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The difference in endolymphatic hydrostatic pressure elevation induced by isoproterenol between the ampulla and the cochlea

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ABSTRACT

Objective: The purpose of the study was to investigate the difference in the responses of endolymphatic hydrostatic pressure to isoproterenol, β-adrenergic receptor agonist, between pars superior and pars inferior.
Methods: The hydrostatic pressure of endolymph and perilymph and endolymphatic potential in the ampulla and the cochlea during the intravenous administration of isoproterenol were recorded using a servo-null system in guinea pigs.
Results: The hydrostatic pressure of endolymph and perilymph in the ampulla and cochlea was similar in magnitude. Isoproterenol significantly increased hydrostatic pressure of ampullar and cochlear endolymph and perilymph with no change in the ampullar endolymphatic potential and endocochlear potential, respectively. The isoproterenol-induced maximum change of endolymphatic hydrostatic pressure in ampulla was significantly ($p < 0.01$) smaller than that in the cochlea. In ears with an obstructed endolymphatic sac, the action of isoproterenol on endolymphatic hydrostatic pressure in the ampulla disappeared like that in the cochlea.
Conclusion: Isoproterenol elevates endolymphatic hydrostatic pressure in different manner between the vestibule and the cochlea.

1. Introduction

Endolymph is believed to be produced mainly by the stria vascularis in the cochlea and the dark cells in the utricle and ampullae and to be finally absorbed by the endolymphatic sac (ES). Obliteration of the ES and the endolymphatic duct causes accumulation of endolymph in the cochlea, the vestibule and the semicircular canals [1]. This condition referred to as endolymphatic hydrops is a characteristic pathological finding in Meniere's disease [2,3]. Endolymphatic hydrops in the cochlea causes deafness [4], and endolymphatic hydrops in the vestibule may cause vertigo [5,6]. Therefore, endolymph regulation is essential for normal hearing and the sense of equilibrium.

The membranous labyrinth includes the cochlea, the vestibule (saccule and utricle), the semicircular canals, and the ES. The ES and endolymphatic duct are connected to the pars superior (utricle and semicircular canals) and the pars inferior (cochlea and saccule) by the utricular duct and the saccular duct, respectively. The utriculoendolymphatic valve (Bast's valve) separates the pars superior from the pars inferior, and may also protect the pars superior [7,8]. Therefore, the hydrostatic pressure in the pars superior and the pars inferior may be regulated by different systems.

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Although the membranous labyrinth is a seamless cavity, the direct current potentials differ in each part of the labyrinth [9]. The current potentials of the ES and ampulla are referred to as the ES potential (ESP) and the ampullar endolymphatic potential (AEP), respectively [10–14]. AEP generated by the dark cells in the utricle and ampullae is independent of the endocochlear potential (EP), and contains positive and negative components like EP [13]. The direct current potential in each part of the labyrinth shows different responses to various agents. Catecholamines reportedly depress ESP via β-adrenergic receptors without changing EP [14,15]. It has been reported that intravenous administration of canrenoate, an aldosterone antagonist, increased AEP [16], and that it decreased ESP with no change in EP [17].

The ES has been hypothesized to regulate the endolymphatic hydrostatic pressure [18,19]. Our recent study [20] demonstrates that intravenous administration of isoproterenol, a β-adrenergic receptor agonist, significantly increased the hydrostatic pressure of cochlear endolymph and perilymph with no change in EP or the hydrostatic pressure of the cerebrospinal fluid in normal guinea pigs, whereas the action of isoproterenol on the hydrostatic pressure of cochlear endolymph and perilymph disappeared almost entirely in ears with obstructed ES. These findings support the regulation of cochlear endolymphatic pressure by the ES.

In order to investigate the difference in the responses of endolymphatic hydrostatic pressure to isoproterenol between pars superior and pars inferior, we examined the responses of the endolymphatic hydrostatic pressure in the ampulla (pars superior) and the cochlea (pars inferior) to isoproterenol.

2. Materials and methods

2.1. Animal preparation and recording techniques

Albino guinea pigs with a positive Preyer's reflex (weight, 300–400 g) were used. The protocols for animal care and use were approved by the Experimental Animal Committee of the Faculty of Medicine at Kagawa University (Protocol #31), in accordance with the principles of the Declaration of Helsinki. During the experiments, the animals were deeply anesthetized by intramuscular administration of a mixture of ketamine (Daiichi Sankyo, Tokyo, Japan) (50 mg/kg body weight) and xylazine (Sigma–Aldrich, Tokyo, Japan) (5 mg/kg body weight) and allowed to breathe spontaneously. The electrocardiographs and heart rates of all animals were monitored throughout the experiments. The body temperatures of the animals were maintained at 36–38 °C using a heating pad. The agents were administered by left jugular vein cannulation.

The head of the animal was fixed in a prone position by using a head holder (SH-15; Narishige, Tokyo, Japan). Glass micropipettes were beveled to a tip diameter of 5–10 μm and filled with 2 M KCl [21]. The bulla of the temporal bone was exposed using a retroauricular approach to record the hydrostatic pressure of ampullar endolymph and perilymph and using a submandibular approach to record the hydrostatic pressure of cochlear endolymph and perilymph. To record endolymphatic hydrostatic pressure in the ampulla, an operating microscope and micromanipulator were used to insert the tip of a micropipette into the endolymphatic space in the superior ampulla through a fenestra in the bony wall [13,16]. The pipette was sealed in place with methyl cyanoacrylate adhesive (Permabond 910; Permabond, NJ) [22]. AEP was simultaneously measured to verify the position of the pipette tip. To measure the hydrostatic pressure of ampullar perilymph, the tip of a micropipette was inserted into the perilymphatic space in the superior ampulla through a fenestra in the bony wall. To record the hydrostatic pressure of cochlear endolymph and perilymph, the tip of a micropipette was inserted into the endolymphatic space in the cochlear scala media and the perilymphatic space through a fenestra in the bony cochlear wall of the basal turn.

The pipettes were connected to a servo-null system (900A Micropressure System; World Precision Instruments, Sarasota, FL). The system was calibrated by lowering a micropipette by 0 and 1 cm into an electrically grounded beaker of normal saline [18].

An Ag-AgCl reference electrode was placed on the neck muscles of the animals. All experiments were performed in an electrically shielded booth.

2.2. Drug administration

As isoproterenol has the strongest potency of any catecholamines for action on the ESP, this substance was used as one of the catecholamines [14,15]. Isoproterenol (L-isoproterenol hydrochloride; Kowa, Tokyo, Japan) diluted in saline was infused through a catheter inserted into the jugular vein by using an infusion pump (STC-521; Terumo, Tokyo, Japan) with an infusion rate of 0.5 ml/min at a concentration of 6.25 $\mu g\ kg^{-1}\ min^{-1}$, which was sufficient to reduce the ESP near the lowest level [23].

2.3. Data analysis

The data obtained from experiments with preinfusion AEP values of less than 2 mV or preinfusion EP values of less than 50 mV were excluded. The values are presented as means ± SE. Welch's t test was used to determine statistical differences. For comparison of more than two groups, analysis of variance (ANOVA) was used.

3. Results

Table 1 lists the pretreatment values of AEP and EP and hydrostatic pressure of ampullar and cochlear endolymph and perilymph in normal animals. The amplitude of AEP ($n = 20$) was similar to the results in the previous reports [11,13,16]. The magnitudes of the hydrostatic pressure in ampullar endolymph ($n = 20$) and perilymph ($n = 5$) were similar to those of cochlear endolymph (scala media ($n = 3$)) and perilymph (scala vestibuli ($n = 3$) and scala tympani ($n = 10$)), respectively. There was no significant difference in magnitude among endolymph and perilymph in the ampulla and the cochlea.

Fig. 1 shows the response of the endolymphatic hydrostatic pressure in ampulla and AEP to intravenous administration of

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