at a fixed dose for a long period, carcinogenicity tests are often not required for permanently implanted medical devices for which biocompatible materials are used. In addition, except for sensitive populations such as pregnant women and neonates, it is not necessary to evaluate carcinogenicity for products in contact with intact skin.

However, if the medical device contains a substance with mutagenicity or reproductive and developmental toxicity, the evaluation for carcinogenicity is considered to be necessary regardless of the contact duration. In this case, it is desirable to perform comprehensive carcinogenicity evaluation by exhaustively collecting toxicity information of chemical substances that may be eluted using literature and public toxicity data collection, together with information of genotoxicity tests, etc.

Since non-genetic carcinogenic substances usually require a long-term exposure time before development of cancer, it may be possible to omit evaluation for products with short-term to medium-term contact.

In addition, in carcinogenicity risk assessment, toxicological screening limit (TSL), tolerable intake (TI), and threshold of toxicological concern (TTC) can be applied (Reference: ISO 10993-17).

5-8. Haemocompatibility test　Reference standard: ISO 10993-4

The test is intended to confirm that a medical device in contact with blood does not cause haemolysis, thrombogenicity, etc. Irrespective of the duration of contact, medical devices with direct or indirect contact with circulating blood should be verified for haemocompatibility. For the apheresis device, artificial heart valve, etc., it is necessary to confirm the haemolytic property due to mechanical causes.

Since there are several test items from the viewpoints of haemolysis and thrombus formation, it is desirable to select tests to be evaluated according to the degree of risk of blood contact in the clinical use to be evaluated.

Generally, for medical devices in direct contact with circulating blood, it is necessary to determine the necessity of evaluation not only by a haemolysis test but also by complement activation and thrombus formation test based on the characteristics of the product. For devices with indirect contact with circulating blood, complement activation or thrombus formation testing is not required and haemolysis testing alone may be sufficient.

As for the sufficiency of test items, PMDA consultation, etc. should be proactively utilized because it may be changed in the method of clinical use of individual medical devices (For application for certification, consult with the certification authority.)

Note 16: Complement activity is a biological defense mechanism that complements the ability of antibodies to eliminate pathogens in the body.

5-9. Other tests

**[Material-mediated pyrogenicity] Reference standard: ISO 10993-11**

Pyrogenicity may be induced by bacterial endotoxins, such as Gram-negative bacteria, or other non-endotoxin product-related substances (such as synthetic polymers or natural biomaterials), Gram-positive bacteria, or other microorganisms. Among them, the pyrogenicity test of medical devices required for biological safety evaluation targets material-mediated pyrogens. This test is not intended to detect endotoxins.

In addition, the pyrogenicity test in rabbits is a test to confirm whether there is any substance in the medical device that causes heat generation including bacterial endotoxin. Therefore, it should be noted that endotoxin should be confirmed during the manufacturing process of medical devices by the "endotoxin test," which is separately required as a risk management, instead of by a pyrogenicity test using rabbits in a biological safety test.

Implantable devices, sterile devices in direct/indirect contact with the cardiovascular system, lymph, cerebrospinal fluid, etc., and products claimed to be nonpyrogenic need to be evaluated in a pyrogenicity test.

Note 17: For bacterial endotoxins, refer to ISO 11737-3, etc.

**[Reproductive and developmental toxicity test]**

For the medical device used for reproductive function, embryonic development, treatment before or immediately after delivery, it is necessary to evaluate the potential effects of reproductive and developmental toxicity.

5-10. Selection method of test samples

If one final product consists of different contact categories or durations, care should be taken in selecting test samples. In particular, if the final product contains non-contact components, the contact (intended) site should be the test article. In addition, even when the implanted and non-implanted components are contained in one product, the implanted and non-implanted components should not be mixed with the same test article because extraction including the non-implanted part may cause dilution of the specimen.

6. Concept of extraction of products Reference standard ISO 10993-12

ISO 10993-12 describes extraction procedures and conditions for medical devices for biological safety evaluation. Test-specific extraction methods are specified for sensitization tests and genotoxicity tests.

In general, it is recommended that polar and non-polar solvents be used as extraction solvents for medical devices, except for new materials that have not been in contact with the body. For example, in situations with indirect contact via a polar solvent without direct body contact, it is not necessary to select a non-polar solvent. If the test design is direct intravascular injection of the extract, polar extracts are sufficient to confirm.

As for the extraction temperature, it is desirable to select the temperature assuming the clinical worst-case as much as possible. However, it should be noted that chemical substances not expected during clinical use may be leached due to deformation of the product if the temperature is too high.

7. Use of chemical property data in biological safety evaluation

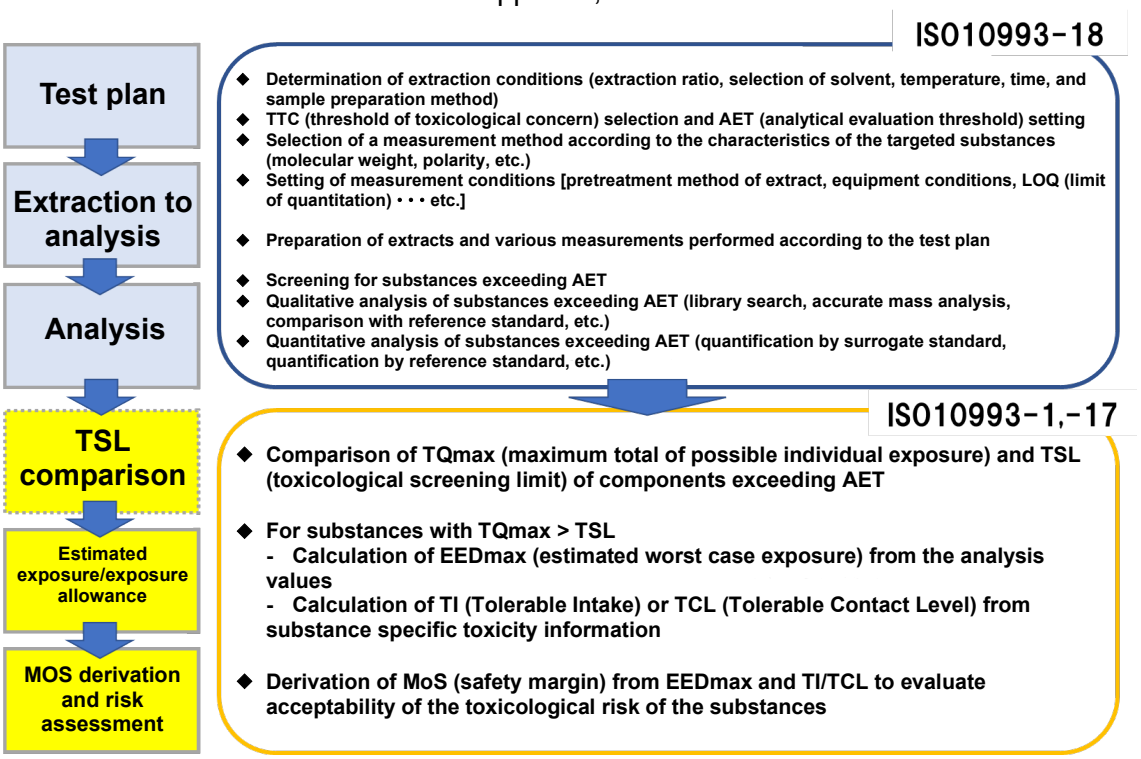
When conducting the "Risk estimation" of toxicological risk assessment according to ISO 10993-1 from the viewpoint of chemical toxicity evaluation, it is useful to understand chemicals that may be dissolved during the manufacturing process to the final product or at the time of clinical use. If there is sufficient information on chemical characterization to estimate the risk of the product, no new chemical analysis is required.

Further, when identifying substances that may leach from the final product, chemical analysis techniques should be used, focusing on ISO 10993-18. In addition, it is necessary to evaluate and discuss whether chemical substances identified as having toxicological concerns may become a problem when exposed to living organisms in clinical use, using ISO 10993-17.

In the chemical equivalence evaluation of pre-approved products, etc. and the evaluation of whether the products to be evaluated have biocompatibility, it is acceptable to identify the chemical substances that may elute from the products to be evaluated using ISO 10993-18 and ISO 10993-17 to evaluate the toxicological effects in the clinical use. These results can be used for evaluation of tests without performing a part of the biological safety tests required for each category of ISO 10993-1.

7-1. How to proceed with toxicological risk assessment using chemical property data

Figure 1. Main flow of chemical characterization in the application for marketing approval, etc.




7-1-1. Requirements necessary for toxicological risk assessment using chemical property data, which are required for the application for marketing approval, etc.

1) The following information shall be made clear in accordance with ISO 10993-1 and ISO 10993-18:
   ・Description of the overview of the medical device (appearance, dimensions, material information, etc.)
   ・Intended use of the medical device (assuming the worst-case for clinical use)
   ・Specific information (e.g. molecular structure and CASRN) and quantity of chemical substances analyzed according to ISO 10993-18

2) The validity of chemical analysis methods shall be explained in accordance with ISO 10993-18.
   The contents to be explained include, for example, the following points.
   ・Validity of analytical methods and analytical devices selected based on the purpose of analysis
   ・Validity of selected extraction solvents and extraction conditions

・Rationale for calculation of AET (Analytical Evaluation Threshold)
・Qualitative method of each substance, details of the qualitative rationale
・Justification of quantification method for each substance

3) The toxicity of each substance shall be characterized based on a systematic review
・Nature of health hazards
・Relationship between exposure and hazards (dose response, route, duration, etc.)

4) Toxicological risks should be assessed for each substance by one of the following:
・It can be judged that the substance do not induce health hazards under the intended use/method
・It can be judged that the total amount (TQmax) of the substance is small, and therefore no obvious health hazard will be induced
・It can be determined that the worst-case exposure levels of substances present in or released from the medical device are below the tolerable contact level (TCL) or tolerable intake (TI)
・It can be determined that the amount released from the medical device is less than the relevant threshold of toxicological concern (TTC)

7-2. Acquisition of chemical property data

In order to clarify the chemical characteristics of substances that can be eluted from the medical device to the body, chemical analysis shall be performed in accordance with ISO 10993-18 and be documented. Extractables expected from the composition of materials, manufacturing process, etc. shall be categorized into volatile organic substances, semi-volatile organic substances, non-volatile organic substances, inorganic elements, and ionic species, and necessary evaluation methods for the extract shall be examined.

NVR and FT-IR are also useful for structural analysis of chemical substances in terms of the physical detachment and elution amount of coatings on the surface of medical devices.

A medical device is a product consisting of various materials and additives before its final product is manufactured. Therefore, when identifying chemical substances that may be eluted during clinical use of the final product, it is not possible to identify and quantify all chemical substances by simply running extracted samples on the analyzer. On the other hand, in many cases, the current analytical chemistry technology cannot identify or quantify all trace substances that may leach from medical devices.

Thus, the analytical evaluation threshold (AET) is available as a threshold for rationally targeting the detection substances to be identified and quantified from the group of substances detected large and small.

The AET is a threshold at which a new extractable substance would not need to be identified or quantified for assessment of the potential toxicity of the medical device under investigation if the amount of chemical substance in the analysis falls at or below that value.

In other words, if it is possible to calculate AET by using TTC in accordance with ISO 10993-18, then no further biological safety evaluation is required for those substances that are below the AET.

For substances that exceed the AET, a toxicological risk assessment of extractables of the medical device can be performed by using ISO 10993-17 as described below. However, it should be noted that it is required to identify and quantify the substances to be evaluated by the chemical analysis methods as described in ISO 10993-18.

Therefore, please note that when planning the chemical characterization, the AET setting should be considered first in addition to the extraction conditions and analytical techniques.

7-2-1. Test plan for obtaining chemical property data

1) Examination of extraction conditions

The extraction conditions required to obtain chemical characterization data that will allow toxicological risk assessment of the device being tested should be examined. Specifically, in conducting toxicological risk assessment, it is necessary to examine referring to ISO 10993-17 and -18 while taking the following into consideration.

- ✓ Are the conditions suitable for the purpose of evaluation such as characterization of extractables under conservative stress conditions or leachables under conditions simulating actual clinical use?
- ✓ Are the extraction conditions that can estimate the amount of exposure and release kinetics according to the category of contact of the specimen with the body and its characteristics?

The basic idea for selection of extraction solvents is to perform extraction using solvents with different polarity (polar, semi-polar, and non-polar) under severer conditions than the clinical use to search for hazards that may be eluted from the device to the body.

In the biological safety test, purified vegetable oil is used as a nonpolar solvent. However, it is not appropriate as an extraction solvent in chemical analysis because a constituent thereof may interfere with detection of constituents. For this reason, it is desirable to use high-purity organic solvents.

In addition, it should be noted that excessive extraction conditions that could alter the sample significantly more than expected in clinical use would interfere with conduct of a reasonable toxicological risk assessment and therefore need to be considered in setting the conditions.

In addition, the rationale for selection of the extraction conditions shall be documented in the chemical analysis report, etc.

2) Setting of AET

AET should be set based on the following calculation formula.

[Calculation formula]

## AET (μg/mL) = DBT (μg/day) × (A / BC) / UF

DBT (Dose Based Threshold) =TTC, etc.
A = Number of medical devices that were extracted to generate the extract (pieces or $cm^2$)
B = Volume of the extract (mL)
C = Clinical exposure to the medical device (number of devices a user would be exposed to in a day under normal clinical practice) (pieces or $cm^2$)
UF = Uncertainty factor

Note 18: Refer to ISO/TS21726 and ISO 10993-17 to select an appropriate TTC as DBT for unknown organic substances. The use of TTC assumes that product materials do not contain CoC (Cohort of Concern). It is desirable to investigate in advance that CoC is not included in the list of materials of the target device. It is said that TTC cannot be applied to inorganic components and AET cannot be set. Therefore, instead, it should be considered that PDE (Permitted Daily Exposure) of ICH-Q3D, etc. serves as DBT and replaces AET setting.

Note 19: A (number or area of devices to be extracted) and B (extract volume) are set based on the set extraction conditions.

Note 20: From a conservative viewpoint, it is desirable to set C (number of devices in clinical use or exposed area) assuming the worst-case.

Note 21: It is necessary to confirm analytical conditions to see if the lower limit of quantitation (LOQ) or lower limit of detection (LOD) that is equivalent to or lower than the established AET can be set.

Note 22: For UF, data should be obtained and set for each testing facility according to the test method to be selected. It is desirable to refer to ISO 10993-18, etc. for the concept of UF calculation. In addition, attention should be paid to excessively large UF settings, which may decrease AET and interfere with targeting of components of reasonable concern.

3) Examination of analytical methods

For analytical conditions, see the following example (Table 6).

Table 6. Methods of chemical analysis for implementation of toxicological risk assessment (example)

| Evaluation target | Test method (example) |
|---|---|
| Volatile, semi-volatile organic substances | HS-GC with FID and/or MS<br>GC with FID and/or MS<br>HPLC with UV, CAD, ELSD and/or MS |
| Non-volatile organic substances | HPLC with UV, CAD, ELSD and/or MS |
| Inorganic elements | ICP-MS, ICP-AES |
| Ion species | IC |
| [Remarks]<br>・For qualitative analysis of a wide range of organic to inorganic extractables, it is desirable to combine GC, HPLC, and ICP-MS (or AES), and to combine the detector of GC and HPLC with MS for a qualitative analysis.<br>・In order to perform qualitative analysis, it is desirable that LC/MS is combined with MS that can obtain accurate mass. | |

7-2-2. Implementation of analysis

1) It is desirable to perform analysis promptly after extraction in order to prevent transformation and adsorption of extracted substances. If long-term storage occurs between extraction and measurement, the storage should be justified.

2) When pretreatment such as dilution, concentration, filtration, and solvent replacement is added to the extract before measurement, the details should be documented. If necessary, the validity of the operation is verified by a spike and recovery test, etc.

2) In the analysis, qualitative and quantitative analyses will be performed for detected components that have exceeded the AET and require toxicological risk assessment. The information obtained by the analysis (CAS No., substance name, composition formula, structural formula, qualitative accuracy, extraction volume, etc.) should be summarized.

7-2-3. Documentation of chemical property data

After evaluation, chemical analysis results shall be summarized and documented with reference to ISO 10993-18 and its attachments, etc.

## 7-3. Implementation of toxicological risk assessment

### 7-3-1 ISO 10993-17: Concept of toxicological risk assessment of 2023 version

The medical device is composed of several materials, and each product has a different level of contact risk to the body, requiring a toxicological risk assessment appropriate for each risk. For this reason, in the 2023 version, instead of uniformly calculating the safe contact level, the concepts and calculation methods to estimate the worst-case exposure level according to the body contact level of each product are presented.

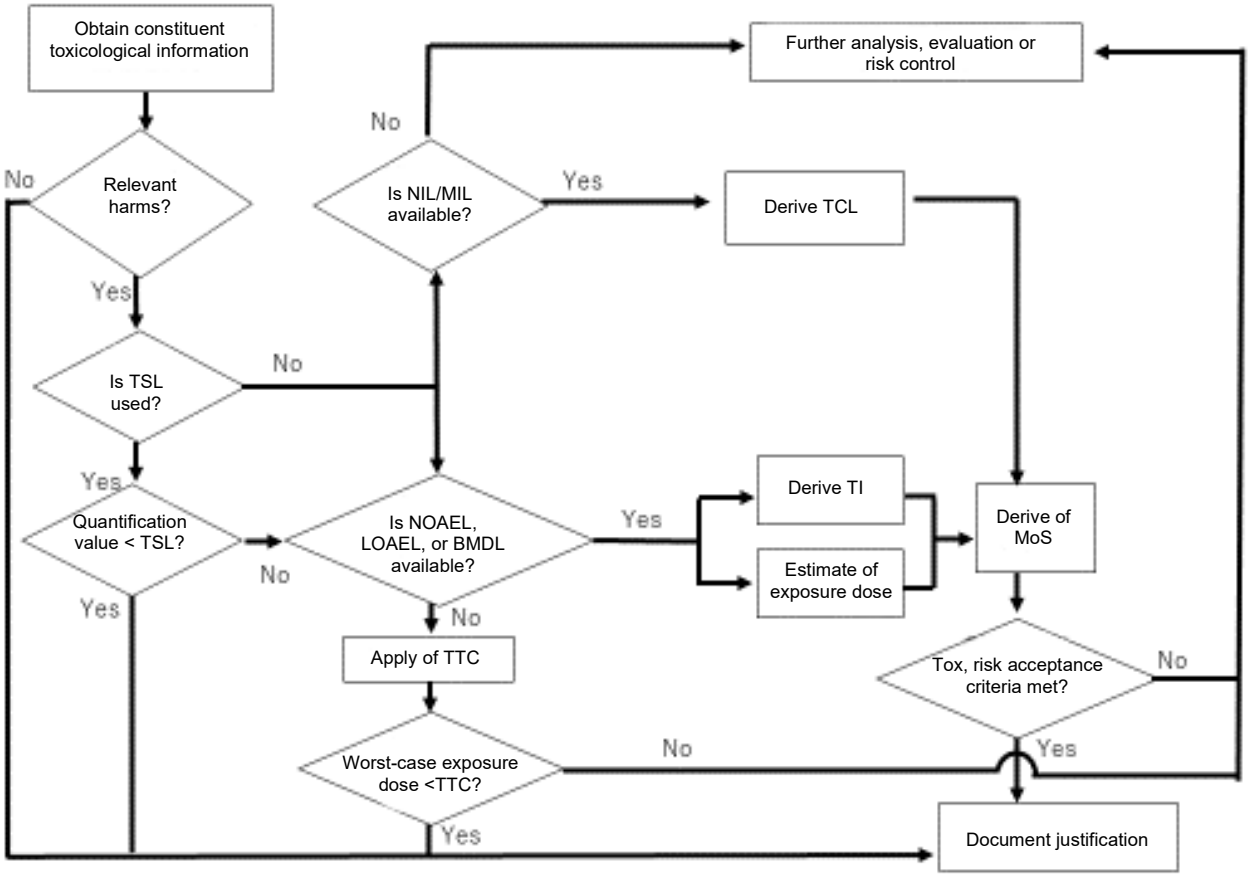
Major changes from the 2002 version to the 2023 version are shown below.

1) Addition of the concept of Toxicological Screening Limits (TSL)
2) Deletion of terms of tolerable exposure (TE) and concomitant exposure factor (CEF)
3) Addition of medical device contact category and estimated duration of contact, and method of estimation based on available data in terms of exposure level as a normative document
4) Clarification of evaluation method for the safety margin MoS (Margin of safety)

7-3-2　Implementation process of toxicological risk assessment ISO 10993-17

The process for conducting toxicological risk assessment is shown in Figure 2.

Figure 2. Toxicological risk assessment process (from ISO 10993-17: 2023)



7-4. Biological safety test that can be evaluated using toxicological information (TCL, TI, TSL and TTC)

Toxicity information necessary for identification of hazards should be systematically searched and collected from multiple sources and databases in accordance with ISO 10993-1. Regarding the following biological safety evaluation, it is possible to perform biological safety evaluation without performing tests by calculating the exposure level and safety margin using toxicological information (TCL, TI, TSL, and TTC). When planning chemical characterization, consideration should be given on the assumption that such toxicological information is used.

- Irritation (TCL)
- Systemic toxicity (acute, sub-acute, sub-chronic, chronic) (TSL, TI, TTC)
- Genotoxicity (TSL, TI, TTC)
- Carcinogenicity (TSL, TI, TTC)
- Reproductive and developmental toxicity (TSL, TI, TTC)

Note 23: If any available toxicological data other than TSL, TI and TTC are appropriate, they might be acceptable, but individual judgment is required

Note 24: TTC concept can be used if TI derivation is inadequate

7-5. Use of TSL

TSL is the cumulative exposure level for a given period of time to an identified substance that does not cause significant health hazard. In accordance with ISO 10993-18, TSL can be used to identify substances that exceed the AET and quantitate TQ (total quantity), then to check whether their dissolved concentrations are sufficient to induce genotoxicity, carcinogenicity, systemic toxicity, and reproductive and developmental toxicity risks. TSL cannot be applied to substances that are irritating, substances of concern of toxicity, or excluded substances, or those for which no identification has been made.

If the TSL is larger than the total amount dissolved or cumulative amount (TQmax) of substances that may be exposed for a specific period of time, the toxicological risk can be judged to be negligible and no further toxicological risk assessment is necessary.

If the total amount of substances extracted exceeds the TSL value, further evaluation by estimating the exposure level during clinical use will be required by reference to ISO 10993-17.

[Calculation of TSL]
TSL is expressed in terms of cumulative exposure level (μg) to a particular substance during a defined period of time that does not cause significant health hazard.

[Calculation formula]

# TSL = TTC × D

Note 25: "D" is expressed per unit day in accordance with ISO 10993-1:2018,5.3.

Table 7. Default toxicological screening limit TTC and D for TSL calculations

| Estimated exposure period | TTC (μg/d) | D (days) | TSL (μg) |
|---|---|---|---|
| Exposure within 30 days | 120 | 1 | = 120 (i.e. 120 × 1) |
| Exposure for more than 30 days | 20 | 30 | = 600 (i.e. 20 × 30) |

**<Example 1>**
Limited or long-term exposure: 100 μg specified is extracted from a single medical device, comes in contact with a single-use medical device within 30 days

TSL = 120 × 1 = 120 μg > TQmax = 100 × 1=100 μg

* No further toxicity assessment is required as TQmax is below the TSL
* For TQmax = TQ × SF, SF was set at "1" based on 1 device that comes in contact with the body/1 piece extracted = 1

**<Example 2>**
Limited or long-term exposure: 100 μg specified is extracted from a single medical device, comes in contact with 2 single-use medical devices within 30 days

TSL = 120 ×1 = 120 μg < TQmax = 100 × 2 =200 μg

* MoS calculation is required because TQmax exceeds the TSL
* For TQmax = TQ × SF, SF was set at "2" based on 2 devices that come in contact with the body/1 piece extracted = 2

7-6. TI (Tolerable Intake)

The TI (tolerable intake) is calculated from NOAEL, LOAEL, $BMD_L$, etc. for exposure level at which it is judged that toxicologically relevant health hazards do not appear when taken by humans for a specific period (e.g., acute, sub-acute, sub-chronic, or chronic). For medical devices, a specific period of time (e.g., estimated daily exposure to a particular substance in systemic toxicity) is used, based on body weight, during which no obvious harm to health is expected, and units for the TI are given in μg/kg/day.

<How to detect TI based on serious adverse event effects>

- Evaluation of toxicity data and identification of serious adverse events
- If the substance induces a serious adverse event, the lowest PoD (Point of Departure) value is specified.
- The appropriate toxicity threshold (TCL, TI) that can be applied to serious adverse events is derived.

[Calculation formula]

## TI = PoD (NOAEL, LOAEL, etc.)/MF

Unit: μg/kg/day

Note 26: MF = UF1 × UF2 × UFn
* Refer to ISO 10993-17: 2023 Annex C for specific UF values

Note 27: PoD (Point of Departure): NOAEL, $BMD_L$, etc. which refers to values as the starting point for toxicity criteria when extrapolating the results of dose-response evaluation obtained from an animal study to humans with low intake and estimating health effects at low dose.

Note 28: $BMD_L$: benchmark dose confidence lower limit: dose-response curve, upper and lower limit curves of 95% confidence limits are depicted from several animal studies. The confidence limit on the safe side (95% lower confidence limit) of a dose (benchmark dose "BMD") where the occurrence of toxicity (e.g., frequency of carcinogenesis) is increased by a certain percentage relative to the control group in a dose-response curve is $BMD_L$ (value that allows for greater safety).

Note 29: UF (Uncertainty factor): When setting the acceptable level of chemical substances for humans from data such as animal tests, UF (Uncertainty factor) is set so that the risk is not underestimated

<Example of UF calculation>

**Uncertainty factor product (MF) = Individual difference (UF1) × species difference (UF2) × use of LOAEL (UFn) × test period (UFn) × factor for type and quality (UFn) of test, etc.**

* UF1: The default is difference between species of the same kind such as adults and children x 10 (may also be ×3)
* UF2: The default is extrapolation of species difference x 10
* UFn: Additional UF

Figure 3. Relationship between TI and NOAEL/LOAEL

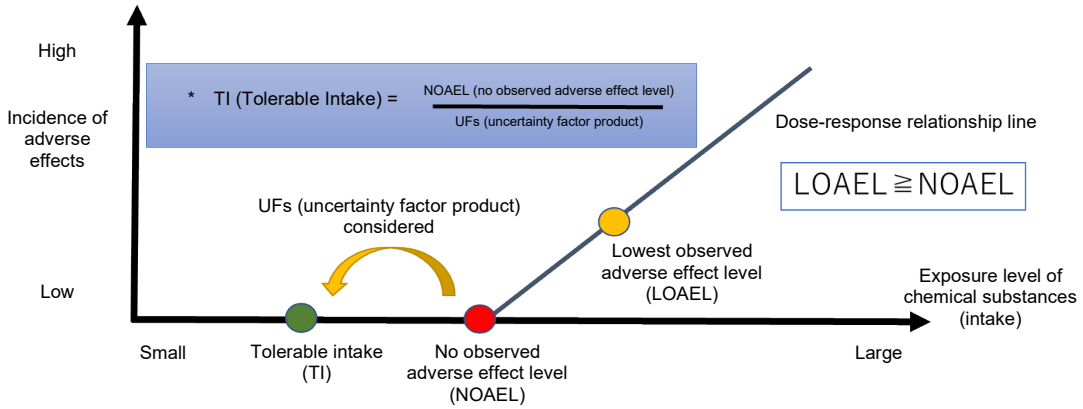
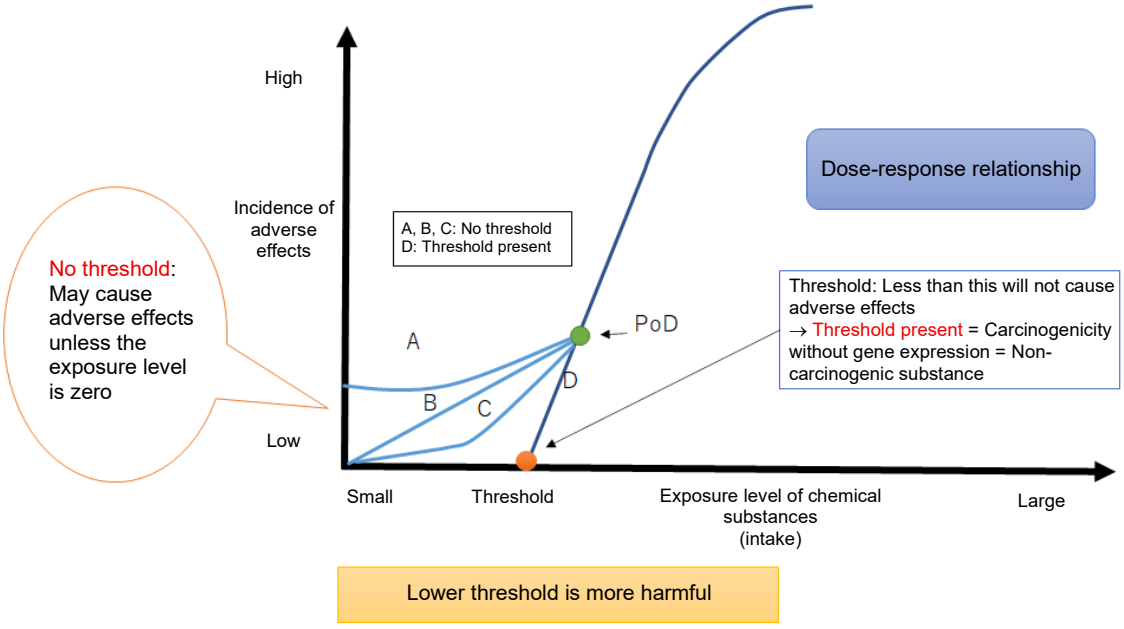



Figure 4. Relationship between threshold and dose response

A-D indicates extrapolation options below the PoD value



7-7. TTC

TTC is a threshold of substance exposure below which there would be no apparent risk to human health. However, it should be noted that there are groups of substances for which TTC cannot be applied, such as CoC and excluded substances.

[Application of TTC]

- TTC is used if TI is not available for specific toxicity information.
- The TTC approach can be used for the evaluation of genotoxicity, carcinogenicity, systemic toxicity (acute, sub-acute, sub-chronic, chronic), and reproductive and developmental toxicity, but cannot be used for other tests.

For example, if a conservative estimation of exposure level shows the value below the TTC, it is considered a toxicologically negligible dose.

[Selection of TTC]

- The lowest TTC value in both the genotoxicity TTC/carcinogenic TTC and non-carcinogenic TTC will be used in the toxicological risk assessment when selecting TTC.
- Hereditary/carcinogenic TTC can be deduced from genotoxic and carcinogenic responses.
- Non-carcinogenic TTC refers to toxicological information in the absence of genotoxicity.
- See ISO/TS 21726 for the TTC value (TTC of medical devices is based on an adult body weight of 60 kg).
- TTC values are expressed in μg/day.

7-8. Estimation of exposure level

For the chemical substances extracted from the product to be evaluated (AET or higher and TSL or higher), it is necessary to estimate the exposure level in assuming the clinical use. In this case, the exposure level (EEDmax) of the substance that contacts (invades) the body in a day is estimated. The exposure level per day should be estimated from one day or multiple days accumulated based on time. The worst-case exposure level should be set assuming the following worst-case conditions for clinical use of the medical device.

(Example)

- ✓ Type of information on exposure to chemical substances related to intended use
- ✓ Maximum quantity or area of exposure of an individual, unless there is a special reason
- ✓ Minimum body weight assumed to be exposed

Refer to ISO 10993-17 for the method of estimation of exposure level, etc.

7-9. MoS

The MoS uses the TI (TCL) and estimated worst-case exposure (EEDmax). It is the ratio between the TI (TCL) and estimated exposure level EEDmax.

[Calculation formula]

## MoS = TI (or TCL) / EEDmax

[Evaluation of MoS]

Conditions under which exposure levels of substances are considered to be clearly no risk/possibly no risk are as follows (the opposite is that there is a risk):

- MoS > 1
- The value for calculating the MoS is selected to conservative lower the MoS

Even if the TI (TCL) and EEDmax are conservative, further risk assessment would be required if any of the following are met. In such a case, the toxicological risk should be addressed again in accordance with ISO 10993-1 and ISO 14971.

- ✓ a MoS is below 1 based on release kinetics and TI or TCL are used,
- ✓ cancer risk of a human carcinogen, or suspected human carcinogen, exceeds 1 in 100 000,
- ✓ the MoS is judged to represent possible toxicological risk (i.e. MoS < 1).

Additional risk analysis, risk evaluation or risk control may consider information that addresses the following matters.

- ✓ Confirmation of dosage of the substance that causes hazards (LOAEL)
- ✓ Confirmation of adequacy of exposure level for clinical use of medical device
- ✓ Confirmation that the risk control is not practical and that it can be demonstrated that the expected benefits outweigh the toxicological risks

# Attachment A　Examples of medical devices by category

In order to make it easy to imagine medical devices that fall into each category, representative medical devices marketed in Japan are listed here.

However, as this list does not cover all products, it is important for applicants to ultimately classify the target product into the appropriate category based on the information in actual clinical practice.

[Medical devices in contact with intact skin]

| | |
|---|---|
| A. Devices with limited exposure | Skin electrode, ultrasound probe, touch panel, sphygmomanometer, pulse oximeter, and massager |
| B. Prolonged exposure | Compression bandage |
| C. Long-term exposure | Orthopedic devices for fixation (splint, corset) |

[Medical devices in contact with intact mucosal membranes]

| | |
|---|---|
| A. Devices with limited exposure | Irrigation tube/catheter, examination tube, ureteral catheter for contrast, flexible gastroscope, intrauterine catheter, condom, flexible laryngoscope, periodontal probe |
| B. Prolonged exposure | Gastrointestinal tube inserted from natural orifice, urinary catheter including urethral catheter, intratracheal tube for short-term use, catheter for oxygen administration, tampon |
| C. Long-term exposure | Contact lenses, long-term indwelling ureteral catheters, some dental prostheses and orthodontic appliances, pessary for pelvic organ prolapse |

[Medical devices in contact with damaged sites other than blood and tissues other than the above such as skin and mucosal membranes]

- Medical devices in contact with compromised skin surfaces
- Medical devices in contact with bones, dentin, body tissues or organs
- Medical devices implanted in tissue or bone

| | |
|---|---|
| A. Devices with limited exposure | Surgical drapes, burn dressings (if the duration of use is 24 hours or less), surgical staple equipment, arthroscopes, single-use intraocular lens inserters, and rigid laparoscopes |
| B. Prolonged exposure | Absorbable suture, antiadhesive agent, endoscopic clip (including those for marking), adhesive bandage, and absorbent pad for wound (duration of single use or cumulative use exceeding 24 hours and within 30 days) |
| C. Long-term exposure | Dressings (if used for more than 30 days), dental root canal filling material, permanently implanted staple/vascular ligation clips, permanently implanted endoscopic clip, non-absorbable ligament prosthesis, cardiac pacemaker, intraocular lens, implantable fixation pin/plate, dental implant, artificial bone matrix implant |

[Medical devices in contact with blood]

- Medical devices in direct or indirect contact with the blood pathway
- Externally communicating medical devices in contact with circulating blood
- Medical devices implanted in a blood vessel, heart, etc. and in contact with circulating blood.

| | |
|---|---|
| A. Devices with limited exposure | Catheter for cardiac surgery, catheter for intravascular examination/diagnosis/surgery/indwelling catheter/sizer/guidewire, etc., membrane plasma separator, membrane oxygenator, drug administration device for intravascular use (injection needle), blood collection device, infusion set, blood bag, general-purpose syringe, winged needle |
| B. Prolonged exposure | Continuous haemofilters, intravascular catheters, infusion filters |
| C. Long-term exposure | Cardiovascular stents, central circulatory cardiovascular patch, implantable haemodialysis catheters, dialysis tubes, hollow fiber dialyzers, artificial blood vessels, pacemaker electrodes, cardiac valves |

# Attachment B　Examples of biological safety evaluation including chemical analysis

## [Example] (Tentative name) "Vascular ligation clip Q"

1. Biological safety evaluation of (Tentative name) "Vascular ligation clip Q"

1-1. Biological risk analysis

(1)　Target of biological safety evaluation

This time, in order to apply for approval (no approval criteria, no clinical studies) of (tentative name) "Vascular ligation clip Q" (hereinafter referred to as "this product"), the final product (EO sterilized) is subject to evaluation.

(2)　Clinical use information and physical/chemical information related to the target of evaluation

This product is used to ligate a blood vessel during surgery, etc. The main material XXXX is a non-absorbable polymer material that remains in the patient body semi permanently after use of this product.

As physical information related to the biological safety of this product, poor ligation and physical inflammatory effects on normal tissues around ligation may have effects on safety. However, the evaluation of these physical effects has no particular concerns based on the results of the performance evaluation test (Attachment XX to the application form).

As for chemical information, since the material of the pre-approved older generation product "vascular ligation clip P" is changed from "OOOO" to "XXXX," there may be some effects when the "XXXX" comes into contact with the ligated site or the surrounding tissues during the period when this product is implanted in the body.

(3)　Information on bioequivalence with pre-approved products

The manufacturing process, sterilization method, shape, and physical characteristics, contact with the body, and clinical use are equivalent to or the same as those of the pre-approved older generation product, except that the main material "OOOO" of the pre-approved product "vascular ligation clip P" will be replaced with "XXXX." However, impurities that may occur in the manufacturing process using "XXXX" cannot necessarily be said to be the same as "OOOO."

(4)　Test and evaluation target

As a result of the risk analysis of the above (1) to (3), it was decided to use the final product sterilized with EO as the evaluation sample, instead of toxicity evaluation of the single material "XXXX," to perform the test or evaluation specified in ISO 10993.

1-2. Category of medical device

・Contact sites: Damaged sites other than blood and tissues other than the above such as skin and mucosal membranes

・Duration of contact: Long-term exposure

<Biological effects for consideration>

Cytotoxicity, sensitization, irritation, acute/sub-acute/sub-chronic/chronic systemic toxicity, implantation, genotoxicity, carcinogenicity

Since this product is a vascular ligation clip, it is unlikely to come into contact with blood during usual clinical use. However, an evaluation of haemocompatibility will be added just in case.

1-3. Implementation of biological safety tests

For the material "XXXX" used for this product, no toxicity information on cytotoxicity, sensitization, and irritation when simulating clinical use environment, including toxicity information at generic name level, were available from the literature information. In addition, since impurities in the manufacturing process could not be identified, cytotoxicity, sensitization, and irritation were evaluated using the sterilized final product as the test sample in accordance with the latest ISO 10993.

The local effects after implantation are evaluated using the results of the vascular ligation study in dogs conducted as a performance study of this product and a simulated use study. The haemocompatibility will be also comprehensively evaluated on the basis of the results of the study, general toxicity information on "XXXX," and clinical use of this product.

For the tests of acute/sub-acute/sub-chronic/chronic systemic toxicity, genotoxicity, and carcinogenicity, the final product of this product will be subject to chemical analysis in accordance with ISO 10993-18 XXXX, and the results obtained will be used to perform toxicological evaluation in accordance with ISO 10993-17: XXXX, thereby omitting the tests.

2. Test and chemical analysis results

2-1. Results of biological safety tests performed

The results of each test performed are shown in a table.

(Results are omitted here.)

In conclusion, the results of the tests of cytotoxicity, sensitization, and irritation conducted were all negative, and no clinically significant risks were found.

The results of vascular ligation study in dogs showed no local effect related to implantation of this product (vascular ligation site and surrounding tissues) and no problem in haemocompatibility related to ligation procedure with this product.

2-2. Chemical analysis results

<Extraction conditions>

E & L extraction: To obtain the extractables profiles, the extractions were performed under stress conditions not affecting the device, in which X g of the final product was exhaustively extracted in water (polar solvent), ethanol (semi-polar solvent) and hexane (non-polar solvent) for 1 cycle at 50°C for 24 hours.

<Type of analysis>

Based on the materials and manufacturing process, elution of organic compounds (volatile, semi-volatile, non-volatile) and metals from the final product cannot be ruled out. Therefore, GC-MS, LC-MS and ICP-MS were performed.

<Analytical results>

(1) Results of metal analysis
Ni X μg/mL and Fe X μg/mL were detected in the water extract measured by ICP-MS.

(2) Results of organic compound analysis
As a result of GC-MS and LC-MS, 2 peaks exceeding the AET were observed in the hexane extract. These peaks were tentatively identified by MS library search, and Compound B and Compound C were identified by confirming matching of the retention time and MS spectrum with the reference standard. The assay results were y μg/mL and z μg/mL, respectively.

<Setting of AET>

(1) Since Ni and Fe are metals and TTC cannot be applied, AET was calculated conservatively with the minimum PDE value of ICH-Q3D as DBT. Since both of the obtained analytical amounts exceeded AET, toxicological safety evaluation was performed in accordance with ISO 10993-17.

(2) For Compounds B and C which are organic compounds, TTC was set as DBT, and UF was determined in accordance with ISO 10993-18 and AET was set. As a result, since all the amounts exceeded the AET, the toxicological safety evaluation was performed in accordance with ISO 10993-17.

<Toxicological evaluation>

For Ni and Fe assayed, the amounts with toxic effects on the body were confirmed based on multiple literature information. The total amount extracted from the final product was not more than one thousandth which is considered to cause health hazards in humans. Therefore, it can be judged that they do not cause obvious health hazards.

The TQmax of Compound B assayed was determined in accordance with ISO 10993-17 and compared with TSL. As a result, for Compound B, TQmax was less than TSL, and therefore it was considered that there was no further toxicological concern.

Since TQmax was more than TSL for Compound C assayed, the MoS was determined. However, since the MoS of Compound C was 150 and was more than 1, it was considered that there was no toxicological concern.

2-3. Appropriateness of omitting the biological safety test based on the chemical analysis results

Presence or absence of health hazards related to this product was confirmed based on the information used for toxicity evaluation of each substance, and a systematic review of the exposure and hazards was performed using toxicity data calculated according to ISO 10993-17. Substances that cause toxicological concerns among those quantified in accordance with ISO 10993-18 were evaluated in accordance with ISO 10993-17. As a result, it was judged that there was no risk such as systemic toxicity or carcinogenicity due to long-term implantation in the body.

Based on the above, the results of this chemical analysis can be used to adequately evaluate the acute, sub-acute, sub-chronic, chronic systemic toxicity, genotoxicity, and carcinogenicity, and thus the implementation of these tests is considered unnecessary.

3. Overall biological safety evaluation

Biological effects for the medical device category of this product are cytotoxicity, sensitization, irritation, acute/sub-acute/sub-chronic/chronic systemic toxicity, implantation, genotoxicity, and carcinogenicity. Also, haemocompatibility was added. The results of the tests required for evaluation (cytotoxicity, sensitization, and irritation) met the acceptance criteria in all tests. The local effects after implantation and haemocompatibility were evaluated based on the results of performance tests, etc., and no problematic results were observed. For acute, sub-acute, sub-chronic, chronic systemic toxicity, genotoxicity, and carcinogenicity tests, toxicological evaluation based on chemical analysis results revealed no risks such as systemic toxicity or carcinogenicity.

Based on the above results, it was considered that the materials of this product and the chemical substances that may be eluted during the clinical use had only a small impact on the body for the required biological effects.

Therefore, no additional risk control is required, and it is considered that biological safety of this product is sufficiently assured.

[Reference]

Here, examples are presented in order to make it easy to imagine. For the appropriateness of the contents of analysis, etc. to be attached to each application, face-to-face consultation with PMDA, etc. should be used where necessary.

# Attachment C　Glossary

● Exhaustive Extraction
- For long-term exposure devices, extraction operation is performed in multiple steps until the extraction volume becomes 10% or less of the initial extraction volume

● Exaggerated Extraction
- It is intended that larger amount of substance than that in actual clinical use is released within the range not causing chemical changes

● AET (Analytical Evaluation Threshold)
- Level below AET that no additional analysis is required
- The analytical evaluation threshold calculated based on the safety margin of toxicological concern such as TTC, that is "the threshold below which a chemical's toxicity evaluation is considered unnecessary." Further biological safety evaluation is not required for the substances below the AET and toxicological risk assessment according to ISO 10993-17 is also not required.

● $BMD_L$ (benchmark dose lower confidence limit)
- The confidence limit on the safe side (95% lower confidence limit) of a dose (benchmark dose "BMD") where the tumor development (carcinogenesis) is increased by a certain percentage relative to the control group in a dose-response curve (value that allows for greater safety).

● DBT (Dose Based Threshold)
- The threshold used to calculate AET, e.g., μg/day as DBT, such as TTC or SCT

● EEDmax (estimated worst-case exposure)
- Estimate of exposure level (EEDmax) representing exposure assuming the worst-case of an extract

● LOAEL (Lowest Observed Adverse Effect Level)
- The lowest concentration or amount of an identified substance that has been shown to cause health hazards to the target organism under the specified exposure conditions
- Minimum dose "without harmful effects such as disease" determined in animal studies, etc.
- Determined from systemic toxicity (long-term toxicity), reproductive and developmental toxicity, etc. in multiple dose groups

● MFs(Modifying factors)
- A factor, determined by the expert judgment of a toxicologist, to allow extrapolation of experimental data to human safety MF = UF1× UF2 × UF3

● MoS (Margin of Safety)
- Set the allowable threshold (TE: Tolerable exposure) based on toxicity-related information from the literature, and judge based on MoS whether the extractable exceeds the threshold
- MoS=TI (tolerable intake)/EEDmax (maximum exposure level*)
  *It may be referred to as Patient Exposure.

● NOAEL (No Observed Adverse Effect Level)
- Maximum dose "below which there are no harmful effects such as disease" determined in animal studies, etc.
- Amount of chemical substances per 1 kg of body weight per day (e.g., mg/kg/day)
- Can be determined based on systemic toxicity (long-term toxicity), reproductive and developmental toxicity, carcinogenicity, sensitization, etc.

● NOEL (No Observed Effect Level)
- Highest dose "at which all biological effects did not show statistically significant changes compared to the control group" determined in animal studies, etc.
- NOAEL is the toxicological value, while NOEL is the "amount of drug"
- Determined from systemic toxicity (long-term toxicity), reproductive and developmental toxicity, etc. in multiple dose groups

● PDE (Permitted Daily Exposure: Acceptable intake of residual solvent derived from drugs)
- Developed to avoid confusion with International Programme on Chemical Safety (IPCS) TDI and WHO ADI
  No-observed-effect level (NOEL) or lowest-observed effect level (LOEL) in the most appropriate animal experiment
- PDE = (NOEL (*) × human body weight)/(F1 × F2 × F3 × F4 × F5)
  *It is desirable to derive from NOEL while LOEL is also acceptable.

● PoD (Point of Departure)
- "NOAEL" and "$BMD_L$" are often used, which refer to values as the starting point for toxicity criteria when extrapolating the results of dose-response evaluation obtained from an animal study to humans with low intake and estimating health effects at low dose.
- Utilize toxicologically important points, substances suspected of being carcinogenic, quantitative data ($TD_{50}$) available for estimating cancer risk
- If no valid data from adverse event assessment are available, a toxicity risk assessment using a structurally similar TTC should be performed.
- If the PoD is a component of the toxicological concept and one or more adverse events are available under the same route of administration, the lowest PoD value can be used to derive the TI value as an effect of serious adverse events.

● SCT (Safety concern threshold)
- A level at which risks of carcinogenicity and non-carcinogenicity can be ignored if the concentration is lower than the SCT

● SF(Scaling Factor)
- Ratio of the amount of medical device in contact with the body (e.g. $cm^2$, g or ml) divided by the amount of medical device used in the extraction test, SF = MDb.c./MDa.r.s.
  MDb.c. is the maximum amount of a medical device that comes in contact with the body at the same time (e.g. $cm^2$, g or ml);
  MDa.r.s. is the amount of medical device used in the extraction test.

● TCL (Tolerable Contact Level)
- Estimated surface contact exposure
- An indication of quantitative and safe amounts of a substance per medical device which simulates the actual clinical use and "the amount of chemical substance per medical device below which it is considered to have no toxic effect on surface contact"

● TDI (Tolerable Daily Intake)
- In humans, "the dose below which there is no adverse effects such as diseases even when taken (exposed) daily by humans for a lifetime"
- Amount of chemical substances per 1 kg of body weight per day (e.g., mg/kg/day)

(Reference) The ADI (Acceptable Daily Intake) is an index used for acceptable daily intake, food additives, etc.

● TI (Tolerable Intake, synonymous with TDI)
- Exposure considered not to cause adverse health effects if taken by human for a lifetime
- Estimates of daily exposure to a specific constituent, based on body weight, for a specific period of time (e.g., acute, sub-acute, sub-chronic, chronic) considered not to cause apparent health hazard
  *Exposure/TI >1 indicates concern about occurrence of adverse events
- TI = PoD (NOAEL, LOAEL, etc.)/(MFs) Unit is μg/kg/day
- Indicators used in ISO 10993-7 and -17

● TQ (Total Quantity)
- Components that are present in or can be extracted from the medical device

● TQmax (Total Quantity (maximum))
- Maximum total of possible individual exposure (cumulative exposure)
- TQ multiplied by a coefficient (maximum amount of device in contact with the body/amount of device used for extraction test), and can be calculated by TQmax = TQ × SF

● TTC (Threshold of Toxicological Concern)
- It is set as the threshold of human exposure to any chemical substance below which no obvious adverse effects appear
- Determined by statistical analysis of historical toxicity data based on structural analysis and chemical similarity to develop a comprehensive method for evaluating the safety of many chemical groups or chemicals with unknown toxicological information

● TSL(Toxicological Screening Limits)
- An indication of quantitative and safe amounts of a substance per medical device which simulates the actual clinical use and the amount of chemical substance per medical device "below which it is considered to have no toxic effect"
- Substances with TQmax below the TSL do not require further toxicological risk assessment; substances above the TQmax need to be evaluated with respect to their Margin of Safety (MoS), etc. according to the flow in ISO 10993-17.

● UFs (Uncertainty factors)
- A factor that is set so that the risk is not underestimated when setting the acceptable amount of a chemical substance in humans based on data such as animal studies, because uncertainty arises in estimating the carcinogenicity of the chemical substance.
  (*Refer to the reference standard: ISO 10993-17: 2023 Annex C for specific UF values)
- Used to calculate TI
- Uncertainty factors (UFs) = Individual difference (UF1) × species difference (UF2) × use of LOAEL (UFn) × test period (UFn) × factor for type and quality (UFn) of test, etc.

* More UFs are associated with less reliability and increased risk.
* UF3: Uncertainty factor (×1 to 100) from acute toxicity data and database