Encapsome®

DESCRIPTION

Clodrosome® is a multilamellar liposome suspension in which clodronate is encapsulated in the aqueous compartments of the liposomes. Encapsome® is formulated and prepared identically to Clodrosome® except that clodronate is not added to the liposomes. The liposomes are filtered through 2 μm polycarbonate membranes to ensure that the larger particles, which may be toxic to animals, are removed from the suspension. Both are prepared and packaged under sterile conditions. When animals or cells are treated with Clodrosome®, phagocytic cells recognize the liposomes as invading foreign particles and proceed to remove the liposomes from the local tissue or serum via phagocytosis. The liposomes then release clodronate into the cytosol, resulting in cell death. Non-encapsulated clodronate cannot cross the cell membrane to initiate cell death.

Control liposomes (Encapsome®) are recognized and phagocytosed by the same mechanism as Clodrosome®. Since the control liposomes do not contain clodronate, the phagocytic cells are not killed. However, phagocytes do respond to the ingestion of control liposomes by cytokine secretion, temporary suspension of phagocytic activity and other responses described in the literature.

FORMULATION INFORMATION

Encapsome® Control Liposome Suspension

Lipid Composition	Concentration (mg/ml)	Concentration (mM)	Molar Ratio Percentage
L-α-Phosphatidylcholine	18.8	24.3	70
Cholesterol	4.2	10.9	30
Total	**23 mg/ml**	**35.1 mM**	**100**

Buffer and Liposome Size	Specification
Buffer	Phosphate Buffered Saline
pH	7.4
Liposome Size	1.5-2 µm

TECHNICAL NOTES

- When animals or cells are treated with Clodrosome®, phagocytic cells recognize the liposomes as invading foreign particles and proceed to remove the liposomes from the local tissue or serum via phagocytosis. The liposomes then release clodronate into the cytosol resulting in cell death. Unencapsulated clodronate cannot cross the cell membrane to initiate cell death.

- Encapsome® control liposomes are recognized and phagocytosed by the same mechanism as Clodrosome®. Since the control liposomes do not contain clodronate, the phagocytic cells are not killed. However, phagocytes do respond to the ingestion of the control liposomes by cytokine secretion, temporary suspension of phagocytic activity and other responses described in the literature.
- The product must be removed from the vial using sterile technique. Do not use if sterility is compromised. This is particularly important if a single vial is accessed multiple times over several weeks. The product should not be used more than 60 days after receipt, even if unopened.
- Liposomes may settle when left undisturbed for more than a few hours. Immediately prior to use, in order to ensure a homogeneous liposome suspension, slowly invert the vial several times until the suspension appears homogeneous by visual inspection. Vigorous or erratic shaking will not damage the liposomes; however, may induce foaming and bubble formation making it more difficult to accurately measure the desired dosage.
- If the personnel performing intravenous injections are not experienced in or familiar with, precautions for injecting larger volumes (~10% animal weight in ml), viscous liquids or particulate suspensions, consider having extra animals available in case serious injection-related adverse events occur. Dose control animals first to become familiar with large volume injections.
- Within hours after systemic administration of Clodrosome®, animals begin to lose important components of their immune system. Standard animal handling and housing protocols are not suitable for immunocompromised animals. Even when such precautions are taken, monitor the general health of each animal for opportunistic infections unrelated to the experimental protocol. There is no inherent toxicity to the product at the recommended dose levels.
- When dosing intravenously, use standard precautions for dosing larger volumes to animals including the following: a) warm product to room temperature prior to dosing; b) ensure that all air bubbles are removed from the syringe prior to dosing. Intravenous injection of air bubbles may result in air emboli which can kill or seriously injure

animals; c) inject product at a slow, steady rate of no more than 1 ml/min; d) decrease infusion rate if animals display any atypical reactions such as unusual agitation.

- Infusion-related adverse reactions usually involve the animal gasping for air or other seizure-like movements. Animals often recover with no apparent permanent injury, but any potential effects on experimental results must be assessed by the researcher.
- Liposomes should be kept at 4 °C and **NEVER** be frozen.

APPEARANCE

Clodrosome® and Encapsome® are both white milky suspensions made of large micro size multilamellar liposomes. Due to their large size, some liposomes might settle to the bottom of the vial. If left sitting idle in the refrigerator, Encapsome® will phase separate and form pellets in the bottom of the vial leaving a clear solution on top. Clodrosome® might do the same only not as severely. Therefore, both should be shaken to form a homogeneous solution prior to use.

STORAGE AND SHELF LIFE

Storage

Clodrosome® and Encapsome® should always be stored at in the dark at 4 °C, except when brought to room temperature for brief periods prior to animal dosing. DO NOT FREEZE. If the suspension is frozen, clodronate can be released from the liposomes thus limiting its effectiveness in depleting macrophages. ENS is not responsible for results generated by frozen product.

Shelf Life

Clodrosome® and Encapsome® are made on daily basis. The batch that is shipped is manufactured on the same day. It is advised to use the products within 60 days of the manufacturing date.

REFERENCES AND BACKGROUND READING

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