1	Application of insects to wounds of self and others in chimpanzees in the wild
2	Alessandra Mascaro ^{1,3} , Lara M. Southern ^{1,2,3} , Tobias Deschner ^{1,4} and Simone Pika ^{2,4,*}
3	¹ Max Planck Institute for Evolutionary Anthropology, Interim Group Primatology, Deutscher
4	Platz 6, 04103, Leipzig, Germany
5	² University of Osnabrück, Institute of Cognitive Science, Comparative BioCognition,
6	Artilleriestrasse 34, 49076 Osnabrück, Germany
7	³ shared first authorship
8	⁴ shared last authorshipx
9	*Please address all correspondence to Simone Pika: <u>spika@uos.de</u>
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19	prosocial behaviour

20 In brief

21	Medical practices characterize human societies and have been suggested to originate
22	from observing the behavior of other animals. Mascaro and colleagues report that
23	chimpanzees apply insects to their own and the open wounds of other conspecifics, thereby
24	adding to the current debate on self-medicative and prosocial behaviours in nonhuman
25	animals.
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Self-medication refers to the process by which a host suppresses or prevents the 31 deleterious effects of parasitism and other causes of illness via behavioural means¹. It has 32 been observed across multiple animal taxa (e.g., bears, elephants, moths, starlings)², with 33 multiple case studies concerning great apes^{1,3}. Although the majority of studies on self-34 35 medication in primates concern the ingestion of plant parts or non-nutritional substances to combat or control intestinal parasites⁴, more recent examples also report topical applications 36 37 of leaves or other materials (including arthropods) to skin integuments³. Thus far, however, 38 the application of insects or insect parts to an individual's own wound or the wound of a conspecific has never been reported in non-human primates 39

40 Here, we report the first observations of chimpanzees applying insects to their own wounds (N=19) and to the wounds of conspecifics (N=3, see table in the SI). Over a period of 41 42 15 months (November 2019 - February 2021), we observed a total of 76 open wounds on 22 43 different chimpanzees (eleven adult males, two adolescent males, one juvenile male, four 44 adult females, two adolescent females, and two juvenile females; see table in the SI). In 19 events, individuals (five adult males, one adult female, one juvenile female) applied an insect 45 46 to one of their own wounds using the following behavioural sequence: (1) they first caught an insect, (2) immobilised it by placing and/or squeezing the insect between their lips, (3) 47 placed the insect to an exposed surface of the wound and moved the insect on the surface 48 49 using their fingertips or lips, and (4) extracted the insect from the wound with the mouth or 50 their fingers (see figure 1, videos S1 and S2 and additional videos at https://youtube.com/c/OzougaSociety). Parts 3 and 4 are often repeated multiple times 51 during each event. Though the insect species(') utilised has not yet been identified, there are 52 several consistencies across all our observations: 1) the insect(s) appears to be a winged, 53 54 flying insect, given the fast motion used to catch it; 2) the insect(s) is caught from under a leaf

or branch; 3) the insect(s) are small in size ~5mm and usually dark in colour and 4) there was
no observation of insect ingestion.

In three other events, we observed different chimpanzees applying or moving an insect not to their own wound, but to the wound of another chimpanzee (see selected event descriptions in the SI). On November 13th 2019, an adult female, Suzee, caught an insect and applied it to an approximately 2cm open flesh wound on the foot of her adolescent son, Sia. Subsequently, she extracted and re-applied the insect two more times using both her mouth and fingers (see video S2). This was the only event of allo-application involving maternally related individuals.

On October 20th 2020, another adult female, Carol, had been grooming around the approximately 5cm open flesh wound on the calf of an adult male, Littlegrey. She then caught an insect, and Littlegrey took it from her fingers, put it between his lips and placed the insect to the surface of his open wound. Subsequently, Carol and another adult male, Thea, used their fingers to move the insect on the surface of the wound. A third adult male, Ngonde, then approached them, took the insect out of the wound, placed it between his lips, and re-applied it to the wound.

On January 29th 2021, another adult male, Arnold, caught an insect, moved it to his mouth, between his lips and then applied it to the surface of an approximately one cm open flesh wound on the right thumb of Littlegrey. He lip-smacked whilst moving the insect on the surface of the wound with his fingertips.

Given the unambiguous context in which the observed behaviour occurred (injured individuals with open flesh wounds), we suggest that these observations may represent another case of medicative behaviour in non-human animals². Moreover, these observations expand the existing knowledge to include allo-medication of open wounds. However, further

systematic research is needed to elucidate the efficacy of the treatment associated with an
improvement in healing of wounds, identification of insect species used, and the distribution
and acquisition of this behaviour in the Rekambo community.

Furthermore, our observations contribute to the current debate on the existence of 82 prosocial behaviours in non-human species (e.g.,^{5,6,7}). Prosocial behaviours refer to actions 83 that are intended to benefit another, and seem to be driven in humans by empathic concerns 84 for each other⁶. Prosocial behaviours have long posed a problem for evolutionary theory, 85 86 because it was not immediately clear why organisms might help others in the face of selection operating in the interest of self. Chimpanzees have been suggested as important candidates 87 for studies into the evolution of prosocial behaviors because they participate in a variety of 88 89 activities that benefit from cooperation, such as territorial patrols, coalitionary aggression, and hunting⁸. However, the literature remains controversial, with some evidence suggesting 90 91 that chimpanzees lack prosocial behaviours involving the type of empathy that is thought to characterize humans (e.g.,⁷), while others argue that their prosocial tendencies exhibit 92 characteristics consistent with empathy⁵. Hence, our observations may add another facet to 93 the ongoing debate on prosocial behaviors and inspire future studies investigating the 94 behaviours surrounding wound care and the potential medicative function of insect-95 application. 96

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108 Author Contributions

TD and SP direct the Ozouga Loango Chimpanzee Project and supervise the administrative
and scientific work. AM and LMS collected the data. AM, LMS, and SP edited the data. AM
wrote the first draft of the paper, and SP and LMS created the detailed event report. SP, LMS,
TD and AM finalized the paper.

Declaration of interest

114 The authors declare no competing interests.

115 Supplemental Information

Supplemental data including methods (the study site description and data collection protocol), detailed descriptions of events and two video clips (see video S1 and S2) are available online at: xxx. Additional videos clips of insect application are available at https://youtube.com/c/OzougaSociety.

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121 Figure 1. Application of insect to a wound

Figure 1 shows the behavioral sequence of an adult chimpanzee male catching and applying 122 an insect to a wound on his left shin in six frames (A-F). Red circles highlight the object or 123 124 action of interest in specified frames. (A) The male captures an insect with his left hand; (B) He transfers the insect from the fingers of his left hand to his lips; (C) Keeping the insect 125 126 between his lips, he moves his head and lips towards the wound; (D) The male applies the 127 insect, which is still between his lips, onto an exposed surface of the open wound on his left 128 shin; (E) He moves the insect on the surface of the wound with the index finger of his right hand; (F) The male closely inspects his wound and continues to groom around it with his left 129 hand. 130

131 Video S1. Capture and self-application of an insect by an adult male, Freddy, to his own 132 open wound

This video involves an adult male, Freddy, who has a day-old wound in the elbow crook of his 133 left arm (for further details see event descriptions in the SI) and is resting close to another 134 135 adult male. He pulls a shrub towards himself with his left foot, reaches out with his right hand 136 and makes a quick catching movement thereby grabbing a leaf with his right hand. He then moves the leaf towards his mouth and uses his lips to pick up an unidentified insect from it. 137 138 He takes the insect from his mouth with the thumb and index finger of his right hand and applies it to the wound on his left arm using his thumb to move the insect on the surface of 139 the open wound. He then moves his head quickly towards the wound and picks up the insect 140 141 again with his lips. Next, he again takes the insect with the thumb and index finger of his right hand from his mouth/lips and applies it again to the wound. He presses it softly with his 142 thumb against the surface of the wound. Subsequently, he moved his fingers of his right hand 143

back to his mouth and seems to move and press the insect between his lips. He then applies
it again to the wound with the thumb and index finger of his right hand. Overall, this video
sequence shows a total of three applications of an unidentified insect to a wound.

147 Video S2. Capture and allo-application of an insect from an adult female, Suzee, to a wound 148 of her adolescent son, Sia

This video shows an adult female, Suzee, resting with her infant, Sassandra, and her 149 adolescent son, Sia (see SI for further information). Sia had a day-old open wound of 150 approximately three cm on his left foot and is lying on his back in proximity to her. Sassandra 151 is swinging playfully around her. Suzee sits up and reaches out with her right hand to catch 152 153 an unidentified insect from under a leaf of a low hanging branch. She looks at it between her fingers and then places it carefully between her lips. Sassandra directly stopps swinging, and 154 jumps towards her mother and observed her behaviour. She stretches out her arms to be 155 156 picked up but Suzee parries her off and moves closer to Sia whilst in parallel grabbing his left 157 foot with her right hand. Sia moves the foot up in the air and disengages Suzee's grip while 158 Suzee is still parrying off Sassandra. She then grabbs Sia's foot with her left hand, hands it over to her right hand, takes the insect out of her mouth with the fingers of her left hand and 159 applies it to the wound. Subsequently, she picks up the insect from the wound with her mouth 160 and appears to keep it tightly pressed between her lips. She then moves the fingers of her left 161 162 hand to her lips, takes the insect and applies it again to Sia's wound. An adult female, Joy, 163 approaches Suzee, Sassandra and Sia to observe the behaviour and her positioning reduces 164 the visibility of the observers. Suzee then again moves the fingers of her left hand from the wound back to her lips and then back to the wound. An adult male, Littlegrey, who had been 165 in proximity approaches them and positions himself quadrupedally to look over Joy's shoulder 166

- 167 and observes Suzee's behaviour. Overall, this video sequence shows a total of three
- applications of an unidentified insect to the wound of a related individual.

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