NATRIOREXIGENIC EFFECT OF DAMGO IS DECREASED BY BLOCKING AT1 RECEPTORS IN THE CENTRAL NUCLEUS OF THE AMYGDALA

The central nucleus of the amygdala (CeA), a critical coordinator of emotion and behavior (Lang and Davis, 2006), hosts important facilitatory mechanisms for the control of sodium intake. Previous studies showed that bilateral electrolytic lesions of the CeA abolished spontaneous sodium intake and sodium appetite induced by subcutaneous injections of the mineralocorticoid deoxycorticosterone, the α2-adrenoceptor antagonist yohimbine, or angiotensin II (ANG II), intracerebral ventricle injections of renin or by 24 h of sodium depletion in rats treated with furosemide (Galaverna et al., 1992; Zardetto-Smith et al., 1994), suggesting that induction of sodium appetite by different excitatory stimuli like ANG II or mineralocorticoids depends on CeA facilitatory mechanisms for the control of sodium intake. Lately, we have also shown that CeA μ-opioid receptor (μ-OR) activation enhances 0.3 M NaCl intake in rats submitted to water deprivation–partial rehydration (WD–PR) or in rats treated with the diuretic furosemide (FURO) (10 mg/kg b.w.) combined with a low dose of the angiotensin-converting enzyme inhibitor captopril (CAP) (5 mg/kg b.w.) injected subcutaneously (FURO/CAP) (Yan et al., 2013).

The CeA produces its actions through extensive efferent projections to the basal forebrain, hypothalamus, midbrain, and brainstem nuclei that mediate fear response, reward behavior, and environmental analgesia (Pitkanen et al., 1997; Swanson and Petrovich, 1998; Davis, 2000). μ-ORs are present in the CeA (Mansour et al., 1995; Poulin et al., 2006; Glass et al., 2009) and the CeA contains intrinsic neurons and axon terminals that contain opioid peptides (Fallon and Leslie, 1986; Cassell and Gray, 1989; Poulin et al., 2006). Activation of μ-ORs in the CeA has shown to inhibit most of CeA neurons (Zhu and Pan, 2004; Chieng et al., 2006). Given the anatomical evidence that CeA cells and their efferent projections are predominantly GABAergic (Swanson and Petrovich, 1998; Sah et al., 2003), it is possible that CeA neurons hyperpolarized by the activation of CeA μ-ORs are mostly GABAergic neurons and at least some of them send GABAergic output projections (Zhu and Pan, 2004). Taken together, opioid could negatively modulate CeA GABAergic projection neurons.
The effects of ANG II acting centrally on regulation of fluid and electrolyte balance and related behaviors are mediated mainly by ANG II type 1 (AT1) receptors located in different regions of the brain, such as the lateral parabrachial nucleus (LPBN), anterior hypothalamic area (AHA), amygdala, subfornical organ (SFO) and organum vasculosum of the lamina terminalis (OVLT) (Fregly and Rowland, 1991; Rowland et al., 1992; McKinley et al., 1996; Morris et al., 2002; Krause et al., 2008). The CeA contains AT1 receptors and has been proposed as a possible site of interaction between ANG II and mineralocorticoids to stimulate sodium appetite (Galaverna et al., 1992; McKinley et al., 2002). Different investigations using whole-cell voltage-clamp techniques have shown that ANG II acting on AT1 receptors may modulate GABAergic synaptic transmission and the effects of ANG II acting on pre- and post-synaptic AT1 receptors are just opposite (Li et al., 2003; Li and Pan, 2005; Henry et al., 2009; Xing et al., 2009). It has been suggested that ANG II acting on pre-synaptic AT1 receptors decreases GABA release and reduces the amplitude of evoked GABAergic inhibitory post-synaptic currents (IPSCs) (Li et al., 2003; Li and Pan, 2005; Xing et al., 2009). In contrast, it was shown that endogenous ANG II acting on post-synaptic AT1 receptors increases IPSCs in sodium-sensitive neurons in the median preoptic nucleus (MnPO), suggesting a post synaptic action of endogenous ANG II that facilitated the effect of the GABAergic input to the MnPO (Henry et al., 2009).

Considering the possibility of opioid negatively modulating CeA GABAergic projection neurons, the effect of the activation of CeA μ-ORs on sodium intake (Yan et al., 2013), and the results of previous studies showing that AT1 receptor activation may modulate GABAergic synaptic transmission (Li et al., 2003; Li and Pan, 2005; Henry et al., 2009; Xing et al., 2009), in the present study we investigated the effects of injections of losartan, the nonpeptide antagonist that selectively binds to AT1 receptors, on the GABAergic input to the MnPO (Yan et al., 2013). It has been shown that ANG II acting on AT1 receptors may modulate GABAergic synaptic transmission and the effects of ANG II acting on pre- and post-synaptic AT1 receptors are just opposite (Li et al., 2003; Li and Pan, 2005; Henry et al., 2009; Xing et al., 2009). In contrast, it was shown that endogenous ANG II acting on post-synaptic AT1 receptors increases IPSCs in sodium-sensitive neurons in the median preoptic nucleus (MnPO), suggesting a post synaptic action of endogenous ANG II that facilitated the effect of the GABAergic input to the MnPO (Henry et al., 2009).

Cerebral cannulas

Following anesthesia with an intraperitoneal dose of chloral hydrate (300 mg/kg b.w.), the rats were secured in a stereotaxic apparatus (SN-2N, Narishige Group, Tokyo, Japan) for bilateral implantation of stainless steel cannulas (23 gauge) into the CeA. The stereotaxic coordinates of the CeA were determined according to the brain atlas of the rat (Paxinos and Watson, 1997) and were: 2.3 mm posterior to bregma, 4.0 mm lateral to the midline suture, and 7.0 mm below the skull surface. The tips of the cannulas were placed 1 mm above the CeA. The cannulas were cemented to the skull bone with dental acrylic resin and jeweler screws and filled with obstructors (30 gauge). After the cerebral surgery, the rats were allowed to recover for 7 days in individual metabolism cages, a part of Feeding-Drinking-Activity Analyser (Cat. No. 4180011121) (UGO Basline Biological Research Apparatus, COMERIO-Varese, Italy), with free access to pelleted laboratory rodent chow, distilled water and 0.3 M NaCl solution before starting injection tests.

Injections into the CeA

Bilateral injections into the CeA were administered using 1-μL Hamilton syringes (Hamilton, Reno, NV, USA) connected by PE-10 polyethylene tubing to 30-gauge injection cannulas. At the time of testing, obturators were removed and the injection cannula (1 mm longer than the guide cannula) was carefully inserted into the guide cannula, and manual injection was initiated 15 s later. The injection volume into the CeA was 0.5 μL in each site and the injection was delivered at a flow rate of 0.5 μL/min. The injection cannulas were maintained in place for 30 s after delivery of the drugs or vehicle to minimize the backflow. The obturators were replaced after the injections, and the rats were placed back into their individual metabolism cages.

Drugs

The drugs including the selective μ-OR agonist DAMGO, the specific AT1 receptor antagonist losartan potassium, the diuretic FURO, and the CAP, were purchased from Sigma–Aldrich (Sigma–Aldrich, Saint Louis, MO, USA). All the drugs were dissolved in sterile 0.9% (w/v) saline solution. Accordingly, the 0.9% saline solution was used as vehicle. The drugs and vehicle solutions were made just before the infusion. DAMGO was administered in the CeA at the dose of 2 nmol in 0.5 μL which has previously been shown to be effective in dose–response study on the effects of CeA injections of DAMGO on sodium intake (Yan et al., 2013). Losartan was injected into the CeA at the dose of 108 nmol in 0.5 μL based on previous studies that have tested the effects of central injections of losartan on sodium and water intake and on the pressor response to ANG II (Gripp et al., 2002; Menani et al., 2004; Da Silva et al., 2011a,b; Roncari et al., 2011). FURO and CAP were administered subcutaneously at 10 mg/kg and 5 mg/kg of body weight respectively as described previously.
0.3 M NaCl and water intake by WD–PR rats

After the cerebral surgery, the rats had free access to food, water and 0.3 M NaCl solution for at least 7 days (period of recovery) in individual metabolism cages mounted on Feeding-Drinking-Activity Analysers where the rats were tested.

The WD–PR protocol is a sequence of water deprivation and partial volume repletion ensured by water intake that precedes the sodium appetite test. In the present study, bilateral-cannulated rats were deprived of water and 0.3 M NaCl solution (WD), with free access to food for 24 h (from 08:00 to next 08:00). Then, food was removed and water was offered for 2 h (PR). Food was not available for the rats during tests.

Following the WD–PR, bilateral-cannulated rats (n = 16) received CeA injections of combinations of saline, DAMGO (2 nmol), and losartan (108 nmol). The rats were randomly assigned to treatment groups to receive either (a) saline + saline, (b) saline + DAMGO 2 nmol, (c) losartan 108 nmol + saline, (d) losartan 108 nmol + DAMGO 2 nmol. Each rat received all four treatments in a counter-balanced design. Bilateral injections of losartan or saline into the CeA were performed 10 min before the injections of DAMGO into the CeA. Following the CeA injections, 0.3 M NaCl was offered, and 0.3 M NaCl and water intakes were automatically recorded by the Feeding-Drinking-Activity Analyser at 15, 30, 45, 60, 90, 120, 150, 180, 210 and 240 min. A recovery period of at least 3 days was allowed between tests.

0.3 M NaCl and water intake by FURO/CAP rats

After the cerebral surgery, the rats had free access to food, water and 0.3 M NaCl solution for at least 7 days (period of recovery) in individual metabolism cages mounted on Feeding-Drinking-Activity Analysers where the rats were tested.

The FURO/CAP protocol is that a subcutaneous injection of the diuretic FURO (10 mg/kg b.w.) is combined with an angiotensin-converting enzyme inhibitor CAP (5 mg/kg b.w.). In the present study, bilateral-cannulated rats received subcutaneous injections of the diuretic FURO (10 mg/kg b.w.) plus CAP (5 mg/kg b.w.) at 08:00 and then were returned to their individual metabolism cages in absence of food, water and 0.3 M NaCl solution. Food was not available for the rats during tests.

One hour after bilateral-cannulated rats (n = 16) received subcutaneous injections of the FURO(10 mg/kg b.w.) plus CAP (5 mg/kg b.w.), the rats received bilateral injections of combinations of saline, DAMGO (2 nmol) and losartan (108 nmol) into the CeA. The rats were randomly assigned to treatment groups to receive either (a) saline + saline, (b) saline + DAMGO 2 nmol, (c) losartan 108 nmol + saline, (d) losartan 108 nmol + DAMGO 2 nmol. Each rat received all four treatments in a counter-balanced design. Bilateral injections of losartan or saline into the CeA were performed 10 min before the injections of DAMGO into the same site. Following the CeA injections, water and 0.3 M NaCl, but no food, were available for rats and intakes of 0.3 M NaCl and water were automatically measured by the Feeding-Drinking-Activity Analyser at 15, 30, 45, 60, 90, 120, 150, 180, 210, and 240 min. A recovery period of at least 3 days was allowed between tests.

Histology

After the completion of each experiment, the rats received bilateral injections of 0.5 μL of 2% Chicago sky blue solution into the CeA. The rats were then deeply anesthetized with a high dose of chloral hydrate and perfused transcardially with saline followed by 10% formalin. The brains were removed, fixed in 10% formalin, cut into 40-μm serial coronal sections on a freezing microtome, and analyzed under a light microscope to confirm the injection sites in the CeA with reference to the atlas of Paxinos and Watson (Paxinos and Watson, 1997).

Statistical analysis

Statistical analysis was performed using SPSS for Windows (version 13.0). According to Fitts (Fitts, 2006), non-cumulative data from induced intakes were utilized for statistical purpose. Data are presented as means ± S.E.M. and were analyzed using an analysis of variance (ANOVA) except when the assumptions for ANOVA were violated, in which case nonparametric statistics were used. Planned comparisons were made using Fisher’s LSD test following a significant F-ratio. The statistical significance was set at P less than 0.05.

RESULTS

Histological analysis

Fig. 1 corresponds to a diagram based on the Paxinos and Watson Atlas (Paxinos and Watson, 1997) showing sequential coronal sections of the areas reached by injections within and outside the CeA. Fig. 2 is a photomicrograph of a transverse section of the forebrain of one rat, representative of the rats tested, showing the typical bilateral injection sites in the CeA. 32 rats were originally used, but in experiments, three rats were dead and four rats’ cerebral cannulas were pulled out by themselves. Injections reaching the medial, lateral and capsular portions of the CeA were observed in some rats and these rats’ number was 16 and the results from these rats were included in the statistical analyses of data presented in Figs. 3 and 4. Results from the other nine rats in which injections did not reach the CeA (misplaced injections) were included in the statistical analyses of data presented in Figs. 5 and 6.

Effects of combined injections of losartan and DAMGO into the CeA on WD–PR-induced 0.3 M NaCl and water intake

In the uncumulated analysis, ANOVA revealed significant main effects of drug treatment [0.3 M NaCl: F(3, 21) =

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postinjection time [0.3 M NaCl: $F(9,63) = 5.061$, $P = 0.000$; water: $F(9,63) = 7.866$, $P = 0.000$], and their interaction [0.3 M NaCl: $F(27,189) = 2.018$, $P = 0.004$; water: $F(27,189) = 2.727$, $P = 0.000$] (Fig. 3, left). Compared to saline injected into the CeA (saline + saline), bilateral injections of DAMGO (2 nmol in 0.5 µl at each site) into the CeA (saline + DAMGO) in WD–PR rats induced 0.3 M NaCl intake at 45, 60, 90 and 120 min of test (Fig. 3, top left) and water intake at

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**Fig. 1.** Diagram based on the Paxinos and Watson Atlas (Paxinos and Watson, 1997) showing sequential coronal sections of areas reached by the injections within (hachured area) and outside (dotted area) the central nucleus of the amygdala. OT, optic tract.

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There were significant main effects of drug treatment 
Effects of combined injections of losartan and DAMGO into the CeA (arrows) reduced the ingestion of 0.3 M NaCl and water intake. Specificity of injections into the CeA on 0.3 M NaCl and water intake. Results from rats that received injections outside the CeA (misplaced injections) were analyzed to show that the effects on 0.3 M NaCl and water intake were due to a specific activation of μ-ORs in the CeA. Misplaced injections were medial, lateral, or dorsal to the CeA (as shown in Fig. 1).

In the uncumulated analysis, in WD–PR rats (Fig. 5, left) or in FURO/CAP rats (Fig. 6, left) that received injections in sites outside the CeA, ANOVA revealed no significant main effects of drug treatment [WD–PR: 0.3 M NaCl F(3,12) = 0.202, P = 0.893; water F(3,12) = 0.494, P = 0.693. FURO/CAP: 0.3 M NaCl F(3,9) = 0.482, P = 0.703; water F(3,9) = 0.311, P = 0.817] and interaction between treatment and postinjection time [WD–PR: 0.3 M NaCl F(27,108) = 0.647, P = 0.904; water F(27,108) = 0.633, P = 0.914. FURO/CAP: 0.3 M NaCl F(27,81) = 0.600, P = 0.932; water F(27, 81) = 0.552, P = 0.958]. There was no significant main effects of drug treatment for the total 0.3 M NaCl intake [WD–PR: F(3,16) = 0.184, P = 0.906 (Fig. 5, top right); FURO/CAP: F(3,12) = 0.532, P = 0.744 (Fig. 6, top right)] and the total water intake [WD–PR: F(3,16) = 0.493, P = 0.692 (Fig. 5, bottom right); FURO/CAP: F(3,12) = 0.429, P = 0.736 (Fig. 6, bottom right)].
FURO/CAP rats that received injections in sites outside the CeA.

DISCUSSION

Similar to previous results (Yan et al., 2013), the present study shows that bilateral injections of µ-OR agonist, DAMGO, into the CeA induce 0.3 M NaCl and water ingestion in WD–PR rats or in FURO/CAP rats, which suggests that DAMGO activates CeA µ-ORs to increase sodium intake. Here, the present results extend the conclusions of the previous study by showing that pre-treatment with bilateral injections of the specific AT1 receptor antagonist, losartan, into the CeA reduce 0.3 M NaCl and water intake induced by DAMGO bilaterally injected into the same site in WD–PR rats or in FURO/CAP rats. However, injections of losartan alone into the CeA did not change 0.3 M NaCl or water intake in WD–PR rats or in FURO/CAP rats. Results from rats with misplaced injections confirm that DAMGO effect on 0.3 M NaCl and water intake is specific to the CeA. The present results suggest that angiotensinergic mechanisms in the CeA are essential for the natriorexigenic responses induced by the activation of µ-ORs with DAMGO injected into the CeA in WD–PR rats or in FURO/CAP rats.

Injections of DAMGO into the CeA did not induce water intake if only water was available (Yan et al., 2013). In the present study the intake of water increased after injections of DAMGO into the CeA when rats simultaneously ingested 0.3 M NaCl, which is
probably as a consequence of the increased plasma osmolarity due to the excessive ingestion of hypertonic NaCl. Injections of DAMGO into the CeA produce no effects on arterial pressure and on renal excretion in WD–PR rats or in FURO/CAP rats (Yan et al., 2013), suggesting that in present study 0.3 M NaCl and water intake produced by DAMGO injected into the CeA is not secondary to cardiovascular responses or increased renal excretion.

It is noticed that pre-treatment with losartan injected into the CeA reduced DAMGO effects on 0.3 M NaCl intake in WD–PR rats or in FURO/CAP rats, implying that endogenous opioid peptides release or their interaction with activated AT1 receptors in the CeA is essential for sodium intake induced by WD–PR or FURO/CAP. If endogenous opioid peptides release in the CeA was important for WD–PR or FURO/CAP-induced sodium intake, similar effects would be expected when losartan alone was injected into the CeA in WD–PR rats or in FURO/CAP rats. However, injections of losartan alone did not show such effects in our experiments. Perhaps any reduction of endogenous opioid peptides effects by losartan was compensated for by changes in the release of other unknown neurotransmitters in the CeA that also modulate sodium intake.

Differences in the control of sodium intake (Galaverna et al., 1992; Zardetto-Smith et al., 1994; Yan et al., 2013). The CeA

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**Fig. 4.** Non-cumulative (left) 0.3 M NaCl (top) and water (bottom) intake in experiment and the total (right) 0.3 M NaCl (top) and water (bottom) intake over the entire 240-min experiment by FURO/CAP rats that received bilateral injections of combinations of saline, DAMGO 2 nmol and losartan 108 nmol into the CeA. *Different from saline + saline; #Different from saline + DAMGO 2 nmol. n = number of rats.
has reciprocal direct connections with the LPBN and the nucleus of the solitary tract (NTS) (Norgren, 1995; Swanson and Petrovich, 1998). Signals from volume, taste and other visceral receptors that participate in the control of sodium and water intake ascend to the NTS and then make a second relay in the LPBN prior to projecting into the amygdala, thus forming a major neuroaxis for the control of taste and sodium appetite (Flynn et al., 1991; Johnson et al., 1999; Geerling and Loewy, 2006; Norgren et al., 2006). This particular pathway may be one of the neuroanatomical circuits explaining the modulation of sodium intake by the CeA (Geerling and Loewy, 2006). Important inhibitory mechanisms for the control of NaCl and water intake have been demonstrated in the LPBN (Edwards and Johnson, 1991; Callera et al., 2005). CeA and LPBN are strongly connected to control sodium appetite and it seems that the increase of sodium intake produced by the deactivation of LPBN inhibitory mechanisms is totally dependent on facilitatory mechanisms present in the CeA (Andrade-Franzé et al., 2010). Given most of CeA neurons are inhibited by the activation of $\mu$-ORs in the CeA (Zhu and Pan, 2004; Chieng et al., 2006), and CeA neurons and their efferent projections are predominantly GABAergic (Swanson and Petrovich, 1998; Sah et al., 2003), it is possible that CeA neurons hyperpolarized by the activation of CeA $\mu$-ORs are mostly GABAergic neurons and at least some of them send GABAergic

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**Fig. 5.** Non-cumulative (left) 0.3 M NaCl (top) and water (bottom) intake in experiment and the total (right) 0.3 M NaCl (top) and water (bottom) intake over the entire 240-min experiment by WD–PR rats that received bilateral injections of combinations of saline, DAMGO 2 nmol and losartan 108 nmol into sites outside the CeA (misplaced injections). $n$ = number of rats.
output projections (Zhu and Pan, 2004). Given the anatomical evidence that GABAergic connections exist between the CeA and LPBN (Jia et al., 2005), based on the present results, it is enticing to propose possible roles for CeA neurons in the opioid-mediated inducing sodium intake. In the CeA, endogenously released opioid peptides, or exogenously administered opioids, would inhibit, through $\mu$-ORs, those GABAergic projection neurons, and reduce their inhibitory effects on the projection targets such as the LPBN. The reduced inhibition from the CeA (disinhibition) could contribute to the deactivation of LPBN inhibitory mechanisms for sodium intake, and then result in 0.3 M NaCl intake. Indeed, $\mu$-ORs agonists also inhibit those CeA neurons by increasing local GABA synaptic transmission (Roberto et al., 2003), and induce c-Fos immunoreactivity in enkephalin-expressing CeA cells (Criado and Morales, 2000). In addition, projections from the CeA have also been hypothesized to reduce the inhibitory activity of the paraventricular nucleus of the hypothalamus (PVN) in the control of sodium appetite (Gray et al., 1989; Zardetto-Smith et al., 1994).

AT1 receptors are present in different areas of the brain including the CeA, and the CeA has been proposed as a possible site of interaction between ANG II and mineralocorticoids to stimulate sodium appetite (Galaverna et al., 1992; McKinley et al., 2002). Intracerebral ventricle injection of ANG II significantly

Fig. 6. Non-cumulative (left) 0.3 M NaCl (top) and water (bottom) intake in experiment and the total (right) 0.3 M NaCl (top) and water (bottom) intake over the entire 240-min experiment by FURO/CAP rats that received bilateral injections of combinations of saline, DAMGO 2 nmol and losartan 108 nmol into sites outside the CeA (misplaced injections). $n =$ number of rats.
ANG II acting on AT1 receptors may modulate AT1 receptors in the CeA is important for the inhibition of sodium intake. In other words, the action of ANG II on mechanisms in the CeA are important to stimulate interactions of angiotensinergic and opioidergic et al., 2009). Taken together, these results suggest that natriorexigenic effect of CeA neurons, thereby facilitating sodium intake produced by the activation of µ-ORs within the CeA. Fig. 7 presents a possible schematic model on how µ-ORs and AT1 receptors may affect the activity of GABAergic projection neurons in the CeA involved in the control of sodium intake. The hypothesis is that both opioid peptides activating µ-ORs and ANG II acting on post-synaptic AT1 receptors produce inhibitory effects on CeA GABAergic projection neurons, then the disinhibition from CeA deactivates LPBN inhibitory mechanisms, etc., which results in a strong ingestion of sodium.

Fig. 7. Schematic diagram showing a possible model for µ-opioid and AT1 receptors may affect the activity of GABAergic projection neurons in the CeA involved in the control of sodium intake. CeA, the central nucleus of the amygdala; LPBN, the lateral parabrachial nucleus.

CONCLUSION

The results of the present study demonstrate that AT1 receptors blockade reduces the natriorexigenic effect of µ-OR activation in the CeA in WD–PR rats or in FURO/CAP rats. These pharmacological data, therefore, suggest that natriorexigenic effect of CeA µ-OR activation is facilitated by endogenous ANG II acting on AT1 receptors in the CeA.

DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the authors.

AUTHOR CONTRIBUTIONS

Junbao Yan, Jianqun Yan conception and design of research; Junbao Yan, Huiling Sun and Qian Wang performed experiments; Junbao Yan, Jianqun Yan analyzed data; Junbao Yan, Jianqun Yan, Huiling Sun, Qian Wang, Ke Chen, Bo Sun, Lin Song, Wei Yan, Xiaolin Zhao, Shiru Zhao, Yuan Zhang, Hu Qiao and Bo Hu interpreted results of experiments; Junbao Yan drafted the manuscript; Junbao Yan and Jianqun Yan edited the manuscript; Junbao Yan and Jianqun Yan revised the manuscript; Junbao Yan, Jianqun Yan, Huiling Sun, Qian Wang, Ke Chen, Bo Sun, Lin Song, Wei Yan, Xiaolin Zhao, Shiru Zhao, Yuan Zhang, Hu Qiao and Bo Hu approved the final version of the manuscript.

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REFERENCES


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