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ORIGINAL RESEARCH

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A method for deconstructing the health encounter in CAM: The social context

Ian Douglass Coulter, Gery Wayne Ryan, Lisa Sachiko Kraus, Lea Xenakis, Lara Green Hilton
RAND Corporation, Santa Monica, CA

ABSTRACT

Background: A critique of complementary and alternative medicine (CAM) is that its effectiveness is all placebo. In trying to deconstruct the placebo, scholars have looked at the context in the health encounter, identifying specific and non-specific effects in the doctor–patient relationship, including communication; cognitive, psychological, and emotional element of the encounter; the meaning response during the encounter; the impact of the context on outcomes; and how the encounter is manipulated through such things as ritual.

Methods: This study used the chiropractic and osteopathic health encounter as socially constructed space exemplars and used focus groups and a rapid ethnographic observation method in clinics to study the context of the encounter. The objective was to develop a systematic, valid, and rigorous methodology for collecting data about the contexts of health encounters.

Results: The study showed that the clinicians and clinical staff consciously construct social settings and the types of interactions that occur within them. This is done consciously and deliberatively.

Conclusion: The method, we describe, provided a rich data base on the context of the health encounter and it shows that these are not non-specific effects. The method could be applied to other health encounters in CAM and health care.

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KEYWORDS

Complementary and alternative medicine; health encounter; context; non-specific effects; placebo

Introduction

As a part of the movement to understand the placebo effect [1–4] in complementary alternative medicine (CAM) and in health care more generally, increasing attention is being paid to "deconstructing" the health encounter (deconstructing in this context means to analyze, break down, and to critique the assumptions about something). In the case of CAM, the definitional part of placebo that has been attributed to CAM is not that of an inert substance but of deceit. As Jonas [1] notes, the placebo label is used often for political rather than scientific reasons and this is prevalent in commentaries about CAM.

A standard assertion of those opposed to CAM is that positive outcomes from CAM are due entirely to the placebo [5]. The assertion has been given evidential grounding by studies showing that homeopathy [6] and acupuncture [7,8], while getting better results than usual care do not do significantly better than placebo. Other authors, however, have pointed out that the *sham* acupuncture used in such studies does not constitute a true sham [9]. As Walach [3] notes in his discussion of the history of blinded trials, "we learn one important lesson: the attempt to isolate the 'true' component of therapy comes at the cost of tearing a therapeutic system apart and partitioning a whole into allegedly separable entities."

The placebo disagreement has fueled efforts to delineate the non-specific and specific elements of the health encounter. There is broad consensus that the health encounter is a social encounter that occurs within cultural, social, and individual history [3]. Recent studies have explored the content of the doctor–patient communication in the encounter [10–12], focusing on the interpersonal elements of the encounter, such as affective communication and instrumental communication [13]. Attention has

Contact Ian Douglass Coulter ✉ coulter@rand.org RAND Corporation, Santa Monica, CA.

also been paid to the belief and expectations of the patients and the impact on outcomes [14] and on the meaning and context reponse [1,3,5,15].

In the process of deconstructing the placebo, scholars have elevated the importance of the context in which the health encounter occurs. Increasingly, there is recognition that it may not be simply termed a non-specific effect. For example, a systematic review by Di Blasi et al. [13] concluded that the doctor–patient relationship significantly affected health outcomes. Others concluded that the patient-centered communication in the encounters also results in improved health status [16], and attributed related physician–patient communication to empowering patients and improved clinical outcomes [17].

The studies on communication and ritual would suggest that the encounter might be highly specific and can be manipulated [18–20]. There is also increasing recognition that the health encounter is a socially and culturally created space [11], should not be "equated with non-specific effects" [5]. Despite this recognition, few studies treat the health encounter as a socially-constructed system—that is a system deliberately created to structure the activities and interactions of various actors with each other. Most studies have focused on the doctor–patient relationship, including the communications and psychological effects, or the doctor–patient dyad. But, health encounters are seldom just a dyad because they involve numerous actors working together within a specifically constructed clinic or site.

The health encounter includes everything that happens from the time patients enter the clinic until the time they leave. The social context is a functioning social system surrounded by cultural meanings, symbols, and a communication system with its own unique language. For the most part, recording communications or self-reported narratives cannot capture this level of complexity. For instance, Coulter [21] compared ethnographic observation studies of the chiropractic health encounter with the picture derived from health services research on the same encounter and concluded that they presented two completely different encounters. As he noted, if this were wildlife observation you would conclude the writers were observing two different species. What is needed is a new way to study the health encounter in rigorous and comprehensive manner [13].

How can we measure patients' health encounter experiences? The encounter can be divided into two parts: (a) the experience of the main treatment intervention (the therapy) and (b) all other experiences before, during, and after the intervention itself (the context). Contextual effects play two critical roles in assessing the efficacy and effectiveness of interventions. First, contextual effects may mediate how well a treatment works (context-as-mediator). Second, context effects may contribute to outcomes directly (context-as-intervention). In a classic random clinical trial, investigators typically want to control for context-as-mediator effects and measure context-as-intervention effects to disentangle what portion of the results are due to the intervention and what are due to the context. But, in comparative effectiveness research [22], investigators are less concerned about controlling for context-as-mediator effects, but they would like to understand what part of the encounter accounts for any positive results. In either case, investigators need to know how to measure the context. Without such measures, it is impossible to assess either type of context effect.

The health encounter is a social event occurring within a constructed social encounter as a social space. There has been a focus on the doctor–patient relationship, the communications, the meanings, and psychological effects. Studies, to date, have often focused mainly on the doctor–patient dyad. But, the health encounter is seldom just a dyad. It is an encounter that can involve numerous actors, that occurs within a clinic that can be specifically constructed with an effect in mind, that includes everything that happens from the time the patient enters the clinic until the time they leave, that may occur in several spaces in the clinic, that occurs over a span of time, and that has a history. It is a functioning *social system* surrounded by cultural meanings, symbols, and a communication system with its own unique language. For the most part, recording communications or self-reported narratives cannot capture this level of complexity. In this study, we set out to observe this social system in all its complexity. The objective was to develop a systematic, valid, and rigorous methodology for collecting data about the contexts of health encounters.

Objective

The objective of the study was to determine if using a structured rapid ethnographic observation methodology would allow us to collect data and identify

the elements of the total health encounter that occur from the time a patient enters a clinic until they leave. Ultimately, future studies might then be able to determine the impact of the context of the health encounter on health outcomes.

Research Questions

This study addresses three research questions: (1) what kinds of contextual factors are patients exposed to during CAM encounters? (2) what measure of contextual factors can be developed systematically via observation and/or patient and provider recall?, and (3) which contextual factors might vary within and across: (a) CAM modalities (i.e., chiropractic *vs*. osteopath); (b) practice sites; (c) providers; and (d) individual patient encounters? To address these questions, we developed a rapid ethnographic observation method to study the context of the chiropractic encounter, and then applied the method to osteopathic encounters as a way to validate the methodological approach. The objective was to develop a systematic, valid, and rigorous methodology for collecting data about the contexts of health encounters.

Methods

We used a multi-staged research design to develop and test a battery of instruments to systematically measure health encounters. These instruments captured five key dimensions of the health encounter, including: (1) *where* patients are (space); (2) *with whom* they interact (social); (3) *what is communicated* between them (communication); (4) *what* patients do or what is done to them (behavior); and (5) for *how long* and in what order 1–4 occur (time). Our work occurred in three phases. In Phase 1, we conducted focus groups with chiropractic and osteopathic patients to identify what features of the health encounter they felt were most important to them. We used the results to draft a set of systematic observational techniques and structured elicitation instruments to measure each of the key elements above. In Phase 2, we piloted the instruments as a part of a rapid ethnographic observation study [23–25] of chiropractic and osteopathic clinics. We used the results of this study to further modify the instruments. In Phase 3, we validated the finalized tools on 124 health encounters in 15 chiropractic clinics from three states in the United States (five clinics per state). We briefly describe each of the methodological phases below and the kinds of instruments that were ultimately developed and tested. This study was approved by RAND's IRB (Human Subjects Protection Committee).

Phase 1—Focus Groups

We conducted three focus groups with 27 participants, who were selected because they had visited a chiropractor or osteopath within the last 3 months. Focus group questions included open-ended discussion items probing patients' experiences with their providers and more broadly investigating health encounters with these providers and the clinic staff. We audiotaped the discussion and took detailed field notes that were used to pile sort the notes into domains to inform field instruments. Key themes and issues emerged that informed the initial data collection instruments and procedures in Phase 2.

Phase 2—Pilot Study

To further develop the health encounter instruments, we conducted 3-day site visits in a diverse sample of nine chiropractic practices and nine osteopathic practices throughout Los Angeles County. Site visits included detailed observations of the clinic, while in operation, interviews with chiropractors/osteopaths, staff and patients; and shadowing patients through their visit. We used these data and the literature to identify key contextual factors and develop appropriate and low-burden measures to be used in the national sample.

During the site visit 2, investigators mapped and photographed the entire facility, conducted general semi-structured interviews with providers and staff about what aspects of the clinic they think most affect patients' encounters (positively and negatively), shadowed patients from the time they arrived (and gave consent to participate in the study) to the time they left, and observed and took detailed notes on the clinic's operation over the course of 3 days.

For each patient that agreed to participate, we were conducting a *pre-encounter interview* to capture a patient's reason for and expectation about the visit. We then shadowed the patient through the rest of the encounter and recorded our observations using a standardized *observation form.* The form allowed us to follow the patient's progression through time and space. We initially divided the form into a series of 5-minute blocks and used it to record with whom the patients interacted, the

Table 1. Patient themes and their measurement in the health encounter.

Trust the patient does not wince or guard at being touched by the practitioner. The patient puts herself in the doctor's hands—e.g., lays down on the table, moves into the position requested—without argument or question other than for clarification.
Validation Doctor is able to reproduce the patient's pain and/or verbally validates the patient's symptoms. For example, "So you are feeling pain here." "I know how that feels and it can be terrible," "So you have limited motion turning your neck to the right."
Listening Doctor demonstrates active listening. Does not talk over or interrupt the patient and demonstrates that he/she heard what the patient said either through acknowledging it and/or responding consistently to the questions asked by the patient.
Comfort The patient remains visibly relaxed, and/or expresses comfort—e.g., that feels good, that feels better. This should NOT be checked if the patient looks like they are stressed, fearful, or experiencing pain during the encounter.
Empathy The practitioner mirrors or demonstrates an understanding of the patient's emotional state and concerns. [According to the Four Habits, empathy is demonstrated by the practitioner encouraging emotional expression, accepting the patient's feelings, identifying the patient's feelings, and displaying good (appropriate—e.g., not laughing at a painful story) nonverbal behavior.]
Knows me In conversations with the patient, the doctor demonstrates prior (remembered) information about the patient's family, vacation, job, or some other aspect of the patient's life not directly symptom related.
Routine Patient goes to a particular area of the room, takes off shoes, or moves into position without verbal indication by the chiropractor.

kinds of interactions they had, what they did, and what was done to them. Researcher also used the form to note evidence of (or lack of) *trust*, *validation*, *listening*, *comfort*, *empathy*, *knows them*, and *routine* using agreed definitions developed from patient focus group data (Table 1).

After the encounter, before the patient left the clinic, we conducted a *patient post-encounter interview* and asked them to describe their experience. We were particularly interested in understanding what the patient thought of the clinic space and ambience, how they felt about their interactions and communications with clinic personnel, and their experience outside of the manual therapy. We also used a *patient post-encounter survey* to learn whether the experience met their expectations and how satisfied they were with various aspects of the encounter. We also developed a *provider post-assessment of the patient* where we asked the provider to provide a prognosis and assessment for recovery for patients whose condition was similar to this patient's condition.

We refined our data collection instruments in an iterative manner modifying them as we moved from one clinic to the next. For example, we reduced the observation time from 5-minute increments to 2-minute increments or if the action changed. We concluded that 2-minute increments better captured the segments of each encounter and provided a more detailed account of the actions, topics, and personnel changes or consistency within an encounter. We also improved the *patient pre- and post-encounter surveys* by adding or modifying questions and response categories. The finalized tools were integrated in the data capture system for each researcher.

Phase 3—Multi-State Study

We used the national phase of our research to examine the feasibility and reliability of the finalized tool. We conducted this phase of the research in Minnesota, Oregon, and Texas. Within each state, we identified a county that had a high density of licensed chiropractors and a purposefully recruited diverse range of 15 clinics (five from each state). We spent 2 days in each clinic and recruited as many patients as we could during that time. In total, recruited 124 patients were: 41 from Minnesota, 38 from Oregon, and 45 from Texas.

During each of the site visits, we captured pictures of the clinic; sketched the clinic and labeled the offices and treatment rooms that were used by the provider and patient; conducted pre- and post-encounter interviews and surveys with each patient; completed a provider post-assessment of the patient; and conducted a provider post-visit interview and survey. Overall, we found that the instruments were quite useful in assessing variation within and across practice sites, providers, and individual patient encounters. In the results below, we demonstrate how the instruments developed can be used to describe the variation in health encounters within and between the 15 clinics, we observed.

Results

A health encounter is influenced by a clinics physical space and its social environment. The physical space includes the size and layout of the clinic, its décor, lighting, and overall atmosphere. The social space includes the personnel with whom the patient engages; the level of engagement

that is encouraged; and the way in which these components commingle, based on each clinic's operational style that can range from formal and professional to relaxed and homey.

The movement of patients through their encounter had a clinic-specific rhythm. Most clinics had minimal wait time and the patients were in tune with the clinic flow. Most varied in length from 15 minutes to an hour. A short observation typically involved the patient checking in with the front desk with minimal wait time, going back to the treatment room to get vitals taken by an assistant, a less than 10-minute diagnosis and treatment, and then a few minutes to pay the bill and check out. A long observation might entail checking in with a minimal wait time, sitting in a therapy chair for 30 minutes, while the assistant takes their vitals, getting traction for 15 minutes, then going in for diagnosis and treatment with the chiropractor, followed with 15 minutes of heat therapy.

In addition, to tracking a patient's movement through the clinic and often and how long patients interacted with the different clinic personnel, the *observation form* allowed investigators to check for evidence of *trust*, *validation*, *listening*, *comfort*, *empathy*, *knowing them*, and *routine* (Table 1). Figures 1 and 2 show how patients' encounters may vary within and between clinics.

The data in Figure 1 portrays a provider who displays high levels of *trust*, *validation*, and *knowing them* during the encounter. The findings are not surprising given that most of the patients in this clinic had been coming to this clinic for over 12 months. In a trusting interaction, the patient does not wince or guard at being touched by the practitioner and puts themselves in the doctor's hands. In a validating interaction, the chiropractor frequently checks on the patient's current condition, testing the range of motion, palpating known sore spots for tenderness, and acknowledges the pain and confirms the patient's feelings. The provider is this clinic demonstrated they knew their patients well by asking about members of a patient's family or friends and even in the cases where the chiropractor did not seem to know much about the patient's social network, would ask about the patient's work or social life. The provider also demonstrated a consistent pattern of

Figure 1. Positive interactions by duration.

Figure 2. Positive interactions by duration.

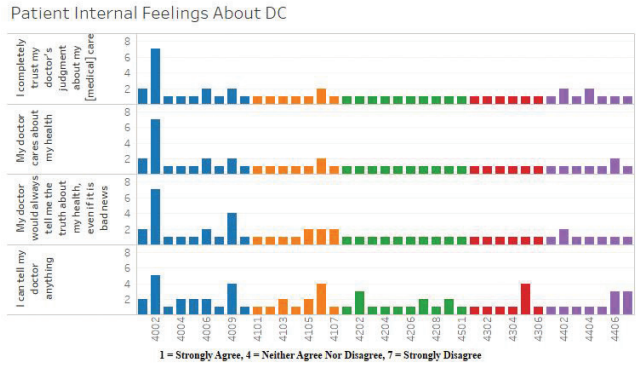

Figure 3. Patient internal feelings about DC.

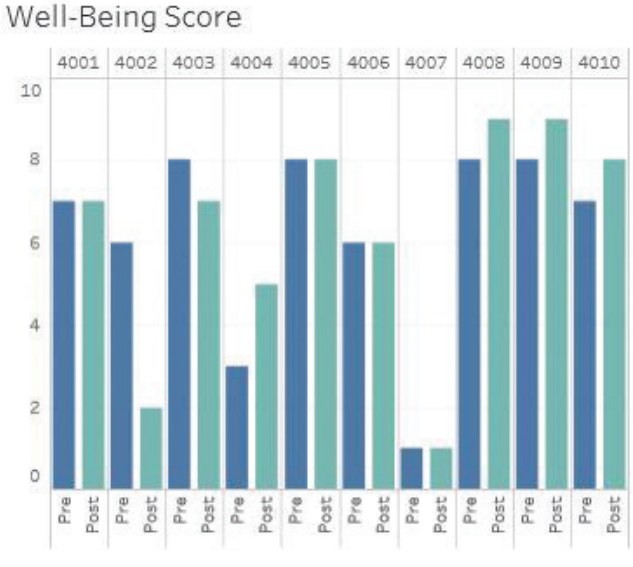

Figure 4. Well-being score.

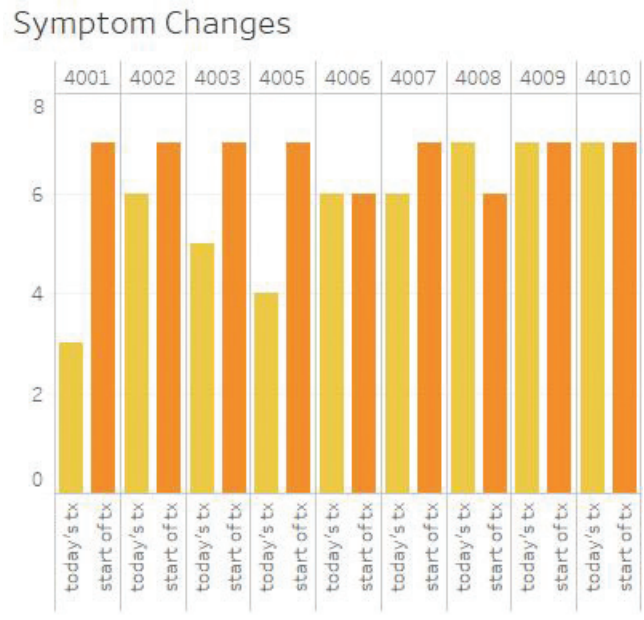

Figure 5. Symptom changes.

Figure 6. Pain score.

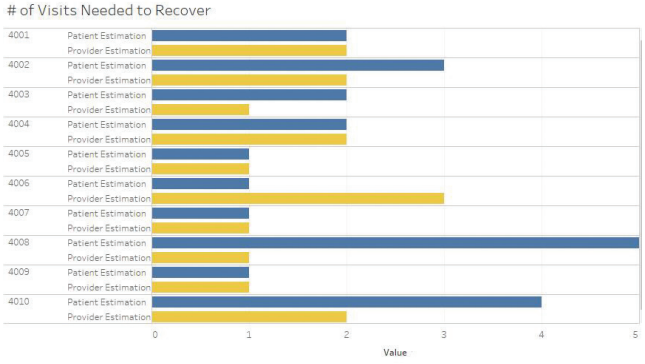

Figure 7. # of visits needed to recover.

active listening acknowledging that they had heard what the patient or responding consistently to the questions asked by the patient.

In contrast, Figure 2 depicts a clinic where encounters were mainly *routine and trusting* and had fewer interactions demonstrating active *listening*, *validation*, and *knowing the patient* than was observed in the previous clinic. A *routine* encounter is one in which there were no surprises and the patient and provider understood and already knew what was going to occur without a lot of instructions. Patients counted on having the same interaction or experience every time they entered the treatment room or office.

In most encounters, we observed that patients demonstrated some level of *comfort*. Patients appeared to be relaxed (often with their eyes closed) and showed no signs of tension or flinching was noted. Although comfort was ubiquitous across encounters, the number of times in which *comfort* was noted varied directly with the length of the encounter.

Unlike some types of interactions that seemed to vary significantly across clinics, *empathy* appeared to vary more by patient than clinic. An empathetic response was demonstrated when the chiropractor recognized and shared in a patient's situation. For example, when patients spoke about their conditions, and the provider would respond with phrases like, "I understand that must be difficult," or "awws" and "mmm" noises, or might share a similar story of their own. Non-verbal indications like pats on the back and hugs were also seen as signs of *empathy*. Internal space was captured through the patient pre-and post-encounter survey and physician assessment of patient. Patients were asked how they felt about their provider. The majority of patients felt positively about the provider and strongly agreed with the statements in Figure 3 (lower is better). There were positive changes in self-reported well-being (Fig. 4), symptom changes (Fig. 5), and pain (Fig. 6). Well-being improved or stayed the same most of the time, symptoms were better (didn't get worse), and pain decreased in most encounters. Patients' estimates of how many visits they would need to recover typically aligned with the number of visits that the provider believed a patient with the similar condition would need to recover (Fig. 7).

Conclusion and Discussion

The goal of this study was to develop instruments to systematically measure the health encounter. Although we applied these measures to chiropractic and osteopathic encounters, we believe that similar instruments could be adapted for type of health encounter. We used a multi-staged and multi-method approach to create tools for mapping and documenting the physical layout of the clinic, shadowing and observing patients, and

conducing structured interviews and surveys with patients, providers, and clinic staff. As a result, we could systematically describe a health encounter, including the physical space of the clinic, the social interactions between patients and providers and staff, time, and duration of patient behaviors, patient and providers beliefs, and expectations.

As noted in the Introduction, the placebo disagreement has fueled efforts to delineate the non-specific and specific elements of the health encounter [1,3,5,10–15]. In the process of deconstructing the placebo, scholars have elevated the importance of the context in which the health encounter occurs. Increasingly, there is recognition that it may not be simply termed a non-specific effect [13]. Others concluded that patient-centered communication in the encounters also results in improved health status [16] and attributed related physician–patient communication to empowering patients and improved clinical outcomes [17].

The previous studies, therefore, suggest that the encounter might be highly specific and can be manipulated. There is also increasing recognition that the health encounter is a socially and culturally created space [11], should not be "equated with non-specific effects" [5]. But despite this recognition, few studies treat the health encounter as a socially-constructed system—that is a system deliberately created to structure the activities and interactions of various actors with each other. This study, therefore, adds to our understanding of the encounter as a functioning social system that is consciously constructed.

When the instruments developed in this study are applied, we show that the encounters are highly structured. The physical and social spaces of a clinic are planned and deliberately constructed. While we observe variation across individual patients, patient encounters are more similar within a single clinic than they are dissimilar. In other words, clinics develop unique styles and standardized routines that affect patient experiences and care. Furthermore, the context of the health encounter is important to patients. Patients are conscious of the context and can articulate what aspects of the encounter they view as positive or negative. It seems inappropriate to consider such elements of the health encounter non-specific if the encounter is deliberately and consciously created and providers and staff and patients are aware of this.

The larger question arising from this study is how context affects health outcomes. This question was beyond the scope of the current study, which was designed to see if the context could be delineated and to see what kinds of data can be collected about the social space. Our data do suggest, however, that from the patient's point of view the context may be highly significant in their choice of CAM for their health problem and possibly plays a significant role in the outcomes from the patient's perspective.

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REVIEW ARTICLE

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Edible insects bio-actives as anti-oxidants: Current status and perspectives

Erhirhie Earnest Oghenesuvwe[1] and Chinwuba Paul[2]

[1]Department of Pharmacology and Toxicology, Faculty of Pharmaceutical Sciences, Nnamdi Azikiwe University, Awka, Nigeria.

[2]Department of Pharmacology and Toxicology, Faculty of Pharmaceutical Sciences, University of Nigeria, Nsukka, Nigeria.

ABSTRACT

Background/Aim: Oxidative stress mediated cell damage is implicated in aging and pathogenesis of several diseases, including inflammation, rheumatoid arthritis, cancer, cardiovascular, and neurodegenerative disorders. Combating the menace of oxidative stress requires improvement in endogenous antioxidant defense system or supplementation with exogenous antioxidants. Although synthetic and plant derived antioxidant food, supplements, and molecules have been discovered, insects, the largest class in the animal kingdom remains an unexploited resource. This present review evaluated insects and their products as sources of natural antioxidants, as wildly documented in literature.
Methods: Information related to this topic was retrieved using PubMed, google scholars, and google electronic databases. Insects' chemical constituents, nutritional and therapeutic benefits in oxidative stress intermediated disorders were emphasized.
Results: Insects possess antioxidant compounds which aid in curbing various pathologies. Out of various categories of insects, termites, beetles, grasshopper, and blowfly evaluated, the bees, *Apis mellifera's* products, royal jelly, and Propolis were the most extensively studied and also displayed promising antioxidant activities compared to other insects. *In vitro, in vivo*, and clinical studies from the aforementioned literatures, especially the bee category validates the fact that edible insects are a potential source of novel bioactive compounds in chronic diseases and aging.
Conclusion: As numerous diseases are oxidative stress mediated, application of edible insects and their products for culinary and therapeutic purposes is crucial. More researches in this field would aid in the development of insect derived antioxidant molecules.

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Introduction

Oxidative stress could be regarded as the excess generation of reactive oxygen species (ROS) [1,2]. It usually occurs when there is a disproportion between production of oxygen free radicals and the antioxidant defense system [3]. Uncurbed oxidative stress could impair lipids, proteins, ribonucleic acid (RNA), and deoxyribonucleic acid (DNA) thereby resulting in aging [4] as well as chronic diseases, including autoimmune disorders, diabetes [5], hypertension, neurodegenerative diseases, cancer [4], and rheumatoid arthritis, among other [3,6].

Several clinical and non-clinical research studies have revealed that the antioxidant supplementation is vital for the treatment and management of oxidative stress-related diseases [7]. In the attempt to overcome the limitations posed by conventional antioxidants, necessary efforts have been made by researchers in discovering alternative agents from natural sources, medicinal plants [8], marine products [9], as well as edible and non-edible insects [10,11].

Insects constitute about 80% of the animal kingdom [12]. In Africa, Asia, Central America, and South America, insects provide a good alternative source of vitamins, minerals, proteins, fats, and calories on a yearly basis [13,14]. Also, insects are

Contact Erhirhie Earnest Oghenesuvwe ✉ erhirhieochuko@yahoo.com Department of Pharmacology and Toxicology, Faculty of Pharmaceutical Sciences, Nnamdi Azikiwe University, Awka, Nigeria.

currently being explored as potentials for the drug discovery [15–17].

While antioxidant potentials of medicinal plants have been extensively reported in various reviewed articles, there exist none of such on insects. To this end, this review was designed to evaluate the antioxidant potential of insects and their products. Sections covered include ethno-medicinal values, nutritional constituents, and antioxidant properties of insects and their products. Literatures that were used in this synthesis were sourced from the articles published in PubMed, goggle scholars, and google electronic databases.

Insects: Brief Overview

Insects, belonging to the class, Insecta, phylum, Anthropoda, and the kingdom, Anamalia are the largest group of invertebrate in the animal kingdom. Their population ranges from 2.6 to 7.8 million [18]. Recent evaluation by Stork [19] revealed that the average global estimate of insect species remains 5.5 million. Insects are divided into 29 orders, with majority under Lepidoptera, Isoptera, Hymenoptera, Coleoptera, Orthoptera, and Hemiptera. Insects are abundant virtually everywhere, including soils, wasteland, forest, trees, water, and deserts [20]. Availability of some insects depends on the seasons of the year [20,21].

Insects have three segments, head, thorax, and abdomen. The head comprises of mouthparts and a pair of antennae. The thorax comprises of three pairs of legs, two pairs of wings, while the abdomen consists of the digestive system and reproductive organs. Insects have the characteristics of jointed chitinous exoskeleton [20].

Although some insects act as pests by causing damages to human environments, researchers have revealed that insects are of numerous relevance in human and veterinary medicines as well as in agricultural [11].

Insects' Consumption

Entomophagy is a term used to describe insects' consumption [22]. In various parts of the world, including Europe, USA, and Africa insects are consumed in various forms, roasted, cooked, and raw [6,10,23]. Prepared insects could also be purchased as snacks or as powdered supplements from the restaurants [24]. Insects, such as winged termites, wasps, beetles, grasshoppers, crickets, maggots, butterflies, beetles, caterpillars, locust, and palm tree larvae, among others, have been reported to provide good sources of proteins, fats, minerals, and vitamins to humanity [25–27]. Zhou and Han [28] opined in their study that insects' proteins are of good quality and can be easily digested. Akinnawo and Ketiku [29] also found that insects are a rich source of vitamins and minerals, especially iron and zinc. According to Kouřimská and Adámková [30], nutrients acquired from meat can also be obtained from the insects.

Generally, insects' protein ranges from 20% to 75%, greater than the protein content of meat [6]. Nutrient analyses of edible insects among South Western Nigerian revealed high amount of crude protein (27%–30%) [25]. Nutrient composition of about 78 Mexican edible insects was found to be 15%–81% protein, 4.2%–77.2% fats, and 77.7% dry-matter basis for carbohydrates [31].

In North-East India, pupae and larvae of *Vespa* sp. (Hymenoptera: Vespidae) is consumed [10]. This specie also has nutritional value of 50.13 g of protein, 13.29 g of carbohydrate, and 25.33 g of fats per 100 g species [10].

Nutritional contents of insects depend on species, season, age, reproductive stage, habitat, and diet. For instance, higher fat content is present in female insects than the male counterpart [32]. Edible insects have higher levels of polyunsaturated fatty acids than fish and poultry sources [33]. Long chain omega-3 fatty acids, such as alpha-linoleic and eicosapentaenoic acids, are predominant in edible insects [34].

According to Afam and Rinah [35], more than 1,900 species of insects serve as a food worldwide. Global statistics in 2014 revealed the proportion of insects, which include beetles (Coleoptera, 31%), caterpillars (Lepidoptera, 18%), bees, wasps, and ants (Hymenoptera, 14%), grasshoppers, locusts, and crickets (Orthoptera, 13%), cicadas, leafhoppers, planthoppers, scale insects, and true bugs (Hemiptera, 10%), other orders (5%), dragonflies (Odonata, 3%), termites (Isoptera, 3%), and flies (Diptera, 2%) [35,36].

Roles of Insects in Traditional Medicine

Entomotherapy, also described as the use of insects in the treatment of diseases is well recognized in various parts of the world, such as Asia, South America, India, Mexico, Korea, Spain, China, Argentina, Brazil, Nigeria, and Ecuador [37,38]. From this viewpoint, researchers have validated the health benefits of insects through clinical and non-clinical approaches. Such benefits could be reflected

by anti-inflammatory, anti-cancer, antimicrobial, anti-ulcer, anti-diabetic, hypolipidemic, and cardio-protective properties produced by insect bioactive compounds [37–39]. Several insect species, such as *Apis mellifera* (*A. mellifera*), *Pseudomyrmex triplarinus*, *Pieris rapae*, *Pseudocanthotermes spiniger*, *Brachystola magna*, and *Simulium vittatu*, among others have been found useful in the treatment of diseases, such as wounds and microbial infections, flatulence, spasm, bleeding, respiratory disorders, paralysis, cough, anemia, rheumatism, cancer, and diarrhea [10,18,39,40]. For instance, boiled termite paste is applied topically on the wounds. It is also used in the treatment of internal hemorrhages in the African traditional system of medicine [41].

Chemical Constituents of Insects

Some factors, such as stage of development, specie, habitat, and season may affect insects' chemical composition [42]. For example, *Tenebrio molitor* and *Zophobas atratus* at maturity were found to contain more protein than their larval counterparts [34]. It is worth mentioning that insects' diet could also affect their chemical constituents. Insects possess several bioactive compounds which propel their biological activities against various diseases [43,44].

Usually, edible insects' exoskeleton is made of chitin, whose carbohydrate contents ranging from 5% to 20% of the dry weight, which provide significant nutritional and health benefits [32,45,46]. Chitin was also isolated from two grasshopper species (*Calliptamus barbarus* and *Oedaleus decorus*) [47].

Insects contain amino acids, such as methionine, cysteine, lysine, and threonine [48]. Essential amino acid contents of edible insects ranged from 10% to 30%, while all the amino acid contents ranged from 35% to 50% [46]. Also, their protein digestibility, usually after the exoskeleton has been removed was found to be 77%–98% [32].

Mineral element analyses revealed that edible insects are rich sources of phosphorus, calcium, manganese, copper, zinc, sodium, potassium, and iron [49]. Edible insects are good sources of carotene and vitamins, such as B1, B2, B6, D, E, K, and C [48,50].

Bee and its products, Propolis, bee wax, royal jelly, and honey possess several phenolic compounds, such as protocatechuic acid, syringic acid, gallic acid, p-coumaric acid identified in Propolis and bee pollen, caffeic acid and ferulic acid identified in Propolis, bee pollen, and royal jelly, artepillin C, chlorogenic acid, and 3,5-dicaffeoylquinic acid identified in Propolis [3,39,51–54].

Also, three amyrins derivatives, α-Amyrin, β-Amyrin, and α-Amyrin acetate were identified in Propolis [46]. Studies have revealed that over 300 polyphenols, terpenoids, steroids, sugars, amino acid compounds are present in Propolis, which was found to be the most dominant antioxidant among all the analyzed bee products. Being the most widely studied in cell and animal research, Propolis possesses significant protective benefits against atherosclerosis, Alzhemier's, and Parkinson's disease [39,55].

Another bee product, royal jelly has been reported to contain mineral salts (copper, zinc, iron, calcium, manganese, potassium, and sodium salts in various proportions), polyphenols, and vitamins (biotin, folic acid, inositol, niacin, pantothenic acid, riboflavin, thiamine, and vitamin E) [39,56]. López-Gutiérrez et al. [57] categorized various flavonoids in royal jelly as flavanones (hesperetin, isosakuranetin, and naringenin), flavones (acacetin, apigenin and its glucoside, chrysin, and luteolin), flavonols (isorhamnetin and kaempferol glucosides), and isoflavonoids (coumestrol, formononetin, and genistein) [39].

Royal jelly was also found to contain oligosaccharides, such as trehalose, maltose, gentiobiose, isomaltose, raffinose, erlose, and melezitose. Varying level of the total protein and sugar content was reported in royal jelly. Its total sugar content varies between 7% and 21.2%, while its total protein content ranges from 8% and 9% [39,58,59]. According to studies by Oršolić [60] and Kolayli et al. [61], carboxylic acids in royal jelly derivatives were found to be 10-Hydroxydecanoic acid (10HDA), Decanoic acid (sebacic acid), 10-Hydroxy-2-decenoic acid (10H2DA), and 4-Hydroxyperoxy-2-decenoic acid ethyl ester (HPO-DAEE). Other of its components include free amino acids, including proline, cystine, and cysteine [62,63].

A study by Chikara et al. [64] revealed the isolation of two flavonol glycosides, quercetin 3-*O*-β-d-galactopyranosyl-(1 → 3)-β-d-galactopyranoside, and kaempferol 3-*O*-β-d-galactopyranosyl-(1 → 3)-β-d-galactopyranoside alongside four known flavonoids from the cocoon of a mulberry white caterpillar, *Rondotia menciana* (Lepidoptera: Bombycidae: Bombycinae).

Gallic acid, vanillic acid, 3, 4-dihydroxybenzoic acid, cinnamic acid, isorhamnetin, naringenin, quercetin, trans-trans, and cis-trans abscisic

acid were found in the honey of the Brazilian bee, *Melipona subnitida* [65]. Activity-guided fractionation of the chloroform extract of *Bruchidius dorsalis* (Bruchidae) larvae produced five new antioxidant lipids, dorsamin-A763, A737, A765, A739, and A767 [66]. Polyphenols (flavonoids and phenolic acids, vitamins), maillard reaction products (melanoidins), carotenoids, and amino acids were extracted from honey bee [67,68]. Insects are rich sources of low polyunsaturated fatty acids, such as linoleic and linolenic acid, which render them to be more stable against free radicals [25].

Secretion from *A. mellifera* contains an emulsion of proteins, sugars and lipids that are enriched with mineral salts, flavonoids, polyphenols, and vitamins [39,56]. Insects' chemical constituents with proven antioxidant properties are summarized in Table 1.

Insects' Endogenous Antioxidant System

Living organisms have natural antioxidant enzymes and substances capable of counteracting ROS [69]. As it applies to other living organism, generation of ROS is not uncommon to insects. Insects with high metabolic rate are meant to produce high level of ROS [70]. Interestingly, insects are endowed with

Table 1. Anti-oxidant properties of insects' products and their isolated compounds.

Specie (family)	Bioactive compound	Method	Reference
Bruchidius dorsalis (Bruchidae)	Dorsamin-A763, A737, A765, A739 and A767	ABTS	67
Chrysomya megacephala (Calliphoridae)	Chitosan	DPPH radical and superoxide anion	86
Calliptamus barbarous (Acrididae) and *Oedaleus decorus* (Acrididae)	Chitosan	DPPH and FRAP assays	47
Holotrichia parallela (Scarabaeidae)	Catechin	DPPH, hydroxyl, and superoxide radical and FRAP.	88
Dactylopius coccus (Dactylopiidae)	Camic acid	DPPH, ABTS, LOX	89
*Apis mellifera (*Apidae)	Fatty acid derivative (4-hydroperoxy-2-decenoic acid ethyl ester (HPO-DAEE)	6-hydroxydopamine- (6OHDA-) induced cell death	64
Propolis: *Tetragonisca fiebrigi, Melipona orbignyi* (Apidae)	Phenolic compounds, aromatic acids, alcohols, terpenes, and sugars Phlobaphene tannins, catechins, chalcones, aurones, flavonones, flavonols, xanthones, pentacyclic triterpenoids and guttiferones	DPPH, ABTS, FRAP, and Oxygen Radical Absorbance Capacity (ORAC) models DPPH	39, 93, 94
Melipona subnitida (Apidae)	Gallic acid, vanillic acid, 3, 4-dihydroxybenzoic acid, cinnamic acid, isorhamnetin, naringenin, quercetin, trans-trans and cis-trans abscisic acid	DPPH, ABTS	66
*Apis mellifera (*Apidae)	10-hydroxy-2-decenoic acid and free amino acids including proline, cystine and cysteine	Cisplatin-induced spermiotoxicity and nephrotoxicity in rats	63
	AMP N1-oxide	Secondary neuronal damage	104
Coccon from butterflies (Papilionoidea)	Flavonoids, quercetin 7-O-β-Dglucoside, kaempferol 7-O-β-D-glucopyranoside, coumaric acid glucoside, 2-hydroxy-nonadecanoic acid and 9,12-dihydroxy stearic acid	Oxidative stress, cardiac enzyme activity and interleukin-6 in murine model	47
Calliptamus barbarous (Acrididae) and *Oedaleus decorus* (Acrididae)	Chitison	DPPH and FRAP	47
Amphiacusta annulipes (Phalangopsidae), *Zophobas morio* (Tenebrionidae), *Agnetina annulipes* (Perlidae) and *Locusta migratoria (*Acrididae)	Peptides	DPPH, $ABTS^{+}$, Fe^{2+}, Cu^{2+} chelation and reducing power activities	6

antioxidant defense systems which contain anti-oxidant enzymes, such as superoxide dismutase, catalase, glutathione transferase, and glutathione reductase that is able to counteract or buffer the disproportions caused by excessive free radical generation [1,71].

For instance, midguts of various grasshopper species produce various antioxidant enzymes, such as superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase, and glutathione transferase peroxidase [72,73].

Male and female red mason bees (*Osmia bicornis* L.), from larval to imago stage were reported to produce elevated levels of antioxidant enzymes, superoxide dismutase, catalase, peroxidase, and glutathione-S transferase in feeding stages-in larvae and inactive imago of both genders [70].

In a study conducted by Weirich et al., total antioxidant capacity (TAC) in honey bee's haemolymph was analyzed in relation to age and exposure to pesticide, imidacloprid (IMD). Their study revealed lowered TAC in 1-day old, except 30-day-old honeybees, indicating that less susceptibility to IMD toxicity is common with older bees which possess higher antioxidants [69]. SOD, CAT, glutathione-S-transferase (GST), and peroxidase were identified as the most important antioxidant enzymes in honey bee's body fluids [74].

In a related study, Sanz et al. [71] investigated the antioxidant enzymes, SOD, CAT, glutathione reductase (GR), glutathione peroxidase, GST, and DT-diaphorase, together with lipid peroxidation in the nymphs of four species of Plecoptera in the superfamily of Perloidea: *Perla marginata* (Perlidae), *Guadalgenus franzi*, *Isoperla curtata*, and *Isoperla grammatica* (family Perlodidae). The study revealed that each insect possesses variable essential enzymatic antioxidant potential with respect to the development period.

Aucoin and Arnason [75] also reported that the presence of antioxidant enzymes, SOD, CAT, glutathione peroxidase, and GR in larvae of three Lepidoptera (*Ostrinia nubilalis*, *Manduca sexta*, and *Anaitis plagiata*) provided defense against toxic oxygen species generated in plant phototoxins.

Although insect endocrine system synthesizes and secretes three major groups of insect hormones and biologically active factors, ecdysteroids, juvenile hormones, and neurohormones [76], adipokinetic hormones (AKH), one of the best investigated group of insects hormones are known to trigger defense reactions responsible for counteracting OS [77]. A review by Dalibor et al. [77] on the role of AKH as principal stress response hormones in insects involved in activation of anti-oxidative stress response pathways revealed that the oxidative stress experimentally induced by various stressors led to significant elevation in AKH levels in insects' body, as an indication of anti-stress reaction. The authors opined that the mechanism may involve both protein kinase C and cyclic adenosine 3′,5′-monophosphate pathways when extra and intracellular Ca^{2+} stores is present. The Forkhead box class O transcription factor (FoxO) could also be involved.

Reports have shown that uric acid exhibited strong free radical-scavenging activity in humans and several insects [78,79]. Tasaki et al. [80] showed that uric acid is the major antioxidant in termites, as antioxidant activities in extracts diminished with reduction in uric acid concentrations. They also found that externally administered uric acid facilitated termite survival under highly oxidative conditions. Study by Hilliker et al. [81] on the fruit fly, *Drosophila melanogaster* revealed that uric acid-deficient mutant *rosy* was vulnerable to high temperatures and oxidative stress. Thus, the antioxidant status of uric acid is known to contribute significantly to the insects' survival and longevity [80].

Reported Antioxidant Properties of Insects

Table 1 depicts anti-oxidant properties of insects' products and isolated compounds, while Table 2 depicts antioxidant properties of insects' products, whose bioactive components were not elucidated in the assays carried out.

In-Vitro Studies

Tasaki et al. [80] reported that Termites, *Reticulitermes speratus*, *Tenodera aridifolia*, *Tenodera aridifolia*, and *Camponotus obscuripes* extracts produced antioxidant activities against 2,2-dyphenyl-1-picrylhydrazyl (DPPH) at 50 µl samples containing 7.5 µg of protein. The high antioxidant activity was found in the bodies of all termite castes except workers.

From the family of Scarabaeidae, rhinoceros beetle larvae, *Allomyrina dichotoma* methanol, water, chloroform, ethyl acetate, and hexane extracts were assessed using DPPH radical scavenging, superoxide anion radical scavenging, and singlet oxygen ($^1O_2$) quenching assays and the results revealed methanol extract (with the concentration of 50% $^1O_2$ quenching, EC_{50} = 0.080 mg/ml) as the most

Table 2. Anti-oxidant properties of insects' products.

Specie (family)	Method	Reference
Reticulitermes speratus (Rhinotermitidae), mantises (Mantidae), *Tenodera aridifolia* (Mantidae) and *Camponotus obscuripes* (Formicidae)	DPPH	81
Rhinoceros beetle larvae, *Allomyrina dichotoma* (Scarabaeidae)	DPPH, superoxide anion and singlet oxygen quenching assays	83
Protaetia brevitarsis (Scarabaeidae)	Singlet oxygen quenching ability, DPPH and ABTS	84
Serrognathus platymelus (Lucanidae)	DPPH and ABTS	85
Vespa affinis (Vespidae)	DPPH, hydroxyl, and superoxide radicals. Antioxidant enzyme (GST and CAT)	10
Royal jelly *Apis mellifera (*Apidae)	DPPH, hydroxyl, superoxide radical reducing power, ABTS, FRAP, and ORAC models	91, 39
Brazilian stingless bee: *Melipona orbignyi* (Apida)	Lipid peroxidation in human erythrocytes	38. 93
Malaysian honey: Acacia (Apis Mellifera*)*, pineapple (Apin mellifera), borneo (*Apis. cerana*) and tualang (*Apis. dorsata*): *Apidae*)	FRAP and DPPH assays	96
Malaysian species of bees, *Apis cerana*, *Apis andreniformis*, *Apis koschevnikovi* and *Apis Nuluensis* (*Apidae)*	DPPH, FRAP, and ABTS	97

active scavenger against DPPH. Interestingly, it was 1.7 times more efficient than the positive control, ascorbic acid [82]. In a related study, Suh and Kang [83] demonstrated the antioxidant activity of *Protaetia brevitarsis* (Scarabaeidae) at various growth stages, larvae, pupae, and imago. Their study revealed that similar singlet oxygen in the quenching ability, DPPH and 2,2′-Azino-bis(3-ethylbenzthiazoline-6-sulfonic acid (ABTS) radical scavenging activities were demonstrated by the larvae and imago extracts with effective concentration, EC_{50} of 0.174 and 0.149 mg ml^{-1}, respectively. Five new antioxidant lipids, dorsamin-A763, A737, A765, A739, and A767, isolated from the chloroform extract of *Bruchidius dorsalis* (Bruchidae) larvae through activity-directed fractionation were found to exhibit ABTS radical scavenging activity better than the control, Trolox [66].

Study by Suh et al. [84] on the antioxidant properties of lucanid beetle, *Serrognathus platymelus* castanicolor (Lucanidae), at three life stages, larvae, pupae, and adult revealed that the pupal methanol extract (PME) produced the maximum DPPH and ABTS radical scavenging activity similar to that of control, ascorbic acid. The $^1O_2$ quenching ability of the PME (EC_{50} = 0.184 mg/ml^{-1}) was comparable to that of ascorbic acid (EC_{50} = 0.167 mg/ml^{-1}).

Chitosan, a partially deacetylated polymer produced from the alkaline de-acetylation of chitin, from the larvae of a housefly and blowfly, *Chrysomya megacephala* (Calliphoridae) were found to produce significant comparable antioxidant activities to that of ascorbic acid in DPPH radical and superoxide anion scavenging assays. Furthermore, the blowfly chitosan exhibited excellent antioxidant activity with IC_{50} value of 1.2 mg/ml. The blowfly larvae were suggested to be a novel alternative source of chitosan and might be used as a natural antioxidant [85].

In addition to antifungal and antiviral activities, chitosan from the larvae of house fly (*Musca domestica* L.) showed scavenging activity against hydroxyl and superoxide radicals. Its antioxidant activity was similar to that of ascorbic acid [86].

In-vitro radical scavenging assay of water and ethanol extracts from a large chafer beetle, *Holotrichia parallela* (Scarabaeidae), which is used traditionally in China and East Asia in gout, tetanus, erysipelas and superficial infection treatments was carried out by Liu et al. [87]. In various assays, linoleic acid peroxidation inhibition, metal-chelating activity, and DPPH radical scavenging assays, the ethanol extract demonstrated potent metal-chelating activity as well as inhibition of lipid peroxidation due to its constituent, catechin. The water extract exhibited significant metal chelating as well as DPPH radicals scavenging activities.

Zielinska et al. [6] evaluated the antioxidant activity of peptides obtained by *in vitro* gastrointestinal digestion of edible insects, *Amphiacusta annulipes*, *Zophobas morio*, *Agnetina annulipes*, and *Locusta migratoria* using DPPH, $ABTS^+$, iron-II (Fe^{2+}), cupper–II (Cu^{2+}) chelation, and reducing power assays. Insects that demonstrated the highest antioxidant

activities in the five models include *Amphiacusta annulipes* (DPPH, 19.1 μg/ml), *Zophobas morio* ($ABTS^+$, 4.6 μg/ml), *Agnetica annulipes* (Fe^{2}+ chelation ability, 58.82% and reducing power activity, 0.652 μg/ml), and *Locusta migratoria* (Cu^{2+} chelation ability 86.05%).

Camic acid, the major compound isolated from the cochineal insect, *Dactylopius coccus* (Dactylopiidae), was found to elicit potent free radical scavenging activity against DPPH, ABTS, and β-carotene bleaching enzymatically induced by lipoxygenase (LOX). The result was comparable to that of ascorbic acid, but it elicited more activity than Trolox [88].

In a another study, Kaya et al. [47] isolated and characterized chitosan from two species of grasshoppers, *Calliptamus barbarous* (Acrididae) and *Oedaleus decorus* (Acrididae) which were found to have antioxidant activities using DPPH radical scavenging and ferric reducing anti-oxidant power (FRAP) assays, besides its potent antimicrobial activity against several strains of fish, clinical and food-borne pathogens. The IC_{50} values for the chitins obtained from *C. barbarous* and *O. decorus* were 10.68 ± 0.27 mg/ml and 10.91 ± 0.96 mg/ml, respectively, which were greater than the value for butylated hydroxytoluene, 0.04 ± 0.01 mg/ml.

Studies have shown that the oxidative stability of sunflower oil was enhanced following the addition of melon bug oil, which resulted in an increase of oleic and a decrease of linoleic acid [89].

A study was conducted by Dutta et al. [10] on the antioxidant potential of the aqueous extract of *Vespa affinis L* (AEVA), a popular edible insect among several tribes in North East India using DPPH, hydroxyl, and superoxide radical methods. Its effect on the activities of antioxidant enzyme (GST and CAT) was determined using both recombinant proteins and human plasma. Their study revealed that AEVA exhibited significant scavenging activities in various models. It also significantly increased the activities of recombinant enzymes (rGST and rCAT) following incubation at 2.5, 5, 7.5, and 10 μg/μl. Its supplementation at 5, 7.5, and 10 μg/μl also enhanced the activities of GST and CAT when incubated with human plasma. The authors concluded that the antioxidant activities of AEVA could mediate its therapeutic activities in oxidative stress-associated health disorders.

Liu et al. [90] investigated the antioxidant properties of the royal jelly (RJ) from *Apis mellifera* using DPPH, hydroxyl and superoxide radical, as well as reducing power models. Based on the larval age, 1-, 2-, or 3-day old and time of harvest after the larval transfer from the queen cell cups to the bee hives (24, 48, and 72 hours), there was DPPH radical-scavenging effect, inhibitory effect on the superoxide radical and hydroxyl radical formation in the youngest larvae (1-day old) transferred into bee hives for the shortest time (24 hours). However, significantly higher SOD level was recorded in the RJ collected at 72 hours after transferring of 3-day old larvae [90].

A study by Inoue et al. [63] on the antioxidant effect of RJ fatty acid derivative, HPO-DAEE on oxidative stress-induced cell death using human neuroblastoma, SH-SY5Y cells revealed a protection against 6-hydroxydopamine- (6OHDA-) induced cell death by increasing the expression of antioxidant enzyme—heme oxygenase-1 (HO-1) mRNA—through Nrf2-ARE signaling following pretreatment.

Eshtiyaghi et al. [91] researched on the protective effect of RJ in the redox state of ovine oocytes matured *in vitro* and embryonic development following *in vitro* fertilization revealed improvement of oocyte maturation supplemented with RJ at 2.5, 5, and 10 mg/ml. An increase in the intracellular glutathione (GSH) content compared to the control group was observed. Increase in mRNA and GPx in both oocyte and cumulus cells as well as SOD expressions in the cumulus cells were observed.

Several literatures have investigated the antioxidant properties of Propolis bee using DPPH, ABTS, FRAP, and ORAC models [39]. Similarly, Propolis from the Brazilian stingless bee, *Tetragonisca fiebrigi* likewise produced antioxidant activity besides its antimicrobial and anti-inflammatory activities. Based on gas-chromatography mass spectrometry (GC–MS) analyses, phenolic compounds, aromatic acids, alcohols, terpenes, and sugars were identified as its major components [92].

The ethanol extract of Brazilian red Propolis (*Melipona* orbignyi) produced significant antioxidant activity better than the positive control, Trolox in DPPH assay. The extract, hexane, chloroform, and ethyl acetate fractions demonstrated activities with IC_{50} values of 8.01, 5.15, 5.20, and 6.01 μg/ml, respectively, when compared to that of Trolox, 14.68 μg/ml. It was found to contain phlobaphene tannins, catechins, chalcones, aurones, flavonones, flavonols, xanthones, pentacyclic triterpenoids, and guttiferones [93].

Aside the antimicrobial and cytotoxic activities, 80% ethanol extract of Propolis produced by Brazilian stingless bee, *Melipona orbignyi* displayed antioxidant activity due to its ability to scavenge

free radicals, inhibit hemolysis, and lipid peroxidation in human erythrocytes incubated with an oxidizing agent [38,92].

Antioxidant activities of honey produced from Brazilian bee, *Melipona subnitida* in DPPH and ABTS models were evaluated. For the DPPH assay, the EC_{50} values ranged from 10.6 to 12.9 mg/ml for pure honey, 108.5–208.6 mg/ml for methanol extract, and 43.5–87.8 mg/ml for ethanol fraction. For the ABTS assay, EC_{50} results varied from 6.1 to 9.7 mg/ml, 21.2 to 53.1 mg/ml, and 13.2 to 33.9 mg/ml for pure honey, methanol extract, and ethanol fraction, respectively. It's chemical constituents include gallic acid, vanillic acid, 3, 4-dihydroxybenzoic acid, cinnamic acid, isorhamnetin, naringenin, quercetin, trans-trans, and cis-trans abscisic acid [65].

In another study, Dor and Mahomoodally [94] evaluated the antioxidant potential in addition to the antimicrobial activities of six honeys obtained from the Republic of Mauritius. For the antioxidant capacity, the following eight assays were conducted; FRAP, iron (II) chelating activity, trolox equivalent antioxidant capacity, hydroxyl (OH), DPPH, hypochlorous acid (HOCl), nitric oxide (NO), and ABTS diammonium salt radical scavenging assays. Their results revealed that all the sample honeys exhibited the same level of antioxidant activity.

Moniruzzaman et al. [95] also evaluated the antioxidant properties of four Malaysian honey viz; acacia (*Apis. mellifera*), pineapple (*Apin mellifera*), borneo (*Apis. cerana*), and tualang (*Apis. dorsata*) using FRAP and DPPH assays. The study revealed that tualang exhibited the best antioxidant activity as well as highest phenolic, flavonoid, and protein contents.

In addition to the acetylcholinesterase inhibitory potentials of four wild honey produced from four Malaysian species of bees, *Apis cerana*, *Apis andreniformis*, *Apis koschevnikovi*, and *Apis nuluensis*. They produce scavenging activities in DPPH, FRAP, as well as in ABTS decolorization assays. Among the samples investigated, 80% methanol extract of the wild *Apis cerana* honey produced the best activity in various assays, DPPH (84%), FRAP (37 mmol/l Fe^{2+}/1 g dry sample), and ABTS decolorization (8 mg AEAC/1 g dry sample) [96].

In Vivo Studies

RJ administered daily for 10 days at doses of 50 and 100 mg/kg was found to reverse oxidative stress in Cisplatin-induced spermiotoxicity and nephrotoxicity in rats due to its component of 10-hydroxy-2-decenoic acid and free amino acids including proline, cystine, and cysteine [62]. The antioxidant effect of RJ at 100 mg/kg and its component on oxymetholone, 5 mg/kg induced liver injury in mice was reported by Nejati et al. [97]. Antioxidant effects of RJ at the following doses have also been reported in animal models of oxidative stress generated by Carbon tetrachloride [98], Azathioprine (50 mg/kg), 200 mg/kg, [99], Bleomycin (10 mg/kg twice weekly, I.P), 100 mg/kg for 48 days [100], Methotrexate (20 mg/kg, I.P), 50, 100 mg/kg, p.o [101], among others [39].

RJ was also found to reduce secondary neuronal damage after experimental spinal cord injury in rabbits by preventing lipid peroxidation and enhancing endogenous enzymic or non-enzymic antioxidative defense systems levels. Apoptotic cell numbers induced by spinal cord injury was ameliorated following the treatment with 100 mg/kg RJ when compared with the control group [102]. Also, the unique compound, adenosine monophosphate (AMP) N1-oxide was suggested to be responsible for the antioxidant activity of RJ following stress induced study in rats [103]. Aqueous extract of Propolis, 100 and 200 mg/kg displayed antioxidant activities in cerebral cortex induced oxidative damage in mice [104].

Clinical Studies

Randomized controlled trials revealed the antioxidant effect of Propolis [105]. Twice daily, 15 drops each time oral administration of commercially available Propolis (Beepolis®) for 90 days on the oxidative status and lipid profile in a human population in Chile were measured. Propolis supplementation resulted in a 67% decrease in the amount of thiobarbituric acid reactive substances as well as 175% elevation in reduced GSH level when compared to the baseline values. A significant increase in the High density lipoprotein (HDL) concentration compared to the baseline value was observed.

According to Jasprica et al. [106], 30-days supplementation with commercially available powdered Propolis extract resulted in a 23.2% reduction in malondialdehyde (MDA) level and 20.9% increase in SOD activity in healthy male, except female volunteers.

In type 2 diabetes mellitus patients, Brazilian green Propolis supplementation at 900 mg/day for 18 weeks caused an increase in serum levels of GSH

and total polyphenols and a reduction in serum carbonyls and lactate dehydrogenase activity [107].

Supplementation of the diet of diabetic patients with RJ at a dose of 1,000 mg daily over a period of 8 weeks resulted in a significant increase in erythrocyte SOD and GPx activities as well as decrease in MDA concentration when compared to control group [108]. In a related study, treatment of diabetic patients with RJ supplemented diet at 1,000 mg thrice daily for a period of 8 weeks caused significant reduction in homeostasis model assessment for insulin resistance and increased total antioxidant capacity when compared to the control group [109].

Discussion

The present review emphasized the relevance of insect products as therapeutic antioxidants in the treatment and management of oxidative stress mediated diseases.

In addition to endogenous antioxidant enzymes, insects also possess non-enzymatic constituents, such as glutathione, ascorbic acid, vitamin E, uric acid, and thioredoxin [110,111].

In recent years, several bioactive compounds discovered from insects have elicited significant bioactivities including anticancer [112–114], anti-hyperlipidemic [115], anti-ulcer [116], cardioprotective [117], anti-diabetic [118], antimicrobial [119,120], anti-inflammatory [121–123].

Following the established facts that the oxidative damage is implicated in the pathogenesis of various diseases [3,6], it is certain that the above highlighted therapeutic benefits of insect products could be attributed to their antioxidant activities.

It is obvious that some *in vitro* antioxidant assays may not be sufficient to translate therapeutic effects [124,125]. Thus, in order to confirm insects therapeutic efficacy, it is crucial to employ *in vivo* studies, such as catalase, superoxide dismutase, glutathione reductase, serum total antioxidant status, erythrocyte MDA, protein carbonyl, and selected serum biochemical using normal subjects or physical and chemical induced oxidative damage in animal models [124,125]. These tests would clearly validate insect derived compounds that elicited significant *in vitro* antioxidant activities.

Among the various insects evaluated, products of the bees *Apis mellifera,* royal jelly, and Propolis have received sufficient scientific exploration using *in vitro, in vivo*, and clinical approaches [39,55]. However, *in vivo* and clinical studies have left much to be explored in other insects, such as Termites, lucanid beetle, blowfly, housefly, and grasshoppers among other. This gaps call for more researches to be done.

In the aspect of safety, edible insects and their products have been found to be safe [126,127]. Although some studies have revealed that poor handling of insects and their products during harvesting, processing and packaging could result in their contamination, mainly from bacteria, fungi, pesticide residue, among other [128]. Thus, it is important to follow acceptable dietary standards in order to reduce exposure to contaminants [129].

Conclusion and Recommendations

This review clearly discovered that insects' derived compounds could play a significant role as potential antioxidants in food and drug industries. Based on the numerous health benefits of edible insects, the populace should be encouraged to invest in insects farming for the production of dietary, commercial, industrial, and pharmaceutical products. Researchers should also give proper attention to insects' products for natural product drug discovery. *In vivo* antioxidant assays should be carried out on insect bioactive compounds which have only received *in vitro* screening.

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